VETERINARY MEDICINE ENHANCING ANIMAL HEALTH AND WELL BEING

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Chapter 1: Animal Management and Care

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Abstract

This chapter describes the humane and sophisticated measures and practices for care and management of animals and form infrastructure of these animal lives in our midst. The five principals should be under consideration in devising any animal care and management program. Animal behavior and physiology are important parameters in determining whether the management and care provided to them are adequate or not.

In case of feed and nutrition (Morais et al., 2020), formulating meals that provide proper nourishment with all essential vitamins, minerals (Alagawany et al., 2021), probiotics and prebiotics to animals according to their requirements. The feed should be fresh, free from contamination and carcinogens (Jin & Zhang, 2020), and stored at optimum and standard conditions. Similarly, the drinking water provided to animals should be free from contamination, available round the clock with optimum temperature conditions.

As far as the housing is concerned, we should build homes not just for enclosures where animals find solace and peace, it should be a place where the breeze carries whispers of safety cccccccc(Beaver et al., 2020). We should regularly perform checkups of animals, give proper treatment if disease outbreak, vaccinate them at proper time (Garcia-Sanchez et al., 2024), manage them with soft hands.

In essence, this chapter is more than just a manual, it's a call to arms, urging us to be not just caretakers, but custodians of their world, a world where compassion reigns supreme and every heartbeat echoes the rhythm of belonging.

Keywords: Animal welfare; Management; Care; Nutrition; Behavior; Housing.

1.1 Introduction:

Animal management and care include critical aspects of providing the well-being and welfare to animals across diverse backgrounds, including zoo, agriculture, research, companion animal, and wildlife conservation (Buller et al., 2020). Zookeepers, farmers, researchers, pet owners, animal trainers, veterinarians and other people who work and live with animals in their daily routine should have proper knowledge to address the nurturing and manage-mental challenges and work within their professions. From farm to field, lab to home we should keep in our consideration and provide the basic freedoms to animals (Browning & Veit, 2021).

Animal care and management is all about looking out for our fluffy, feathered, and scabby friends, ensuring they live their best lives under our care and consideration. It looks like being a parent, but instead of kids, we're responsible for the well-being of a diverse range of creatures, from lovely pets to royal and marvelous zoo animals (Dwyer et al., 2021; Forrest et al., 2023). It represents our commitment to ensure the well-being of the (animals under our consideration, regardless of whether they are domesticated pets, farm animals, or inhabitants of zoos and wildlife locality. Normal and natural behavior of animals indicate the level of animal welfare practiced there (Miller et al., 2020).

It's a responsibility to improve boundaries and cover a range of duties (Balzani & Hanlon, 2020), from providing food and shelter to nurturing their physical and mental health. Essentially, it's about how they live a happy and healthy life in the environments we've provided for them. Before devising any program regarding animals, a prescriber, provider, designer must define what includes animal well-being (Applebee et al., 2024). When animal management and care is up to the mark, an animal seems to be comfortable and healthy, the level of management and care provided to animals is assumed sufficient.

1.2 Five Freedoms:

In animal care and management there came following five freedoms (Grandin, 2015); which an owner, trainer, farmer, veterinarian, zookeeper should provide to animals (Tobin et al., 2022) from welfare point of view:

1. **Freedom from Hunger and Thirst:** The first and basic freedom for animals is freedom from hunger and thirst. It is also important from animal welfare point of view and against ethics (Webster & Margerison, 2022). One should provide enough feed to animals that left animal with no hunger and water is the basic need of all living organisms, without water the existence of life is nearly impossible, we should provide clean, fresh water to animals round the clock without any termination.

2. **Freedom from Discomfort:** Animals should be kept in places that provide comfort and peace to animals, animals should feel safe and secure in the premises of our supervision (Vigors et al., 2021). For instance, in case of pet owners should be guided how to provide safe and comfortable environment to animals, pets should be kept in optimum environment conditions, where noise is minimum, in farm animals scenario, noise pollution, unethical handling should be avoided to prevent discomfort, similarly in zoos animals should be provided with enough space to show their natural behavior and there should be a shelter or hiding space where animal can hide and protect itself from humans.

3. **Freedom from Pain, Injury, or Disease:** Freedom from pain (Steagall et al., 2021), injury and disease is also an important freedom from welfare point of view. Animals

should be checked at regular intervals in weeks or months for timely diagnosis and treatment of infectious and lethal diseases. If animals are not regularly checked for wounds or injury, animals will continuously suffer with pain, this leads to unethical approach and welfare issues related to discomfort.

4. **Freedom to Express Normal Behavior:** Animals should be provided with enough space, so they show their natural behavior (Dawkins, 2023). Animal behavior is a good indicator for assessing the management and care facilities provided to animals. If animals show abnormal behavior, it indicates that the facilities and management policies devised for animals are not up to the mark.

5. **Freedom from Fear and Distress:** Animals feel scared and stressed, especially if they're in loud or unfamiliar places. So, it's important to make sure they feel safe and comfortable in their environment. Protection from predators or anything that frightens animals should be our responsibility. Moreover, wildlife management should also be taken into consideration (Kadykalo et al., 2021).

These freedoms are all about making sure animals are treated with kindness and respect. They live happily under our supervision as they live in natural environments.

1.3 Management and Care:

Animal care and management (Krausman & Cain, 2022) are organized well in hierarchy as human needs (Montag et al., 2020). According to his theory animal needs are listed as: (1) Physiologic needs (2) Safety needs and (3) Nutritional needs (4) Behavioral needs. Of above mentioned needs the physiological needs are well understood and considered the most basic or lowest in this list.

1.4 Physiological Care:

The basic animal need (Otteman et al., 2022) is its normal functioning, as physiologic needs of animals are known well, we should meet with these needs reasonably well. If we are lacking in meeting these basic needs animals suffer from stress and this can affect their production directly and indirectly in sense, transferring nutrients from productive phase to maintenance, changing route of resistance against infection. Animal physiology is closely related to the environment provided to it (Mota-Rojas et al., 2021). Recent studies are performed to check the influence of environment on animal health and physiology. From now onwards we should make heroic efforts to provide animals with optimum environmental conditions like temperature, humidity, light duration and intensity (Deep et al., 2012) and social environment (Becker et al., 2020) (Lacetera, 2012).

1.5 Safety needs:

It's a bitter reality that to date physical handling of animals is both unethical and baseless. Agricultural animals are treated badly and poorly (Mota-Rojas et al., 2022); their safety needs are not attended properly like they should be attended as like physiologic needs. We should take into consideration that animals are living organisms, and we should treat them with soft temperament. As mentioned above, it's an animal's right to provide them with an environment in which they feel peace and comfort and have freedom from fear and distress.

Animals are not adapted to a wide range of climate variations so (Skuce et al., 2013); we should take adequate measures to reduce these variations to a tolerable range to avoid health issues. For instance, if the environment control shed system collapses it can lead to ventilation failure and increase in temperature to drastic level (Das et al., 2016). However, animals provided with primary and secondary environmental adjustments were found more protected from environmental extremes than those who are provided with natural environment. Similarly, we should design equipment which is environmentally friendly and animal friendly (Kul & Sağlık, 2023). There is huge need in modifying the equipment and facilities for animals and it can be done in coordination with companies and animal welfare organizations (Bendel, 2018).

1.6 Nutritional Management:

All the animal needs and requirements known so far, none can be clearer and well understood than their nutritional requirements (Bratosin et al., 2021). A wide range of feed formulations that are according to respective animal according to its requirement and need is the proof of this statement. Moreover, nutritional requirements (Gasco et al., 2020; Mordenti et al., 2021) are also modified according to animal's workload and environmental influence. The basic guidelines related to nutrition is that feed is according to animal's need, it should be prepared according to international standards, it should be a balanced diet for animals, it is free from contamination, free from fungi, bacterial etc., free from carcinogens, should be palatable, should have normal odor, color, taste, appearance, texture etc. (Owen et al., 2012; Yang et al., 2020; De Blas & Mateos, 2020).

1.7 Behavioral Care:

We can assess the level and success of management and care provided to animals by observing their behavior (Debauche et al., 2021), if animal behavior is normal then the management and care provided to animals is considered and declared adequate (Panzera, 2013) and if animal don't show normal behavior, it indicates that the facilities provided are not adequate, we should provide such environment in which animals feel comfortable and show their natural and normal behavior. Human mismanagement and maltreatment include misconduct, neglect and distress (Mota-Rojas et al., 2022).

Misconduct or abuse is considered active barbarity like omitting animal safety needs and treating it badly and beating with stick. Neglect is considered passive barbarity and was practiced when animal's basic needs like nutrition, water, health, shelter etc. is compromised (Nurse, 2017). Misconduct and neglect occur seldom from business as well as humane point of view and must not be tolerated when practiced (Guazzaloca, 2020).

Distress or deprivation occurs when animal's behavioral needs are compromised. It comprises rejection or exclusion of certain environmental parameters that are thought to be not significant than physiologic or safety needs of animals from animal's care and management point of view.

In simple words it is a form of misconduct or maltreatment difficult to recognize (Browning, 2020).

1.8 Recent Advances:

From 2000 to 2016, nearly 360 animal diseases outbreaks were recorded in 116 countries, leading to adverse economic losses especially in third world countries. Of these outbreaks two-thirds were caused by just five diseases, including Avian Influenza, Foot and Mouth Disease.

Scientific progress and emerging techniques like cell therapy and new generation of vaccines have provided greater opportunities to control severe outbreaks that lead to economic losses and diagnose them at initial stages. The modern technology like AI help to improve animal welfare, help in animal management and care and provide healthier and long lives to pets in houses.

Development of new vaccines that cover various diseases like in canine vaccine "DHPPIR", this vaccine covers Canine Distemper, Hepatitis, Rabies, Peritonitis etc. at the same time. Similarly, Biofel pch is feline vaccine which covers Feline Panleukopenia, Calicivirus and Feline Herpesvirus. New advances in vaccination lead to mRNA vaccines (Le et al., 2022), heat resistance vaccines etc.

Biotechnology also plays role in improving nutritional value (Bonciu, 2020) of animal feed and by improving the digestibility of low-quality feed like roughages. The role of biotechnology in animal feed and nutrition is achieved by enhancing gut ecosystem and improving the activity of gut microbes to perform specific functions and introducing new strains into gut. Also, metabolic modifiers (Adetunji et al., 2022), probiotics, ALFRA box dosing system, formulating novel feeds, SRC nutritional are recent advances in animal feeds.

1.9 Role of Veterinarian:

Veterinarians provide a vital role in the care and management of animals used in research programs, maintenance of population like in zoos, agricultural animals, pets etc. (Hernandez et al., 2022). Animal management and care is the primary responsibility of veterinarian but, it's a team event which helps enhance the life of animals and covers all aspects of their care and management. This team comprises of Institutional officials, Animal care committee, Scientist, Veterinarian, Animal facility manager, Office manager, administrative staff. Each member in the team offers his/her duty and plays his/her role in managing and providing optimum environment and habitat to animals in zoos, farms, homes, research labs etc. Some developed countries made standards for veterinary care, these countries include USA, Canada, UK, European Union etc. (Couto & Cates, 2019). The goal of these standards is not only to ensure timely and quality veterinary care services but also to meet the desires of research, reproduction (Itze-Mayrhofer & Brem, 2020).

A veterinarian has eventual responsibility of maintaining and providing day to day facilities to animals and veterinarian himself is responsible for animal health and well-being. These responsibilities are better defined in "Guide for Care and Use of Laboratory Animals.". The term "Adequate Veterinary Care" varies from country to country and administration to administration. Adequate veterinary care defines veterinary access to animals and their records to formulate plans and set the objective. For instance, in farm a veterinarian analyzes the milk record if its decreasing there must be some issue in farm whether there might be a disease outbreak, feed contamination or imbalanced feed or human error, so, a veterinarian go through the whole procedure and set a goal accordingly, if there's disease due to which milk yield is decreased he/she diagnose and treat it, if there's feed issue, he/she check feed for contamination and nutrients etc.

Moreover, a veterinarian should be trained, experienced and have basic knowledge of animal normal and abnormal behavior to assess the level of management and care provided to animals because animal behavior is an obvious indicator to check the status of management and care facilities provided to animals at farms, labs, houses, zoos, etc. (Panzera, 2013).

1.10 Challenges in Management and Care and Their Possible Solutions:

Today we are still facing challenges in managing and providing care to animals (Fernandes et al., 2021) some usual challenges include:

Ensuring animal health and well-being is still a challenge in most Third World countries, which comprises housing, feeding, veterinary services and enrichment practices. Providing proper space, ventilation, temperature, humidity, biosecurity, feed free from contamination, fresh and microbe-free water, proper and timely veterinary checkups, all these practices nullify the well-being and health issues of animals. Assessing animals' behavior helps in assessing management and care levels.

Zoonotic diseases (Rahman et al., 2020) like rabies, brucellosis, avian influenza etc. have drastic effect on animals as well as humans and can only be prevented by standardized and proper management and care of animals, proper and right time vaccination of zoonotic diseases prevent outbreaks and helps in managing these diseases, moreover, active and effective surveillance (Sharan et al., 2023), proper biosecurity measures (Robertson, 2020), proper disinfection, quarantine of newly bought animals and coordination between veterinary and public health agencies help to combat this challenge.

Abrupt climate change effects directly animal (Cheng et al., 2022), habitats and food availability (Godde et al., 2021), these variations in climate change led to drastic and severe weather events like hurricanes, wildfires which greatly disturbs animal populations and habitats. Conservation of endangered species by developing zoos, storing and performing artificial inseminations for protecting endangered species to become extinct by coordinating with OIE, WOAH and other wildlife conservation organizations, devising programs to promote plantation. All these measures help to minimize the effect of harmful climate variations.

Illiteracy and unethical research using cruel and inhumane practices is also a major issue in management and care of animals. In Third World countries the standards set by OIE are not followed because most farmers, trainers, pet owners don't even know what OIE stands for and why this organization developed for! Educating veterinarians, farmers, trainers, owners, zookeepers for animal well-being and welfare is important.

OIE is World Organization for Animal Health, established in 1924. The main objective of this organization is to improve living standards of animals and to improve animal health and welfare by developing standards (Petrini & Wilson, 2005) for diagnosis, treatment and prevention of lethal diseases. Following these international standards helps in minimizing current challenges regarding animal welfare, management and care.

1.11 Future Perspectives:

The future advancements (Boyle et al., 2022) using in animal care and management will prove helpful in many aspects, advanced AI techniques (Zhang et al., 2024) and robotics will help in management, automatic feeding system, remote monitoring of animal health, machine milking, dairy management software, etc. these all modern technologies will help in managing animals regarding their welfare (Kaur et al., 2021). Recording of animal behavior helps in manipulating the welfare offered to animals and assessing the level of management and care of farm animals (Neethirajan, 2020). Modern technology also helps in educating the owners, farmers, trainers, zookeepers and veterinarians at global level (Buller et al., 2020).

1.12 Conclusion:

In conclusion, animal management and care are not a single person's responsibility, it's a teamwork for providing animals with habitat and environment in our supervision that is near to nature. We should provide natural, balanced (Mordenti et al., 2021), hygienic feed, water free from heavy metals, bacteria, sewage, waste disposal etc., provide housing which protect them from harsh climate changes, prevent heat/cold stress, should provide soothing effect to animals and should not be the cause of fear and distress. We should provide such an environment to animals in which they show their natural behavior. Animal behavior is a good indicator in assessing the degree of care provided to animals and to which extent efforts were made for their management (Panzera, 2013). According to animal welfare there are five basic freedoms we must provide to animals. We should fulfill the safety needs of animals; we should devise plans for their betterment and well-being. The role of veterinarian in incredibly significant in animal management and care, a veterinarian must know the international standards for animals organized by OIE and play his/her role in implementing these standards by educating farmers, pet owners, trainers, etc. There are still challenges in third world countries, Artificial Intelligence and modern technology is playing its role in making up this gap, moreover, animal welfare organizations like OIE are providing its services in ensuring the stewardship and care of animals at various levels farms, homes, training centers, zoos, etc.

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Chapter 2: Veterinary Ethics in Research and Clinical Practice

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Abstract

This article provides an in –depth analysis of ethical issues in veterinary research and clinical practice, focusing on beneficence, non-maleficence, non-maleficence, justice, and autonomy. It discusses topics like informed consent, pain management, end of life decisions, and conflicts of interest. The article critically evaluates veterinary ethics policies, identifying areas for improvement. The essay also discusses new moral conundrums brought up by developments in veterinary technology, including telemedicine, artificial intelligence, and gene editing. It emphasizes how moral principles must be continuously discussed and adjusted to maintain social values and moral integrity while keeping up with scientific advancements.

Keywords: Veterinary ethics, Clinical practice, Animal welfare, Ethical issues, Research ethics

2.1 Introduction

Legislation reflects the moral significance of vivisection (Festing and Wilkinson, 2007; Chapouthier, 2013; Global Animal Law Association, 2022; Ashall *et al*., 2023). The Council Directive 86/609/EEC was created in 1986 with the intention of safeguarding vivisection trial and other biological reasons. Contemporary European rules governing application of test animals back track to the 1980s (Louhimies, 2002).

Twenty-four years later, the European Parliament revised this directive, which now applies to all EU member states, as Directive 2010/63/EU (Chlebus *et al*., 2016). A few governments that are part of the European Economic Area (EEA), including Norway, have been modified their national laws to comply with this directive. The Animals (Scientific Procedures) Act (A(SP)A) 1986 was enacted in the United Kingdom (UK) to replace the 'Cruelty to Animals Act,' which was introduced in 1876. It involved the provisions of the Directive 86/609/EEC and established new regulations for the safety of animals other than humans used for research purposes.(Cruelty to Animals Act, 1876; Rankin, 1986).

The laws mentioned above govern the application of laboratory animals (having both vertebrate animals and cephalopods) in biological investigation; however, some procedures may be exempt from these laws, as stated in Article 1 of the Directive 2010/63/EU. These exemptions include any procedure that is considered "non-correlational" in analytic or agricultural veterinary application, any procedure that is improbable to pain agent, danger and hardships that is comparable to, or greater than, that which would result from introduction of a hypodermic needle, and veterinary correlational evaluation that are necessary for the trading permit of a therapeutic products for animals.

Figure 1 Steps involved in clinical practices of veterinary ethics

According to European and UK law, veterinarian clinical research on animals owned by client that fits under the category of correlational veterinary practice" is exempt from ethical scrutiny. Put differently, as per Clutton (2009), Ashall *et al.* (2023), and the World Medical Association (2013), legislation in Europe (including the UK) govern the use of lab animals for scientific operations, but not those pertaining to clinical study with veterinary patients.

This does not, however, mean that simply because a clinical practice or piece of research can be carried out lawfully without ethical review, it should be exempt from ethical review procedures. Furthermore, starting of September 1, 2022, non-routine veterinary treatments in the UK, a broad term that includes clinical trials must undergo an ethics review, according to the RCVS (RCVS, Routine veterinary practice, and clinical veterinary research, 2023). Previous several decades have seen a rise in ethical consciousness in society, which is reflected in the constant push to form committees with the goal of controlling application of animals in non-experimental (also known as non-(A(SP)A)/non-2010/63/EU) research (Fordyce and Mullan, 2017).

Furthermore, irrespective if the research material supplied research lawfully carried out without an initial moral perspective majority of scientific journal particularly, state in their rules for authors, adherence to legal and welfare of animal’s principles as well as approval from a separate ethics committee. To assist authors and journals in determining the minimal data required to be reported in disclosure in vivo research were modified and reintroduced in 2020 (Kilkenny *et al*., 2010; Percie du Sert *et al*., 2020a; Percie du Sert *et al*., 2020b).

Comparably, all forms of animal research and testing are intended to be covered by the

PREPARE recommendations (Planning Research and Experimental Procedures on Animals: Recommendations for Excellence), which also cover animal facility management (Smith *et al*., 2018). This heightened consciousness of ethics and animal welfare, along with the desire to apply these values to non-laboratory animals, is somewhat reminiscent of the evolution of several standards of conduct for human in medical research since the conclusion of World War II. Examples of this include the UK's National Health System Ethics Committees and Clinical Ethics Network, the Declaration of Helsinki (1964), the Nuremburg Code (1947), the Belmont Report (1978), and several international laws and regulations (Goodyear *et al*., 2007, Algahtani *et al*., 2018, Holm, 2020, Harnett, 2021, Clinical Ethics Network, 2023).

This article's goal is to talk about specific moral issues surrounding veterinarian medical studies on non-experimental, animal owned by the client. We want to draw attention to the ethical and welfare principles that all researchers should uphold, no matter what is origin, cultural heritage, religious beliefs, or personal individual mindset toward animals. This is because their research is conducted in a country where laws governing animal welfare are in place. Important steps involved veterinary ethics in research and clinical practice has been shown in Fig.1

2.2 The rights and obligations of the parties participating in the study.

In clinical veterinary research, the "moral agents" should be consider in addition to the well being (and rights) of the moral animals. Every moral agent, the customer or proprietar the formal places where the planned research will be carried out, the investigators, and the participating veterinary team has rights and obligations. These rights must encompass both the "moral rights" of each, and every party engaged as well as "legal rights," which are a component regarding the social compact including in the laws allowing the utilization animal for scientific purposes. Legal rights establish minimal moral requirements, but taking animal rights into account is crucial when defending the morality regarding the ethical agent.

There are various who contend that animals are entitled to be free from needless suffering and medical intervention (Browning and Veit, 2022; Martin, 2022). Utilitarianism, a philosophical philosophy that holds that all sentient beings, not just humans, should be taken into consideration and not be subjected to discrimination, is credited with first conceptualizing this (Spector, 1963, Warren, 2000). Nonetheless, 1975 saw the publication of the first philosophical treatise that served as the foundation for the contemporary animal rights movement (Singer, 1975). The concept of "the idea that animal have morality is known as animal right standing and ought to be free from human cruelty throughout their lives (Singer, 2001; Regan, 1980).

Animals are regarded by most legal systems worldwide as "property." Since animals are legally regarded as property, their owners fully and accurately have the legal entitlement aware the advantages and danger of the studies and treatment being carried out on their land including any possible harm the animals may suffer, as well as the financial ramifications of participating in the study, if any. Additionally, owners have the freedom to withdraw their consent at any moment, to amend it when information changes, and to avoid being forced to participate. In terms of responsibilities, proprietors are moral obligation to put the wellbeing of their animals first and to put those interests ahead of their own. Their desire regarding the clinical state of their pets to improve shouldn't be used as an excuse for ineffective, perhaps hazardous, or nonexistent care.

The Institution has the right to defend itself and keep itself from being linked to studies or therapies that could damage its reputation due to unethical or illegal choices made by the researcher. On the other side, the responsibility of the Institution (as well as the researchers) is to follow applicable laws and refrain from offending society by causing needless harm.

Regardless the matter whether the results are favorable or adverse, researchers have the freedom to share them if they follow ethical and welfare guidelines and employe sound technique. The responsibilities of the researchers include furthering veterinary science for the benefit of future veterinary patients, prioritizing the interests of the owner and the welfare of the animals, being devoted to identifying unexpected harm early on and stopping the trial if it occurs, using reliable scientific techniques that guarantee the reliability of the findings and not doing more harm than lastly understood intervention

Finally, the support staff, veterinarian nurses, for example, have an obligation and a right to uphold their professional conduct codes and oaths. Should the study treatments cause harm, such as animal suffering due to a violation of professional codes or laws, the support staff is also obligated to report the matter to the researchers or the institution.

2.3 The arrangement of ethics committees inside organizations

To guarantee trials in clinical practice would continue to be carried out in compliance with morality and welfare principles, universities established many departmental or institutional ethics committees (Moon, 2019). But academic institutions are not the only organizations with ethics committees. Several professional orders and associations, including the Royal College of Veterinary Surgeons (RCVS) in the UK and several corporate veterinary employers, also have ethics committees.

Members of the committees should have a range of related skill sets; in general, people with a background in science who are knowledgeable about ethics and animal welfare are regarded as crucial members. According to Kuyare *et al*. (2014), individuals possessing proficiency in animal surgery and anesthesia are considered exceptionally equipped to offer insights on the extent of invasiveness of the procedures carried out, as well as the degree of analgesia and actinociception provided by the scheduled rescue action. The composition of committees may differ depending on the members' specific expertise, but at the very least, the person the senior employee with experience on review boards should serve as the chair, a demonstrable research background, and a basic understanding of the veterinary (clinical and non-clinical) procedures that are routinely performed on animals.

Ethics committees may utilize a variety of criteria to inform their evaluations of study proposals. Certain committees may deem it appropriate to demand that the participating researchers submit documentation of their professional skills, scientific, and technical experience proficiency as well as show that there is adequate scientific background and knowledge to bolster the inquiries for study and, consequently, validate the rationale behind the suggested research.

Committee decisions must, in the end, adhere to the principles outlined in codes of professional behavior as well as the applicable national legislation, regardless of their criteria.

2.4 Techniques and Protocols

As indicators of a strong and reliable technique, a research study should prioritize an effective experimental design and the appropriateness of the suggested statistical methodologies. Ethics committees should consider factors including statistical methodologies, the relevance and acceptability among the protocol presented, and the use of verified resultant metrics, even though a comprehensive examination of methodology may be outside the purview of their job (Kilkenny *et al*., 2009). This is because using animals in research is only morally permissible when it is anticipated to yield reliable results that can be used to support new scientific theories. Furthermore, it is considered unethical to do research using procedures that are not reproducible (Baker, 2016). The sample size calculation is a crucial component of statistical approaches since it directly influences the number of animals that participate in the study. Large-scale client-owned animal enrollments in clinical trials, which entail treatment variations from the norm, should only be supported by well-executed sample size calculations grounded in reasonable hypotheses and, ideally, estimations derived from pilot or previously released data. However, if there are insufficient animals in the study, there is a greater chance of type II error, which could lead to the research not being able to find an actual statistically significant difference (Columb and Atkinson, 2016).

All operations carried out upon animals it must be thoroughly detailed in the description of to project assess the level regarding intrusiveness and determine whether there is a significant divergence from accepted clinical veterinary work. If the assignment of treatments is to be randomized, this could present further difficulties. Randomization of treatments during a medical product trial in the United Kingdom necessitates Veterinary Medicines Directorate authorization (Animal Test Certificate, ATC, 2023), which can be expensive and time-consuming. Only procedures carried out for research reasons, as opposed to the animal's benefit, that have the potential to cause harm comparable upto or greater than a hypodermic needle insertion are permitted under both 2010/63/EU/A(SP)A).

Blood sample is a common example of this, even when it is done via an already-placed indwelling catheter. This is because drawing blood without a need is considered a relatively invasive process, even if it requires the insertion of a needle percutaneously. The amount of blood that has to be drawn be raised inadvertently for this purpose, remaining blood collected for clinical use (e.g., regular diagnostic applicant of biochemistry) may be subjected to additional analysis for study- related purposes. However, this can be a gray area because many would contend that the volume- dependent nature of blood collection from a catheter inside the body determines how invasive the procedure is. For the reasons mentioned above, For the same causes mentioned above, injection- based false therapy. In most of Europe, injection-based placebo therapies are considered inappropriate due to the same reasons mentioned above;

however, in the UK, they might be allowed as per A(SP)A regulation. Since 2022, the UK has allowed the delivery of placebos by non-invasive methods (such as oral administration), while European law does not expressly govern this.

The project description should specify exactly what steps and measures are to be followed if unexpected harm occurs. Rescue processes should ought to be possible to identify and also note any unanticipated damage. A common illustration of this would be a thorough rescue analgesia strategy for when pain breakthroughs during invasive procedures occur, even if those treatments are a component of a veterinarian therapeutic therapy that the client is receiving for the benefit of their animal. Similar protocols should be in place to end the experiment should harm from the study or treatment turn out to be evident, as well as to share this information if it does.

If initiatives have the potential to endanger the psychological and/or physical health of the people working on them, then these risks should be recognized, and precautions taken to reduce them.

2.5 Benefit-harm trade-off analysis.

The project's possible advantages and disadvantages should always be weighed against the animal population that are same in their natural habitat, ailment that is the subject of the investigation as well as the individual study of animals. A key component of the regulation of animal research, an ethical precaution known as harm benefit analysis comprises a thorough assessment of the potential benefits outweighing any potential risks, as well as the nature, severity, and risk of any harms. (Petkov *et al*., 2022).

However, these authors would contend that, as a "prima facie" principle, harm-benefit evaluation should also apply to the context of research carried out as part of "recognized veterinary practice," from the viewpoint of the individual animal, veterinary practice, in contrast the scenario for research executed under A(SP)A 1986 or 2010/63/EU, where harm-benefit evaluation proceeds from a large utilitarian viewpoint of society.

In the late 1980s, the idea of harm-benefit analysis in relation to moral laboratory animal care became more and more well-known. It was improved in more recent years and reexamined in 2023 to become a part of the "12 Rs framework," a unified set of ethical guidelines for animal research

(Bateson, 1986, Pound and Nicol, 2018, Brink and Lewis, 2023). It is the duty of researchers to make sure that any harm caused will be as minimal as feasible and that there will be some gain from the planned research (Foëx, 2007; RSPCA, Harm-gain Analysis, 2023). According to DeGrazia and Sebo (2015), one of the requirements that must be met for research with animals to be ethically permissible is the anticipation in order to study enough tangible advantages.

It is appropriate to assume that procedures carried out patients who are veterinarians involved in clinical research conducted outside of the frameworks of A(SP)A 1986 or 2010/63/EU should still adhere with the appropriate laws in the legal authority, and that standard moral judgment structures should be used to help with the achievement of ethically acceptable outcomes, even though no specific law has been developed to regulate such procedures. In this regard, directories and code of behavior from the UK other than the A(SP)A 1986 ought to be considered as well These include the Animal Welfare Act 89–544 of 1966, 2023, the Veterinary Surgeons Act (VSA), (1966), and the Veterinary Medicinal Products Directive regulation 2001/82/EC.

2.6 Information and permission from the owner

Open and truthful interaction with pet owners is another vitally important ethical component. An extensive information document outlining the pertinent background data, the objectives of the study, the treatments to be performed on the animals, and any possible risks should ideally be sent to animal owners. While there is universal consensus that certain incentives, like discounted hospital expenses, are morally appropriate when used to encourage enrollment, caution must be exercised to avoid coercing customers into participating against their will.

The customer need to be notified of their entitlement option out from the research at any moment having an impact on the care they receive going forward, and engagement in the research completely willingly. In addition to withdrawal at the client's discretion, the trial may be terminated by the researchers at any time, either for specific animals or for both, if the study causes discomfort to the animal or its owner, or both; if the project develops and results in an unanticipated, possibly negative results for the animals (RCVS, 2022).

While getting consent from the owner of the animal is a required moral considerations, it should be noted that consent from the person who owned the study animals differs significantly from that of capable adult human suffer who chooses to participate in a clinical trial (Ashall *et al*., 2018). Animals are the legal property of their owners, but just as children and incompetent adults are considered vulnerable subjects because they are unable to protect their own interests or make decisions for themselves, so too should animals be given the same consideration.

2.7 Inherent disputes of interest

A possible disputes of interest could emerge from affiliations - or animosities - to industries, organizations, or groups that could sway the researcher's decisions or behavior. When these interests are private and could lead to financial or personal advantage, the matter becomes much more delicate (Yeates *et al*., 2013). Because of this, many scientific publications demand a conflict-of-interest declaration in which all funding sources, including the support of pharma companies, must be disclosed as contributions to the research.

Certain publications might also demand accreditation on their contributor argument for publication, that the study is free from contributions that would prohibit the researchers from publishing any results, no matter how positive or negative, or that prohibits them from publishing their work without first receiving permission from the sponsor.

2.8 Data retention, deletion, and storage

Only recently has data protection evolved into a moral and legal need. Other than the data that law enforcement and intelligence organization manage, the General Data Protection Regulation (EU GDPR 2016/679) is a European Directive that was established in 2016 and lays out the fundamental rights, obligations, and principles for most of the processing's of personal data (General Data Protection Regulation (GDPR), 2023). The UK Data Protection Act (DPA) went into effect in May 2018. It was modeled after the EU GDPR, which was already in place in the country prior to that date, but with modifications intended to better tailor it to the UK environment.

According to EU GDPR regulations, client anonymity must be guaranteed, and safeguards must be establish to safeguard the animal's owner's identity if delicate information is to be collected. Furthermore, private information needs to be kept safe. For electronic files, this means a locked drawer; for paper files, it means a password-protected access. Only the investigators should have access to this sensitive material.

An additional consideration is how research data is disposed of. Delicate information must be kept securely as required amount of time additionally, only for as long as reasonable and essential is necessary and appropriate, as per the EU GDPR, before being safely destroyed. Federal rules in the United States govern the preservation of data related to studies using human participants; however, the position is less clear when clinical trials include animals rather than humans. The US Department of Health and Human Services (2021) mandates, for instance, that research data be kept on file for a minimum of three years following the conclusion of trials (45 CFR 46). However, Health Insurance Portability and Accountability Act (HIPAA, 1996) standards apply to research involving identifiable health information, requiring records to be the best of the authors' knowledge, no regulations have been created expressly to control how research data is disposed of. While it does not specify a time limit, the DPA provides guidelines for the preservation of research files that contain sensitive or personal data. The investigators will determine this based on how long the data will be required for their goals, but proper security measures must always be in place to protect people.

In general, it's thought to be a good practice to save data for at least five years, or until there is no plausible chance that it may be needed to refute claims of scientific misconduct. Sensitive information may still be included in research data after anonymization; this is especially true for data pertaining to animals, such as rare breeds or traits of the research animals. It is possible to preserve scientific data and truly anonymized data indefinitely, according to the RCVS Ethics Review Panel (ERP) Guidance on storing personal data in Clinical Veterinary Research (RCVS, 2021). Lastly, it should be remembered that when research is sponsored, the sponsors may have requirements.

2.9 Conclusion

The ethics surrounding animal experimentation have developed over the previous few decades, and current century is seeing an ethical enlightement in both the scientific and societal arenas. Contractarianism, a moral theory that asserts that only people who can comprehend and choose to participate in moral rights can be attributed to an agreement and hence denies animals any kind of moral rights, is no longer regarded as morally acceptable (Gross and Tolba, 2015, Baldwin, 2023). The formation of suitably constituted local ethics committees is essential without European union law that particularly regulates research carried out on non-protected animals A(SP)A 1986 or 63/2010/EU.

To safeguard the security of interest of owners and research animals, these committees should provide advice regarding the desirability, morality and legality of the suggested study. They should also work to prevent reputational harm to the institution and the researcher, as well as public concerns regarding the public usage of animals. The veterinary community believes that there should be a complete, unified framework of behavior and ethical norms that apply regarding the use of non-laboratory animals owned by client in research, even though there isn't currently a lawful process of that kind.

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Chapter 3 : Role Of Nutraceuticals In Veterinary Medicine

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Abstract

This chapter delves into the growing significance of nutraceuticals in veterinary medicine, focusing on their potential to enhance animal health and complement standard treatments. Nutraceuticals, which include products like vitamins, minerals, herbs, and dietary supplements, are gaining recognition for their beneficial effects on animals. The chapter explores the mechanisms by which these compounds operate, such as boosting the immune system, reducing inflammation, and aiding metabolic functions. It book chapter the effectiveness of nutraceuticals in treating common veterinary conditions, including joint issues, digestive problems, and skin disorders. Additionally, the chapter addresses the challenges of regulation, quality control, and the need for more comprehensive scientific research to ensure the safety and efficacy of these products. Through a detailed examination, this chapter underscores the importance of integrating nutraceuticals into veterinary care to improve the health and quality of life of animals.

3.1 Introduction

A nutraceutical is a dietary supplement that provides a concentrated form of a biologically active substance found in food, presented outside its natural food context, aimed at improving health.

Nutraceuticals are widely used in animals for preventing and treating diseases, much like they are in humans. The growing preference for nutraceuticals over synthetic drugs is driven by their cost-effectiveness, safer nature, and broader availability.

Nutraceuticals come from food sources and are thought to offer extra health benefits beyond basic nutrition. Within legal boundaries, these products might claim to prevent chronic diseases, enhance overall well-being, slow aging, increase lifespan, or support the structure and function of the human body.

Dietary supplements contain a range of ingredients known as 'dietary ingredients,' which encompass vitamins, minerals, botanical extracts, amino acids, as well as substances like enzymes, organ tissues, glandular extracts, and metabolites. These supplements are commonly available in concentrated forms like extracts and are offered in different formats such as tablets, capsules, softgels, liquid formulations, or powders.

These substances interact with the body's processes to potentially improve health and prevent diseases by supporting immune function, reducing inflammation, enhancing joint health, improving digestion, and providing antioxidants. They achieve these benefits by influencing biochemical pathways and metabolic functions, offering more than just basic nutritional support.

Nutraceuticals for animals struggle with varying quality and effectiveness due to sourcing and manufacturing differences. Regulatory complexities across regions complicate approval. Ensuring accurate dosing is challenging due to animal variability. Limited scientific evidence of efficacy across species hinders acceptance by veterinarians and owners.

3.2 Composition of Nutraceuticals

Nutraceuticals are defined as meticulously crafted formulations, designed to meet targeted dietary needs and provide prophylactic healthcare. These products consist of specific nutrients intended to prevent and treat certain illnesses, as well as complement a balanced diet.(Carrasco-González et al., 2017). Natural bioactive compounds encompass a wide range of chemical structures and properties, offering a rich reservoir of molecules suitable for the development of nutraceuticals, functional foods, and food additives.(Joana Gil-Chávez et al., 2013). Edible mushrooms, which are food items with low fat content and high protein levels, have been cultivated in China since 600 CE. The large-scale cultivation of these mushrooms, particularly the Agaricus bisporus, was not successful until the 17th century in France. Subsequently, the cultivation of the "oyster mushroom" (Pleurotus ostreatus) was introduced in the United States in 1900 (Zhang et al., 2013a).

3.3 Edible Mushroom And Hemp Seeds As A Source Of Nutrition

Currently, China stands as the leading global producer of edible mushrooms, accounting for 64% of the total production (FAOSTAT, 2016).(Carrasco-González et al., 2017). The hemp seeds (Cannabis sativa L.) have served as a significant nutritional resource in Chinese and European societies for millennia. Presently, despite abundant clinical data emphasizing their vast health-promoting attributes, there remains a lack of awareness among individuals regarding their nutritional and nutraceutical advantages. The remarkable abundance of crucial polyunsaturated fatty acids in hemp seed remnants contradicts their oxidative stability.(Crescente et al., 2018). Nutrients encompass substances with confirmed nutritional roles, including vitamins, minerals, amino acids, and fatty acids. Herbals refer to herbs or botanical products in concentrated or extracted forms. Dietary supplements consist of reagents sourced from various origins, like pyruvate, chondroitin sulfate, and steroid hormone precursors, targeting specific purposes such as sports nutrition, weight-loss aids, fortified traditional foods, and meal substitutes(Chauhan et al., 2013).

3.4 Herbal nutraceuticals

A substantial level of energy is found within the herbal nutraceutical tablet. The newly developed herbal tablets were found to contain iron, sodium, potassium, phosphorus, magnesium, iodine, copper, zinc, and calcium. Additionally, a considerable quantity of water-soluble and fat-soluble vitamins were detected in the tablets.(Debnath et al., 2024) Nutraceuticals encompass a variety of sources including natural food items, spices, herbs, and their derivatives, as well as processed products like fruits, vegetables, cereals, tubers, dairy products, marine resources, genetically modified foods, and more.(Riar & Panesar, 2024).

Various high-value chemicals and bioproducts can be derived from the waste of fruits and vegetables. The waste generated from the processing of fruits and vegetables serves as a sustainable bioresource for the nutraceutical and functional food industry, as well as a promising feedstock for biorefinery. Therefore, this chapter presents an overview of recent trends in utilizing fruit and vegetable waste for recovering bioactive compounds and incorporating them as nutraceutical and functional ingredients.(Pradhan et al., 2023). A nutraceutical bolus designed for commercial use comprises herbal extracts (Echinacea purpurea, Silibum marianum) and synthetic compounds (carnitine and vitamin E) and serves as a proactive measure against diseases in transition cows. The primary objective of this product is to function as a catalyst for intermediate metabolism, achieved through several mechanisms: firstly, the immunomodulatory properties of Echinacea purpurea (Seckin et al. Citation2018;McNeil et al. Citation2023); secondly, the hepatoprotective effects of silymarin derived from Silybum marianum (Hermenean et al. Citation2015; Ulger et al. Citation2017); thirdly, the antioxidative capabilities of vitamin E (Haga et al. Citation2021; Xiao et al. Citation2021), leading to a decrease in the occurrence of mastitis and reproductive disorders, as well as alterations in the fatty acid composition of colostrum and milk (Weiss Citation2017); and lastly, the promotion of energy metabolism by Carnitine, which aids in the prevention of fatty liver in dairy cows experiencing negative energy balance (Ringseis et al. Citation2018).(Esposito et al., 2024).

The by-products generated in the course of processing exhibit a high content of oil, carbohydrates, proteins, and various essential nutrients. Scholars are progressively investigating the application of residual materials to mitigate environmental contamination and associated challenges. Historically, botanical by-products have been acknowledged as a reservoir of bioactive substances, such as fiber, phenolic compounds, and numerous other bioactive agents.(Routray & Orsat, 2017).

3.5 Green Pea (Pisum sativum L.) as Ingredient in Nutraceutical Formulations

Pea (Pisum sativum) is classified as a plant that thrives in cool seasons and is a member of the Fabaceae family, with a widespread cultivation across the globe and traditionally believed to originate from Southern Europe. The consumption of pea seeds has been historically significant due to their rich nutritional composition consisting of substantial levels of proteins, starch, fibers, minerals, and vitamins. Nevertheless, it is important to note that pea seeds do contain antinutritional elements like phytic acid, which has been associated with hindering mineral absorption.(Castaldo et al., 2021).

3.6 Mode of Action of Nutraceutical

"We explore various nutraceutical classes for ruminants, categorizing them by their mode of actions (probiotic, prebiotic, phytonutrient, and polyunsaturated fatty acid).

3.7 Probiotics to modulate immune response

Probiotics mainly work by fighting harmful microbes and boosting gut immune defenses. Probiotics that boost butyrate can change gut defenses.Top of Form

Butyrate, a short-chain fatty acid, nourishes gut cells and enhances barrier function, promoting gut health(Böcker, Nebe et al. 2003, Peng, Li et al. 2009).

Probiotics in mature ruminants affect both the rumen and lower digestive tract, with some able to bypass initial compartments and still offer benefits to the lower gut.(Tkalcic, Zhao et al. 2003).

Oral probiotics' effects on gut health are simpler to grasp compared to how they enhance immunity in non-mucosal tissues like the mammary gland. The mucosal immune system is a complicated network covering the GI, respiratory, and urogenital tracts. Immune responses from one mucosal site can move to others, but probably not to the mammary gland. Unclear mechanisms weaken the argument for oral probiotics improving mammary gland health.(Rainard and Foucras 2018)

Directly applying probiotics, like lactic acid bacteria or Bacillus sp., to inhibit mastitis-causing pathogens in a teat dip is a more promising way to boost mammary gland health. Injecting live lactic acid-producing bacteria into the mammary gland increased the release of proinflammatory cytokines such as interleukin-8 (IL-8) and IL-1, attracting neutrophils to the site.(Beecher, Daly et al. 2009) Key mechanisms include boosting rumen bacteria, balancing populations, utilizing lactic acid, averting acidosis, and maintaining low oxygen levels.(Newbold, Wallace et al. 1995, Chaucheyras-Durand and Fonty 2001)

3.8 Prebiotics

Prebiotics, like oligosaccharides and fructans, boost animal health by fueling beneficial bacteria in both the rumen and lower gut.(Liu, Li et al. 2017).Galactooligosaccharides are in soybeans and milk, while fructooligosaccharides come from plants. Prebiotics from yeast cell walls, like mannan-oligosaccharides (MOS) and β-glucans (BGs), vary in composition and affect animal health based on their form in feed or extract(Ganner, Stoiber et al. 2013).

β-Glucans, derived from fungi, can modulate immunity. They bind to Dectin-1 receptors on immune cells like monocytes, macrophages, and neutrophils, and to a lesser extent, on dendritic and T-cell surfaces.(Taylor, Brown et al. 2002).

Activation of leukocyte responses through Dectin-1 receptor ligation is seen with fungal extract supplementation, explaining the positive immune response(Davis 2018).

3.9 Antoxidant activities of plant nutraceuticals

Nutraceuticals possess qualities that include acting as antioxidants, contributing to anti-aging efforts, displaying anti-cancer properties, and enhancing biochemical processes and bodily structures (Nasri et al. 2014). Nutraceuticals have the potential to help prevent and treat conditions like heart disease, neurodegenerative disorders, oxidative stress-related illnesses, and modern lifestyle diseases such as diabetes, hypertension, and cancer(Das et al. 2012; Sachdeva et al. 2020). Studies have shown that nutraceuticals are safe to consume, despite some active ingredients potentially having toxic profiles. Controlled dosages ensure they do not cause toxicity, while their beneficial bioactive compounds synergistically enhance health impacts (AlAli et al. 2021). Nutraceuticals are popular due to their effective immune enhancement, widespread availability, affordability, and high individual tolerance (Kim and Kim 2010).

3.10 Function of Nutraceutical in Veteinary Medicine

The term nutraceutical originates from the fusion of 'nutrition' and 'pharmaceutical', with numerous definitions available in various dictionaries. In a broader context, a nutraceutical refers to a type of food that not only provides nutritional value but also offers health benefits, containing bioactive compounds that interact with animal physiology at different levels. Probiotics, prebiotics, fatty acids, peptides, amino acids, secondary plant metabolites, and essential oils represent a non-exhaustive compilation of nutraceutical compounds. This review will specifically concentrate on probiotics ,secondary plant metabolites and prebiotics.(Colitti, Stefanon et al. 2019)

3.11 Nutraceutical against OTA (ochratoxin A) Kidney Oxidative Stress:

Nutraceuticals are biologically benign, renewable, and conveniently accessible commodities distinguished by a specific ease of disintegration and preservation. These attributes could offer a significant benefit in utilizing natural compounds to enhance animal well-being and, consequently, human health [58]. Nutraceuticals encompass various bioactive compounds targeting multiple sites, enabling their extensive application in numerous animal and human ailments.

They have the capacity to induce both physiological and pharmacological impacts, such as safeguarding against reactive oxygen species and displaying antioxidant properties [18]. As previously stated, lipid peroxidation, recognized as a critical outcome of mycotoxicosis, is linked to the production of reactive oxygen species, whose imbalance induces functional and structural modifications in the kidney, the principal organ accountable for OTA metabolism [59].

Various studies emphasize that OTA-triggered nephrotoxicity and carcinogenicity might result from the creation of reactive oxygen species like superoxide anion ($O2^{-}$), hydroxyl radicals (OH^{-}), and peroxide (ROO^{-}), leading to significant damage to cellular components and compromising antioxidant defenses [60]. Considering the crucial role of oxidative stress in an OTA-exposed kidney, numerous antioxidants have been employed due to their advantageous role in regulating the production and neutralization of reactive oxygen species/reactive nitrogen species, as well as preserving equilibrium.

In this circumstance, nutraceuticals exhibiting antioxidant properties could serve as notable instances of natural substances that could be integrated into feeds suspected to be contaminated with OTA. Presently, the scientific community, encompassing veterinarians, has acknowledged the significance of nutraceuticals in animal welfare, with numerous research studies assessing their efficacy in averting and shielding against OTA-induced nephrotoxicity and oxidative stress, recognized as primary modes of action of this toxin [61].

This section concentrated on antioxidant mechanisms that could play a pivotal role in averting and alleviating the detrimental consequences of both reactive nitrogen and reactive oxygen species imbalance throughout OTA exposure.(Longobardi, Ferrara et al. 2022)

3.12 In Poultry:

The nutraceutical industry generates an annual revenue exceeding 250 billion dollars, with a higher prevalence of nutraceutical use in animal diseases compared to human diseases due to their economical nature and safety (Gupta et al., 2019). The application of nutraceuticals in the diet and rearing of poultry is extensive. It is crucial to note the incorporation of a novel range of feed supplements, such as nutraceuticals, into poultry diets, It is possible to purchase chicken products (meat and eggs) that have been augmented with biologically active substances including vitamins, microelements, and polyunsaturated fatty acids.

(Alagawany et al., 2019a, Alagawany et al., 2019b, Dhama et al., 2014a, Dhama et al., 2014b, Lee et al., 2019, Michalak et al., 2020, Yadav et al., 2016).(Alagawany, Elnesr et al. 2021).Organic acids, probiotics, prebiotics, and exogenous enzymes are examples of nutraceuticals, are utilized as substitutes for antibiotics due to their impact on the intestinal microbiota modulation (Dhama et al., 2008; Dhama et al., 2014a, Dhama et al., 2014b; Yadav et al., 2016; Yadav and Jha, 2019). These nutraceuticals play a crucial role in safeguarding the host against infectious diseases (Cencic and Chingwaru, 2010) while also enhancing the gut microbiota, immune responses, and growth promotion in poultry (Sugiharto, 2016).

Their impact on the microbiota in the stomach helps improve the digestion, absorption, and metabolism of nutrients as well as the general health and performance of chickens (Yadav and Jha, 2019). They exhibit significant roles in combating gastritis and protecting the gut (Sangiovanni et al., 2018).(Alagawany, Elnesr et al. 2021)

3.13 Quality of nutraceuticals on veterinary animals

Nutraceuticals have proven to be advantageous not just for humans but also for veterinary animals like cows, horses, dogs, and cats, among others. Various nutraceutical medicines and supplements have been employed in veterinary care for many years. They contribute to enhancing meat production, reproductive rates, disease treatment, improving the quality of life for animals under medication, and boosting egg production in poultry(Zhang et al., 2017).

Various commercial probiotics are utilized to enhance the productivity and quality of cattle milk. Supplements like vitamin E, vitamin A, copper (Cu), iron (Fe), manganese (Mn), and selenium (Se) function as antioxidants, effectively extending the shelf life of milk(Olchowy et al., 2019).

Additionally, casein and whey have been identified as ant oxidative agents, with some formulations even being patented for their efficacy(Castillo et al., 2013). Moreover, dietary manganese-methionine supplements prolong the lifespan of eggshells in laying hens, thereby preventing breakage and mitigating significant commercial losses(Li et al., 2018). hydroxy selenomethionine is used for increasing the production on meat quality of yellow feather broiler. Omega-3-enrichedtherapeutic diets, Green-lipped mussels and Fish oil are used as a treatment ofNutraceuticals in Canine and Feline Osteoarthritis.(Barbeau-Grégoire et al., 2022)

3.14 Nutraceuticals effect on egg laying hens:

The nutrition of breeders is a critical element affecting chick quality, as the chick embryo depends on the nutrients present in the egg for its growth and development. Moreover, the egg provides essential antibodies crucial for the chick's well-being during the initial weeks of life. Brown algae contains numerous bioactive compounds, and incorporating algal extracts into the diet has demonstrated enhancements in gut health and immune responses in both pigs and poultry.(Li et al., 2018)

Ensuring eggshell quality is a significant priority not just for egg producers but also for consumers. The commercial processing and marketing of eggs often result in a notable incidence of cracked or broken eggshells.manganese-methionine is an important supplement for making the eggshell of hens. Supplementation of manganese at a dosage of 100 mg/kg in Hy-Line Grey layers yielded positive effects on eggshell quality characteristics. This was evidenced by an enhancement in the synthesis of glycosaminoglycan and uronic acid within the eggshell glands (ESG).(Li et al., 2018)

3.15 Nutraceuticals effect on production of milk in cows:

Various commercially available probiotics are designed to enhance the quality of milk. These probiotics contain beneficial microorganisms that can positively influence the microbial balance within the digestive system of dairy cows these probiotic includes.*Lactobacillus acidophilus*, *Lactobacillus casei, Lactobacillus plantarum* etc. By applying supplements dairy cows with vitamin E can have neutral or even adverse effects on mastitis incidence, particularly when plasma concentrations of vitamin E at dry-off are high, exceeding 14.5 micro mol/l.(Olchowy et al., 2019)

Other supplements such as copper (Cu), zinc (Zn), vitamin A, iron (Fe), and selenium (Se) also serve as antioxidants and activators of various enzymes, contributing to increased milk productivity(Castillo et al., 2013). These supplements play roles in enhancing the activity of enzymes like Superoxide dismutase, which converts superoxide to hydrogen peroxide, β-Carotene, which prevents the initiation of fatty acid peroxidation chain reactions, Catalase, primarily found in the liver and responsible for converting hydrogen peroxide to water, and

Glutathione peroxidase, an enzyme that also converts hydrogen peroxide to water. By supporting these enzymatic activities and providing antioxidant properties, these supplements contribute to overall milk production enhancement in dairy cows.(Castillo et al., 2013)

3.16 Nutraceuticals effects on Canine and Feline Osteoarthritis:

Given the prevalence of osteoarthritis among pet animals, a diverse range of natural health products is easily accessible for its management. Osteoarthritis (OA) stands as a common musculoskeletal ailment in pets. With no curative treatment available, veterinarians aim to manage its symptoms, primarily focusing on alleviating joint pain and enhancing motor function to enhance the affected animals' quality of life.(Barbeau-Grégoire et al., 2022)

Various categories of diets have been tested for the treatment of osteoarthritis in pets. These include:

Category 1: Omega-3-enriched therapeutic diets (e.g., Green-lipped mussels, Fish oil)

Category 2: Omega-3-based nutraceuticals (e.g., Green-lipped mussels, Fish oil)

Category 3: Collagen-based nutraceuticals (e.g., Collagen, glucosamine hydrochloride, and chondroitin sulfate; Collagen-derived gelatin; NEMO, vopet, Movoflex)

Category 4: Cannabinoid-based nutraceuticals (e.g., Cannabidiol) and other various kind of nutraceuticals diet are tested.

Results indicate that all nutraceuticals are effective against osteoarthritis in pets. However, omega-based nutraceuticals appear to be more effective compared to other categories in managing osteoarthritis symptoms in pet animals.(Barbeau-Grégoire et al., 2022)

3.17 Challenges Faced in Nutraceuticals

3.17.1 Regulatory Uncertainty:

Nutraceuticals often fall into a regulatory gray area between food and pharmaceuticals, complicating classification, approval processes, and compliance with international regulations (Singh & Bhatnagar, 2020).

3.17.2 Scientific Validation:

Establishing robust scientific evidence to support health claims for nutraceuticals requires rigorous clinical trials and research, which can be costly and time-consuming (Giudetti et al., 2021).

3.17.3 Quality Control and Standardization:

Ensuring consistency in quality, potency, and purity across nutraceutical products is challenging due to variations in raw materials and manufacturing processes (Riaz et al., 2020).

3.17.4 Consumer Education and Perception:

Nutraceuticals face consumer skepticism and confusion regarding their benefits, safety, and proper usage, necessitating effective education and transparent labeling (Gahche et al., 2020).

3.17.5 Market Competition and Saturation:

The nutraceutical market is highly competitive, with numerous products claiming various health benefits, making it difficult for new entrants to differentiate themselves (Papoutsis et al., 2020).

3.17.6 Cost Considerations:

Developing and marketing nutraceuticals involves substantial costs related to research, development, regulatory compliance, and marketing, impacting product affordability and accessibility (Freedman & Brochhausen, 2021).

3.17.7 Ethical and Sustainability Issues:

Ensuring ethical sourcing of ingredients and sustainability in production practices are increasingly important but challenging aspects for nutraceutical manufacturers (Hashmi & Khan, 2020).

3.17.8 Health and Safety Concerns:

Despite their natural origins, improper use or dosage of nutraceuticals can lead to adverse effects, emphasizing the need for accurate labeling and safety monitoring (Fugh-Berman, 2020).

3.18 Conclusion:

Summarize the identified mechanisms through which the nutraceutical composition exerts its effects. This could include processes such as ant oxidation, anti-inflammatory activity, modulation of gene expression, or interaction with specific cellular receptors. Discuss the various physiological functions influenced by the nutraceutical composition. This may encompass improvements in metabolic health, immune function, cognitive performance, cardiovascular health, or other relevant areas based on the scope of the study. Evaluate the quality of the nutraceutical composition used in the study, considering factors such as purity, potency, stability, and bioavailability.

Assessing quality ensures the reliability and consistency of the observed effects. Explore the potential of the nutraceutical composition in promoting recovery from specific health conditions or physiological stressors. This could include recovery from exercise-induced fatigue, oxidative stress, injury, or illness. Present the key observations derived from the study, including any statistically significant findings, dose-response relationships, time-course effects, or adverse reactions observed during the intervention period.

Discuss the implications of the study findings for both scientific understanding and practical applications. Consider how the identified mode of action and observed functions of the nutraceutical composition could inform future research directions or therapeutic interventions.

Acknowledge any limitations of the study, such as sample size, study duration, potential confounding variables, or limitations in measurement techniques. Addressing limitations provides context for interpreting the results and suggests areas for improvement in future investigations. Suggest potential avenues for further research to build upon the current findings. This might involve exploring different formulations or dosages of the nutraceutical composition, investigating its effects in different populations or clinical contexts, or elucidating additional aspects of its mode of action.

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Chapter 4: Global Veterinary Chronicles: Confronting Challenges, Embracing Solutions

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Abstract

"Global Veterinary Chronicles: Confronting Challenges, Embracing Solutions," the chapter's topic, examines the myriad problems and inventive fixes that arise in the field of international veterinary medicine. It begins by enumerating significant concerns, including emerging infectious diseases, antibiotic resistance, climate change, limited financial and resource availability, and safe and secure food. Through in-depth examination, it shows how these issues impact the environment, animal welfare, and human health. The chapter's following section explores innovative methods and advancements in veterinary technology, including telemedicine, precision veterinary treatment, and remote monitoring. It is emphasized that addressing complex health concerns that impact the well-being of humans, animals, and the environment requires a solid foundation in the One Health idea. Policy, legislation, education, and capacity building are necessary for the progress of veterinary practices as well as for effective health interventions. Through case studies and success stories, the chapter offers instances of effective tactics and collaboration that have advanced veterinary research, strengthened the battle against zoonotic diseases, and implemented sound regulations. Examples such as the containment of the Ebola virus in West Africa, the battle against rabies in Latin America, and the animal welfare laws enacted in New Zealand serve as examples of the need for concerted efforts and innovative thinking. The overarching theme of the chapter underscores the vital role veterinary care plays in preserving world health, stressing the need for continual innovation, collaboration, and education in order to overcome impending challenges. It concludes with a call to action for adopting the One Health concept, integrating communities, and utilizing state-of-the-art technologies in order to safeguard the health and wellness of both people and animals.

Keywords: Global Veterinary, Challenges, Solutions, Veterinary Diseases, Advanceent in veterinary

4.1 Introduction:

About 1.3 billion people are employed worldwide in the livestock industry, which also directly supports the livelihoods of about 600 million smallholder farmers in developing nations.

Livestock systems make up around 30% of the planet's ice-free terrestrial surface area (Thornton, 2010). As a result, the production of livestock is essential to global agriculture and is highly valued since it provides breeders with a living. The idea that no two breeds or animals are alike forms the basis of all animal breeding programs. Numerous characteristics, including fertility, size, stress tolerance, resilience to disease, grazing habits, and mothering style, vary between and within breeds. Productivity is a mean indicator of production efficiency. The main factors influencing a breeding herd's productivity are the breeding females' reproductive success, the sires' breeding values, the mortality rate, the growth rates of the young animals to maturity, and the amount of milk produced (De Leeuw et al., 1991). According to Huettel et al. (2011), (Huettel et al., 2014)productivity is a significant economic element that affects both biological and financial rewards. Production efficiency is crucial for maintaining increases in animal output over the long term and is a major component in livestock farmers' capacity to make a living. One of the main things that prevents cattle production systems from being as effective as they could be is low productivity (Brumby & Trail, 1986). According to Ozoje et al. (2005) and Lamy et al. (2012), the majority of cattle germplasm in sub-Saharan Africa has excellent adaptation potential but poor productive performance. These papers state that animals from Africa mature later, grow more slowly, have lower mature body weights, and have much lower overall performance efficiency (Ozoje, 2018). Claimed that because native livestock breeds are highly adapted to difficult environmental conditions, their genetic improvement is extremely beneficial. Enhanced adaptation typically comes at the sacrifice of production potentials, even though adaptable features are significant contributors to overall output. Therefore, in order to maximize output, these traits must be compromised. The concept of productive adaptation is a great way to use environmental pressures (diseases and heat) and an animal's productive response to precisely anticipate the relative degree of production potential in unfavorable, hot weather. The way these variables interact determines how productive a particular breed of animals performs. The environment, human health, and animal health all depend on veterinary care. It covers topics including food security, climate change, and antibiotic resistance in addition to newly developing infectious diseases. This chapter examines these issues and presents creative solutions and global success stories (Ozoje, 2018).

4.2 Major Challenges in Global Veterinary Medicine

4.2.1 Emerging Infectious Diseases

The resurgence of ancient diseases or the introduction of new ones in animals should not come as a surprise in this era of increased globalization (Williams et al., 2002). The world's ecosystem is changing quickly due to the presence of pathogenic microorganisms, their vectors, and hosts that can also change quickly. While some of these infections have the potential to seriously harm wild species, in other instances the wild animals may act as reservoirs for pathogens that do not directly cause illness in their natural hosts. Global health is significantly impacted by emerging infectious diseases (EIDs) as COVID-19, avian influenza, and H1N1 (Brown, 2000). A strong veterinary reaction is required because these diseases can spread from animal populations to human populations.

H1N1 Influenza:

This started in pigs and in 2009 spread over the world as a pandemic. caused a large number of deaths and financial losses for the cattle and healthcare industries. emphasized the necessity of enhanced monitoring and quick reaction systems (McArthur, 2019).

Avian Influenza:

First identified in 1997, it mainly affects birds but can sometimes infect people. causes the poultry business to suffer significant financial losses. demands immunization schedules and strict biosecurity protocols(McArthur, 2019).

COVID-19:

It believed to have started in bats, with pangolins acting as possible intermediate hosts. sparked a worldwide pandemic, highlighting the connection between animal and human health. shown how important One Health methods are for controlling zoonotic illnesses (Brown, 1999).

Table 1 Notable Emerging Infectious Diseases and Their Impact

Disease	Year Detected	Primary Host	Human Impact	Economic Impact
H1N1 Influenza	2009	Pigs	Global pandemic, high mortality	Billions in healthcare costs, livestock losses
Avian Influenza	1997	Birds	Sporadic human infections	Poultry industry devastation
COVID-19	2019	Bats, possibly pangolins	Global pandemic, high mortality	Trillions in economic losses

Antimicrobial Resistance

One urgent global health concern is antimicrobial resistance (AMR). One of the main causes of this issue in veterinary care is the overuse and misuse of antibiotics. It can be more difficult to cure diseases in humans when resistant bacteria are present in animals.

Causes of AMR:

The overuse and over prescription of antibiotics in animals. Inadequate regulatory supervision in certain areas. Inadequate procedures for infection control(Iwu & Patrick, 2021).

Consequences of AMR:

Higher rates of morbidity and mortality due to infections with resistance. Increased medical expenses as a result of protracted sickness and more involved therapy. The loss of cattle poses a threat to food security.

Global Initiatives:

The Global Action Plan on AMR by the World Health Organization (WHO). The guidelines for the responsible use of antibiotics set forth by the World Organization for Animal Health. National policies and guidelines in different nations (Brown, 1999; Iwu & Patrick, 2021).

4.3 Climate Change

Climate change has profound effects on animal health through shifts in disease patterns, habitat changes, and extreme weather events (Ozoje, 2018).

1. **Effects on Disease Patterns:**

- Changes in temperature and humidity affect the distribution of vectors like mosquitoes and ticks.
- Increased spread of vector-borne diseases such as Lyme disease and West Nile virus.

2. **Habitat Changes**:

- Loss of natural habitats forces wildlife into closer contact with humans and livestock.
- Increased risk of zoonotic disease transmission.

3. **Extreme Weather Events**:

- Floods, droughts, and storms disrupt veterinary services and animal health.
- Livestock deaths and reduced food security.

Table 2: Climate Change Impacts on Veterinary Medicine

Impact	**Description**	**Example**
Vector-borne diseases	Changes in temperature and humidity affect vectors	Increased spread of ticks and Lyme disease
Habitat loss	Destruction of natural habitats forces wildlife into closer contact with humans and livestock	Increased human-wildlife conflict
Extreme weather	Floods, droughts, and storms disrupt veterinary services and animal health	Livestock deaths, reduced food security

Economic and Resource Constraints

Financial and resource constraints frequently affect veterinary care, particularly in underdeveloped nations. The inability to implement preventive measures, deliver quality treatment, and handle medical emergencies is impacted by this imbalance.

4. **Funding Limitations:**

- Insufficient funding for veterinary infrastructure and services.
- Challenges in maintaining a skilled workforce.

5. **Resource Disparities:**

- Differences between developed and developing regions in access to veterinary care.
- Impact on animal health and productivity.

6. **Solutions:**

- International aid and development programs.
- Public-private partnerships to improve resource allocation (Lamy et al., 2019).

4.4 Food Security and Safety

Veterinary medicine is critical for ensuring food safety and security. Challenges include disease outbreaks, contamination, and the safe use of veterinary pharmaceuticals (Smith & Kelly, 2008).

1. **Disease Outbreaks**:

- Control and prevention of diseases like foot-and-mouth disease and African swine fever.
- Impact on livestock production and food supply.

Foodborne Pathogens:

- Ensuring the safety of food products through rigorous inspection and testing.
- Management of pathogens such as Salmonella, E. coli, and Listeria.

2. **Veterinary Pharmaceuticals**:

- Safe use of drugs in livestock to prevent residues in food products.
- Regulatory frameworks to ensure drug safety (Okocha et al., 2018).

Table 3: Major Foodborne Pathogens and Their Sources

Pathogen	**Primary Source**	**Human Health Impact**
Salmonella	Poultry, eggs	Gastrointestinal illness
E. coli	Beef, leafy greens	Severe diarrhea, kidney failure
Listeria	Dairy products, deli meats	Meningitis, septicemia

4.5 Innovative Solutions and Strategies

Advancements in Veterinary Technology

Recent developments in nanomedicine and worldwide efforts to foster nanotechnology research demonstrate how products based on nanotechnology are transforming both human and animal modern medicine. In order to improve the quality of human life, scientists from a variety of fields, including engineering, material science, food science, biomedical sciences, environmental science, agriculture, energy, and information technology, should stay up to date on nanotechnology and use it appropriately to further their research and development (Zhang et al., 2024). Veterinary medicine is about to undergo tremendous changes as we approach a new era of health-related technologies. Our new capacity to detect, control, and arrange matter at the nanoscale level has been a fundamental factor in these transformations. Since nanotechnology produces minute structures, they might be utilized. Technological advancements are transforming veterinary medicine, improving diagnostics, treatment, and disease management (Dilbaghi et al., 2013).

Telemedicine:

Remote consultations and diagnostics.

Increased accessibility to veterinary care in remote areas.

Precision Veterinary Medicine:

Genetic testing and biomarkers for accurate diagnosis.

Tailored therapies based on individual animal genetics.

Remote Monitoring:

Every year, new illnesses that endanger the health of animals arise in the modern world. Currently, there is a dearth of accurate, reasonably priced diagnostic tests for the early identification of illnesses in livestock raised for food. Biosensing technologies can potentially alleviate these issues by creating novel diagnostic instruments that enable prompt identification of significant health risks in the livestock and agri-food industry (Dilbaghi et al., 2013). Food security is impacted by a multitude of factors that impact food production globally. Food demand is predicted to rise by 70% by 2050, while meat output is predicted to rise by 50%. As a result, agri-food and the livestock industries will be crucial to future growth. Wearable devices for continuous health tracking. Early detection of health issues (Neethirajan et al., 2017).

4.6 Biomedicine or Biomarker:

Veterinary medicine is a rapidly expanding area that has arguably assiduously absorbed innovative advancements emerging from other health domains, including biomedicine.

Veterinary Medicine Enhancing Animal Health and WellBeing

Veterinary practitioners strive to iuzammprove animal welfare as well as human public health outcomes, and they have a great deal of responsibility for the prompt diagnosis, treatment, illness prevention, and nutrition of animals (Blancou, 1990; Council et al., 2005). The diagnosis of veterinary diseases was primarily based on clinical indicators just a few decades ago. A small number of laboratory tests and microbiological cultures were used for confirmation, and radiography and ultrasound imaging were added later. These confirmation diagnoses frequently take several days, and they can call for specialized knowledge, outsourcing, or referral. This not only postpones treatment, but in the case of infectious diseases, diagnosis timing is crucial since the spread of a disease can lead to severe consequences for industry and public health, including large cullings and enormous financial losses (Scott, 2007). Regular, reliable, and early diagnostic tests are being created and implemented into practice more frequently in order to address those welfare and financial concerns. Consequently, biotechnology is currently having a significant impact on the veterinary industry, with prospective uses including animal reproduction and the identification and management of animal illnesses. There are still many obstacles to overcome in the process of identifying biomarkers and translating them for clinical use, even though the value of emerging biotechnologies for veterinary diagnostics is already acknowledged and there are several examples of their effective integration into everyday practice. The area of biotechnology began with the twentieth-century discoveries in microbiology and cellular biology, which led to the creation of vaccines, serum, and antibiotics as well as the selective breeding and cross-breeding of animals and plants. At this point, pathogen identification also became feasible, mostly through the use of microscopy, biochemical testing, and microbial culture techniques. Rapid, precise, and sensitive diagnostic tests are therefore necessary to identify disease pathogens because these approaches can be laborious and call for specialized tools and knowledge(Scott, 2007). The field of biotechnology has significantly broadened recently with the advent of molecular biology, encompassing gene engineering, DNA manipulation, and numerous academic, medical, and agricultural applications (Ko & Abatan, 2008). The human genome's sequencing was a significant accomplishment. This has made possible a new era in biotechnology where large datasets in almost any tissue, cell, or species can be created, in addition to the ongoing characterisation of other species' genomes, thanks to mass spectrometry (MS)-driven "omics" platforms such as proteomics, lipidomics, metabolomics, transcriptomics, epigenomics, and genomics (Manzoni et al., 2018). Large volumes of information regarding the molecular characteristics of biological systems are being revealed by these new technologies, which are constantly developing in capability and becoming more widely available (Manzoni et al., 2018). The assessment of numerous health indices has seen the application of biomarkers, which has been recognized as a growing trend in the animal health sector. It is especially helpful in clinical settings as it is used to diagnose diseases, follow and/or anticipate how a patient will respond to treatment, and assess the toxicity or failure of an organ. The phrase "biomarker" is a shorter form of the term "biological marker," which first appeared in literature in the late 1960s [7]. By the 1990s, it was being used more frequently, albeit in a variety of inconsistent ways [8]. While the term "biomarker" has multiple definitions in human medicine , the BEST glossary's expanded definition of biomarkers and associated terms is the most pertinent and useful in veterinary medicine. "A distinguishing feature that is measured as an indicator of pathogenic processes, normal biological processes, or biological reactions to an exposure or intervention, including therapeutic interventions," according to the definition. According to this definition, biomarkers are categorized as "molecular, histologic, radiographic, or physiologic characteristics." Additionally, it is suggested that biomarkers be divided into seven categories: safety, prognosis, predictive, pharmacodynamic/response, susceptibility/risk, diagnostic, monitoring, and prognostic (Figueroa & Rawls, 1969). A planned search for a biomarker for a particular disease, the accidental discovery of a marker (protein, lipid, metabolite, etc.) that could serve as the target for a new biomarker test, or a comparison to currently used biomarkers in human medicine are some examples of how opportunities to develop new biomarkers can arise. Much of the biomarkers commonly seen in normal biochemistry and hematology profiles have been around for a while and were derived from human medicine (Sarko & Pollack Jr, 2002). This also holds true for a large number of the more recent additions to molecular diagnostics, which will be covered later. The development of new technologies has made it feasible to carry out extensive research aimed at finding novel biomarkers that can be used to diagnose or forecast a particular ailment. As veterinary care enters a new era of precision and high-quality practice, biomarker development is currently on the rise. More recently, there has been a growing tendency in discovering markers for survival and response to treatment, whereas older biomarkers tend to focus more on diagnosing disease processes and organ malfunction. This is crucial when dealing with relatively uncommon diseases or outcomes that present a major medical challenge because there is currently no way to detect these conditions early and they are usually diagnosed after patients have reached the symptomatic stage, which lowers the likelihood of survival and decreases the effectiveness of treatment (Osei et al., 2021). This holds true not only for clinical conditions but also for other biological processes that are crucial to animal husbandry, management, and reproduction—all of which promote public health and animal welfare. Consequently, we think it's critical to look at the biomarker invention process. The most successful biomarker research process, according to the review by Myers et al. (Sarko & Pollack Jr, 2002), involves carefully defining the target population and clinical utility first, then choosing and validating the potential biomarker(s) and related analytical method(s) by iteratively going back and forth through the process. Particularly in the veterinary area, new diagnostic biomarkers are being developed for the early discovery of disease, when it may be easier to treat or reversible, and susceptibility/risk biomarkers are being produced to identify animals with enhanced resistance or susceptibility to disease. (Osei et al., 2021). Blood and bodily fluids like urine, saliva, and endometrial secretions are most frequently used to create these tests. Molecular markers inside individual cells are also becoming possible as technology advances. Somatic cells leak distinct chemicals into extracellular tissue or bodily fluids in response to specific biological changes or diseases. As a result, the samples can be used to detect variations in the substance of interest's concentration. Since blood plasma travels throughout the entire body and is easily extracted from the patient, it tends to contain transudates from practically every type of tissue. These factors make it usual practice to use patient samples that are simple and routine to obtain, like serum and blood plasma, for diagnostic purposes . The more well-established science of enzymatic testing is being replaced by emerging technologies such as nanoscience and "omics" approaches, which include multivariate large-scale analysis at the level of DNA, miRNA, RNA, proteins, lipids, and other metabolites, in the search for new biomarkers and the validation of their predictive value. These methods identify the most representative marker for the condition by taking a snapshot of the cells, tissues, or body fluids as they are at that moment (Ceciliani et al., 2014).

Table 4: Benefits of Precision Veterinary Medicine

Benefit	**Description**
Improved Diagnostics	Genetic testing and biomarkers for accurate diagnosis
Personalized Treatment	Tailored therapies based on individual animal genetics
Enhanced Monitoring	Wearable devices for continuous health tracking

One Health Approach

1. The One Health concept places emphasis on the interdependence of environmental, animal, and human health. Taking a holistic approach is crucial when dealing with difficult health issues (Clifford & Coppolillo, 2009) It is imperative that the One Health strategy be put into practice as soon as possible to combat zoonotic illnesses. Understanding how the environment, animal health, and human health are intertwined will help us prevent and address new risks from infectious diseases. The significance of conducting research on zoonotic illnesses is discussed in this review paper, particularly in Indonesia where there is currently a dearth of such studies. The National Zoonosis Committee and the One Health Coordinating Unit were established by the Indonesian government as part of its One Health implementation efforts, albeit the process has not gone well. Critical parts of the community's implementation are the focus of the urgency and difficulty. The fact that zoonotic disease outbreaks have occurred multiple times in Indonesia, the country's high rate of environmental deterioration, and the antimicrobial(Clifford & Coppolillo, 2009).

   Concept:

   - Integration of efforts across human, veterinary, and environmental health sectors.
   - Collaborative approaches to disease prevention and control.

2. **Case Studies**:

   - Rabies control in Latin America through coordinated vaccination campaigns.
   - Successful reduction of zoonotic disease transmission.

3. **Benefits**:

   - Improved disease surveillance and response.
   - Enhanced public health outcomes.

Veterinary Medicine Enhancing Animal Health and WellBeing

Case Study : Control of Rabies in Latin America Through well-coordinated dog vaccination campaigns and public health education initiatives, rabies transmission was successfully reduced throughout Latin America as part of a One Health project.

1. **Policy and Legislation**

- Legislation and policies that are effective are essential for developing veterinary medicine and tackling issues with global health.

2. **International Cooperation**:

- The role of organizations like the OIE and WHO in setting standards and guidelines.
- Examples of successful international collaborations.

3. **Regulatory Frameworks**:
   - Policies for the safe use of veterinary pharmaceuticals.
   - Standards for animal welfare and disease control.
4. **Impact**:
   - Improved animal health and welfare.
   - Enhanced food safety and public health.

Table 5: Key Veterinary Policies and Their Impact

Policy	Region	Impact
EU Animal Health Law	European Union	Strengthened disease prevention and control
USA Veterinary Feed Directive	USA	Reduced antibiotic use in livestock
OIE Standards	Global	Improved international animal health practices

Education and Capacity Building

To solve the worldwide scarcity of veterinary specialists, veterinary medicine needs to be strengthened through education and training.

1. **Training Programs**:
   - Initiatives like Veteducation Africa to enhance skills and knowledge.
   - Online courses and resources for continuing education.
2. **Capacity Building**:

   Attempts to alleviate the scarcity of veterinarian specialists. The significance of ongoing professional growth.

3. **Outcomes**:

- Enhanced veterinary services.
- Improved animal health and welfare.

Table 6: Veterinary Education Initiatives

Initiative	Description	Outcome
VetEd Africa	Training programs for African veterinarians	Enhanced skills and knowledge
Global Veterinary Education Project	Online courses and resources for veterinarians worldwide	Increased accessibility to education

Community Engagement and Awareness

Advocating for animal health issues and involving communities are essential components of effective remedies.

1. **Public Health Campaigns**:

- Education initiatives to promote understanding of animal health.
- Community-led programs for disease prevention and control.

2. **Partnerships**:

- Collaborations with local communities and stakeholders.
- Building trust and ensuring sustainability of veterinary programs.

3. **Success Stories**:

- Community-led animal health programs in India.
- Effective reduction of livestock diseases through participatory training.

Case Study: Indian Community-Led Initiatives for Animal Health Using local resource mobilization and participatory training, community-led projects have significantly decreased the incidence of livestock diseases in India.

4.7 Case Studies and Success Stories

Combating Zoonotic Diseases

The importance of coordinated efforts and creative approaches is demonstrated by successful interventions in the fight against zoonotic diseases.

1. **Ebola Virus Control in West Africa**:

- Surveillance, community engagement, and rapid response measures.
- Effective containment of the outbreak.

2. **Rabies Eradication**:

- Coordinated vaccination campaigns in dogs.
- Public health education programs.

3. **Brucellosis Control**:

- Vaccination and testing programs in livestock.
- Improved public health outcomes.

Case Study: Control of the Ebola Virus in West Africa Rapid response techniques, community involvement, and surveillance all contributed to the containment of the Ebola virus outbreak in West Africa.

Innovative Research and Development

Breakthroughs in veterinary research are paving the way for new treatments and preventive measures.

1. **CRISPR Gene Editing**:

- Precise genetic modifications for disease resistance in livestock.
- Potential to enhance productivity and reduce disease burden.

2. **mRNA Vaccines**:

- Novel vaccine technology for the prevention of viral infections.
- Applications in both human and animal health.

3. **Artificial Intelligence**:

- AI-driven diagnostic tools for early detection of diseases.
- Improved accuracy and efficiency in veterinary care.

Table 7: Recent Advances in Veterinary Medicine

Breakthrough	**Description**	**Application**
CRISPR Gene Editing	Precise genetic modifications	Disease resistance in livestock
mRNA Vaccines	Novel vaccine technology	Prevention of viral infections

Breakthrough	Description	Application
Artificial Intelligence	AI-driven diagnostic tools	Early detection of diseases

Effective Policy Implementation

1. Case studies of successful policy implementation show how standards and regulations can significantly improve the health and welfare of animals.

   Animal Welfare Reforms in New Zealand:

- Comprehensive animal welfare legislation.
- Significant advancements in the treatment and care of livestock and companion animals.

2. **Antimicrobial Stewardship Programs**:

- Policies to reduce antibiotic use in livestock.
- Improved management of antimicrobial resistance.

3. **Disease Surveillance Systems**:

- Implementation of robust surveillance systems for early detection of outbreaks.
- Enhanced response capabilities.

Case Study: Reforms in Animal Welfare in New Zealand. The extensive animal welfare laws in New Zealand have resulted in notable progress in the handling and maintenance of companion and livestock animals (Ceciliani et al., 2014).

4.8 Conclusion

Although veterinary medicine has many obstacles in the world, great strides are being made in this area thanks to innovation, teamwork, and education. Successful veterinarian offices must incorporate the One Health concept, cutting-edge technologies, sensible policies, and community involvement. Maintaining the health and welfare of both humans and animals will need ongoing efforts in these areas, as well as tackling upcoming concerns.

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Chapter 5: Veterinary Medicine Promoting Optimal Animal Health and Welfare

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5.1 Introduction

Veterinary medicine is a branch of science that deals with the prevention, diagnosis, and treatment of diseases, disorders, and injuries in animals (Ettinger, Feldman et al. 2016, Jones and Koolmees 2022). The goal of veterinary medicine is to promote animal health and support the human-animal bond (Adams, Conlon et al. 2004). Veterinarians work to ensure the well-being of all animal species, from pets like dogs and cats, to farm animals like cattle and poultry, to wildlife and zoo animals (Tomlinson, Black et al. 2021). They play an important role in public health by detecting and controlling zoonotic diseases that can spread between animals and humans (King, Control et al. 2006). Veterinarians also contribute significantly to economy through their work in agriculture and biomedical research (Council, Earth et al. 2003). A veterinarian's responsibilities involve different aspects of animal care including routine vaccinations and wellness exams, surgical procedures, dentistry, diagnostic imaging and lab testing, therapeutic treatments, euthanasia and end-of-life care, and client education (Christiansen, Kristensen et al. 2015). They work collaboratively with animal owners and provide guidance on nutrition, parasite prevention, behavioral issues, and overall care best practices (Overgaauw, Vinke et al. 2020). Veterinarians operate in various settings including private companion animal practices, shelters and animal welfare clinics, zoos and wildlife parks, farms and dairies, biomedical research facilities, the military, government agencies, and academia for teaching and research (Hernandez, Llonch et al. 2022). Some specialize in areas like oncology, dermatology, ophthalmology, dentistry, or exotic/wildlife medicine (Hemming 2003, Plumb 2018). The growth of veterinary medicine in both knowledge and scope of services allowed has greatly improved animal welfare and longevity over the years (Walker, Diez-Leon et al. 2014). Looking ahead, emerging technologies and continual research promise further advancements in disease diagnosis and novel treatments to enhance all animal lives and health (Neethirajan 2017).

5.1.1 Goal of veterinary care - enhancing animal health and well-being

The overarching goal of veterinary medicine is to promote and maintain animal health, prevent illness and injury, alleviate suffering, and improve overall well-being and quality of life (Pinillos 2018). Achieving this requires veterinarians to utilize their scientific knowledge and clinical skills to diagnose issues accurately, develop treatment plans, perform surgeries and procedures skillfully, and educate clients (Hodgson and Pelzer 2017). While veterinarians aim to cure animals, when possible, their focus is broader than just curing disease (Huth, Weich et al. 2019). Preventative care like vaccines, parasite control, dental care, nutrition advice, and behavioral guidance helps animals avoid medical problems in the first place (Angliss, Hedge et al. 2021). Early detection of health changes through diagnostic testing allows conditions to be managed proactively before they worsen (Schaer, Gaschen et al. 2022). Pain management is also a priority, as unrelieved pain and distress can seriously compromise welfare (Couto and Cates 2019). Veterinarians help ensure animals are kept comfortable through their illnesses, injuries, and at end of life (Bishop, Cooney et al. 2016). They may recommend interventions like acupuncture, hydrotherapy, or medications to minimize discomfort. Beyond physical health, veterinarians consider an animal's psychological wellness (Ramey and Rollin 2008, Scanlan 2024). They address issues like anxiety, fear, stress, and abnormal behaviors that impact quality of life. Ensuring animals live in healthy, stimulating environments per their species' needs supports overall well-being (Brando and Buchanan-Smith 2018). The human-animal bond is another important consideration. By keeping pets happy and prolonging their lives through geriatric care, veterinarians enhance bonding experiences for owners (Hosey and Melfi 2014). They provide grieving support during euthanasia and end of life as well. Ultimately, the goal of all veterinary efforts is to help animals experience the highest attainable state of physical, emotional and environmental wellness through prevention, diagnosis, treatment and education (Griffin 2021). The primary objective of veterinary medicine is to improve the health and welfare of animals by using a compassionate, holistic approach. This entails applying clinical expertise and scientific understanding to correctly identify health problems, create efficient treatment programmes, and expertly carry out operations or procedures while giving pain management a priority (Willis, Monroe et al. 2007). In order to avert problems or maintain the comfort of their patients, veterinarians also prioritise quality of life, early identification, client education, and preventative care (Florian, Skurková et al. 2023). In order to fully comprehend each patient's distinct demands, they collaborate with owners, advocates, and other specialists while taking into account the entirety of the animal's surroundings (Julius, Beetz et al. 2012, Schaer, Gaschen et al. 2022).

5.2 Historical Development of Veterinary Medicine

5.2.1 Early History of Animal Medicine

The earliest evidence of rudimentary veterinary practices can be traced back over 7,000 years to agricultural civilizations that relied heavily on animals (Köhler-Rollefson 2023). In ancient Mesopotamia around 5000 BC, livestock provided critical resources like wool, meat and labor (Goulder 2018). Illnesses affecting valued cattle, sheep and goats were documented on clay tablets to identify patterns and pass knowledge between herders (Scurlock and Andersen 2010, Attia 2018). Similarly, ancient Egyptian priests around 3000 BC used medicinal herbs and wound care techniques detailed on papyrus scrolls to treat important farm animals like donkeys and oxen used for plowing fields and transporting goods (Scurlock and Andersen 2010, Avalos 2019). Moving east, India witnessed the publication of what is considered the first veterinary textbook in 1000 BC called the Shalihotra, containing advice on horse health, anatomy and natural remedies influenced by Ayurvedic medicine (Giri and Hedayetullah 2020, Prasad, Prasad et al. 2023). During this same period, the great Greek philosophers Aristotle and Plato authored seminal works containing observations on comparative animal anatomy, physiology, diseases and husbandry practices (Morange 2021). Their contributions advanced zoological understanding. By 100 BC in the Roman Empire, veterinary treatises referenced various livestock and poultry ailments along with suggested animal husbandry standards, wound closure methods and even dentistry performed on prized stallions kept at large estates (Mitra, Bhattacharya et al. 2019, Jones and Koolmees 2022). As mounted cavalry grew in importance during the European Middle Ages, farriers emerged as skilled specialists focused on horsemanship and farriery, especially meticulous hoof care critical for warhorses (Beale 2021). Over millennia, informal folk remedies evolved slowly through oral tradition passed down by farmers, herders and animal attendants worldwide until more formal practices could develop (Jones and Koolmees 2022). In addition to written knowledge contained in ancient texts, early civilizations developed rudimentary medical tools and surgical techniques still utilized by modern veterinarians. Clay tablets depict Mesopotamian healers performing dentistry on livestock using stone picks and forceps (Upson-Saia 2023). Egyptian papyrus scrolls illustrate wound closure via suturing with flax thread and needles forged from bone or metal (Giri and Hedayetullah 2020). The Romans manufactured specialized scalpels, lancets, probes and speculums from bronze and iron to examine orifices and conduct minor procedures (Bliquez 2014). Farriers crafted a diverse array of hoof knives, files, nippers and rasps to shape horse hooves and apply therapeutic shoeing (O'Grady and Ovnicek 2020).

5.2.2 Establishment of Modern Veterinary Schools and Practices

It was not until the 18th century that the first formal institutions of veterinary education were established to systematically train and certify veterinarians (Edition 2018). In 1762, the world's first veterinary school was founded in Lyon, France with a standardized curriculum focused on equine medicine and husbandry (Edition 2018). Throughout the 19th century, visionary pioneers in both Europe and the United States worked to professionalize clinical standards and licensing requirements for veterinarians through the establishment of dedicated colleges and regulatory boards. A prime example was the historic opening of America's first veterinary program at the University of Pennsylvania in 1863, laying the foundation for modern veterinary medicine in North America (Smith 2010, Jones and Koolmees 2022). Concurrent advancements in bacteriology, anesthesia and surgical asepsis in the late 1800s allowed veterinary practice to move from an empirical field into a science-based profession (Lees, Bäumer et al. 2022). These transformative developments were further enabled by breakthroughs throughout the 20th century, including expanded demand driven by post-World War II economic growth and increased globalization of livestock production and meat consumption following the war (Reardon, Echeverria et al. 2019). Together, these momentous occurrences allowed veterinary medicine to evolve from its artisanal roots into the standardized, evidence-based field that ensures public and planetary health today through specialized training facilities worldwide (Bresalier, Cassidy et al. 2021).

As the professionalization of veterinary medicine progressed, early curriculums predominantly focused on large animal species integral to agriculture and transportation (Dolby 2022). Over time, small animal medicine became emphasized as the human-companion bond strengthened (Nelson and Couto 2019). International collaboration also intensified across agencies like the OIE and FAO to address global challenges at the human-animal-environment interface including emerging diseases (Berrian, Mekuria et al. 2024). More recently, innovative models have aimed to increase access to care through community clinics, mobile services, and public-private alliances particularly for remote regions with shortages (Gizaw, Astale et al. 2022).

5.2.3 Evolution of Veterinary Specializations

The late 19th and early 20th centuries marked the beginning of specialization within veterinary medicine, with focused areas of practice emerging to address the individualized healthcare needs of major agricultural species (Bonnaud and Fortané 2021). Equine medicine and farm animal clinicians became some of the earliest specialists. Throughout the mid-20th century, companion animal medicine expanded rapidly in tandem with rising pet ownership trends (Bresalier, Cassidy et al. 2021). By the 1950s, veterinarians began specializing in small animal fields like canine and feline practice (Gardiner 2021). Today, veterinary specialties have proliferated to include internal medicine disciplines like cardiology, oncology and ophthalmology as well as areas like exotics, lab animal science and aquatic animal medicine (Sharkey, Radin et al. 2020, Datt, Rai et al. 2021). Currently, research specializations are delving into gene therapy, stem cell regeneration, cutting-edge vaccine development, epigenetics influencing disease susceptibility, and other pioneering fields with potential to transform prevention and treatment of both human and animal ailments. This continuum of deeper specialization has allowed veterinarians to provide highly advanced, targeted healthcare that improves quality and length of lives across all species.

5.3 Preventative Healthcare

Preventive care is crucial for animal health and disease prevention, with 1/3 of all dog and cat veterinary appointments in the UK focusing on it. However, owner education on preventive care topics and infectious disease risks may be limited, misunderstood, or perceived as less valuable by clients (Force 2011, Day, Horzinek et al. 2016). Research on the quality and quantity of preventive care discussions between veterinary staff and clientele in clinics is sparse. In Canada, educational discussions on preventive care formed a minor component of wellness visits, and the content varied depending on the pet's age and vaccination status. There may be a disconnect between what veterinary teams perceive as valued by clients and what clients truly regard as important or essential (Robinson, Brennan et al. 2016). Research on pet owner knowledge, attitudes, and practices (KAPs) related to preventive care is needed to identify education gaps and target areas for improved communication, ultimately reducing infectious disease risks and improving companion animal health (Belshaw, Robinson et al.

2018). The Partnership for Healthy Pets (PHP) was established in 2011 by the American Animal Health Association (AAHA), American Veterinary Medical Association (AVMA), and other stakeholders to provide resources for communicating the importance of preventive care to pet owners. The concerns of pet owners towards their pet's preventive care becoming gradually less. A contributing factor to these changes was thought to be increasing pet owner reliance on non-veterinarian-approved internet-based information sources instead of directly from veterinary teams.

In Canada, pet infectious disease exposure and risk related to pathogens, such as tick-borne and Leptospira spp., have changed dramatically in recent years, particularly in regions of the Maritimes and Eastern Canada. Individual pet-specific risks, such as animal travel or involvement in group animal settings, may increase a pet's infectious disease risks further through exposure to regional pathogens during travel or an increased number of potentially infectious animals and contaminated fomites in a group setting (Evason, McGrath et al. 2021). Zoonotic pathogens affecting companion animals have direct public health impacts, especially for household members at risk due to age, pregnancy, or immunocompromising. The veterinary team plays a central role in influencing KAPs of pet owners with likely direct impacts on pet and owner health. There is a critical need to determine clients' preventive care practices and investigate possible modifiable factors influencing uptake of veterinary recommendations. By assessing client knowledge of preventable infectious diseases and preventive care options, veterinary health professionals can target knowledge gaps or correct misinformation by providing credible sources of information regarding preventive care for their pets (Evason, McGrath et al. 2021).

Preventative healthcare has been a component of animal medicine since antiquity. Ancient Chinese texts first documented variolation techniques using materials from cowpox or smallpox lesions to inoculate against potential future infection (Gurushankara 2021, Jones and Koolmees 2022). Such practices were also noted in early African and Indian cultures (Boylston 2012). In the late 18th century, Edward Jenner demonstrated the protective effects of vaccinating humans with cowpox, establishing the scientific principle behind immunization (Khardori, 2022).

5.3.1 Monitoring wellbeing

Improving animal health and welfare in livestock systems depends on reliable proxies for assessment and monitoring. Animal health goes beyond the mere absence of sickness since it also attempts to safeguard the welfare and dignity of animals. At the farm level, objective evaluations are required to confirm the efficacy of the measures. Indicators of animal health and wellbeing are combined with those based on resources or management in modern methodologies. The European Union-funded Welfare Quality® project created procedures and methodologies for evaluating the welfare of sheep, goats, pigs, chickens, and cattle. These extensive protocols comprise evaluations of the animal as well as its surroundings and husbandry. Nonetheless, they necessitate on-farm visits and labor-intensive manual data collecting. Data-based techniques that offer important insights into animal health and welfare include the MulTiViS approach and the Animal Health Barometer. As electronic records and large databases become more widely available, the potential for these kinds of data-based solutions is growing quickly. Precision Livestock Farming (PLF) technologies' "smart farming" data, which may be utilised to track the wellbeing and health of animals, is likewise covered by this (Thomann, Würbel et al. 2023).

5.3.2 Vaccinations

Veterinarians provide vaccinations as part of preventative veterinary care during wellness checkups. They take into account variables like lifestyle, geographic location, and individual risk factors while creating a personalised regimen for every pet based on industry standards. Puppies and kittens receive their first vaccination series as early as 6–8 weeks of age, and mothers transfer protective antibodies to them throughout pregnancy and colostrum (Virhia, Gilmour et al. 2023). In some situations, active immunisation lessens the amount of pathogen shedding and can shield dogs and cats against the serious effects of infection with a range of different pathogens. Live microorganisms that have been attenuated, inactivated, or parts of these organisms are contained in vaccines. They might also contain adjuvants and preservatives (Sykes 2021). A course of booster doses spaced three to four weeks apart provides active immunity. Basic immunisations are essential for both public health and legal compliance because they guard against dangerous, communicable diseases like rabies and distemper (Pettini, Pastore et al. 2021). Throughout an animal's life, vaccinations against adenovirus, parvovirus, and distemper are usually required, with booster intervals ranging from yearly to every one to three years (Harrus 2020, Sykes 2021). Veterinary associations examine immunity duration on a regular basis prior to prescribing booster doses. Non-core vaccinations target particular risks such as kennel cough in boarded or housed pets or leptospirosis in animals exposed to standing water or wildlife (Barker and Costa 2023). In order to achieve community-wide and personal preventative healthcare goals, full protection against vaccine-preventable diseases, both core and non-core, requires collaboration between clients and veterinarians.

5.3.3 Parasite prevention

Internal (like worms) and external (like fleas and ticks) parasites deplete the host's nutrition and can lead to illness (Kustritz 2022). The Veterinarian's Oath states that veterinarians are acknowledged as public health experts. In keeping with historical patterns and precedents, their function in public education and communication is changing. Since the 1990s, the role of veterinarians in the discourse surrounding parasitic zoonoses has changed. Although a One Health-centered strategy has been articulated, it is nevertheless interesting to discuss how veterinarians specifically fit into these discussions (Cagle, McGrew et al. 2022). Veterinarians are committed to reducing parasite illnesses in animals through the use of pharmaceutical treatments and environmental education. During inspections, they look for parasites like fleas, ticks, and mange mites on pets, and they examine faeces under a microscope to look for intestinal worms (Cagle, McGrew et al. 2022). 75% of newly discovered infectious diseases are zoonoses, which are illnesses or infections spread from vertebrate animals to humans. Globally, the effects of these diseases may be catastrophic for both human and animal health. Protozoa and helminths are examples of zoonotic parasites that can alter their ecological niches and epidemiological patterns in response to dynamic biotic and abiotic factors. Their spread and expansion will be greatly impacted by climate change and globalisation. A well-known illustration of the One Health concept is the spread of parasitic zoonoses, which affects and is influenced by humans, animals, and the environment. Certain zoonotic parasites can infect humans through a variety of routes, including consuming undercooked meat or coming into contact with sporulated oocysts in cat faeces. After the host consumes an infectious pathogen or comes into contact with an arthropod vector carrying a zoonotic pathogen, other diseases may arise (Cagle, McGrew et al. 2022). They advise either seasonal or year-round parasite prevention programmes depending on the lifestyle and environmental exposures of each pet. Age, fertility, and the existence of wormplus all affect the deworming process (Baiyasi, Juárez et al. 2022). To stop the spread of heartworm, roundworm, and hookworm, preventative measures are frequently given. Pets kept indoors are nevertheless vulnerable to parasite illnesses. Regular showering and grooming are examples of good hygiene that contribute to home protection. In order to give personalised prophylactic advice, veterinarian clinics stay up to date on regional trends in parasites. They employ a variety of tactics to shield animals from parasitic and viral illnesses. In order to maintain general health and lower the chance of zoonotic parasite transmission, they employ a variety of tactics to shield animals from infectious and parasitic illnesses (Noack, Harrington et al. 2021, Kustritz 2022).

5.3.4 Nutrition counseling

In companion animals, proper nutrition is essential for their health, lifespan, and defence against disease. It expedites healing, shortens the time needed for release, lowers the risk of infection, raises tolerance for invasive procedures, and improves hospital outcomes. Nutrition plays a critical role in patient care since it prevents and manages diseases linked to diet, regardless of a patient's stage of life. Studies indicate that providing veterinary patients with nutritional support improves their hospital outcomes, lowers their susceptibility to infections, and speeds up their recuperation (Lumbis and De Scally 2020).

By offering dietary and feeding recommendations, veterinarians play a critical role in preventive pet care. During wellness visits, they evaluate the general health, food history, way of life, and underlying medical conditions of each animal (Wortinger and Burns 2024). To make sure the food is appropriate for the pet's needs, they evaluate the meal's quality and amount. They can offer nutritional guidance to growing pets according to their maturation stage and can help elderly and weight-conscious pets. It may be suggested to follow a therapeutic diet to manage an ailment. Veterinarians can offer guidance on feeding schedules and practices, emphasising the need of maintaining a healthy weight and minimising dental risks and obesity. In order to lower dental risks and obesity, they also go over snacking, sharing food at the table, and chews and treats. In general, veterinary nutritionists assist pet owners in maintaining the health of their animals by offering age- and balance-appropriate. In general, veterinary nutritionists assist pet owners in maintaining the health of their animals by offering age- and balanced-appropriate foods as well as responsible feeding techniques that are specific to each species (Wortinger and Burns 2024).

5.3.5 Behavioral modification

Every patient brought to the veterinary hospital must have their clinical status evaluated by the veterinarian. The veterinarian must identify whether the reason of a behaviour issue is behavioural or medical. Veterinarians should generally perform a basic blood analysis, as well as a physical and neurological assessment, on any pet that presents with behavioural problems. Animal behavioural disorders are emotional conditions unrelated to training. Pets who are unruly or do not recognise or obey cues or directions are considered to have training issues. In behaviour, an animal trainer's job is to mentor and instruct animals and their owners in fundamental etiquette and training. In order to prevent major problems before they worsen, veterinarians collaborate with owners to evaluate and treat the behaviour and temperament of their animals. They provide specific treatments, including as desensitisation methods, environment adjustments, and positive reinforcement, for ailments like accidents during housetraining, separation anxiety, or excessive barking or jumping (Martin and Martin 2023). Additionally, they recognise behaviours that point to illnesses that require more attention, such discomfort or cognitive dysfunction, and they offer guidance on how to handle, play, exercise, and enrichment methods for the species (Brando and Norman 2023). For more specialised modification, veterinarians can recommend private trainers for animals exhibiting scared, aggressive, or intolerant behaviours (Martin and Martin 2023). In general, treating behavioural problems in animals through training and habitat adjustment benefits their mental and preventive health equally to that of their physical health. Pet owners and veterinarians working together to improve pets' quality of life (Martelli and Krishnasamy 2023).

5.3.6 Spaying and neutering

Veterinarians support sterilisation surgery as a crucial population control and animal welfare measure to avoid undesired litters. Veterinarians address the advantages of spaying and neutering young pets during wellness checkups. Veterinarians neuter men in order to reduce the incidence of testicular cancer and prostate problems in later life. Spaying females breaks heat cycles and undesired reproduction while preventing uterine infections and breast cancer (Imogen 2021, Furthner, Fabian et al. 2023). Additionally, fixing pets reduces their inclination to roam and mark and makes their behaviour more consistent. Veterinarians evaluate joint growth plates in large breeds and adhere to evidence-based guidelines about the best ages for surgery (Martin 2023). Early sterilisation prevents unintended pregnancies and health problems in females connected to heat (Furthner, Fabian et al. 2023). It gives an owner's preventative plan more structure and aids in the prevention of pet overpopulation. Veterinarian advice following surgery helps patients heal while educating owners about how sterilisation affects their pets' natural behaviours. All things considered, this proactive strategy significantly enhances community pet welfare and lessens the workload for rescues and shelters. Veterinary wellness recommendations and spaying/neutering go hand in hand (Martin 2023).

5.4 Diagnostics

Diagnostics is a crucial process in both human and veterinary medicine, but the sociology and social science of diagnosis have not received much attention. This is especially valid for animal diagnoses. But in the social sciences, especially in "more-than-human" studies and the Sociology of Science and Technology, the profile of veterinary diagnostics, its evolving professional and social context, and its relationship to emerging technologies have all increased recently. The ability to diagnose accurately supports the knowledge and scientific aptitude of veterinarians and other paraprofessionals employed in the animal care sector (Buller, Adam et al. 2020). While baseline information is provided by physical examinations and the gathering of medical histories, objective evidence supporting clinical findings is provided by diagnostic testing. Among the many diagnostic techniques used by veterinarians are simple laboratory tests on blood and urine, microbiological cultures, faecal examinations, radiography, ultrasound, endoscopy, biopsy, and, where necessary, more sophisticated techniques like CT, MRI, and PET scans (Mattoon, Sellon et al. 2020, Verocai, Chaudhry et al. 2020). Veterinarians adopt an integrated strategy, using numerous diagnostic instruments customised for each patient, because no single test can provide all the answers. In order to maximise medical outcomes, this thorough diagnostic evaluation helps identify underlying diseases and abnormalities, supports the creation of treatment programmes, and helps identify any new problems sooner. The foundation of evidence-based veterinary practice is diagnostics, which enables practitioners to provide the best possible medical care (Hendrix and Robinson 2022).

5.4.1 Physical exams

Infectious disease diagnostic tests fall into two general categories: direct and indirect. Every organisation that offers medical care ought to make an effort to keep a minimal amount of laboratory supplies and equipment for diagnostic testing in-house. Based on their resource requirements, the knowledge and expertise needed to conduct the tests and interpret the results, and their viability for being conducted within the resource constraints of the majority of animal shelters, common diagnostic tests have been divided into core, primary, secondary, and diagnostic laboratory tests (DiGangi 2021). Any veterinarian diagnostic evaluation begins with a comprehensive physical examination. An animal's general disposition, mentation, bodily condition, coat/skin quality, vital signs, secondary sex traits, mobility, orthopaedic difficulties, oral cavity, and more are all carefully observed by the veterinarian during a hands-on assessment (DiGangi 2021). Palpation is a useful tool for examining internal organs and structures for anomalies, discomfort, oedema, or lumps. Any asymmetry, peculiar smells, or discharge are noted by veterinarians. Auscultation is also performed on heart rate and respiration. While thorough tests identify hidden issues, focused physical exams guided by presenting indicators help with diagnosis. The breadth of wellness checks varies from focused exams for certain concerns (Englar and Dial 2022). Physical examinations are extremely important for detection, surveillance, and response monitoring because of the ability to discover small abnormalities through observation and touch (Balajee, Salyer et al. 2021). Veterinarians can better prioritise potential issues by getting a thorough history from owners in addition to the physical examination. When taken as a whole, these diagnostic processes direct auxiliary testing strategies to obtain conclusive diagnoses quickly and efficiently. Thus, physical examinations function as nonverbal communication channels with patients and as sensitive screening instruments (Hammersla 2020).

5.4.2 Laboratory tests

Diagnostic laboratory testing is an increasingly crowded arena within veterinary medicine. Veterinarians can objectively assess a variety of an animal's health and organ function parameters using blood, urine, and other sample types in clinical pathology labs. Complete blood counts, coagulation panels, thyroid tests, cytologies, microbiological cultures, and serum biochemistry profiles are examples of common testing (Lustgarten, Zehnder et al. 2020). The goal of a complete blood count is to identify anaemia, infection, inflammation, and problems with clotting by assessing red blood cells, white blood cells, platelets, and other produced elements (Walton and Lawson 2021, Tvedten 2022). Biochemistry profiles look at waste, nutrition, enzyme, and electrolyte levels; among other things, they reveal information on liver and kidney function (Loi, Pilo et al. 2021). A urinalysis finds variations in the content, composition, sediment, and crystals of the urine, which might point to illnesses such kidney disease or bladder infections (Yadav, Ahmed et al. 2020). Blood gas analysis, medication levels, tumour indicators, heart markers, and other specialised testing are available (Gray 2023). Veterinarians can select efficacious antibiotics by using microbiology culture and sensitivity testing to determine the causative organisms for infected wounds, tissues, and bodily fluids. Cancer cells can be found in fluid and mass sample cytologies (Sharkey, Radin et al. 2020). Antibiotic sensitivity is measured and samples are cultured to identify bacterial or fungal infections. This influences the choice of the best antimicrobial treatment (Allerton and Nuttall 2021).

A high white blood cell count suggests inflammation or infection that is ongoing. Specific disorders can be identified by abnormal amounts of parameters such as neutrophils, lymphocytes, monocytes, or eosinophils. To rule out clotting diseases, platelet levels are also measured (Walton and Lawson 2021, Tvedten 2022). Cytologies aid in the detection of abnormalities in cells (Sharkey, Radin et al. 2020). Fluid analysis can identify tumours, infections, or inflammation in organs (Vap and Sprague 2020). Veterinary labs, both in-house and commercial, can run customised panels and serial monitoring affordably with automation and low volume needs. More precise diagnosis and treatment of complicated medical diseases are made possible by this objective data (Englar and Dial 2022).

5.4.3 Imaging modalities

Traditionally, veterinary anatomy relied on meticulous dissections to provide anatomical representations. However, with the advent of contemporary imaging techniques, this has changed, and now living animals may be quickly and non-invasively visualised for therapeutic and research purposes. The number of available locations and annual studies utilising computed tomography, ultrasonography, fluoroscopy, and magnetic resonance imaging in veterinary medicine is increasing (Yitbarek and Dagnaw 2022, Becker and Garibotto 2023).

Radiography is quick and affordable, but has limitations in visualizing soft tissues. Digital radiography provides higher resolution images than traditional films. Contrast studies use barium, iodine or gastrografin to highlight organs for better visibility. Radiography (x-rays) provide invaluable views of bone and tooth structures without surgery. This is a first-line method for trauma cases and orthopedic evaluation. Contrast studies may outline organs. This kind of data offers fresh research opportunities as well as new challenges. These imaging modalities are mostly found in the field of specialty veterinary medicine, which is becoming a more and more well-liked source of care. These specialised centres offer excellent opportunities for future collaboration as well as advanced research and training (Yitbarek and Dagnaw 2022).

Ultrasound is widely used due to its mobility and lack of radiation. Its images in real-time and can guide needle aspirations. Color Doppler mode detects blood flow patterns. Echocardiograms evaluate heart structures and function. Endocavity transducers improve cranial and rectal exams. Ultrasound uses high frequency soundwaves reflected by tissues and their echoes to safely visualize internal abdominal organs, heart, muscles/tendons, and fetal growth without radiation. It detects masses, bladder stones and foreign bodies.

CT produces extremely detailed cross-sectional views without obstructing structures. CT angiography highlights blood vessels. Low dose protocols minimize anesthesia risks. Multislice scanners reduce imaging times. Computed tomography (CT) provides cross-sectional views of soft tissues and bone with high resolution. CT is especially useful for brain, chest and abdominal imaging in small animals.

MRI has unparalleled soft tissue contrast without ionizing radiation. It is well-suited for neurologic (i.e. spinal cord and brain imaging), orthopedic and oncologic exams. Open magnets reduce feelings of confinement compared to human scanners.

Nuclear scintigraphy traces radiotracer distribution to identify physiological abnormalities. Bone scans detect cancer metastasis. Liver-spleen scans evaluate hematopoietic tissue function. PET/CT marries anatomical and molecular data for cancer staging. Nuclear scanning techniques like PET (positron emission tomography) show tissue metabolic activity and function to detect abnormalities. PET is commonly used for oncology (Yitbarek and Dagnaw 2022). One unique kind of diagnostic imaging is nuclear medicine. Over the past 20 years, its application in veterinary care has grown. It is similar to how it is used in human health, although it is far behind. It is believed that practically any diagnostic or therapeutic treatment used in human medicine may be performed in animals (Yitbarek and Dagnaw 2022). Veterinarians integrate various imaging modalities for fully informed diagnoses, treatment planning and postoperative management. Continual technological advances enhance clinical capabilities. Imaging advances non-invasive diagnosis and aids minimally invasive surgical/treatment options for improved patient care and outcomes (Yitbarek and Dagnaw 2022). By supplying precise anatomical and pathophysiological information, identifying diseases, and enabling research, advancements in imaging technologies have greatly enhanced veterinary diagnosis, treatment planning, and preclinical research (Yitbarek and Dagnaw 2022).

5.4.4 Endoscopy

Endoscopy allows inspecting respiratory and gastrointestinal tracts. Rigid endoscopes provide sharper images while flexible ones access tighter spaces (McCarthy 2021). Endoscopy allows internal visualization of organs and structures through thin, flexible viewing tubes inserted via natural body openings. This minimally invasive technique aids veterinary examination and diagnosis. Rigid endoscopy using rigid endoscopes equipped with a camera provides sharp views of areas like the trachea accessed directly through the mouth or nose. Flexible endoscopes are threaded into the gastrointestinal tract via the esophagus to inspect the stomach and intestines. Veterinary bronchoscopy and gastroscopy help diagnose inflammatory, foreign body and mass conditions (De Groen 2017). Gastroscopy and colonoscopy reveal mucosal abnormalities, foreign bodies, bleeding and intestinal diseases (Hong and Baek 2023). Bronchoscopy permits lung evaluation. Cystoscopy inserts through the urethra for bladder and ureter examinations. Arthroscopy inserts an endoscope fitted with a camera into joint spaces like the stifle through small portals, allowing observation of cartilage and ligaments. Most endoscopic procedures do not require incisions, minimizing post-procedural pain (Boese, Wex et al. 2022). Biopsy forceps can retrieve tissue samples through the endoscope for histopathology without full surgical exploration. Diseases diagnosed include esophagitis, gastritis, inflammatory bowel disease, diskospondylitis, bladder masses and respiratory illnesses. Endoscopy guides interventional procedures and provides visual feedback for procedures like esophageal foreign body removal. It enhances diagnosis and treatment of many internal veterinary conditions (Boese, Wex et al. 2022).

5.4.5 Biopsies

A biopsy involves surgically removing a small tissue sample to be analyzed microscopically by a pathologist (Kamstock, Ehrhart et al. 2011). It provides a definitive diagnosis for many conditions when other tests are inconclusive. Fine needle aspiration biopsies use thin needles to collect fluid or cells from lesions. They yield rapid results with minimal invasiveness but only detect 80% of cancers. Core biopsies extract solid tissue cores for detailed histopathology. Incisional biopsies remove part of a lesion. Excisional biopsies remove an entire growth. Biopsies guide diagnosis of skin conditions, eyes, lymph nodes, organs and masses. Common sites include the liver, mammary tissue, gastrointestinal tract, lung and bone marrow. Image-guided biopsies like ultrasound- or endoscopy-assisted techniques increase sampling accuracy. Histopathology reveals normal vs. abnormal cellular structures and infiltrating infectious agents at a microscopic level. Immunohistochemistry utilizes antibody staining to identify specific cell types like tumor markers. Electron microscopy magnifies tissues further. Findings from biopsies often dictate treatment protocols for conditions like cancer, IBD, proliferative dermatopathy and storage diseases (Tseng, Matsuyama et al. 2023). Serial biopsies also aid monitoring responses to therapies over time. In many cases, biopsy provides the most definitive diagnostic information for veterinarians (Chibuk, Flory et al. 2021).

5.5 Common Medical Conditions

A wide variety of medical conditions affecting various body systems can be identified and evaluated through the diagnostic testing performed in veterinary medicine. Cancers are often initially diagnosed through physical examination findings of abnormal masses, with radiography, ultrasound and biopsy providing definitive confirmation of malignancy and cell type. Infectious diseases rely heavily on laboratory diagnostics like microbiology cultures and cytology to identify the causal pathogen. In addition to imaging techniques like ultrasound and biopsy for tissue classification, biochemical profiles and enzyme assays from bloodwork are frequently used to assess organ disorders of the liver, kidneys, and pancreas. Endoscopy is done for many reasons for the diagnosis of inflammatory bowel illness, parasitism, and foreign bodies in the digestive tract, endoscopy and biopsy are frequently used to evaluate digestive problems. A variety of diagnostic techniques, such as cytology of the airways, bronchoscopy, and chest radiography, are commonly used to evaluate respiratory disorders. In addition to these well-known medical specialties, skeletal radiographs can clearly show orthopaedic issues, and otic cytology and skin biopsy analysis are helpful in the diagnosis of dermatological conditions. Echocardiograms are the main tool used to assess cardiovascular function, and thyroid and glucose testing procedures are frequently used to diagnose endocrine disorders. Veterinary medical treatment thus requires an integrated strategy combining several specialised diagnostic methods.

5.5.1 Infectious disease

A multitude of diagnostic instruments are available to veterinarians for the purpose of identifying infectious diseases that impact animals. By isolating bacterial, viral, and fungal pathogens from sample microbiology cultures, medical professionals can direct focused antibiotic therapy. The examination of fluid, tissue, and sample smears by cytology also makes it possible to see infectious organisms like bacteria, fungus, and parasites up close. By identifying pathogen-specific antibodies generated following exposure to particular viruses or bacteria, serologic testing contributes to diagnosis (Hobbs, Colling et al. 2021). The sensitive and quick detection of genetic material from infectious pathogens is made possible by polymerase chain reaction. Histopathology, serum biochemistry tests, blood counts, and epidemiological techniques are among the diagnostic modalities available for infectious disorders. These techniques aid in the tracking of infections and inflammation-related organ failure, the identification of pathogens in biopsied tissues, and the investigation of epidemics by taking into account animal exposures to one another. Veterinarians can use parts of the clinical history, serology, culture, cytology, and advanced molecular tests to systematically detect and treat diseases (Beaufrère and Vergneau-Grosset 2021).

5.5.2 Orthopedic issues

Veterinarians assess animals with orthopaedic problems using a range of diagnostic tools. Advanced imaging methods such as CT and MRI distinguish between soft tissue injuries, while radiography is the initial screening test for abnormalities related to the bones and joints. By enabling direct visualisation into the knee and elbow joints, arthroscopy validates damage seen by other imaging modalities (Baltzer 2020). Physical orthopaedic tests assess swellings or anomalies, pain response, and range of motion. Septic arthritis and other inflammatory disorders are diagnosed by joint fluid cytology (Belshaw, Dean et al. 2020). Systemic anomalies in severe orthopaedic disorders may be revealed by bloodwork. Surgery biopsies or arthroscopy are used to find primary bone cancers or problems with implants. The efficacy of nonsurgical treatments is monitored over time with imaging modalities or follow-up radiography. Veterinarians can diagnose and treat a broad variety of orthopaedic issues with the aid of this all-encompassing diagnostic technique (Krishnamurthy, Lang et al. 2021).

5.5.3 Gastrointestinal disease

Veterinarians frequently deal with gastrointestinal disorders. The oesophagus, stomach, and intestines can all be biopsied or endoscopically imaged to directly visualise abnormalities of the mucosa and get tissue samples for histopathology. This helps with the diagnosis of foreign bodies, intestinal masses, gastric ulcers, and inflammatory bowel disease (Krishnamurthy, Lang et al. 2021). By identifying thicker intestinal walls, larger lymph nodes, or alterations to the pancreas and liver linked to gastrointestinal inflammation, ultrasonography is a useful adjunct to endoscopy (Worley 2023). Intestinal parasite identification is guided by the analysis of stool samples and involves faecal flotation, centrifugation, and microscopic examination. Blood tests for liver and pancreatic enzymes may reveal underlying organ disease (Verocai, Chaudhry et al. 2020). Radiographs show abnormalities in the pattern of the gut wall and abdominal organs associated with specific disorders, as well as blockages of the intestines or oesophagus. Contrast studies provide further information about anomalies. In chronic gastrointestinal illnesses, serial monitoring with imaging, haematology, serum biochemistry, and abdominal palpation helps assess treatment response (Raut, Naphade et al. 2020). Together, direct visualisation, biopsy, imaging, and laboratory analysis enable veterinarians to detect a wide variety of gastrointestinal disorders.

5.5.4 Respiratory disease

Respiratory disease is a commonly diagnosed problem area in veterinary patients. Veterinarians utilize a variety of diagnostic tools to thoroughly evaluate pets presenting with respiratory signs such as coughing, breathing difficulties or exercise intolerance. Initial diagnostic steps include obtaining a detailed clinical history and performing a physical examination, with auscultation of the chest to assess breath sounds (Johnson 2020, Clercx 2022). Thoracic radiography allows inspection of the entire lung fields and associated structures, revealing evidence of infections, masses, pulmonary edema, pulmonary thromboembolism or foreign objects. Pulmonary ultrasonography provides additional visualization of real-time moving images of the lung surfaces and pleural space, detecting consolidations, pleural effusions or nodules not apparent on standard radiographs. Endoscopy of the trachea and bronchi permits direct visual inspection of the lower respiratory tract and collection of samples for cytology, culture and histopathology. Analysis of tracheal washes or bronchial alveolar lavage fluid retrieved endoscopically can identify inflammatory cells, bacteria, fungi or virus's indicative of conditions like pneumonia, allergic bronchitis or pulmonary fibrosis (Downes, Bouso et al. 2020, Johnson 2020). Together, these clinical, radiographic, endoscopic and fluid analysis diagnostic modalities help veterinarians diagnose a diverse range of lower airway diseases. Identification of the underlying cause is crucial for initiating the most effective treatment plan in each case.

5.5.5 Dermatological conditions

Veterinarians have a variety of diagnostic tools that can be used together to achieve accurate dermatological diagnoses. Initial examination involves inspecting the skin and hair coat for abnormalities such as alopecia, scaling, crusting, erosions or superficial or deep pyoderma. Skin scrapings collected from multiple areas are analyzed microscopically for evidence of mites, bacteria or fungal elements like demodex mites or malassezia yeast clusters that could be contributing to underlying dermatological disease (Bond, Morris et al. 2020). Deeper punch biopsies of abnormal skin permit histopathological evaluation of epidermal, follicular and dermal aspects to characterize inflammatory patterns and identify less readily apparent infectious agents or conditions such as folliculitis or furunculosis (Mueller, Rosenkrantz et al. 2020). Allergen-specific intradermal testing further aids in vetting atopic dermatitis as a differential by assessing immediate hypersensitivity reactions to numerous environmental and food allergens. Microbial culture of specimens retrieves bacterial, yeast or dermatophyte isolates for antimicrobial susceptibility profiling (Ortiz 2023). Response to initial therapeutic trials incorporating antimicrobials, anti-fungals, anti-parasitics, antihistamines or fatty acid supplements provides insight into treatment efficacy. Collectively, these clinical examination findings and diagnostic test results guides veterinarians toward targeted dermatological diagnoses and optimized management plans.

5.5.6 Ophthalmic disorders

Veterinarians rely on a thorough ophthalmic examination to accurately diagnose various eye conditions seen in small animal patients. A complete eye exam evaluates both structure and function through tests such as menace response, pupillary light reflexes, slit lamp biomicroscopy, tonometry, and direct or indirect ophthalmoscopy (Gelatt, Gelatt et al. 2021). Ophthalmoscopy allows veterinarians to examine the retina and optic nerve for degenerations, retinal detachments, or signs of glaucoma, diabetes mellitus, or inflammation (Hamor 2022). Additional tests such as Schirmer tear testing, gonioscopy, and fluorescein staining aid detection of disorders like dry eye, glaucoma susceptibility, or corneal ulcerations, respectively (Terhaar, Henriksen et al. 2022). Together, findings from the ophthalmic exam and use of adjunct diagnostic tests helps ophthalmologists diagnose a wide range of ocular diseases, facilitating early and appropriate treatment.

Table 8 Overview of the various ophthalmic tests, their descriptions, and purposes

Test	Description	Purpose
Slit Lamp Biomicroscopy	High magnification examination of anterior segment structures (cornea, iris, crystalline lens, anterior chamber angle).	Detailed inspection to visualize inflammation, masses, ulcerations, and other abnormalities (Sakti 2021).
Tonometry	Objective test to measure intraocular pressure using applanation tonometry.	Screening for glaucoma by determining intraocular pressure levels (Da Silva and Lira 2022).

Ophthalmoscopy	Indirect ophthalmoscopy uses a special lens and mydriatic eye drops to examine the retina; direct ophthalmoscopy requires no lens.	Examination of the retina for degenerative/inflammatory diseases, detachments, vascular abnormalities, and tumours (Şirin 2020).
Schirmer Tear Test	Test strip placed in the lower eyelid fold to measure natural tear production over a set time.	Diagnosing dry eye conditions like keratoconjunctivitis sicca by measuring tear production (Nogradi, Szentgáli et al. 2020).
Fluorescein Staining	Dye highlights defects in the cornea such as ulcers, scratches, or foreign bodies.	Differentiating types of corneal ulcers and guiding appropriate treatment (Ferreira, Warth et al. 2020).
Gonioscopy	Special gonio lens visualizes the iridocorneal angle anatomy.	Detecting narrow or closed angles which can predispose to acute glaucoma attacks (Rodrigues 2022).

5.5.7 Dental/oral disease

Veterinarians perform thorough oral examinations to comprehensively evaluate dental and periodontal health in small animal patients. A meticulous intraoral inspection assesses the condition of gums, teeth, tongue, lips and oral cavity for signs of gingivitis such as inflammation, reddening, swelling or bleeding (Uusitalo 2023). Dental radiography utilizes specialized x-rays to reveal deep periodontal disease, fractures, receding gums, retained roots, and periapical lucencies from dental infections not visible on oral exam alone (Uusitalo 2023). Oral hygiene indices provide objective scoring of gingival margin position, calculus presence and other factors to gauge gingivitis severity. Bacterial cultures identify specific pathogenic bacteria like Porphyromonas and Prevotella species that drive inflammation and periodontal breakdown. Response to cleaning, scaling and polishing followed by recheck exams indicates periodontal therapy effectiveness (Uusitalo 2023). Together, these clinical, radiographic, bacteriological and therapeutic monitoring methods equip veterinarians to definitively diagnose and appropriately treat common dental diseases. Advanced imaging such as CT may reveal subtle abnormalities. Oral biopsies are used to identify the causes of stomatitis and gingivitis. Blood tests are used to screen for systemic effects such as anaemia and organ dysfunction caused by chronic oral infections, as well as inflammation. During dental cleanings, plaque and tartar biofilms are physically removed using scrapers and ultrasounds (Bastendorf, Strafela-Bastendorf et al. 2021). Prognosis and treatment planning, including management of underlying systemic disease, nonsurgical periodontics, extractions, and follow-up oral health exams and therapies to help pets achieve optimal oral function and quality of life, are guided by multiple diagnostic tests and monitoring periodontal response.

5.5.8 Neurological disorders

Veterinarians rely on a thorough neurological examination to localize and characterize neurological abnormalities in small animal patients. A careful cranial nerve, gait, postural reaction and proprioceptive assessment is performed looking for abnormalities' indicative of nerve, spinal cord or brain lesions. In-depth images of the brain and spinal cord structures are provided by advanced anatomical imaging modalities such as MRI and CT scans, which can be used to identify congenital abnormalities, hydrocephalus, herniated discs, and space-occupying lesions (De Lahunta, Glass et al. 2020). Analyses of cerebrospinal fluid look for indications of meningitis, shunt malfunction, or inflammation by counting cells, measuring protein, and measuring glucose (de Lahunta 2020, Partridge and Rossmeisl Jr 2020). EMG evaluates individual muscle membrane changes and conduction velocities to diagnose conditions causing peripheral neuropathy or myopathy. Biopsy of nervous tissues may be needed to diagnose neoplastic, infectious or inflammatory diseases. Bloodwork screens for systemic diseases with neurological symptoms. Response to treatments such as anti-convulsant, analgesics, steroids or surgery helps differentiate functional disorders from structural abnormalities.

5.6 Surgical Procedures

Veterinary surgeons perform various medical and orthopaedic procedures for small animal patients, including spaying and neutering, orthopaedic surgeries, soft tissue surgeries, dental extractions, ophthalmic procedures, gastrointestinal surgeries, urinary surgeries, neurosurgical interventions, and cardiothoracic procedures (Casas-Alvarado, Mota-Rojas et al. 2020, Niemiec, Gawor et al. 2020, Monnet 2023). Careful surgical asepsis and case planning are crucial for successful postoperative outcomes, ensuring the well-being and health of the animal. These procedures aim to prevent pet overpopulation, reduce cancer risks, and address various medical and orthopaedic conditions.

5.6.1 Oncology/tumor removal

Excision, diagnosis and treatment of tumors is procedure proceed by veterinary surgeons. Prior to surgery, an incisional or fine needle aspirate biopsy is performed to determine if a mass is benign or malignant and aids in staging. The goal of cancer surgery is complete removal of all visible tumor tissue with clear surgical margins. Common tumor types excised include lipomas, mast cell tumors, melanomas, and carcinomas of the skin, digits, and internal organs. Surgical approaches are tailored based on location, with superficial masses accessible through a skin incision. Thoracic and abdominal tumors may require more extensive surgery with meticulous hemostasis and achieving wide tumor-free margins. Sentinel lymph node biopsies provide staging information for cancers known to metastasize to regional lymph nodes like certain mast cell tumors. Adjuvant therapies such as chemotherapy or radiation are sometimes recommended post-operatively for tumors with high-risk histologic features or close, incomplete margins to prevent recurrence. Long-term follow up exams monitor surgical sites and screen for metastases. With defined margins and localized disease, surgery offers the potential for cure of certain benign and malignant tumors. Thorough pre-surgical planning and careful technique optimize oncologic outcomes (Castejón-González and Reiter 2022, Culp and Ehrhart 2022, Kudnig and Séguin 2022).

5.6.2 Ophthalmic surgery

Ophthalmic surgeons perform a variety of procedures to treat ocular diseases and injuries in small animal patients. Cataract extraction using phacoemulsification and implantation of an intraocular lens is a common sight-restoring surgery (Ong, Peh et al. 2020). Glaucoma is managed through trabeculectomy to create an outflow pathway for aqueous humor or cyclo-destructive procedures like cyclo-cryotherapy that lower intraocular pressure (Webb 2021, Sacchi, Monsellato et al. 2022). Entropion and ectropion corrections reposition the eyelid margin through techniques like tarsal stapling for turned eyelids. Protruding third eyelid glands or "cherry eyes" are repaired by removing the prolapsed gland and replacing it with a scleral graft (El Toukhy , Mitchcell and Oliver 2021). Corneal ulcers undergo superficial keratectomy to remove diseased layers of the cornea. Enucleation is performed when the eye has been severely traumatized or develops painful atrophy to alleviate discomfort. Vitrectomy may be used to remove opacified vitreous gel in cases of retinal detachment or distention (Shue, Wong et al. 2020). Post-operative care focuses on topical antibiotics, anti-inflammatories, bandage contact lenses, and patching to promote healing and prevent infection. These specialized techniques help veterinary ophthalmologists preserve and restore vision in small animal patients.

5.6.3 Dental surgery

Dental surgeons perform a variety of procedures to treat oral diseases and improve dental health in small animal patients. Tooth extractions are commonly performed to remove fractured, diseased, or severely decayed teeth that cannot be restored and are causing pain or infection. Oral surgeries address other conditions like oral cysts, tumors, or bone fractures within the jaws and mouth region. Endodontic therapy involving root canal treatment aims to save teeth with exposed or infected pulps by removing the infected tissue from within the root canal. Periodontal surgeries including gingival flap operations treat gingivitis, periodontitis, and gingival recessions. Dental radiographs provide valuable diagnostic information to identify retained root tips, lesions, cysts or periapical lucencies below the gumline. Oral exams under general anesthesia allow a thorough visual examination and full-mouth dental cleaning or scaling for many pets. Advances in digital dental radiography provide high-resolution images of underlying bony structures. Laser or ultrasonic dental tools minimize tissue trauma (Niemiec, Gawor et al. 2020, Miller, Rhodus et al. 2023). Post-operative fluoride treatments help strengthen exposed root surfaces. Comprehensive dental treatment improves oral pain, function, chewing ability and overall health.

5.6.4 Soft tissue procedures

Soft tissue surgery is a comprehensive approach to treating non-orthopaedic conditions in small animals. Common surgeries include total excision of skin masses, lumps, or growths for biopsy and to relieve symptoms, with care taken to obtain clear margins. Elliptical incisions allow full excision of skin masses with primary closure, while deeper subcutaneous growths may require dissection through muscle fascia layers depending on their size and location (Mullins and Skandalakis 2021). Haemostasis using electrocautery or sutures controls any bleeding from surrounding tissues once the mass is removed. The excised growth is submitted for histopathology to determine its benign or malignant status, aiding prognosis and further treatment planning (Corr 2012).

Repair of abdominal wall hernias is an essential soft tissue procedure to stop organ protrusion and intestine strangling. Veterinarians place the animal under anaesthesia, find the hernia sac, and delicately return any trapped organs to the abdominal cavity (Pratschke 2002). Gastropexy is a corrective operation used to prevent gastric dilation/volvulus (GDV) in dogs with deep chests and small waists. It staples the stomach serosa to the body wall, providing support and preventing twisting on its vascular pedicle (Volvulus 2001).

Two common abdominal organ removal surgeries are splenectomy and routine spay/neuter procedures. Splenectomy involves surgical removal of the spleen, indicated for cases of splenic tumors, rupture from trauma, or infectious causes compromising spleen function (Petroianu 2011). Ovariohysterectomy in females and castration in males are among the most frequently performed surgeries, removing reproductive organs to prevent unplanned litters (Petroianu 2011, De Cramer and May 2015). Both procedures provide health benefits while eliminating unwanted breeding behaviours.

Perianal fistula surgery aims to repair chronic draining tracts in the sensitive perineal area (Akiba, Rodrigues et al. 2016). Under anesthesia, the veterinarian cleans and explores the tract with a probe to delineate ramifications. Debridement uses scalpels or lasers to remove unhealthy, necrotic tissue from open wounds or pressure sores unable to heal. Thorough cleansing debrides nonviable tissue, contaminants, and debris to stimulate viable cells. After cleaning and ensuring hemostasis, the defect may be closed primarily or packed (Culp, Cavanaugh et al. 2012).

Skin flap surgery is a reconstructive technique used to close defects created by traumatic wounds, open fractures, or tumor excisions. Local or regional flaps rotate or transpose islands of viable cutaneous and subcutaneous tissue into the defect area (Khundkar 2019). Colorectal procedures address diseases of the intestines and rectum such as intussusception or prolapse. Intussusception surgery manually reduces the telescoping of one portion of bowel within another under anesthesia, while resection may be needed if reduction fails. Prolapsed portions of colon or rectum are cleaned, manually replaced, and sutured to prevent recurrence. Both skin flap surgery and colorectal operations utilize tissue transfer techniques to close defects and resolve conditions affecting the GI and integumentary systems (Elbetti, Granchi Zanieri et al. 2009, Dunn and Madoff 2016).

5.7 Pain Management

5.7.1 Analgesics

Proper pain management is a critical component of soft tissue surgery recovery. Nonsteroidal anti-inflammatory drugs are commonly prescribed to provide both analgesia and reduce postoperative inflammation (Constantinescu, Campbell et al. 2019). Opioid analgesics such as buprenorphine, fentanyl, hydromorphone, and morphine are often utilized in conjunction with NSAIDs to control more severe pain, employing a multi-modal approach. Local anesthetic infiltration of incisions or field blocks can also aid in localized pain relief. Preemptive use of analgesics prior to surgery has benefits of preventing central sensitization from surgical stimuli. Aggressive initial pain management is important as acute pain may sensitize the nervous system, potentially contributing to chronic pain issues. Extended-release formulations of some analgesics allow less frequent dosing for around-the-clock coverage. Multimodal analgesia using two or more analgesic classes at lower doses provides synergistic effects while mitigating side effects from heavier narcotic usage. With appropriate analgesic protocols and client education on home care, patients can experience improved recovery and comfort levels following soft tissue procedures (Fox 2013, Alamrew and Haben Fesseha 2020).

5.7.2 Local and general anesthesia

Proper use of anesthesia is necessary for performing soft tissue procedures and protecting patient welfare. Most invasive operations require general anesthesia to control pain and movement during surgery. Inhalation agents like isoflurane are commonly used, though some hospitals also employ intravenous inductions with propofol and maintenance with ketamine/midazolam infusions. Local and regional nerve blocks can supplement certain operations when lidocaine or bupivacaine is injected to provide surgical anesthesia and postoperative analgesia, such as dorsal penile blocks for scrotal surgeries (Marongiu 2012). Infiltrating local anesthetic around incisions enhances intraoperative pain control as well. Close physiological monitoring under general anesthesia, including vital signs, temperature, and ECG, ensures patient safety. Regional techniques paired with lightened general anesthetic administration also reduce cardiovascular and respiratory depression risks compared to higher doses. Smooth postoperative recoveries are supported through continued infusions, analgesic provisions, and monitoring by the veterinary team until patients are comfortable and stable (Duncan, Ross et al. 2015).

5.7.3 Alternative therapies

In order to enhance traditional care during the postoperative healing phase, a number of complementary therapies may be used. Mentioned in table. 2.

Table 9 A concise overview of various therapeutic methods and their respective details

Therapy	Details
Laser Therapy	Uses low-level lasers to reduce pain, swelling, and promote tissue healing through photobiostimulation (Gupta and Hamblin 2013).
Acupuncture	Stimulation of acupuncture points to provide pain relief, relaxation, and supplement analgesics during recovery (Castro 2005).

Hydrotherapy	Utilizes underwater treadmill to promote earlier return of range of motion after procedures involving orthopedic tissues or body walls (Tomlinson 2012).
Massage Therapy	Increases circulation and range of motion, decreases muscle spasms/tension, aiding in wound healing and recovery comfort (Mahaseth and Raghul 2021).
Nutraceuticals	Supplements like glucosamine, chondroitin, and omega-3s support joint and soft tissue integrity during healing (Hall and Kalten 2013).
Herbal Medicines	Anti-inflammatory herbs such as arnica and calendula aid in the resolution of bruising and swelling (Hall and Kalten 2013).
Nutritional Supplementation	Ensures patients receive optimal nutrients to facilitate healing and provide energy for rehabilitation sessions (Marchegiani, Fruganti et al. 2020).
Behavioural Techniques	Methods like music therapy, pheromone therapy, and therapeutic touch promote relaxation and relieve pain/stress (Vroegindewey and Kertis 2021).

5.8 Client Communication and Education

Soft tissue surgery outcomes depend on effective client communication and education. Veterinarians should provide comprehensive explanations about the procedure, risks, recovery period, and possible outcomes. They should also offer preoperative and postoperative pain control techniques. Discharge instructions should cover incision care, activity restrictions, medication administration, and follow-up exams. Regular check-ups are essential for early detection of issues. Open communication is crucial for clients' recovery at home, and a well-organized home care plan is essential for long-term care (Kanji, Coe et al. 2012). Relationship-cantered veterinary care is a collaborative partnership between a veterinarian and a client, fostering mutual understanding and recognition of the client's perspectives and expertise. This model is crucial for an ideal healthcare system, as it emphasizes the importance of relationships in delivering high-quality care. Adherence to this model involves intentional choices about treatment regimens based on the diagnosis and client's beliefs about the illness and treatment options (Kanji, Coe et al. 2012).

5.8.1 Treatment plans and compliance

Creating a well-defined treatment strategy is essential to having successful surgical results. All postoperative care procedures, including wound monitoring, bandage changes, exercise restriction, and scheduled recheck appointments, must be described by the veterinarian. Clients can administer therapy appropriately when drug schedules, dosing instructions, and refill authorizations are communicated. Encouraging further therapies offers complete support for healing. Feedback is obtained to confirm comprehension and capacity to adhere to the plan, and any possible obstacles to compliance are addressed before they arise. Post-operative goals are stated to assist owners in monitoring their pet's milestones, and routine check-ins evaluate progress and resolve any problems. For the first few crucial times, strict adherence is required because non-compliance could compromise the outcomes. Unexpected obstacles in the actual world may still call for flexibility as long as they are handled quickly. Veterinarians may best guarantee that patients undergoing soft tissue surgery receive the best possible aftercare to optimise results by engaging in ongoing, client-centred planning that is both meticulous and ongoing (Kanji, Coe et al. 2012).

5.8.2 Behavior modification plans

Behaviour modification programmes post-soft tissue surgery aim to prevent undesirable behaviours like licking, scratching, or rubbing from damaging incisions. Elizabethan collars, soothing drugs, and pheromone sprays are used to deter undesirable behaviours. Alternate toys and activities are provided to keep pets busy and encourage wound healing. These programmes aim to encourage suitable replacement behaviours rather than preventing recovery (Sari, Cobre et al. 2023). Veterinary behaviour plans provide a comprehensive strategy for dog recovery, including owner supervision, positive reinforcement training, and environment adjustments. These plans discourage harmful behaviours like wound-licking and scratching by limiting access to designated areas (Hammerle, Horst et al. 2015). Positive reinforcement is used to develop desired replacement behaviours, such as quieting. Targeting sessions reward calm replacement activities with incentives or praise. Clients are taught to monitor their pets for prohibited behaviours, and early redirection to trained behaviours can be achieved. Plans also address persistent behaviours, such as putting pets on a leash or applying sour or bitter sprays to deter licking. A target completion date is set to align with wound healing, and advice on avoiding relapses is given (Hedges 2021). Consistent follow-through is crucial, as the most consistent results come from a family's joint commitment to upholding substitution behaviours and constraints during therapy.

5.8.3 Preventative care recommendations

Veterinarians advise pet owners to prepare ahead of time for the care of their animals following soft tissue surgery. Wellness exams should be performed annually to monitor overall health and spot any chronic soft tissue problems close to the surgical site (Shanan 2011). To maintain joint and tissue structure, prophylactic supplements such as glucosamine, MSM, or omega-3s are advised. It is advised to regularly evaluate food, activity, and weight to relieve stress on problematic areas. For short-haired breeds, frequent brushing and combing is recommended to prevent matted fur. Surgery or re-injury can be avoided in the future by identifying new issues early. In order to avoid new cuts or rips close to surgery areas, proper nail care is required. It's critical to advise against rough play in order to preserve recovered tissues. It is advised that maintenance programmes include hydrotherapy or continuous physical therapy. Periodic diagnostic imaging follow-up may be necessary for recurrent conditions. It is recommended that clients notify the veterinarian right once if they notice any new lumps, bumps, or injuries. By preventing difficulties in the future, these long-term preventative actions guarantee that patients who have surgery stay healthy for the rest of their lives (Kanji, Coe et al. 2012).

5.9 Conclusion

So, in conclusion from prehistoric times veterinary medicine practiced greatly by the farmers and herders and potentially enhanced the health and wellbeing of animals. In 18th century with the establishment of veterinary schools, institutions and certification procedures the veterinary practice elevated to the level of science-based profession. Because of the advancement in many fields i.e. bacteriology, anaesthesia, and surgical methods, the veterinary professional are now equipped better to identify and cure illnesses. In order to promote the physical, mental and environmental well-being of all animals the veterinary professionals use their clinical experience and knowledge. This provide holistic care to all animals which is necessary for their wellbeing. The veterinary professionals place strong focus on approaches that promote good health and prevent sickness. This includes programs that provide vaccinations to boost immunity, guidance on healthy eating habits, treatment to control parasites, and medical screening tests. Working together across disciplines improves diagnostic and treatment planning. Future technological developments promise much more progress. All things considered, the veterinary field has significantly increased animal longevity and quality of life while safeguarding public health. Future-focused innovative care models seek to expand access globally, and cutting-edge fields of study like regenerative medicine have the potential to revolutionise patient outcomes and preventive care for both human and veterinary populations. Veterinarian care is positioned as an essential component of fair One Health solutions through ongoing advancement.

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Chapter 6 : One Health Approach: Integrating Human, Animal, and Environmental Health

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6.1 Introduction

The One Health approach is a holistic strategy that recognizes the interconnectedness of human health, animal health, and environmental health. It emphasizes the collaborative efforts of professionals from various disciplines to address global health challenges and promote sustainable well-being for all living beings. This chapter explores the principles of the One Health approach, its significance in safeguarding the health of populations and ecosystems worldwide, emerging challenges, and opportunities for innovation and collaboration.

6.2 The Interconnectedness of Human, Animal, and Environmental Health

Human health is intimately linked to the health of animals and the environment. Zoonotic diseases, which originate in animals and can be transmitted to humans, represent a significant public health concern. Examples include diseases like Ebola, avian influenza, and COVID-19, which have demonstrated the potential for devastating global impacts. Additionally, environmental factors such as pollution, deforestation, and climate change can directly affect human and animal health, leading to the emergence of new diseases and exacerbating existing health issues.

The concept of the One Health approach recognizes that the health of humans, animals, and ecosystems is interconnected and interdependent. It emphasizes the need for collaborative efforts to address health challenges at the interface of these domains. By adopting a holistic perspective that considers the complex interactions between humans, animals, and their shared environment, the One Health approach seeks to promote health and well-being for all living beings.

6.3 Principles of the One Health Approach

1. Collaboration: The One Health approach emphasizes interdisciplinary collaboration among professionals in human health, veterinary medicine, environmental science, and other relevant fields. By working together, experts can leverage their respective knowledge and skills to address complex health challenges more effectively. Collaborative efforts may involve joint research projects, surveillance programs, and policy development initiatives aimed at preventing and controlling disease outbreaks and promoting environmental sustainability.

2. Prevention: One Health promotes proactive measures to prevent and control disease outbreaks at the human-animal-environment interface. This includes surveillance programs to monitor disease trends in humans, animals, and the environment, as well as vaccination campaigns and other interventions to reduce the risk of disease transmission. Prevention-focused strategies aim to identify and address underlying drivers of disease emergence, such as habitat destruction, wildlife trade, and climate change, to mitigate future health risks.

3. Sustainability: One Health recognizes the importance of sustainable practices in promoting health and well-being for current and future generations. This involves balancing economic, social, and environmental considerations to ensure the resilience of ecosystems and the health of communities. Sustainable development principles are integrated into One Health initiatives to address underlying drivers of disease, such as poverty, inequality, and environmental degradation, and promote long-term health outcomes.

6.4 Collaborative Efforts to Address Global Health Challenges

Numerous initiatives and organizations around the world are dedicated to advancing the One Health agenda and addressing pressing health issues through collaborative action. These include:

- The World Health Organization (WHO), the Food and Agriculture Organization of the United Nations (FAO), and the World Organisation for Animal Health (OIE), which jointly promote One Health approaches to address zoonotic diseases and food safety. These organizations collaborate on surveillance, research, and capacity-building initiatives to strengthen health systems and improve disease preparedness and response.
- Research institutions and universities that conduct interdisciplinary studies to better understand the dynamics of disease transmission and develop innovative solutions for prevention and control. These institutions bring together experts from diverse fields, including epidemiology, microbiology, ecology, and social sciences, to address complex health challenges and inform evidence-based interventions.
- Non-governmental organizations (NGOs) and community-based initiatives that engage local stakeholders in One Health interventions, such as community health education programs and wildlife conservation efforts. These initiatives empower communities to take ownership of their health and well-being and build resilience to health threats through education, capacity-building, and sustainable development initiatives.

6.5 Emerging Challenges and Opportunities

While the One Health approach has gained recognition and momentum in recent years, there are still significant challenges to its widespread implementation. One such challenge is the need for improved coordination and communication among diverse stakeholders, including government agencies, non-governmental organizations, academic institutions, and local communities. Efforts to promote interdisciplinary collaboration and knowledge sharing can help overcome barriers and facilitate more effective responses to health threats.

Additionally, the rapid pace of globalization and urbanization has led to increased human-animal-environment interactions, creating new opportunities for disease emergence and spread. Urbanization, in particular, presents unique challenges for One Health practitioners, as dense human populations and close proximity to wildlife can facilitate the transmission of pathogens between species. By integrating One Health principles into urban planning and public health strategies, cities can better manage risks and promote healthier environments for residents and wildlife alike.

Furthermore, climate change poses a significant threat to global health, with impacts ranging from extreme weather events to shifts in infectious disease patterns. Rising temperatures, changing precipitation patterns, and habitat destruction can alter the distribution of disease vectors and reservoirs, potentially leading to the emergence of new zoonotic diseases and exacerbating existing health disparities. Addressing the health impacts of climate change requires a coordinated, multi-sectoral approach that integrates environmental conservation, public health interventions, and community resilience efforts.

Despite these challenges, the One Health approach also presents numerous opportunities for innovation and collaboration. Advances in technology, such as genomic sequencing, remote sensing, and big data analytics, are transforming our ability to monitor and respond to health threats in real time. For example, genomic surveillance enables researchers to track the evolution of pathogens and identify emerging variants with pandemic potential. Remote sensing technologies provide valuable data on environmental changes, such as deforestation and land use, which can inform disease risk assessments and conservation strategies.

Moreover, One Health initiatives can lead to co-benefits beyond health, including biodiversity conservation, food security, and economic development. By recognizing the interconnectedness of human, animal, and environmental health, and investing in collaborative research and capacity-building efforts, we can build more resilient and sustainable societies that prioritize the well-being of all living beings.

6.6 Conclusion

The One Health approach offers a comprehensive framework for addressing the complex health challenges of the 21st century. By embracing interdisciplinary collaboration, prevention-focused strategies, and sustainable development principles, we can build a healthier, more equitable world for current and future generations.

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Chapter 7: Advances and Epic Challenges in Veterinary Food Security: Animal Health and Protection

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Abstract

Animal production must be increased to meet the upcoming necessities of a growing population. The upcoming demand for shortage of food supply has been a matter of great concern. Fears about food quality at both administrative and consumer levels has been increased. This Book chapter highlights the both environmental and climate change which greatly impacts on livestock. The use of plants in maintaining veterinary health and the use of phytotherapy as a veterinary drug has been also discussed. With certain other herbs, the therapeutic herbs are used for additional treatment rather than conventional medicines. We explore the challenges and future trends of Veterinary Public Health. We also discussed the Zoonotic diseases which should be taken as a serious issue as they raise many infections and infectious diseases. This chapter also highlights the AMR link with high commercial costs due to its healthcare load, as it raises diseases and deaths. The significance of veterinary food security is important for Animal Health and Protection. One Health theory is also learned at the end which stresses the environmental interactions among human, animal, and ecological health.

Keywords: Antimicrobial resistance (AMR), Livestock, One Health theory, Phytotherapy. Therapeutic herbs, Zoonotic disease.

7.1 Recent food-related concerns

One of the extreme challenges in front of this generation is food safety. Approximately 925 million persons live with starvation. Strangely, food safety is a perplexing issue that opposes the veterinary profession and many other financial, administrative, technical, and social disciplines. (Kelly, A. M, et al., 2020) The objective is to find solutions to food safety by associating human health, animal health, food manufacturing, and biodiversity protection. The professional training of veterinarians of the future needs to include an understanding of how the survival of humans and wildlife is speedily changing in this larger universe, mainly in unindustrialized nations, where the changes are driving a necessity for veterinarians.

7.1.1 Harmful chemicals in animal food

Animal feedstuff must be healthy and free from impurities, whether it is for dogs, poultry, or farm animals. We must look after six poison manufacturers. These are Remnants, Genetic modifications (GM), Biological impurities, Mycotoxins, Dioxins, and Heavy metals. (Chen, K., & Kan, J. 2021). The performance and health of animals significantly depend on a healthy diet, similar to humans. The quality of food is basic feature of success in farm animals. Food is subject to strict guidelines to ensure this therefore, Regular examinations are important.

1. **Remnants:** Similarly, as meat for human consumption, animal food is subjected to strict controls, to produce meat. However, it could happen that remains of veterinary drugs, antibiotics or hormones are present in the meat. ELISA Tests can detect a great quantity of such elements. Also, composts or remains of plant security products might get into animal food.
2. **Genetic modifications (GM):** Genetically modified soybeans produce Soy meal which is generally used in food manufacture. Rice Corn and canola are frequently made from genetically modified yields.
3. **Biological impurities**: Polluted food can be the origin of contamination in animals. High-protein food is mostly at risk of contamination because it offers a very favorable atmosphere for Salmonella. This is mostly difficult for farm animals because Salmonella may grow into the milk, meat, or eggs of animals. Further bacteria as well as molds and yeasts can also cause infection in animal food. Total bacteria count and the Checking of pathogenic microbes is a significant task in the production of animal food.
4. **Mycotoxins:** Poisonous metabolites created by molds are called Mycotoxins. Even in small concentrations, they can be the reason for growth disorders in animal lateral flow tests Mycotoxins could be identified rapidly and simply. Mycotoxins hardly occur in animal food today, through wide controls and precautionary methods. (Rehman, H. U et al., 2022)
5. **Dioxins**: Dioxins are carbon-based contaminants that occur universally in waters, soils, animals, and plants.
6. **Heavy metals:** Heavy Metals get into feedstuff through dust, fertilizers, or extracts. It can be dangerous for animals and humans and might cause chronic infections.

7.1.2 Preventive Controls for Animal Food

A food protection scheme that examines threats and risk-based preventive Measures, Must be implemented (Bianchini, A. 2019). The instruction sets for food safety plan should include:

1. **Risk examination:** The main step is risk identification, which must consider physical and chemical hazards. These risks could exist as naturally or accidentally, or purposely introduced for commercial advantage if they disturb the security of food.
2. **Precautionary measures:** These measures are necessary to confirm that dangers requiring precautionary control will be reduced or prevented and these steps are necessary to ensure that precautionary measures are effective. Each facility must have a recall plan that produces animal food with a risk requiring precautionary measures.

1. **Verification:** To confirm that precautionary measures are regularly applied and effective these actions are mandatory. It includes verifying with scientific evidence that measures successfully controlling a recognized risk. Environmental monitoring and Product analysis are possible for verification activities but they only require the nature of the precautionary measures, and the role of controlling those measures in the facility's food security scheme. (Surareungchai, S, et al., 2024)
3. **Improvements and Corrections**: A short problem that happens during animal feed manufacture is timely reorganization and are corrected in the step of Corrections. Correction activities must be documented with records. Correction activities include actions to detect a problem with precautionary measures implementation and estimation of affected animal feed for protection.

7.1.3 Food Safety Implementation

To prevent food from any infection or pollution all methods should be undertaken, this is called Food Safety Implementation**.** For the Execution of protection of animal food, public health veterinarians are involved. Procedures for food safety have proceeded from traditional control bases to more targeted food protection schemes in recent years. (Adamchick, J., & Perez, A. M. 2020). Many food industries use a preventive risk-controlling approach (HACCP) to increase food security. Veterinarians can ensure the production of safe food at food manufacturing factories.

At farmstead level: The main criteria for any food manufacturing plant is safe food for animals. Food animals are generally found in disciplined farms where infectious diseases are not likely to happen in present food manufacturing companies. Veterinarians play an important part in guaranteeing the manufacture of healthy animals through their proper cooperation with professionals and farmers.

At Food Manufacturing Factories: The examination of live animals plays a significant part in guaranteeing the safety of meat for their future uses. During major production, most reported food-borne infections are due to the uncleanness of foodstuffs. Veterinarians play a significant title role in the examination of these epidemics and also implement remedial methods once the infection has been recognized. Further main Facility is to approve health certification for worldwide trade.

Environmental Protection: New fears for the health of animals have been produced due to infectious and biochemical contamination of water sources and significant changes in the use of land. Alarms are increasing about waste and nutrient management and antimicrobial resistance of pathogens. Wildlife populations and Food animals are linked to certain environmental glitches. Veterinarians are performing a role in the field of environmental health.

7.1.4 Significance of Traceability for Consumer Protection

The traceability of animal foodstuffs increased importance to customers in addition to the organizations that facilitate the needs of customers because marketing and food production have been removed from direct control of the customer. New international companies with limited control of production or handling were formed to market foodstuffs. (McKean, J. D. 2001).

Fears about quality and food safety have increased at administrative and customer levels in the last two years. Attention has mainly focused on the control of parasites, microbes, viruses, and harmful chemicals that can be dangerous to individuals during foodstuff supply. Product traceability necessities increase as data supervision machinery becomes less expensive and more powerful. Private and Public divisions should grab these chances to improve public health. These fears have been extended to include animal health, in several nations. The primary goal of organizations is the capability to preserve the distinction of these foodstuffs based on customer wishes and requirements. Private and Public sector efforts to improve food safety fears through modifications of processing and manufacturing techniques. The traceability of animals affected by these alterations from farmstead to dealer is an important part of this information transfer mechanism. (García-Díez, J, et al., 2021) This information transfer mechanism may be used to improve safety. The traceability necessities will significantly heighten the complications of these schemes.

7.2 Animal Health and Disease Management*

Disease Management and Animal Health programs are much more focused on the early stage of disease recognition, risk assessment of health, and crowd monitoring. In recent times, extra importance has been put on the concerns of manufacturing procedures and the quality of products like meat and milk. Every linkage should deliver the next linkage with protections concerning the product supplied is an important element in production chains.

7.2.1 Emerging and Re-Emerging Infections

Contaminations that seem in recent times within a populace or those whose geographical range or frequency is speedily growing are called emerging diseases. Re-emerging diseases are known as infections that significantly progress their occurrence or either shift their geographical setting or increase their host range. (Wang, W. H, et al., 2021) Control and Prevention will need unique strategies as zoonotic diseases are different. Simultaneously, as new pathogens and viruses have consecutively arisen in current years, well-known diseases and infections for which treatments are usually present have re-emerged in mostly infectious new creations together with multidrug-resistant tuberculosis. In 2010, the World Health Organization highlighted Emerging infections and pathogens with high epidemic potential to protect people from more contaminations during epidemic outbreaks. (Roberts, S. L. 2023) A variety of infectious diseases are given following:

Microbial diseases: Bacterial-borne contaminations across populations have had an unbelievable historic influence on animal health. A water-borne infection bacterium Vibrio cholera causes **cholera**. Epidemics of cholera were widespread in Yemen in 2015 **Tuberculosis** affects the public health around the universe. Nowadays, effective treatments and remedies are present for these bacterium-based diseases.

1. **Viral Infections:** The majority of increasing infections and pathogens that establish pandemic infections include: influenza, monkeypox, and Zika virus. The appearance of new viral pathogens and viruses signify high-risk vectors for possible universal epidemics, Dissimilar to bacterium-based infections. Emerging Viral Infections include SARS-CoV-2

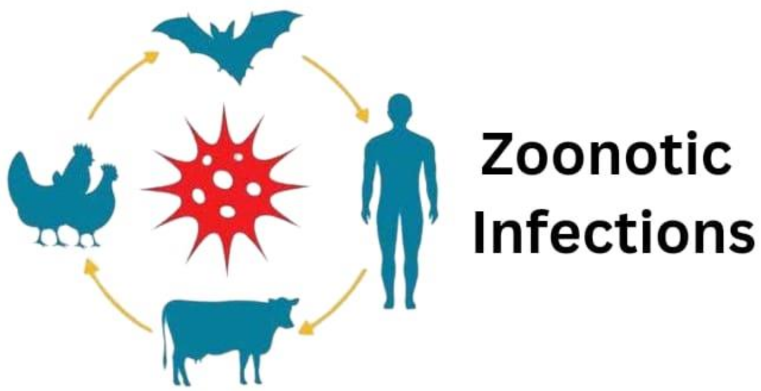
and the Mpox virus. (Hoffman, S.A, et al., 2024) Re-emerging viral Infections include the Ebola virus, Dengue virus, and Chikungunya virus. Current research highlighted the high-level epidemic risks related to emerging virus-related contaminations.

7.2.2 Need for Vaccination and its implementation

The shift of mankind from a hunter to an agricultural lifestyle is one of the major conversions in history. Numerous animal virus infections affecting both food safety and public health can be also transferred to humans (zoonotic infections) by direct interaction with infected animal tissues and fluids. Control and elimination of them have to be implemented as farm animal infectious agents, which can also cause infections in humans.

7.2.3 Prevention and Control of Zoonoses

Disease that can be transferred from animals to humans in natural circumstances, is called zoonoses. An enormous task for veterinarians is controlling and monitoring the entire variety of zoonoses and the wide range of vulnerable animal classes. (Bernstein, A. S, et al., 2022) Several of these infections are possibly to become global problems and spread over long distances. First of all, a disease must have the potential to grow into a main social contagious disease. Several zoonoses like anthrax and brucellosis, can cause very significant human diseases and a great number of viral and bacterial infections. Certain infections also cause diseases in farm animals.



7.3 Role of animal Health programs in Economic Development

Unindustrialized nations frequently face severe animal health difficulties, with numerous widespread infections. Social expenses as well as a lot of trade difficulties are due to animal infections which frequently exceed the private expenses to livestock manufacturers. One can generate good earnings through finance development and better maintenance of livestock by improving animal health. Economic analyses of animal health programs have been often measured.

7.3.1 Benefits and Risks Associated with Animal Health Programs

Animal health programs might be concern with the reduction, prevention, and outbreak controls of livestock infections. The benefits and costs of animal health programs were classified into consumers, government, and producers. We can add livestock traders to this list, meanwhile, in livestock industries there are classically numerous traders and organizers. For Example, Trade benefits as it is included in both social and private welfare. Additional income may be increased by the private division some of which will be taken by the administration, in the form of charges and taxes, hence it becomes a social rather than private profit. (Shi, R, et al., 2022) Manufacturers can gain prices by stocking vaccines and medicines, further employment, and veterinary facilities. Animal health programs can include a variety of expenses. Greater charges for animal foodstuffs might be faced by Consumers due to increased exports. Requirements such as Vaccine manufacturing plants should be recognized. Vaccines should be delivered to livestock holders at sponsors charge, and the need for computer tools for data entry should be also full filed. In animal health programs most of the public security programs are without any hazard. If the program is more striving, the chances of risk will be greater. If livestock holders do not co-operate energetically with it the reduction of infection can fail. Domestic and provincial vaccination programs are unlikely to be effective if a satisfactory treatment level is not achieved.

7.3.2 Animal Welfare and Ethics

The term Animal Welfare is normally used to define an ethical view that agrees with how animals should be treated, whether animals should be used by humans, and under which circumstances animals should be used by humans. (Hernandez, E, et al., 2022) People's observations about animal welfare are determined by their ethical observations hence Animal welfare is a argumentative problem. Several dissimilar ethical opinions about the use of animals conflict with each other, which clarifies why people might not agree about animal health. To produce large amounts of animal protein at a reasonable price Agriculturalists face increasing pressure which has placed some stress on preserving health standards.

7.4 Role of plants in maintaining veterinary health

Many things are prepared from plants like bathing soaps, decoctions, and extracts. The usage of vegetation in veterinary medication is slightly insufficient because the provisions for animals are not offered presently. Metabolic alterations of foodstuffs of primary metabolism create secondary or specialized metabolites. Primary metabolism Includes carbohydrates, fatty acids, and amino acids. Systematically interconnected plants typically produce chemically alike, but not equal metabolites. Secondary plant metabolites are composed of very multipurpose chemical compositions and dissimilar pharmacological actions. Hence Pharmacological effects cannot replace one another but they have similar effects.

7.4.1 Usage of Phytotherapy as a veterinary drug

The most commonly spread treatment method is Phytotherapy, which is based on plant usage. Research on unfamiliar species of plants are scientific outcomes on the application of some planning's. Phytotherapy, prepared from plants is a form of veterinary medicine. (Davidović, V, et al., 2012) It includes remedial effect, and inhibits the beginning of infections in animals. Medicinal and other plants have supportive remedies which are used in veterinary health care, for defensive resolutions, or as a whole treatment. Phytotherapy is very useful in treating slighter infections such as recurring infections. Different plants are used to stimulate different tasks in animals and it increase the immune system's action.

7.4.2 Antimicrobial actions of plants

Sub-therapeutic dosages of antibiotics increase the development of beneficial microflora and kill a great number of pathogens. Researchers have found alternate results for the usage of antimicrobials substances in recent times. Numerous groups of extracts like enzymes, prebiotics, and antioxidants, are being used these days so, It is necessary to induce food digestion by using beneficial supplements and inhibit the development of pathogenic microbes. As herbal preparations contain active substances so, it has many Benefits. They are non-toxic and have antimicrobial effects. Thus, the quality of foodstuffs like eggs and meat can be improved. Fragrant herbs have antimicrobial actions and are rich in oils. Thus plants are used as natural disinfectants and antiseptics in veterinary medicines. For Example sweet basil, lemon balm, and yarrow. Pekin ducks contributed to the reduction of disease-causing pathogens by adding prebiotics and herb mixtures like sweet basil and oregano to their foodstuffs.

7.4.3 Treatment of the gastrointestinal system by plants

A vast number of plants are used in the cure of diseases of the gastrointestinal tract, whose active principles contain bitter ingredients. Plantain, marshmallows, and nettle are used for the cure of diarrhea as veterinary medicines. (Tan, N, et al., 2020) Boiled seed of dock are used in farm animals for the cure of diarrhea. If inflammation occurs in cattle and sheep Klamath weed boiled in water is administrated through drinking water. Some Plants are used in the inhibition of infections and management of several body systems like respiratory systems, and gastrointestinal systems.

7.5 Veterinarians' role in one health

One health is a methodology of uniting human, animal, and environmental mechanisms to speech worldwide health challenges. An individual loyal to the advantage of humanity, relieving the suffering of animals, and stimulating animal welfare is a veterinarian. (Kaba, T, et al., 2017) Further roles of Veterinarians in domestic Animal Health are to promote animal protection, Prevent disease outbreaks, Capacity for Disaster Management, and Deliver population and medical health expertise for all wildlife. Veterinarians in environmental health include: Protecting biodiversity, controlling animal resources, Disease surveillance and prevention in wild animal populations, preserving natural resources, conservative medicine, and Climate change adaptation.

7.5.1 Challenges of Veterinary Public Health System of Livestock Production, Health Problems, and Globalization of Trade are challenges in veterinary public health. The use of Antimicrobials in food-producing species for improved production is associated with risk. The most important challenge is given below:

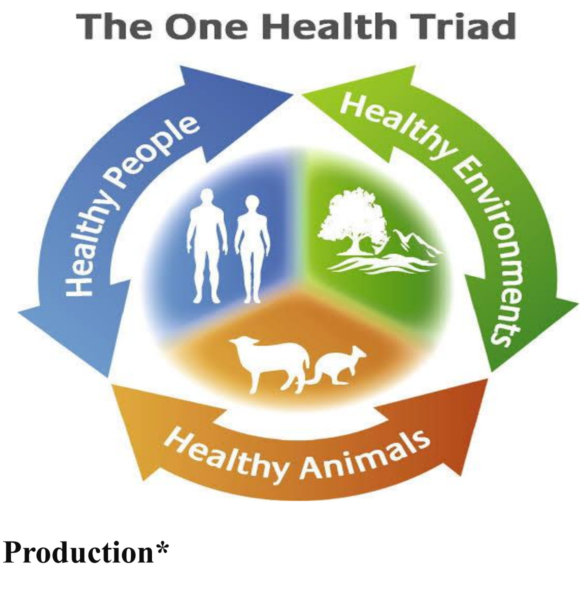
Lack of food supply veterinarians: Fears have been increasing that the supply of veterinary medicine profession may face many changes. These changes will affect both the demand for their facilities and the supply of employers. The profession is also changing because of the Globalization of the food system and the growing requirement to protect food animals from infection. Skills related to Public health are becoming more significant. On the front line of these demands are Veterinarians who have roles in the private food supply veterinary medicine profession. (Haakuria, V. M, et al., 2020) Students from inner-city backgrounds are entering the institutions of veterinary drugs hence, the supply of students entering the profession has also significantly changed. Several demands within academies and institutions of veterinary medicine, and students are ready to take supply veterinary medicine roles to make important contributions to protecting animal health.

7.5.2 Upcoming Trends of Veterinary Public Health

Inside the universal health agenda, the role of Veterinary Public Health will boost actions that contribute in the achievement of human and animal health. (Hernandez, E, et al., 2022) Diseases spread through animals, water, and food. Infections caused by environmental pollutants, and the misuse of antibiotic in the present are developing health fears. The Structures that are disturbing these fears include the universal movement of individuals, controlling of the composite foodstuff and fiber system. Its environment, and ecological deviations which are influencing universal safety.

7.5.3 Intersections between human-animal health

If we apply the One Health approach to the micro biome it reach a decision with the concern of both non-pathogenic and pathogenic infections transfer among humans, animals and surroundings. In Modern studies of such transmissions, molecular and arithmetic procedures are being used. **(**Allore, M. 2021). The effects of such micro biome relations on social health are reviewed in this chapter. The micro biome of animals as well as the Ecological micro biome in close interaction can disturb the social health consequences. Transmission of Such micro biome can occur at the domestic level along with the workplace location.



Sustainable Livestock Production*

For the sustainability of deprived and marginalized ranchers Farming contributes importantly. Variations of external features increasingly influences animal production. There are increasing fears about livestock and its health significance at the same time. For improvement in animal production, new skills and technologies seem to offer several chances. In both industrialized and unindustrialized nations Animal production progress has important mitigation effects. (Enahoro, D, et al., 2021) Livestock is a major benefit for weak populations also it plays a significant title role in an ecological food organization. Improving livestock organization is an essential role of "One Health" method which is designed to optimize human-animal and environmental health.

7.6.1 Environmental impact of livestock farming

There are various influences on the environment of the livestock industry. It includes low temperatures, wind, and rain or wet snow. These factors can affect the animal's heat balance. Students have almost no awareness of the ecological effect of the foodstuff they consume. (Koytcheva, M. K, et al., 2021) Today, it is essential to promote awareness of the ecological and health effects which are affected by the livestock industry. In different educational training programs, an introductory course on ecological discipline must be included.

7.6.2 Climate change and its impact on livestock production

From animal farming, the main significant greenhouse gases are methane and nitrous oxide. Methane is mostly produced by manure storage and enteric fermentation. (Fawzy, S, et al., 2020) A gas with a global warming potential 265 times greater than carbon dioxide is Nitrous oxide. Its radiations are produced when organic and inorganic composts are applied to the soil. Nitrous oxide arises from compost storage and the use of inorganic and organic composts. (Jain, R., & Prajapati, D. 2023). Food manufacture together with the related soil carbon dioxide and nitrous oxide radiations is another important concern for the livestock area as well as greenhouse gases (Pollard, A. E., et al., 2021).

7.7 Antimicrobial Resistance and Stewardship*

Antimicrobial stewardship programs decreases prices, decrease AMR, and enhance beneficial consequences. As various studies are descriptive the understanding of which methods are more effective is limited. A severe risk to universal public health is AMR. (Butzin-Dozier, Z, et al., 2020) The usage of antibiotics in COVID-19 increases microbial resistance and eventually leads to more deaths. Antimicrobial stewardship (AMS), is planned to encourage, expand, observe, and estimate the balanced usage of antimicrobials to preserve their upcoming efficiency, along with the safety of public health. (Majumder, M. A. A., et al., 2020) The "One Health" approach, is also required to address Antimicrobial Resistance increasing risk.

7.7.1 One-Health Method to Face Antimicrobial Resistance

The cooperative struggle of numerous health science professions globally is to achieve the best health for people, local animals, wildlife, and the surroundings is known as One-Health. (Steele, S. G., et al., 2021) The highest actions taken by the One-Health method consist of infection control strategies and greater awareness in the human population. In the environment sector, The One-Health activities include the proper handling of manufacturing, public, and homes waste, to decrease the whole spreading of the AMR traits among regions. In the animal sector, it concentrates on the actions stimulated by the One-Health method.

7.7.2 Role of Companion Animals in AMR Spread

The companion animal population has intensely improved in the established nations in recent decades. The enlarged "contact" between animals and human beings can cause a greater threat of contamination and the cross-transmission of Antimicrobial Resistance characters. Currently, several epidemiological studies are being applied to observe the occurrence and spreading of Antimicrobial Resistance. Only abuse and misuse of antibiotics are not sufficient for such a huge transmission of resistant Antimicrobials among human beings and domesticated animals. (Galarce, N, et al., 2021) Numerous examinations are being discovered, for example, human-animal transmission is being detected. Moreover, it is assumed that observing non-pathogenic samples and their potential ability to attain resistance traits is a favorable scheme to prevent upcoming resistant strains.

7.8 Conclusion and Future Directions

Since the beginning of the 21st Century, there has been a fast change in administrative, social, and financial realities. The control and prevention of animal diseases, zoonoses and giving new tasks to the veterinary profession are also upgrading universally. In the veterinary profession, Leadership is necessary to move onward with effective strategies and corresponding planned actions. Veterinarians can ensure the production of safe food at food manufacturing factories and farmstead levels. Using the One Health methodology, Veterinarians can decrease antibiotic-resistant infections. Inhibit epidemics of zoonotic infections in humans and animals while increasing food security and protection. An adjacent relationship between Public Health and Veterinary Medicine professionals is important to achieve the best public health. People in industrialized and unindustrialized countries are raising more awareness and are concerning how food is manufactured as Ethical fears in growing industry. If veterinarians are enthusiastic to face ethical tasks arising from the demanding foodstuff animal production systems, their role in the discovery of suitable results can be improved. It is necessary to report upcoming worldwide changes for animals, humans, and the environment. If current situations continue and there are no planned actions that change the current glitches, the food supply veterinary medicine profession will realize the growing demand for veterinary facilities over the next several years. History is full of examples of animal health and zoonotic disease disasters that have been countered. Our increasingly integrated food supply system and developing bio-security fears make this ancient fact progressively significant.

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Chapter 8: Global Crisis And Challenges And Holistic Solutionfor Sustainable Veterinary Management

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Abstract

Effective responses to catastrophes that impact both people and animals are crucial for worldwide veterinarian crisis management. Veterinarians are inevitable in such situations as chemical explosions, fires, and floods. Prior studies have demonstrated that coordination with well-thought livestock evacuation strategies is important in minimizing some of these problems. For an effective crisis management plan, veterinary specialists, animal owners, and crisis responders must work together for the welfare of animals during evacuation. There should be a close collaboration between veterinary and human medicine, focusing on a One Health approach toward interlinked matters to have sustainable veterinary practices. Collaboration between public and private sectors would be excellent for reducing environmental damage caused by veterinary activities such as drug pollution. To develop sustainable livestock development challenges emanating from food insecurity due to global warming must be handled. It is essential that a sophisticated understanding of bioethics and moral decision-making is achieved to solve ethical dilemmas pertaining to veterinary treatment especially where an equilibrium must be established between animal rights and financial gains. Bio marker-based devices and the practice of remote health care are among those factors that have contributed to the change in veterinary diagnostics and interventions, eventually leading to significant improvement in animal health. Precision Livestock Farming can contribute positively to animals' well-being, environmental pollution mitigation, and wealth maximization. On a global scale, the solutions must rely on modern science and technology so that public health is not put at risk and a stable future is ensured.

Keywords: Medicine, social support, stress buffering theory, direct theory, resilience, holistic solution

8.1 Understanding the urgency

8.1.1 Overview of Global Veterinary Crisis Management

Besides the people, the animals are affected by catastrophes like floods, jungle fires, chemical blasts, or earthquakes. Many people, like veterinarians, are trying their best to solve these problems to save the lives of humans as well as animals.

Linnabary and other veterinarians explained the consequences of a study of the crisis discharge of dairy cattle. This study was for the farmer's behavior towards evacuation or discharge, the accessibility of apparatus and persons, discharge time, evacuated cattle, protection of evacuated cattle, and any other possible option if cattle were not moved. The result was that there was a lack of economic weakness among farmers. In consequence, the writer planned a timely and explained analysis of a crisis after preplanning the case of an emergency.

In the USA, the Emergency Operation Center is often overburdened by invites from animal holders, who know that they are under shifting but are not confident about what to do about shifting or discharging their animals. The study explains, how the preparation and sleek procedures between crisis responders and the people can lead to a successful strategy to carry out the conclusion-making process for animal evacuation (Wijnker et al., 2018).

Therefore, all these studies help to create an animal shifting plan, for both the animal and its owners. In this way, farmers can know how to manage the time and accessibility for animals' evacuation (Chilonda & Van Huylenbroeck, 2001).

Data taken from the farm animals tells us that the finance provided to the farm of animals is very low to give the facilities (Howe, 1988).

Animal welfare can be under threat if we do not follow the five rules: liberty from hunger or thirst, physical comfort, liberty from pain or injury, liberty from fear, liberty for normal behavior (Mogoa et al., 2005).

8.1.2 Rationale for Holistic Solution

For sustainable veterinary management, there are some responsibilities for veterinarians. Nowadays, veterinarians working in the manufacturing of animal food should consider the attentiveness of food companies, sellers, users, and the community (van Herten & Meijboom, 2019). The Dutch proposed the rule for animals, which states that veterinarians have a responsibility to safeguard animals.

Figure 2 (Responsibilities veterinarians should think)

The significance of animal welfare also becomes an influential task for the Dutch. They think about themselves to be also in charge of food protection, public health, and growing the money-spinning benefit for farmers (Voogt et al., 2023).

One health gives veterinarians some extent guidance to solve the problems. To find common ways to fix the problem, we start from the first level where there is agreement. It needs closer collaboration between veterinary and human medicine (van Herten & Meijboom, 2019).

8.2 Understanding Global Veterinary Management Crisis

8.2.1 Economic Factors and Resources Allocation

There is a need to appreciate the attitude and resolution phenomenon of minor-scale farmers in the health management of animals to guide the resolution of the plan related to delivering the animal health facilities. Besides this, the participation of the private sector is also helpful in improving the regulation of providing veterinary facilities (Holden, 1999).

Agricultural menage modeling allows the induction and trying out of the theory related to veterinary services, while qualitative models are very suitable for analyzing the factors beyond the farmer's choice. Spending on veterinary services and medicines is a small component of farm financial costs, which greatly benefits farmers (Webber et al., 2014).

8.2.2 Environmental Impact of Veterinary Practices

Medicines are crucial for curing and protecting animals and humans from disease. Besides this, accidental impacts on the environment are also noted. Large quantities of veterinary medicines like antibacterials, antifungals, and parasitic drugs also have a stress impact on the environment (Boxall, 2004).

Veterinary drugs, given to treat and prevent disease in animals, represent the origin of environmental pollution. These drugs can enter the environment and affect the plant's growth (Bártíková et al., 2016).

8.2.3: Social Implications on Animal Health

Social Support is the main key to gaining the physical as well as physiological welfare of animals. For example, as social support is necessary for humans dealing with AIDS, it is also important for animal welfare. Social attitude is an important factor in animal welfare (Boissy et al., 2007).

A scientist made two major theories to define the idea of social support: one is the main effect theory, which explains that social support benefits animals whether they are facing challenges or not. The second is the stress-buffering theory, which explains that social support minimizes the effects of stress (Rault, 2012).

Figure 3 (Theories of social support)

8.3: Emerging Diseases & Pandemic Threats

8.3.1: Zoonotic Diseases: Origin & Threat

Zoonotic origin, infectious agent, or infection giving a threat to the health of humans as HIV. Urbanization and the destruction of natural habitats are the major causes of zoonotic diseases. Factors that can cause exposure to pathogens in humans, wildlife, and other animals can be transmitted by Morse, Schrag & Weiner, etc. (Slingenbergh et al., 2004).

Zoonotic diseases can cause illness as well as severe infections that can lead to death. The transmission of zoonotic pathogens can be caused by close contact with an animal, usually through inhalation or any other process that can cause contamination of the mucous membrane, damaged tissue, etc. (Saba & Balwan, 2021).

8.3.2: Veterinary Epidemiology in a Globalized World

Veterinary epidemiology tells us the effects and causes of interest, like disclosure of the chemical. In this way, veterinary epidemiology helps us to prevent diseases and their cures, which transfer from animals to human beings (Stevenson, 2008).

A veterinary epidemiological study looked into the spread of Salmonella typhimurium from cattle that weren't given growth-promoting medicines to people who were in close proximity to sick animals (Timoney, 1978).

8.3.3 Challenges in Disease Surveillance & Control

The National Disease Control Program has a limited budget to control diseases in different countries, based on a limited number of clinics, hospitals, private sectors, and NGOs. There is a lack of ability to cope with and manage the data, so this is also the reason to analyze the disease. So, there is a need to tell people and train them on how they can handle the disease to reduce the social and economic burden.

Local health workers are also responsible for the diseases that are so infectious and require several days or months of treatment, but local health workers don’t tell the individuals. To some extent, there is late treatment and diagnosis of disease due to the missed management of data. Moreover, local area clinics or local health workers have low capacity to diagnose the disease (Buckee et al., 2018).

8.4: Climate Change & Veterinary Medicine

8.4.1 Climate Change Impact on Animal Health

There are several environmental factors, like water, soil, plants, food quality, and air, that have a considerable impact on animal health. Society and the economy are also other factors that are related to animal health. All these factors can have severe effects on crops, which can lead to bad animal health (Forman et al., 2008). There are different ways of disease occurrence, such as warm-linked illness and density, severe meteorology incidents, when animal manufacturing systems alter the environment, and exposure to or restoration of contagious illness (Lubroth, 2012). There are environmental pathogens that cause disease in animals. We should improve the sanitation system. Manage the resources of inheritance, food security, and animal homing. The effect of climate change is correlated with a number of universal factors that are changing landscapes (Kuraz et al., 2021).

Classification	Disease	Causal agent	Vector

Vector-borne diseases	African-horse sickness Rift valley fever Chagas disease	Orbivirus Phlebovirus *Trypanosoma cruzi*	Anopheles Mosquitoes Bed bugs
Disease-associated with flooding or stagnant water	Anthrax	*Bacillus anthracis*	No vector
	Leptospirosis	*Leptospira interrogans*	No vector
	Cryptosporidiosis	*Cytosporidium spp*	No vector
	Fascioliasis	*Fasciola hepatica*	Snails of *Lymnea*

Table 10 Animal Diseases Affected by Climate Change (Sánchez Mendoza etal., 2020)

8.4.2 Resilience & Adaptation Strategies in Veterinary Practices

For good health in veterinary medicine, one key factor is resilience. We can explain resilience as “a category of procedure distinguished by positive results as well as some severe risks to alteration and also usually known as the capability to rebound from difficulty”. So, we can conclude from the definition that distinctive means for resilience comprise self-belief and inspiration, and circumstantial means comprise applying the crutch, promoting well-being, talk therapy, and grabbing in contemplative practice (McArthur et al., 2021). So, resilience is an energetic phenomenon where different master plans are applied to create an alternative response. We can calculate resilience by using common resilience scales, like the Brief Resilience Scale or the Cannor-Davidson Resilience Scale, or by measuring the elements of resilience. For the use of distinctive means for flexibility in veterinary practice, the Veterinary Resilience Scale Personal Resources (VRS-PR) has been designed (Lubroth, 2012).

8.4.3: Sustainable Livestock Management in a Changing Climate

Food issues and livestock problems are caused by global warming, which produces heat pressure and greenhouse gases (GHG). So, to solve the food issues and livestock problems, we have to implement preventive procedures. The threat of food issues and livestock problems is giving us the signal of necessity for feasible livestock presentation in relation to a climate-smart livestock system that protects us from food problems and our population in the coming days. So it is important to make a system that provides sustainable livestock management and also protects our food (Park, 2022).

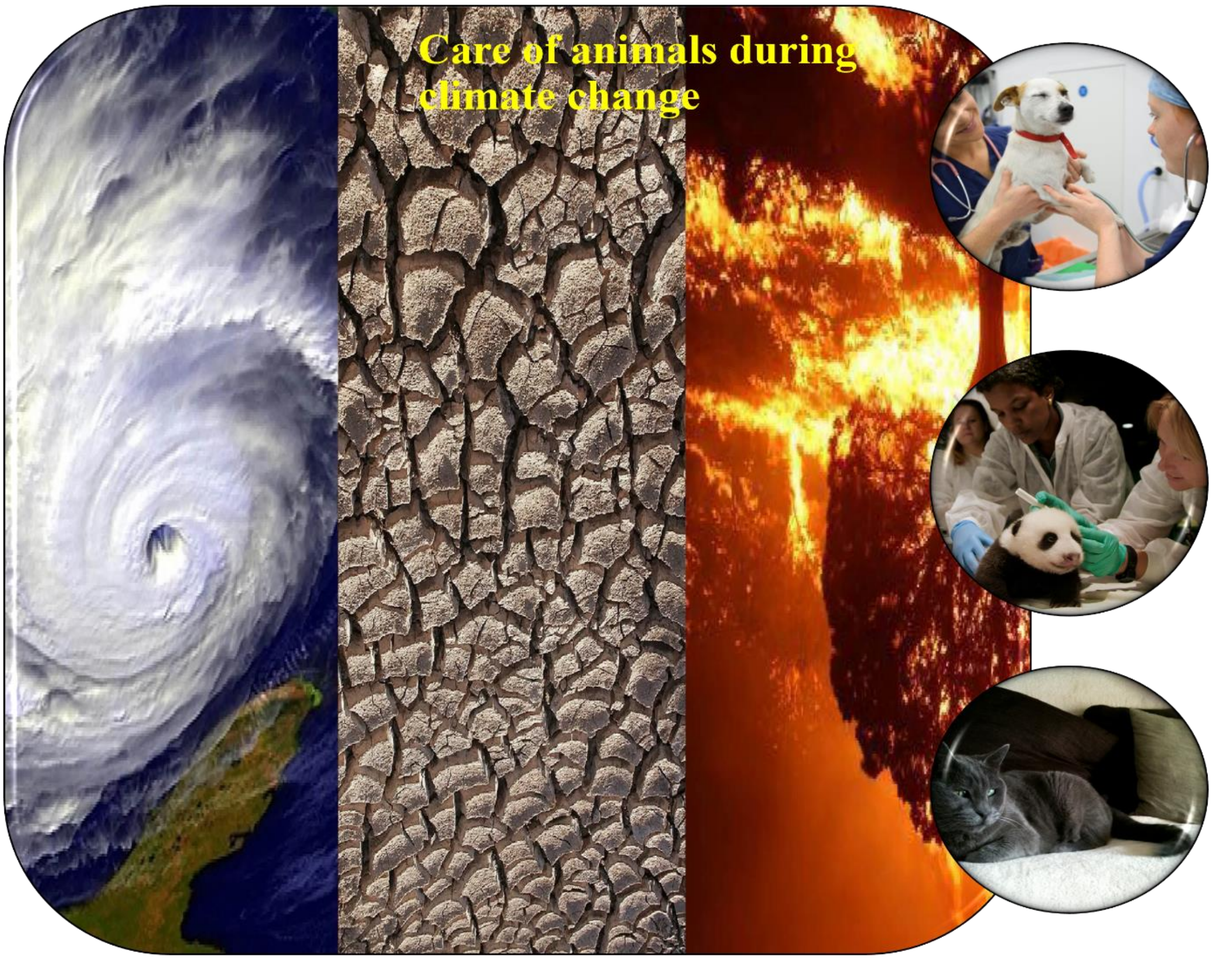

Figure 4 (Animal care during changing climate) Systems:

The important elements of a food system are models and sensors. For obliging the flightiness of important livestock production processes, the models are used (Koytcheva et al., 2021).

8.5: Biodiversity Loss & Conservation

8.5.1: Veterinary Role in Biodiversity Preservation

Veterinarians are trying to improve the fitness and comfort of imperial species and improve animal safety and welfare through advanced vaccination and other different pharmaceuticals (van Herten & Meijboom, 2019).

There is an important role for wildlife diseases in wildlife preservation. While advancing in wildlife development, there is a great role in global environmental changes. If on one side a misplacement of biodiversity has been causing the increased risk of imperial species, then on the other side some opportunistic taxa have enlarged their ampleness (Lanfranchi et al., 2003).

8.5.2: Conservation Medicine: Integrating Wildlife & Livestock Health

In an environmental competition, the idea of fitness for both animals and humans should be appreciated. The preservation of medicine has a focus on ritualizing and developing this cross-functional outlook. Conservation medicine helps us to improve animal and human health through several practices (Patz et al., 2004).

Advocates for the preservation of medicine contend that only an environmental view can support healthcare providers in comprehending the phenomenon of illness, following a predictive example can give profit to environmentalists (Norris, 2001).

The preservation of medical science is a field that is yet attempting to explain itself. On the other hand, the preservation of medicine is helpful in the protection of illness and natural systems (Patz et al., 2004).

8.5.3: Mitigating Human-Wildlife Conflicts

Studies used a number of types of mediation to minimize these human-wildlife disputes. Eight types are classified as direct (minimizing the number of meetings with wildlife) and five types as indirect (enhancing human forbearance) mediation (Treves et al., 2009). Studies are examining the reason-and-effect connection to clarify the main point of mediation for every type. We should handle wildlife and humans by making plans for mediation or intervention in a systematic way, using merit criteria, and enhancing the involvement of experts, legislators, and impacted communities. Studies expect that if we handle all these steps in a systematic way, then the result will be more effective (Massei & Cowan, 2014).

8.6 Ethical Dilemmas in Veterinary Care

8.6.1 Animal Welfare VS Economic Interest

For the advancement of farm animal welfare, we assess four attitudes by viewing the threats from an environmental perspective. These four attitudes are judicial enterprise, enterprises handled by producers, consumer selection, and food industries. For good advancements in animal welfare, the union of all four attitudes is necessary.

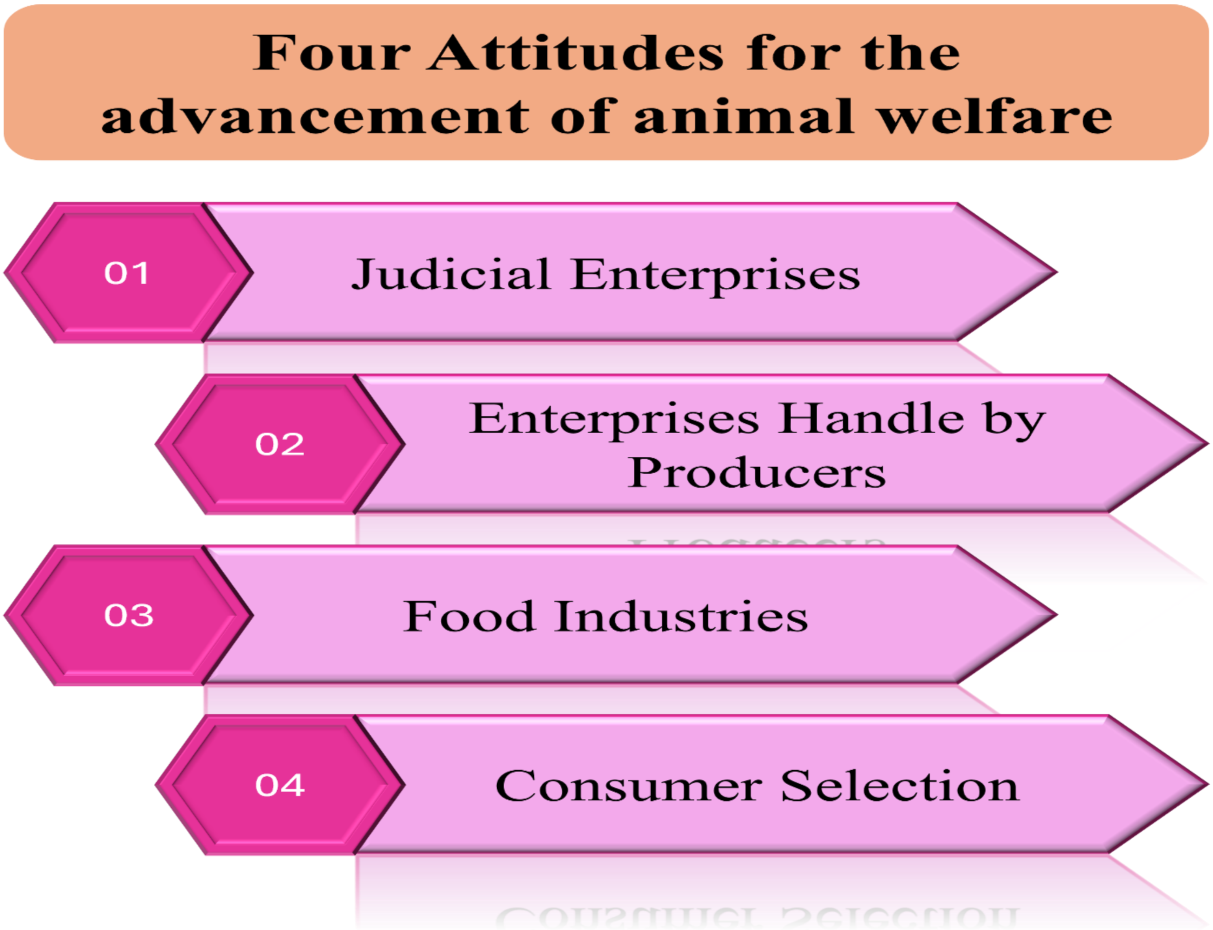

Figure 5 (Four attitudes for the advancement in animal welfare)

To understand the welfare opportunities enterprises, handle for producers, appreciating the human-animal relationship is necessary. For the best use of enterprises handled by consumer's selection, we see not only the animal welfare but the other welfare extents observed as necessary by veterinarians also (Christensen et al., 2019).

8.6.2: Bioethics in Veterinary Decision Making

Veterinary moral principles are a combination of rules that employ morals, ethics, and judgments in the application of a veterinary occupation. In veterinary morals, there is a combination of veterinary professional morals and the view of animal morals. Several moral problems arise in a business entity in veterinary application or exercise (Vettical, 2018).

8.6.3 Ethical Decision:

To sort out moral dilemmas, it is necessary to understand the ethical theories. During moral dilemmas in a veterinary practice, a powerful dedication to character, the corporation's dedication to morals, and employee support can help to reduce the extent of ethical stress when facing moral dilemmas in veterinary practice.

For making the actual decision, a better understanding of moral problems is important for connection, good task achievement, and protecting a good social outline for both sole and veterinary applications (Vettical, 2018).

8.7 Technological Innovations & Veterinary Medicine

8.7.1 Advancements in Diagnostic Techniques

- **Biosensors:**

  It can measure the specialized framework that covers an animal's physiology and examine its habitat. Biosensors also play a key role in farm animal handling; they can detect disease, observe health, detect reproductive phases, and analyze the environment. The data we will collect can help the farmers improve their animal production in the future (Neethirajan et al., 2017).

- **Advances in Engineering Research:**

  With the development of engineering research, there has been a decrease in the price of electric technologies, resulting in the exposure of sensing solutions that help to connect and provide an easy way to collect and analyze data (Croney et al., 2018).

8.7.2: Telemedicine & Remote Consultations

As telemedicine sets off the most conventional career in veterinary medicine, it is necessary to appreciate when and how to use it properly. This will help to prop up the utilization of veterinary e-medicine for telehealth and apply the frippery and m-Health apparatuses to monitor animal welfare. Telemedicine is specifically affected in areas where there is less access to care because of the economy, finance, geography, or limited resources. Overall, veterinary telehealth and telemedicine can yield positive results (Hess, 2017).

8.7.3: Precision Livestock Framing

Precision Livestock Farming (PLF), if properly applied, will improve animal welfare in industry, minimize greenhouse gas (GHG) release, appreciate the social presentation of farms, improve livestock retailing of farm animal products, minimize outlaw marketing of farm animal products, and improve the firmness of rustic areas (Banhazi et al., 2012).

The following counseling is applied to make PLF possible in farms: make a new helping hand industry; find, reveal, and make the benefits of PLF public; connect the attempts of several industries and organizations; appreciate the trading sectors; and connect with occupationally controlled product advancement (Banhazi et al., 2012).

8.8 Globalization of Food Supply Chains

8.8.1 Food Safety & Security Challenges

Global shifts comprised of social change, flourishing, and maturing populations, development, and enhanced wealth will develop food safety challenges and produce new demands on producers, constructors, traders, sellers, and controllers (Borras & Mohamed, 2020). Advancements in scientific technologies like whole genome sequencing, smart packaging, advancements in detecting and following technologies, details calculating technology, and large data examining can minimize the challenges and complete requests, but they can also produce new challenges. For underdeveloped countries, developed economies, and big food industries, it is very difficult to sort out these challenges (King et al., 2017).

8.8.2 Supply Chain Resilience & Risk Management

The focus of this study is to examine the connection between precursors of supply chain flexibility consisting of threat handling culture, supply chain joining, evidence, cooperation, and nimbleness, their effect on supply chain flexibility, and eventually on unyielding presentation. By using this literature, a model has been developed.

The main purpose of this model is to improve supply chain flexibility by making the threat-handling culture better, improving threat knowledge in people, and managing regular threat examination exercises.

By embracing knowledge and transmission technologies and other tools for fast reply, companies can maintain flexibility in food chain supply and connection and minimize supply chain destruction (Kumar & Anbanandam, 2020).

8.9 Socioeconomic Disparities in Animal Health

8.9.1 Access to Veterinary Care in Developing Regions

The government did not make a policy of instructions for veterinary prevention, but the private companies made a policy for laboratory animal doctors. This policy includes instructions regarding the suitable qualifications for the animal doctors that are doing work on laboratory animals, the power of lab animal doctors, and the clear character of a veterinary doctor in research occupations, and the analysis of agreements requiring lab animals (Zurlo et al., 2009).

8.9.2: Community Engagement & Empowerment

Social significance in analyzing environmental progression and advancement has been understood. Still, small experimentation has literally gazed into how environmental capital characterizes and what part it will play in public advancement enterprises (Rudito et al., 2022).

8.10 Conclusion

Disease Control Programs have a limited budget to control diseases in different countries, based on a limited number of clinics, hospitals, private sectors, and NGOs. There is a lack of ability to cope with and manage the data, so this is also the reason to analyze the disease. So, there is a need to tell people and train them on how they can handle the disease to reduce the social and economic burden.

The threat of food issues and livestock problems is giving us the signal of necessity for feasible livestock presentation in relation to a climate-smart livestock system that protects us from food problems and our population in the coming days. So, it is important to make a system that provides sustainable livestock management and also protects our food.

Systems:

If the role of the system is to maintain food production for animal health, the examiner has to measure the input values, which help to maintain growth and food criteria. The important elements of a food system are models and sensors. For obliging the flightiness of important livestock production processes, the models are used.

For sustainable veterinary management, there are some responsibilities for veterinarians. A modern veterinarian should take into account the global outlook and take responsibility for taking care of animal welfare as well as the welfare of humans. In the present day, veterinarians in animal grazing should think about the attentiveness of food companies, users, sellers, and the community. The Dutch proposed the animal law, which declares that veterinarians have a responsibility to protect animals. In day to day life, veterinarians have to cope with problems where animals and humans are in a fight and no simple solutions are accessible. The significance of animal welfare has also become a crucial topic for the Dutch. They think of themselves as being in charge of food protection, human health, and growing the money-spending profits of farmers.

Fulfilling all these responsibilities, which are necessary for animal as well as human health, is not easy. One Health gives veterinarians some guidance to solve the problems. To find common ways to fix the problem, we start at the first level, where there is agreement. There is a need for closer collaboration between veterinary and human medicine.

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Chapter 9 : Emerging Technologies And Innovations In Aquatic Veterinary Practices

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Abstract

The veterinary sciences have advanced significantly in the last few decades thanks to the development in technology a deeper understandings of animal biology and a growing emphasis onanimal welfare. Groundbreaking discoveries in veterinary sciences are transforming the field and altering the way that animals are cared for and treated. This chapter provides a comprehensive review of the most recent break throughs in the filed with a focus on improvement in treatment modalities, diagnostics tools and preventive health care. We examine how automation and robotsare revolutionizing the field of aquatic veterinary animals' research by enabling precise disease detection and state of the art therapeutic strategies. The employment of cutting-edge imaging andtracking technologies to collect chemical and physical data on aquatic creature submerged in watergreatly improves data rectification. This chapter also covers advance in veterinary surgery such asuse of robotics aid and less invasive techniques that improves outcomes and reduce recovery time.We discuss the development of aquatic veterinary medicine, the rise of individualized medicine for specific animals, and the development of innovative vaccines and biologics in the therapeuticsindustry. By increasing accessibility and diagnostic accuracy, the integration of digital technologies such as telemedicine and artificial intelligence is further modernizing veterinary operations. We also discuss the significance of the One Health concept, which emphasizes the connection between animal, human, and environmental health and promotes teamwork in addressing zoonotic illnesses and global health issues. This chapter is to give veterinarians, researchers, and students a thorough understanding of the future directions in veterinary sciences while emphasizing the ongoing efforts to improve animal welfare and health via scientific and technical developments.

Key words: Technological advancement, Robotics and Automation, challenges and future direction, solution

9.1 Introduction to aquatic veterinary practices:

Aquatic veterinary practices involve the healthcare of aquatic animals, including fish, amphibians,reptiles, and invertebrates. It includes preventive care, diagnosis, treatment, and management of diseases and injuries in aquatic environments(Farkas, Mannion et al., 2020). These professionals often work in aquariums, aquaculture facilities, research laboratories, and government agencies toensure the health and welfare of aquatic species(Narwal, Katyal et al. 2023). Key areas include water quality management, nutrition, disease prevention, and surgical interventions tailored to aquatic species(Volpe, Errani *et al.*, 2023).

9.2 Evolution of Aquatic veterinary medicine:

The evolution of aquatic veterinary medicine has been shaped by various factors, including advancements in veterinary science, the growth of aquaculture, and increased understanding of aquatic ecosystems. Here's an overview of its evolution(Volpe, Errani *et al.*, 2023):

9.2.1 Early Beginnings:

Aquatic veterinary medicine has roots in the early efforts to manage the health of fish populationsin aquaculture systems. Historically, the focus was primarily on disease prevention and basic husbandry practices. To interpret pathology in aquatic patients, veterinarians must perform timelyand thorough necropsies, paying attention to histopathology differences such as: B. Absence of distinct adrenal glands or pancreas, or identification of pelvic organs or rectal glands. Diagnostic interpretation is another challenge when working with aquatic patients, and all veterinary studentsshould be familiar with the clinical chemistry, histopathology, and hematology of common finfish.(Farkas, Mannion et al. 2020, Volpe, Errani *et al.*, 2023).

9.2.2 Expansion of Aquaculture: As aquaculture expanded globally to meet growing demand for seafood, the need for specialized veterinary care for aquatic species became more apparent. This led to the development of dedicated aquatic veterinary programs and organizations(Sood, Pradhan et *al.*,2024).

9.2.3 Advancements in Veterinary Science: Advances in veterinary diagnostic, therapeutic andsurgical techniques have given aquatic veterinarians access to a wider range of tools and treatments for the health of aquatic animals.

9.2.4 Disease Challenges: The emergence of new diseases and spread of existing ones in aquaticenvironments posed significant challenges for aquatic veterinary medicine. This spurred research into disease prevention, detection, and control strategies(Shanmuganathan, Sibtain Kadri *et al.,* 2023).

9.2.5 Regulatory Frameworks: The development of regulatory frameworks for aquatic animal health, such as the Aquatic Animal health Code by World Organization for Animal Health(OIE), helped standardize practices and promote collaboration among countries in managing aquatic animal diseases(Narwal, Katyal *et al.,* 2023).

9.2.6 Interdisciplinary Collaboration: Aquatic veterinary medicine increasingly involves collaboration with other disciplines, including fisheries biology, ecology, and environmental science. This interdisciplinary approach is essential for addressing complexissues affecting aquatic animal health and welfare(Narwal, Katyal *et al.,* 2023, Cheng, He*et al.*, 2024).

9.2.7 Technological Innovations: Advances in technology, such as aquaculture automation, remote monitoring systems, and genomic sequencing, have enhanced the ability of aquaticveterinarians to diagnose diseases, monitor water quality, and improve overall managementpractices(Lennox, Aarestrup *et al.,* 2017, Zheng, Zou *et al.,* 2024).

9.2.8 Focus on Conservation: With growing concerns about biodiversity loss and habitat degradation, aquatic veterinarians are increasingly involved in conservation efforts to protect endangered aquatic species and their ecosystems(Sood, Pradhan *et al.,* 2024).

9.2.9 Education and Training: The establishment of specialized educational programs and certification courses in aquatic veterinary medicine has helped train a new generation of professionals equipped to address the unique challenges of caring for aquatic animals.

Overall, the evolution of aquatic veterinary medicine reflects a growing recognition of theimportance of aquatic ecosystems and the need to ensure the health and welfare of aquatic animalsfor sustainable food production, conservation, and ecological balance.

9.3 Cutting-edge technologies in aquatic health monitoring:

Cutting-edge technologies are revolutionizing aquatic health monitoring, offering new ways to monitor, diagnose, and manage the health of aquatic animals. Here are some examples:

9.3.1 Remote Sensing and Monitoring: Remote sensing technologies, such as satellites, drones,and underwater vehicles equipped with sensors, allow for real-time monitoring of water quality parameters like temperature, pH, dissolved oxygen, and algae levels. This data helps identify potential stressors and disease outbreaks before they escalate (Zheng, Zou *etal.,* 2024).

9.3.2 Biosensors: Biosensors are devices that detect specific biological molecules or markers that indicate the presence of health or disease. When monitoring the health of aquatic organisms, biosensors can be used to detect pathogens, toxins, or biomarkers of stress in water samples or directly in aquatic animals. (Rowan and Kalacska 2021).

9.3.3 Genomic Sequencing: Advances in genomic sequencing technologies enable rapid and accurate identification of pathogens in aquatic environments, helping aquatic veterinariansdiagnose disease, track the spread of pathogens, and develop targeted treatment strategies.

9.3.4 Aquaculture Automation: Automation technologies, such as automated feeding systems,water filtration systems, and monitoring platforms, streamline aquaculture operations while minimizing stress on aquatic animals. These systems can also provide real-time dataon feeding behavior, growth rates, and water quality parameters(Lai, Peng *et al.,* 2024).

9.3.5 Imaging Techniques: Imaging techniques like ultrasound, CT scans, and MRI are increasingly used in aquatic veterinary medicine for diagnosing internal injuries, tumors, and reproductive conditions in aquatic animals. These non-invasive techniques allow for accurate diagnosis and treatment planning.

9.3.6 Environmental DNA (Edna) Monitoring: Edna monitoring involves collecting and analyzing environmental samples to detect traces of DNA shed by aquatic organisms. This non-invasive approach can be used to assess species diversity, detect invasive species, andmonitor changes in aquatic ecosystems.

9.3.7 Artificial Intelligence (AI): AI algorithms are being developed to analyze large datasets generated from aquatic health monitoring efforts. These algorithms can identify patterns, predict disease outbreaks, and optimize management practices for improved health and productivity(Lai, Peng *et al.,* 2024).

9.3.8 Microfluidics: Microfluidic devices enable precise manipulation and analysis of small volumes of liquids, making them valuable tools for monitoring the health of aquatic environments. These devices can be used for rapid diagnostic testing, drug screening, andto study microbial communities in aquatic environments.

9.3.9 Block chain Technology: Block chain technology provides a secure and transparent wayto track and record data related to monitoring the health of aquatic organisms, such as waterquality measurements, disease spread and treatment records. This helps ensure dataintegrity and traceability in aquaculture and conservation efforts.

These cutting-edge technologies are transforming aquatic bio health monitoring practices, providing new insight into the health and well-being of aquatic animals and ecosystems, helpingto advance sustainable aquaculture and conservation efforts. (Bharathi S, Perdana *et al.,* 2024).

9.4 Recent advancement in Aquatic Animals Welfare

One of the planet's most important ecosystems, the ocean contains the majority of the biosphere. It is essential to the cycling of mater and energy as well as the survival of a wide variety of livingorganisms (Purkis, S. and Chirayath, V. 2022). Changes in human behavior directly effects changesin marine aquatic environment and the behavior of marine creatures since social production is necessary for both. Thus, to ensure the preservation of robust and healthy marine ecosystem, specific and targeted interventions techniques based on a thorough understanding of the maritimestate are needed. (Duarte, C. M. 2014, von Storch, H. 2004, Cognetti, G. and Maltagliati, F. 2010).Animals' movement are driven by their basics need for migration, breeding, avoiding natural predators, and predation. (Bowler, D. E. and Benton, T. G. 2005).

For instance, whale sharks migrate to warmer water in response to seasonal variation in sea temperature, freshwater eels swim deeper through their reproductive cycles, and whale sharks favor warmer tropical seas where they may reproduce and feed. These motions aid in the flow of nutrients, biomass changes, and the transformation of dynamic energy across the ecosystem to preserve its health and identify the maximum amount of aquatic productivity. But in the modern globalized and industrialized era, human activity has improved transportation, seafood processing,fishing, and other related enterprises (Erbe, C. *et al.*, 2019). In addition, problems such as pollutions spills and overfishing may have detrimental long-term effects on the maritime environment. For example, the release of contaminants such as heavy metals can have detrimentaleffects on the quality of water and the subaquatic ecosystem (Yuan, G. L. *et al.,* 2011, Gong, X. *etal.*, 2021). Global warming and overfishing can subtly impact the natural habits of marine animals(Wright, P. J. and Rowe, S. 2019).

Using a variety of sensor to gather data on environment and physiological parameters is one of themost crucial strategies to reduce the influence of human activities on biological processes. Problems found after data gathering are looked into and resolved. To that end, seawater tags haveeither been sunk into the animal's flesh or permanently affixed to its surface (Cooke, S. J. *et al.,* 2004) Animal tracking and electronic tagging technologies can also provide information on an animal's location, range, and health (Block, B. A. *et al.*, 2011). Because marine animals can naturally perceive places that are inaccessible to traditional sensors, using them as a sensing platform has several benefits.

9.4.1 Technological Advancements in Wearable Flexible Sensors and Implantable Tags forAquatic Species

Sophisticated electronic technologies have been extensively used in the last few decades to build a wide range of intricate maritime environmental monitoring systems (Sun, K. *et al.,* 2021). By analyzing the data transmitted from the monitoring system to the control center, people can quicklydetermine and put into practice mitigation strategies that can greatly lessen the impact of the pollution release and adverse weather on the marine environment. (Sun, K. *et al.,* 2021, Xu, G. *etal.,* 2019). Electronically tagged marine animals can be used by researchers as marine sensing platform to study the physiology and ecology of the species, in addition to the animals themselves(Gallo, A. *et al.,* 2022) but also, be used as in situ oceanographic measurements to monitor the environmental parameters of the ocean (Johnson, M. P., and Tyack, P. L. 2003).Traditional markers, including satellite-linked transmitters and pop-up archive (PAT) tags, are fixedly attached to organisms, which raises the risk of equipment and data loss, and thus offer fewer application alternatives.. (Hammerschlag, N. *et al.*, 2011). Due to the complex traditional fabrication process,it may be difficult to capture analytes since rigid sensors are rigid (Teo, S. L. *et al.,* 2009, Gallagher,

M. J. *et al.,* 2022, Keates, T. R. *et al.,* 2020, Bailleul, F. *et al.,* 2015) perhaps leading to insufficientsignal transduction (Liu, E.*et al.,* 2023). These kinds of sensors placed on the surface of fish and other aquatic species may have an effect on their overall health and mobility. Nonetheless the useof flexible electronics which can function with moving parts and on any kind of surface helps to reduce signal interference brought on by the material characteristics of the device, producing exceptional output quality (Han, S. T. *et al.*, 2017).

Furthermore, flexible sensing technology preserves the animals' biological compatibility while reducing the invasiveness of electronic tags on animals. The ocean has enormous potential value for the biodiversity. Insufficient marine data leads to under sampling, which further hinders researchers' ability to fully understand biodiversity (Costa, D. P. *et al.*, 2012, Jacob, C. *et al.,* 2020).Flexible wearable sensors are currently being used in the field of marine ecology to gather multi-technology data (Cheng, R. *et al.*, 2020). Because of its remarkable flexibility and scalability, flexible electronics like traditional integrated circuit (IC) technology, us primarily driven by manufacture methods and equipment. Technologies like as micro-electromechanical system (MEMS), flexible hybrid electronics and biological telemetry have influenced the design of the device's construction and method of detection. Electronic devices, both inorganic and organic, areintegrated onto thin metal or flexible polymer substrates (Bao, M. and Wang, W. 1996).

Biotelemetry approaches cover a wide range of biomarkers in animal of all sizes, and information gathered by monitoring devices can be Utilized to evaluate the causes and consequence of biological movement (Hussey, N. E. *et al.*, 2015). The gadget must be kept as waterproof as feasible due to the complex and demanding underwater environment in order to guard against potential damages from seawater corrosive properties and to guarantee excellent sensor performance when used frequently. An essential component of using and exploiting marine resources in marine monitoring. The development of wearable flexible sensing technologies for maritime environment monitoring will be aided by these technologies.

9.4.2 Flexible Electronic Tags for Tracking Physical Data Underwater:

Most biological wearable flexible sensor used for under water parameters monitoring are found onthe surface of marine animal. While the animal is immersed these sensors consciously monitor itssurroundings collecting data in real time. The marine environment is primarily composed of three elements: depth, temperature, and conductivity. Sensors that measure conductivity, temperature, and depth (CTD) are commonly used to keep an eye on these three variables. A CTD sensor can be mounted on an animal's surface to provide qualitative firsthand data on the physical propertiesof the habitats it lives in and feeds (Lydersen, C. *et al.*, 2002). The incorporation of CTD sensors with flexible and elastic materials enhances the compatibility of electronic tags with aquatic animalsurfaces.

Consequently, the notion of Marine skin was presented. Nasar et al., reduced the size, weight ad intrusiveness of devices peripherals while integrating Bluetooth a memory chip and high-performance sensor (Nassar, J. M. *et al.*, 2018). To preserve the devices durability and stability, they also covered it in hydrophobic materials. The sensing system is made up of a salinity detecting module that uses the traditional conductivity measurement concept. A pressure senso with a parallel plate capacitance structure for depth sensi g and a resistance temperature detector (RTD) for temperature sensing. The integration of these sensor modules with a low-power Bluetooth transceiver and a Programmable System-on-a-Chip (PsoC) allows for effective energy conservation (Figure 2a). Even though salinity and water density can have an effect, underwater pressure is associated with depth. Temperature and salinity cause saltwater's density to alter gradually, therefore data processing has to take this into consideration when calculating depth measurements. The data recorded by an animal wearing a tag is periodically put to sleep and is momentarily stored in the internal flash memory of the PsoC when the animal submerges. Data is sent to a surface-level floating receiver only when the animal swims in a horizontal plane, whichis when the Bluetooth transmitter activates. The earlier study states that the retractable soft jacketpackage incorporates electronic skin to obtain a very small size and weight (less than 0.5 g in the air) (Figure 2b), allowing for the detection of smaller fish without the use of biological adhesive. (Shaikh, S. F. *et al.*, 2019). This work aimed to optimized many sensing modules. For instance, the original electrode structure of the salinity sensor was replaced with an interdigital electrode structure. The PDMS elastomer, which serves as the dielectric layer, was made thicker and had its raw material ratio changed to increase compressibility. Nearly two kilometers of water make up the Red Sea, and the entire sensor system's sensitivity is many times more than it was in the previous iteration, allowing it to function reliably for four weeks (Ullo, S. L., and Sinha, G. R. 2020).

Sensitivity can be increased by changing the electrode material used in the sensor layer. He and his colleagues replaced the copper/polyimide (PI) bottom electrode of the pressure sensor with a c arbon nanotube (CNT)/PDMS composite (Figure 2c) (He, Q. et al., 2022). This update has improved the wearable CTD electronics tags sensing capabilities. It was created specifically for aquatic animals. The kirigami structure of a waterproof biological electronic tag was designed to ensure the device's stretchability and sensitivity. It was intended to operate continuously at pressures of up to 5 Mpa (Figure 2d) (He, Q. *et al.*, 2023)

9.4.3 Flexible Electronic Tags for Tracking Subsurface Chemical Data

In addition to real time physical information monitoring underwater chemical information monitoring is extremely important. Currently beings discovered are many additional orts of chemical information, surveillance equipment. With an emphasis on wearing real-time monitoringsystems for aquatic species, research in this field is still in its early stages.

9.4.3.1 Ph Sensors

Whole the fields underwater flexible Ph sensor research and development I still in its early stages.One possible application for these sensors could be wearable electronic tags that can react instantlyto Ph variations in the ocean. Glass electrodes were originally often used as Ph sensors, but because of their brittleness and incapacity to reduce flaws, they were not appropriate for the addition of flexible tags (Karastogianni, S. *et al.,* 2016, Salvo, P. *et al.,* 2018). Gao et al., developeda Ph sensor using a microfluidic chip and a 133holera133 field-effect transistor (Gr-FET) embedded on a flexible PI substrate (Gao, J. *et al.,* 2020). A flexible sensor that can efficiently measure seawater Ph in real time and replace glass electrodes has been developed thanks to its innovativedesign. However, the presence of various ions in seawater and their varying ionic strengths alwaysaffect the sensing system's performance.

9.4.3.2 Metal Ion Sensors

Monitoring of heavy metal contamination is necessary to protect the marine ecosystem. This is why scientist are constantly working to create flexible sensor that are capable of tracking differentmetal ions in seawater. The goal is to use these sensors as marine electronic skins, tracking the liquid environment surrounding moving animals. Hui et al. selected Bi-Sn alloy as the modificationmaterial for the flexible multifunction sensors' metal ion-sensitive layers' working electrode surface. (Hui, X. *et al.,* 2022). The micro-dendritic microstructure of the Bi-Sn alloy and Sn, whichpossesses properties similar to Bi, can both boost the stability of Bi. (Osório, W. R. *et al.*, 2013). This increases the surface area of the sensor layer, which facilitates the provision of additional places for the detection of the adsorbate metal ions Pb2+ and Cd2+.

9.4.3.3 Dissolved Oxygen Sensors

In addition to being a necessary nutrient for marine life Dissolved oxygen is a source of energy that kept living things healthy and active. The dynamic fluctuations of dissolved oxygen can be monitored in real time to provide insight into the environmental conditions that promote biologicalviability. Three electrode devices or organic electrochemical transistors (OECTs) can be utilizedfor this purpose. OECTs combine electrochemical reactions with organic materials in a low power,extremely sensitive electronic device. Ion implantation in electrolytes is frequently used as a channel material, employing hydrophilic and ionic permeability polymers to control the total conductivity of organic semiconductor channels. OECTs' built-in signal amplification capabilitiesmake them a great option for small ocean sensors (Gueskine, V. *et al.,* 2020). Additionally, they are extremely sensitive in identifying certain metabolites in water.

9.5 Robotics and Automation in Aquatic Veterinary Practices

Robots are being transformed from rote machines into cognitive collaborators by the sensor-driven revolution of today. They must develop into an essential component of a dynamic continuum thatincludes people, various machines, and the digital settings in which they work. Robots have greatlybenefited our businesses by making it possible to produce a wide range of devices, household appliances, vehicles, and processed foods in an efficient and cost-effective manner. Unquestionably, the twenty-first century has brought about incredible advancements and discoveries. Robots has emerged from science fiction to real-world global events, performing complex tasks and changing the world we live in (Khatib, 1986). Recent advancements in roboticshave produced a number of ways that humans may coexist with robots. The introduction of robotswill probably have a significant impact on how productive a business enterprise is. Robots are meant to provide additional automation capabilities to the business and have the ability tocompletely change the way organizations operate (Ogurtsov *et al.,* 2014).

This is a quick overview of the areas in which robots are very useful as well as how they might beintroduced and used in the veterinary industry. It also provides information on the specifics of robot applications in various nations. Thus, a very significant advancement in the current generation can be made if robots are implemented in the veterinary area in India. According to Khatib (1986), robots should not only be used as human companions but also as pet-friendly devices. Sensor-based devices called robotic fish are used to measure and examine many aspects of water quality, such as temperature, turbidity, dissolved oxygen, carbon dioxide, and so forth. The fish’s heart rates are also measured by the robotic fish. They may be linked to a smartphone, which makes it simple for us to gather data. The robotic fish autonomously navigate through the water while keeping an eye on every aspect of the water’s quality (Ogurtsov, et al., 2014). The useof robots and automation in aquatic veterinary procedures is transforming and opening up new avenues for managing the health and welfare of aquatic species. This chapter highlights cutting- edge advancements and their implications on the area of aquatic veterinary medicine while examining the many applications, benefits, and challenges of automation and robots.

9.6 Applications of Robotics and Automation in Aquatic Veterinary Medicine

9.6.1 Automated Health Monitoring Systems

Aquatic creatures are now able to have sophisticated health monitoring systems because to roboticsand automation. These systems continuously monitor vital signs, identify anomalies, and send out early alerts about possible health problems thanks to sensors and artificial intelligence (Kumar etal., 2024).

9.6.2 Water Quality Monitoring:

- Automated systems measure parameters such as Ph, temperature, dissolved oxygen, andsalinity.
- Real-time data helps maintain optimal living conditions and prevent disease outbreaks.

9.6.3 Behavioral Monitoring:

- AI-driven cameras and sensors track the movement and behavior of aquatic animals.
- Changes in behavior can indicate stress, illness, or environmental problems.

9.6.4 Robotic Surgery and Treatment

In order to improve outcomes and lessen the need for human intervention, robotic technologies arebeing used to precisely operate on and treat aquatic creatures.

9.6.5 Minimally Invasive Surgery:

- Robots perform delicate operations with high precision, minimizing tissue damage andrecovery time. Examples include fin repair and removal of tumors.

9.6.6 Automated Medication Delivery:

Systems deliver medications directly to individual animals or through water treatmentEnsures accurate dosing and reduces stress on the animals.

Table 11: Comparison of Traditional vs. Robotic Surgery in Aquatic Veterinary Medicine

Aspect	Traditional Surgery	Robotic Surgery
Precision	Moderate	High
Recovery Time	Longer	Shorter
Stress on Animals	Higher	Lower
Human Intervention	Required	Minimal

9.6.7 Automated Feeding Systems

Automation in feeding systems ensures consistent and efficient nutrition delivery, optimizinggrowth and health of aquatic species.

9.6.8 Precision Feeding:

- Automated feeders dispense the right amount of food at optimal times.
- Reduces waste and ensures all animals receive adequate nutrition.

9.6.9 Customized Diets:

- Systems can be programmed to deliver specialized diets based on species, age, and healthstatus.
- Enhances growth rates and disease resistance (Kumar et al., 2024).

9.7 Benefits of Robotics and Automation in Aquatic Veterinary Medicine

9.7.1 Enhanced Efficiency and Productivity

Automation and robotics simplify a number of procedures in aquatic veterinary clinics, greatlyincreasing output and efficiency.

9.7.2 Labor Reduction:

Automated systems reduce the need for manual labor, allowing veterinary staff to focus on morecritical tasks.

Decreases labor costs and improves operational efficiency.

9.7.3 Increased Precision and Consistency:

- Robots perform tasks with high precision and consistency, reducing errors and variability.
- Leads to better health outcomes and higher quality of care (Bohara et al., 2024).

9.7.4 Improved Animal Health and Welfare

The use of robotics and automation enhances the health and welfare of aquatic animals throughcontinuous monitoring and precise interventions.

9.7.5 Early Disease Detection:

Early detection of health problems by automated systems allows for timely treatment and lowers mortality rates. Enhances productivity and general health of the herd.

9.7.6 Stress Reduction:

Minimally invasive robotic procedures reduce stress on animals during medical interventions. Automated feeding and monitoring systems create a more stable and comfortable environment(Bohara et al., 2024).

9.8 Challenges and Considerations

9.8.1 Technical and Operational Challenges

Despite the numerous benefits, the adoption of robotics and automation in aquatic veterinarypractices comes with several challenges.

9.8.2 Technical Complexity:

- Implementing and maintaining advanced robotic systems requires technical expertise andongoing support.
- Potential for system failures and the need for regular maintenance.

9.8.3 Integration with Existing Systems:

Integrating new robotic technologies with existing aquaculture systems can be challenging. Requires compatibility and seamless data integration.

9.8.4 Economic and Ethical Considerations

Economic and ethical factors must be considered when adopting robotics and automation in aquatic veterinary practices.

9.8.5 Cost of Implementation:

High initial costs for purchasing and installing robotic systemsNeed for cost-benefit analysis to justify the investment.

9.8.6 Ethical Implications:

Considerations around the use of automated systems in animal care.

Ensuring that automation enhances rather than compromises animal welfare.

Table 12: Key Challenges in Implementing Robotics in Aquatic Veterinary Practices

Challenge	Description	Potential Solutions
Technical Complexity	Requires technical expertise andmaintenance	Training and support programs
Integration Issues	Compatibility with existing systems	Standardization of technologies
Cost of Implementation	High initial investment	Financial planning and grants
Ethical Considerations	Impact on animal welfare	Ethical guidelines and oversight

9.9 Case Studies and Success Stories

9.9.1 Case Study 1: Automated Health Monitoring in Salmon Farming

A salmon farming operation implemented an automated health monitoring system, resulting insignificant improvements in fish health and reduced mortality rates.

9.9.2 Implementation:

- Sensors were installed to monitor water quality and fish behavior.
- Data analytics provided insights into health trends and early warning signs.

9.9.3 Outcomes:

- Early detection of health issues and timely interventions.
- Increased overall productivity and profitability (Werkman, 2008).

9.9.4 Robotic Surgery in Sea Turtles

A veterinary clinic utilized robotic systems to perform minimally invasive surgeries on injured seaturtles, improving recovery times and survival rates.

9.9.5 Implementation:

- Robotic surgical tools were used for precise operations.
- Automated systems for post-surgery monitoring and medication delivery.

9.9.6 Outcomes:

Reduced stress and quicker recovery for the turtles.

Enhanced surgical outcomes and lower mortality rates (Werkman, 2008).

Investments and maintenance on robotic fish cages are much higher (Zhenyu et al., 2002)

9.10 Biotechnology and Genetic Engineering in Aquatic Veterinary Sciences

Genetic engineering (GE) is showing promise in aquaculture as a means of producing moreresilient fish and shellfish as well as new feed sources and disease-fighting vaccinations. However,there are ethical, moral, and financial issues with the use of GE in aquaculture. Horizontal genetransfer could theoretically happen from DNA in feed or vaccinations to a recipient genome or byfeces to the environment. For example, genetically modified (GM) feed could spread to the aquaticenvironment and be absorbed by other marine creatures (Aumaitre et al., 2002). Several studieshave reported positive outcomes using DNA vaccine, including resistance against viral diseases.However, there may be unintended consequences that call for more research, such as the activationof genes other than those essential to immune defense processes. Prior to the large-scaleintroduction of GE products, such as DNA/GE vaccines and GM feed in commercial fish farming,it is imperative that the necessary scientific research and ethical concerns be completed in order toaccomplish the sustainable introduction of GE.

This could provide a strong foundation forpreventing possible harmful consequences on human health and the environment (Boudinot et al.,1998). The potential for favorable market and customer reactions increases if GE is able to assist in transforming aquaculture into a sustainable sector. How to safeguard the environment while also achieving financial gains is the main obstacle to GE implementation in a sustainable setting. This task entails a proactive examination of the suggested advantages and potential drawbacks within a research and policy agenda that promotes long-term, comprehensive thinking (Corbeil etal., 2000).

9.10.1 Challenges and limitations:

Expensive and have limited wireless communication. Robotic Fish Cages Ocean Farm Technologies, Inc. of Searsmont, Maine, has developed self- propelled spherical aquaculture cagessuitable for use in rough, open ocean conditions and designed to support a wide variety of aquaticspecies. The robotic fish cage can be fully or partially submerged in water(Zhenyu et al., 2002). The robotic cage is very strong against the strong current. In normal cages, the main problem is ahole in the net but in robotic cages, this problem can be solved by robots, which examine cages and if necessary, then repair the net. In a robotic cage, all activities of fish are monitored automatically by robots. In a robotic cage, various types of cameras and sensors monitor the cages.The price of robotic cages is very high. But if you buy and install it at sea then it will be very beneficial for you through the production increment and less input cost (https://thefutureofthings.com/6281-robotic- fish-cages/). Investments and maintenance on robotic fish cages are much higher (Zhenyu et al., 2002).

9.11 Biotechnology and Genetic Engineering in Aquatic Veterinary Sciences

Genetic engineering (GE) is becoming a very effective technique in aquaculture for producinghealthy fish and shellfish as well as for creating alternate feed sources and disease-fightingvaccinations. However, the application of GE in aquaculture presents ethical, ecological, andfinancial issues. For example, genetically modified (GM) feed may find its way into the aquaticenvironment and be consumed by other marine organisms. Moreover, horizontal gene transferfrom vaccinations or feed DNA to a recipient genome or from feces to the environment is aplausible scenario (Aumaitre et al., 2002). Several studies have reported positive outcomes usingDNA vaccine, including resistance against viral diseases. However, there may be unintendedconsequences that call for more research, such as the activation of genes other than those essentialto immune defense processes. Prior to the large-scale introduction of GE products, such asDNA/GE vaccines and GM feed in commercial fish farming, it is imperative that the necessary scientific research and ethical concerns be completed in order to accomplish the sustainable introduction of GE. This could provide a strong foundation for preventing possible harmful consequences on human health and the environment (Boudinot et al., 1998). The potential for favorable market and customer reactions increases if GE is able to assist in transforming aquaculture into a sustainable sector. How to safeguard the environment while also achieving financial gains is the main obstacle to GE implementation in a sustainable setting. This task entailsa proactive examination of the suggested advantages and potential drawbacks within a research and policy agenda that promotes long-term, comprehensive thinking (Corbeil et al., 2000).

9.12 Nanotechnology Applications in Aquatic Health Management

In recent years, aquaculture has become one of the food businesses with the quickest rate of growthworldwide, contributing to food security and elevating the status of the world economy. The careless disposal of untreated or inadequately managed waste and effluents from a variety of sources, such as food processing industries, manufacturing facilities, and healthcare facilities, releases a range of contaminants into the environment, including unmetabolized antibiotics, bioactive compounds, and organisms resistant to antibiotics (Fernández-Díaz et al., 2017).

Because of their extensive usage in aquaculture, these emerging contaminants (Ecs), particularly antibiotics, have the potential to contaminate the environment, especially the aquatic ecosystem. This could result in a variety of toxicological impacts on aquatic creatures as well as long-term persistence in the environment. But now, many kinds of nanotechnology-based technologies are being investigated to help existing remediation technologies increase sustainability, productivity, and efficiency. Systems based on nanotechnology are currently being employed in a variety of ways to increase sustainability, productivity, and efficiency. There have been recent developmentsin the areas of water purification, seafood processing and preservation, fish and shellfish growth enhancement by nutritional supplementation with nutraceuticals, and health management. According to Fernández-Díaz et al. (2017), nanotechnology is therefore crucial to the advancement of aquatic resource efficiency and environmental impact. Prior to delving deeper into the toxicityand durability of these pollutants in aquatic habitats, scientists need to ascertain the ways in whichantibiotics interact with other emerging toxins. Without a question, nanotechnology is crucial to aquaculture’s expansion and long-term viability. To date, a number of nanotechnology-based strategies have been employed to improve the main tenets of fishing and aquaculture (Saha & Bandyopadhyay, 2019). But there are a lot of restrictions when it comes to using nanotechnology in fish medicine. Different types of nanoparticles, including liposomes, nanotubes, nanocapsules, and dendrimers, may one day prove helpful in the study of fish illnesses.

Investigations into the antifungal, antiviral, and antifungal qualities of NPs against fish illnesses are also ongoing. If the application of nanotechnological advances is to be widespread, further investigation into fish vaccine development is needed. Few studies have examined the applicationof nanoparticles in aquaculture for the diagnosis of mycotic and bacterial illnesses (Fenaroli et al.,2014). Given the demonstrated promise of NPs, more targeted research into their application in a range of fish healthcare research topics is required in order to provide more effective fish illness diagnostics and therapies and meet the growing demand for aquatic animal health. Future production processes for nanoparticles will need to include biological techniques, which are easyto use and provide nontoxic, affordable, and environmentally friendly goods. In this way, nanotechnology would benefit aquaculture producers of all stripes. All things considered, aquaculture's long-term viability may be greatly enhanced by nanotechnology (Felis et al., 2020). Automation and robotics are revolutionizing aquatic veterinary processes, which has numerous benefits for animal care, precision, and efficiency. As long as these technologies are developed and integrated, aquatic veterinary medicine appears to have a bright future, despite the challengesthat lie ahead. By implementing these technologies, the industry may significantly improve the wellbeing and health of aquatic animals, which will result in more sustainable and fruitful aquaculture practices.

9.13 Sustainability and Environmental Consideration in Veterinary Medicine

However, concerns over aquaculture's sustainability and consequences on the delicate aquatic ecology have been raised by the industry's explosive expansion. One such concern is the use of veterinary medications, which are allowed to freely enter bodies of water (Martinez-Porchas M. and Martinez-Cordova LR., 2012).

Pharmaceuticals are classified as xenobiotics with anthropogenic origins, or PPCPs (pharmaceuticals and personal care products) in the literature. Moreover, this class of substances contains disinfectants (antiviral and antibacterial), whose usage has surged since 2020 as a result of the SARS-CoV-2 epidemic (Phonsiri, V. *et al*., 2019) (Adhikari, S. *et al*., 2022) The reality that their existence in soils and reservoirs of water has a detrimental effect on aquatic life and human health is the reason for this increased interest (Anand U. *et al*., 2022) Numerous PPCPs travel swiftly through the environment, but because of their extensive usage, they produce pseudo- persistence in aquatic habitats, which has an adverse effect on aquatic life. (Yang, Y. *et al*., 2017)

Medications utilized in veterinary and medical fields include disinfectants, antibiotics, antiviral, antifungal, and antiparasitic medications (Tong, A. *et al*., 2012). Throughout the world, a large range of antibiotics are used as medications to cure in humans, animals, or plants, or as supplements to fish and animal feed to encourage development. (Carvalho, P.N. *et al*., 2014) (Watanabe, N. et al., 2010) (Yang, Q. *et al*., 2021).

9.14 Telemedicine and remote monitoring in aquatic health care

The most contentious topic in the healthcare industry on a global scale has been telemedicine. Despite the fact that many nations, particularly developed nations, frequently seek telemedicine services with the goal of enhancing healthcare accessibility and reducing healthcare costs

(Schwamm, LH. Et al., 2014).

9.14.1 Internet of Things (IoT) devises

The Internet of Things has become common in recent years and are used in aquaculture to monitorand modify water levels and pumps in real-time (Kassem, T. *et al*., 2021). (Anand and Regi 2018)remotely monitored the water levels in rearing tanks using a Narrow Band (NB)-IoT system. Aquaculture may benefit from this technology's inexpensive prices, low power consumption, andextensive coverage. As an illustration, consider an Internet of Things-based system to monitor salinity, Ph, temperature and dissolved oxygen in water (Dahn, L.V.Q. *et al*., 2020). Aquaculture IoT systems usually employ WSN, which has strong scalability and a large coverage area (Pule, M. *et al*., 2017). In WSN, Zigbee is extensively utilized for monitoring water quality. Sensitive systems, however, need a lot of energy for long-distance transmission and are prone to failure. A conventional agricultural approach may be transformed into smart farming with the application ofIoT and digital technology (Narwane, V.S. *et al*., 2022).

9.14.2 Measurements of Water Quality Parameters

Table 13: Water quality parameters that are often monitored in RAS (Recirculating Aquaculture Systems), along with examples of their analytical techniques and equipment.

Parameter	Analysis	Equipment	References
PH	Electrochemical analysis Online Monitoring Portable instruments	ProMinent Oxygard Pacific AquaMesh network	Zhu et al. (2010) Odey and Li (2013) Pulkkinen et al.(2018)
Salinity	Ion chromatography, ICPortable instruments	Oxygard Pacific YSI 30 Salinity Meter Dionex DX-500	Danh et al. (2020) Lindholm-Lehto et al. (2020, 2021)
Temperature	Electrochemical analysis Online-monitoring Portable instruments	Temperature Meter Oxygard Pacific	Danh et al. (2020) Bergheim andFivelstad (2014) Kolarevic et al.(2014)
Carbon Dioxide (CO_2)	Quick spectrophotometric tests Online-monitoring	Franatech Hach Method 8223	Pulkkinen et al.(2018) Davidson et al. (2019)
Alkinity	Standard titration Method (ISO 9963−1:1994) Hach Method 8203–Sulfuric Acid Digital Titration	TitraLab AT1000, Hach	Lindholm-Lehto et al. (2021) Davidson et al. (2017) Pulkkinen et al. (2018)
Nitrate-N	Quick spectrophotometric tests Portable instruments Online- monitoring	Multi 3410 (WTW GmbH) LCK340, LCK341,	Lindholm-Lehto et al. (2020, 2021) Pulkkinen et al.(2018)

Nitrite-N	Ion chromatography, IC	Dionex DX-500 (Dionex)	Chun et al. (2018

9.14.3 Uses of Biosensors in disease identification:

Vibrio species are a group of bacteria that have been associated with both aquaculture and humaninfections on a global scale. These bacteria can be found in various aquatic and marine habitats, either in a free-living state, attached to biotic or abiotic surfaces, or in a symbiotic or host-pathogenrelationship with other organisms due to their metabolic versatility (Montánchez, I. *et al*., 2019). Analytical tools called biosensors interpret biological and/or chemical interactions into measurablephysicochemical signals proportional to the analyte concentration (Szunerits S. and Boukherroub R., 2018) Sensor-based *Vibrio* research, based on electrochemical, resonance, light-scattering, lateral flow, and colorimetric apt sensor systems, provides enough information about the sensor mechanism and is useful for further steps toward diagnosis and differentiation (Zhang, Z. *et al*., 2019) (Laczka, OF. *Et al*., 2014)

Table 14. Combining sensor-based diagnosis with the number of different applied biotechnological methods involving vibrio species in research

Sr. no.	Sensor based diagnosis study	Techniques	References
1	Nanoparticle-based biosensor combinedwith multiple cross displacement amplification	Lateral flowbiosensor	(Wang Y. *et al*.,2017)
2	Target moleculeidentified andcapture onto the electrode surface	Electrochemical Biosensors	(Zhang Z, Zhou J,and Du X., 2019)
3	Polystyrene-co- acrylic acid (PSA) latex nanospheres-gold nanoparticles composite	Electrochemical Biosensors	(Rahman M. *et al*., 2017)
4	Diagnostic and quality control to *V. parahaemolyticus*	Magnetic resonance biosensor	(Hash S. *et al*.,2019)

9.14.4 The Use of Unmanned System Technologies for Aquaculture Farm Management and Monitoring

Unmanned vehicles can save operating costs, increase mission repeatability, and improve mission safety (Verfuß, U.K. *et al*., 2019). Drones have proved effective in gathering information about fish behavior and the surrounding environment in aquaculture sites nowadays for monitoring purposes (Chang, C.C. *et al*., 2021). Using the data from the cloud, drones can determine the location of the target items, making it more intelligent than standard drone navigation systems. It develops into an intelligent flying robot capable of capturing important data and far-off objects. (N.A. Ubina and others, 2021). The integration of UAVs with IoT is highly advantageous in extending coverage to remote or rural locations, especially with the advent of 5G technology.As aflying gateway, the drone has a lightweight antenna for data collection and LTE cellular networks connected to base stations. UAVs act as the middleman node, gathering information from sensorsand forwarding it to the right place. The drone then flies to the location of the IoT devices to givethe aquaculture farm more coverage or support in the event that there are problems with the wiredconnection of the equipment. Sensor data can be received by the gateway, which can then transportthe gathered data to the servers (Moheddine, A. *et al*., 2019).

9.15 Challenges and future directions in Aquatic veterinary innovations

Fish health plays a vital role in aquaculture and fisheries, however there are several threats to the well-being of fish populations. Fish health faces several challenges, including environmental stresses including temperature fluctuations, habitat damage, and water pollution. Fish immune systems may be weakened by these stressors, making them more vulnerable to diseases (Martos- Sitcha, JA. *Et al*., 2020).

Solution

9.15.1 Through Vaccination

The best method for preventing and treating illnesses brought on by bacteria and viruses is vaccination. (N, Kushnir. *Et al*., 2012). Moreover, concerns about potential hazards to other speciesin aquatic environments used for fish farming impede efforts to develop live vaccines. Alternatively, some research has concentrated on creating DNA vaccinations (Lepa, A. *et al*., 2010)(Kurath, G. *et al*., 2008) (Lorenzen N and LaPatra., 2005).

Table 15 Vaccine developed for marine fish species

Fish species	**Disease**	**Vaccine Type**	**Delivery**	**Country**	**Reference**
Red seabream	Red seabream iridovirus	Inactivated	IP	Japan/South Korea	(Hwang J.Y. *et al.*, 2020)
Grouper	Nervous necrosis virus	Inactivated	IP	Japan	(Matsuura Y.*et al.*, 2019) (Yamashita H. *et al.*, 2005)
Olive flounder	Viral hemorrhagic septicemia virus	Inactivated	IP	South Korea	(Hwang J.Y. *et al.*, 2020)
Salmon	Pancreas disease virus	DNA	IM	Norway, Chile, UK	(Ma J.*et al.*,2019) (Brudeseth B.E. *et al.*,2013)
Salmonids	Infectious hematopoietic necrosis	DNA	IM	Canada	(Ma J.*et al.*,2019) (Brudeseth B.E. *et al.*,2013)

9.15.2 Through Probiotics

Probiotics have an effect on aquatic species by improving the animal's immune system, which raises the organism's capacity for disease resistance and keeps harmful animals from infecting the organism (Dawood and Koshio., 2016).

Table 16 Probiotics tested against viruses infecting marine fishes

Viral Pathogens	Abbreviation	Probiotics	References
Viral hemorrhagic septicemia virus	VHSV	Bacillus subtilis	(Han S.R. *et al*., 2021)
Singapore grouper Iridovirus	SGIV	*B. subtilis 7K*	(Zhou S. *et al*., 2019)
Betanodvirus	NNV	*Shiwanella* spp. *Strain 0409*	(Wu Y.C. *et al*.,2020)
Gibel carp Herpesvirus	CaHV	*Clostridium butyricum* (Cb)	(Li T. *et al*., 2019)
Viral hemorrhagic septicemia virus	VHSV	Bacillus subtilis	(Han S.R. *et al*., 2021)

9.16 Future Directions

Using high-throughput DNA sequencing techniques, metagenomics enables the comprehensive investigation of numerous microbial species related to fish environments, including bacteria, viruses, and fungi (Petrosino, JF.*et al*., 2009). These technologies improved sensitivity andspecificity that allow for the detection of specific disease or biomarkers at lower quantities. Miniature biosensor sensors might be used to track the health of fish in aquaculture systems in realtime. Combining nanomaterials with biosensor technology enables the creation of highly efficientdiagnostic devices for detecting diseases, toxins, and environmental stressors in aquatic environments. (Patra, JK. *Et al*., 2018). Artificial intelligence (AI) has the potential to significantlyincrease the speed, accuracy, and efficiency of disease identification and management when usedto molecular and immunological diagnostic techniques for fish health (Mandal A and Ghosh AR.,2023). AI algorithms analyze massive information, including fish behavior, health indicators, andwater quality parameters, to deliver real-time insights into the condition of aquatic ecosystems. (Ditria, EM. *Et al*., 2022).

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Chapter 10 :Avian Influenza; A Zoonotic Threat to Public Health

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Abstract:

Avian influenza (AI) is a poultry disease caused by infection with type A influenza viruses, specifically avian influenza viruses (AIV). Influenza A viruses are zoonotic organisms that pose a recurring challenge to the world each year. Historically, there have been recurring human pandemics caused by influenza A viruses. Aquatic birds in the wild serve as carriers for all forms of influenza virus A subtypes, making them the most likely natural reservoir for all influenza A viruses. While avian influenza viruses in their natural bird reservoir are often not very pathogenic, certain strains have become more virulent through genetic changes following transmission and adaption to vulnerable gallinaceous chickens. The major focus of this chapter is to demonstrate our present understanding of the cellular foundations of influenza virus disease and its progression, which are responsible for serious complications, and the preventive and therapeutic approaches now in use to combat influenza virus infection. It also underscores the crucial role of continued research, such as developing new vaccine technologies and studying virus mutations, in enhancing vaccination efficacy and understanding the virus, offering hope for future advancements. This chapter will also elaborate on the epidemiological aspects of avian influenza

Keywords:
Influenza Infection, Vaccination, virus strains, sporadic outbreaks, Seasonal Pathogenesis

10.1 Introduction:

The poultry sector is one of the largest sectors of agriculture and fulfills the world's protein demand. The growth of the poultry industry is being affected by several issues, e.g., antibiotics issues (Samad et al., 2022; Samad et al., 2024; Talib et al., 2024), disease stress (Samad et al., 2022b), price fluctuation, and seasonal influence. In this chapter, we are going to bring light to a very important disease of poultry named Avian Influenza, which affects birds and humans due to its zoonotic effect.

From the influenza virus discovery in 1933, it became an essential topic in the research field, with particular attention paid to its structure, biochemistry, replication mechanism, clinical and pathological characteristics, epidemiological research, antigenic variation, animal reservoirs, immunology, and vaccine development (Crosby, 2003). Viruses that affect the respiratory system (Orthomyxoviridae family members) cause influenza, also called the flu (Mubareka & Palese, 2011). An extremely transmittable organism, the influenza virus can trigger pandemics and periodic epidemics that cause substantial global deaths and disabilities (Gurushankara, 2021). Three primary categories of Influenza viruses are A, B, and C.

Viral infections in humans are caused mainly by influenza A and B viruses. However, influenza C usually produces less severe breathing problems (Yuen et al., 1998). Based on two surface proteins, haemagglutinin (H) and neuraminidase (N), influenza A viruses are further divided into subcategories (Guo et al., 2018). Eight single-stranded negative-sense RNA segments make up the genome of influenza A and B, while seven segments, which are more physiologically unique, make up the genome of the other genera (Noda & Kawaoka, 2010). Each genome segment codes for a single protein, except for segments 7 and 8, which instead encode two proteins by alternative splicing. 13,588 nucleotides comprise the genome segments, numbered according to their acrylamide mobility, from slowest to quickest (Lamb & Choppin, 1983). With a dimension of X1–120 nm for spherical forms, the virions are pleomorphic and are enclosed.

The impact and transmission of the virus are primarily determined by these differences, like H1N1 or H3N2 (Richard & Fouchier, 2016). An infected person's cough, sneeze, or speech produces airborne particles that cause the spread of the influenza virus. Furthermore, it is transmitted through skin contact with infected surfaces. Viral infection of the cells lining the airways causes irritation and respiratory signs after entering the organism through the respiratory tract (Reed, 2015).

The common symptoms of influenza are fever, congestion, sore throat, cough, body distress, and exhaustion. Gastrointestinal tract (GI) disorders, including vomiting and diarrhea, can also occur (Javanian et al., 2021). The intensity of the disease can vary from moderate to severe, particularly in those at most significant risk, like young kids, older individuals, pregnant women, and people with existing medical challenges (Albert et al., 2012). Influenza virus can undergo DNA changes through mutations, which constitute some distinctive features of this virus. When the protein coating of the virus gradually alters over time, it is known as "antigenic drift," which causes variations to arise. Instead, antigenic shift happens when two distinct influenza viruses invade the same host and pass their genes, giving rise to a new strain of the virus that has the potential to become a pandemic (Kim et al., 2018).

In wintertime in temperate regions, influenza viruses are most active, as they are known to exhibit seasonal fluctuations. An enormous financial load and pressure on medical facilities can be caused by the virus during yearly outbreaks (Furhmann, 2010). The best method for avoiding influenza infections and minimizing the effect of illness on public health is vaccination. Every year, vaccines against the most common types of influenza viruses predicted to resurface during seasonal flu are created as seasonal vaccinations (Treanor, 2023). By targeting particular viruses and their proteins, including hemagglutinin, these vaccines induce the body's immune response to make antibodies that either prevent infection or lessen the level of disease (Ogra et al., 2001). Several precautions and vaccines are essential in limiting the growth of the influenza virus. The effective ways to stop the transmission of the disease are to wash your hands often, wear a mask during coughs and sneezes, stay at home when sick, and keep your distance from sick people (Harper et al., 1984). Antiviral drugs like oseltamivir (Tamiflu) are frequently used in case of serious viral infections or for high-risk persons to decrease consequences and reduce the severity of disease. Individuals with a higher risk of developing a severe illness should prioritize receiving antiviral medication earlier (Greene et al., 2013). The influenza virus presents difficulties in treatment, management, and prevention; It is a serious issue for public health.

To effectively reduce the outcome of influenza on people and communities, it is imperative to comprehend the viruses, patterns of transmission, and physical symptoms of the virus (Qualls, 2017). The main reason behind this chapter is to bring light to complications and solutions of Avian Influenza by adopting preventive measures and awareness regarding avian Influenza

10.2 Epidemiology:

Two sub-groups of the Orthomyxoviridae family (influenza A and B) cause infectious respiratory illnesses. These illnesses have the potential to generate yearly outbreaks of variable intensity, from minor signs of illness to life-threatening lung injuries. Strains of influenza viruses exhibit a broad range and are categorized based on their antigenic characteristics. These strains are found in several mammals, chickens, and wild birds. In cold locations, influenza outbreaks typically occur during winter because of the favorable temperature and low humidity levels that facilitate virus replication and propagation (Wu et al., 2014).

Influenza outbreaks are unclear in subtropical and tropical areas, making repeated infections possible throughout the year. Some chronic and pandemic influenza virus isolates from humans are thought to have a specific "reservoir" on the Asian continent (Lowen et al., 2007). Each year, seasonal influenza causes substantial illness, impacting adults (about 10%) and children (20%) (WHO, 2015). Seasonal influenza caused significant medical and financial consequences in America from 2003 to 2009, accounting for 3.8% of all hospitalizations for respiratory disorders. Every 20 to 30 years, global epidemics occur, and they are typically accompanied by acute symptoms, which may raise the death ratio (Ortiz et al., 2013).

Typically originating from bird-like reservoirs, pandemic IAV viruses are just getting into an immunologically immature human population. As the primary source of influenza viruses, avian species are also a common host for influenza-associated viruses (IAVs) (Tanner et al., 2015). Additional species identified as secondary hosts for IAVs include horses, dogs, and aquatic mammals like elephant seals (Goldstein et al., 2013). The influenza A viruses (H5N1, H7N922), frequently discovered in commercial fields and shown to cause serious illness in humans, are currently the main hazards to human health from avian influenza (Uyeki & Cox, 2013).

Since the swine breathing mechanism exhibits avian and human influenza receptors, pigs can serve as "integrating hosts" for recombining IAV strains from various sources (Janke, 2014). The 2009 "Swine," a global epidemic produced by a combination of avian, pig, and man strains, verified a pandemic danger associated with these strains. This was the first global pandemic of the 21st century (Pappaioanou et al., 2010).

The global population's health is seriously affected by influenza epidemics, and authorities closely monitor new strains of circulating viruses in animals and humans. To evaluate any potential emerging viral exposure, WHO divides the transmission of a global crisis into six phases, starting with the initial wildlife transmissions in Phase 1 and ending with the outbreak across a wide geographic area in Phase 6. Centers for Disease Control and Prevention (CDC) provided a new influenza risk assessment Technique (IRAT) that is based on an evaluation of possibilities for pandemic events caused by novel strains on several factors, including epidemiology and ecology of the variant, the host response to the virus, and virus properties (Trock et al., 2015). Reports of influenza outbreaks have been documented since the late 1800s, and the features of the virus and the host reactions that accompany it differ from one outbreak to the next. The earliest recorded pandemic, however, with less evidence, was the "Russian flu" (H3N8) (Valleron et al., 2010) that occurred in 1889. It was accompanied by the 1918 "Spanish flu"; H1N1 (1918), the "Asian flu"; H2N2 (1957) (Trotter et al., 1959), the "Hong Kong flu"; H3N2 (1968) (Lindsay et al., 1970), and most recently, the "swine flu"; H1N1 (2009). Curiously, the H1N1 strain was the source of two most recent influenza pandemics, the swine influenza of 2009 and the Spanish flu, which caused a pandemic in 1918. Spanish flu occurred in many outbreaks and caused morbidity of at least 50 million people in less than one year. Therefore, according to Taubenberger and Morens, it is also known as "the mother of all pandemics." It affected approximately 1/3 of the world's population (Taubenberger & Morens, 2006). The cause of the 1918 influenza pandemic has been well-studied, but little is known about the virus traits that led to the pandemic and the high death rate, particularly among people in the 20–40 age range (Sheng et al., 2011). However, compared to less virulent viruses, rat experiments have demonstrated that the Spanish variant elicits a more fantastic immune reaction, particularly in recruiting neutrophils and macrophages (Perrone et al., 2008).

Over 200,000 morbidity rates due to respiratory complications and 80,000 death rates due to heart disease are estimated to have been caused by the 2009 H1N1 virus. The 2009 “swine flu,” which eventually came to be known as “novel influenza A virus (H1N1)”, led to 18,500 confirmed cases. A significant proportion of these fatal cases—more than 80% of affected individuals were under the age of 65. By suppressing the prior innate immune response of the host, the 2009 virus variant appears to be able to spread more widely and be more harmful than the 1918 strain (Mukherjee et al., 2011).

10.3 Transmission of Influenza Virus:

A common barrier to pandemic outbreak avoidance and alleviation techniques has been a lack of awareness about influenza dissemination. Various transmission mechanisms, such as big droplets and droplet nucleic components, are mainly a topic of intense scientific research. Revising evidence-based infection control recommendations for homes, workplaces, schools, and healthcare settings after the 2009 A (H1N1) pandemic requires a clear understanding of the respective significance of various transmission mechanisms (Brankston et al., 2007). The spread of human influenza viruses by conjunctivitis is still unknown, despite evidence from tropism tests using pandemic H1N1 (Cha et al., 2010) and human outbreaks of avian H7 viruses indicating the existence of transmitters in the eye.

Additionally, recent research has demonstrated that an aerosol is a live, domesticated virus that can enter the eye cavity and travel to the nasopharynx. Unlike the bird-to-human transmission mode, there is inadequate proof to support the fecal-oral route in humans (Webster et al., 1978). It is commonly acknowledged that there are three ways in which human influenza infections spread (Brankston et al., 2007):

Droplets are little particles that can adhere to the mouth and nose, two soft tissues of the respiratory system. Although they can enter the body through breathing, they cannot enter the lungs due to their large size (>10lm).

Figures 1 and 2 show the transmission route and agents causing avian influenza.

Droplet nuclei: These particles, also known as aerosols, are small enough to enter the lower part of the respiratory system through inhalation (less than 5l m). They can also accumulate on the surface of the upper respiratory tract (URT).

Contact Transmission: Through direct and indirect contact with an infected person and item, particles can enter the URT and spread to the mucous membranes.

Examining the above-mentioned transmitting routes makes it clear that host, environmental, and virus-related factors have a significant role in the transmission of the virus. Comprehending these components is crucial for making and executing efficacious influenza control strategies, such as infection control, social distance, and personal hygiene, as they can all impact the active transport pathways (Reissman et al., 2006).

10.4 Viral Replication:

By attaching to haemagglutinin (HA) receptors on host cell surfaces, the influenza virus enters the cell by endocytosis and becomes uncoated within endosomes. When the coating is removed, two essential processes take place. Firstly, the matrix (M1) protein separates the protons from M2 channels and the ribonucleocapsid protein (RNP) because the inside of the virion becomes acidic. Second, the RNP is released into the cytoplasm when the viral membrane merges with the endosomal membrane due to a conformational shift of the HA caused by a low pH. RNPs go through nuclear pores to enter the nucleus after they have reached the cytoplasm. Nucleus-activated transcription necessitates "cap-stealing," or removing host caps to activate the viral mRNA. Therefore, at their 5' ends, viral mRNA has a cap structure of host origination consisting of 12–13 nucleotides, and at their 3' ends, it is polyadenylated. The complementary RNA (cRNA), which acts as a precursor for producing copies of viral RNA (vRNA), is likewise encapsulated by the NP protein for transcription and replication. Single-stranded RNA (ssRNA) is transcribed from vRNA without primers. PBl, a molecule that polymerizes ribonucleotides; PB2, an endonuclease and cap-recognizing molecule; and the PA protein, which is involved in genome replication, make up the RNA polymerase complex (Nakagawa et al., 1996).

Large amounts of non-structural (NS1) protein are generated throughout an infection. This protein serves several purposes related to the inhibition of interferon functions. The M1 protein attaches to RNP later in the infection to stop transcription by entering the nucleus. Subsequently, the NS2 protein links the RNP that enters the cytoplasm, so it interacts with actin polymerization before the assembly of the plasma membrane. Budding is the last stage of virion assembly before newly synthesized virions are released. Likely, interactions between the M1 protein and luminal outgrowth of HA and NA proteins inside the plasma membrane contribute to driving and guiding the process. A set of RNPs is present in every virion's genome.

10.5 Viral Adaptations:

Through effective proliferation and interspecies transmission, IAVs must have adapted to humans as a new host (Zhao et al., 2019). The epithelium of the airway in both humans and pigs is the site where IAVs usually attack. It takes changes to the replication of viruses for an adaptation to be successful. When the virion attaches to sialic acids (SAs) produced in the respiratory tract, it is the initial stage of imitation of avian influenza in the human host. This process is facilitated by the viral surface protein HA. After attachment, the virion enters the endosomal pathway, and the breakdown of vRNP from the M1 protein occurs (Bui et al., 1996). The ability of human and avian IAVs to sufficiently release the viral genetic material into the host cell has been linked to the corresponding changes in the HA protein, which is significantly related to the optimum pH, which results in this membrane fusion (Gerlach et al., 2017). Viral proteins and freshly created RNPs eventually aggregate at lipid raft regions during virus assembly, which causes budding, scission, and the release of newly generated virions. This process occurs at the epithelial cell membrane (Nayak et al., 2013).

10.6 The Host Response to Influenza:

Lung autopsy frequently reveals extensive alveolar inflammation in influenza virus-infected individuals who die, yet only a tiny percentage of patients have viral RNA (Bautista et al., 2010). IAV infection-related mortality could be more likely to be caused by an excessively heightened immune response than by an unchecked viral transmission. Numerous types of immune cells have been shown to impact host responses negatively, and specific fundamental molecular pathways have recently been identified, offering possibilities for treatment therapy (Short et al., 2014).

The primary focus for IAVs is epithelial cells, which produce a defense mechanism when various pattern-receiving receptors identify pathogen- or danger-associated molecular patterns (PAMPs/DAMPs).

After interacting with RIG-I, viral unbound 5′-triphosphorylated RNA interacts with MAVS, TRIM25, and IPS-1, triggering IRF-3 and IRF-7-dependent transcription as well as translation of types I and III IFNs (Gack et al., 2007). Additional IAV infection detection domains comprise the NLRP3 inflammasome and endoscopic toll-like hormone receptors (TLR3 and TLR7) (Wright et al., 2012).

According to recent data, DAMPs like S100A9, HMGB1, or purine metabolites play a critical role in influenza disease after viral infection. After identifying the viral pattern, macrophages residing in the lungs serve as the first line of defense and start an immune response. They are a primary source of type I IFNs and other cytokines and chemokines, and they eliminate viral particles and infect apoptotic cells (Kumagai et al., 2007).

Numerous studies have demonstrated the critical role of macrophages residing in the alveoli in reducing influenza virus infection-related complications, death, and viral transmission. In addition, stimulating CD8+T cells triggers innate and adaptive immunological responses, possibly leading to autoimmune diseases (Kim et al., 2007).

Early on in an IAV infection, neutrophils are heavily attracted to the alveoli and have a conflicting function. Early neutrophil depletion during the illness is linked to unchecked viral propagation and the development of a severe influenza-induced illness (Tumpey et al., 2005). One of the leading causes of lung injury is the increased activity of neutrophils, which is seen in highly infectious H1N1 and H5N1 infections (Narasaraju et al., 2011).

It has been observed that inhibiting TRAIL signaling and migrating monocyte-derived macrophages from the alveolar cavity reduce influenza-induced lung injury (Sica & Mantovani, 2012). Furthermore, research indicates TRAIL prevents edema resolution following IAV-induced lung damage in mice (Hogner et al., 2013). Influenza viruses can also infect lung endothelial cells, influencing epithelial damage in mouse influenza models (Teijaro et al., 2011).

In addition to expressing adhesion molecules and chemokines, which promote monocytes and lymphocyte lung intra- and extravasation, they also discharge inflammatory mediators such as TNFα and IL-6, which further enhances the proinflammatory environment during IAV infection (Ishiguro et al., 2003). Improved outcomes following infection with H5N1 and H7N9 in humans and enhanced epithelial cell protection are closely correlated with elevated blood levels of ACE2 following IAV infection (Zou et al., 2014).

10.7 Clinical Manifestation:

Through airborne transmission, influenza viruses propagate among humans (Bridges et al., 2000). An influenza virus (IAV) can cause an infection with severe viral pneumonia that can lead to multiorgan failure or worsen pre-existing medical disorders. In rare instances, however, an influenza virus may appear passive or mild with an upper respiratory tract illness (WHO, 2009). Seasonal immunodeficiencies and underlying chronic diseases like heart or lung illness are the groups that seasonal IAVs precisely target (Van Kerkhove et al., 2011). Clinically, symptoms correlate with increased viral replication when they appear suddenly 24 to 48 hours after the virus is allowed to incubate. The following symptoms, including influenza-like illness (ILI), can last two to eight days: fever, chills, headache, myalgias, malaise, and warmth.

Additionally, it has been reported that gastrointestinal illnesses characterized by vomiting or diarrhea, particularly in children, can be brought on by pandemics and, to a lesser extent, seasonal IAV. In severe cases, the virus spreads to the alveolar compartment, which expresses both α2,6- and α2,3-SA, with the latter being prominently upregulated on type II pneumocystis (Shinya et al., 2006). Alveolar infection results in widespread diffuse damage to the alveoli, histologically similar to other types of acute respiratory distress syndrome (ARDS) with intra-alveolar hemorrhagic edema accumulation, fibrin deposition, significant leukocyte infiltration, extensive apoptosis of alveolar and bronchial epithelial cells, and hyaline membrane formation (Mauad et al., 2010). These patients initially exhibit increasing respiratory failure, which includes hypoxia, tachypnea, dyspnea, and radiological evidence of diffuse bilateral pneumonia. Later, they develop ARDS, which may be fatal.

The sickness course sometimes worsens by coinfection with colonizing bacteria (Rello & Pop-Vicas, 2009). The most common causes of pneumonia-associated deaths during pandemics are shown to be a combination of Streptococcus pneumoniae and Staphylococcus aureus pneumonia, which have been seen to occur often (Rynda Apple et al., 2015). Evidence of bacterial pneumonia in the lung was found in the majority of instances when the funeral procession of 68 soldiers who perished throughout the Spanish flu pandemic occurred. 38 cases in 2009, coinfections predominated in a previously healthy younger population without comorbidities, according to a cohort study conducted in 14 Australian ICUs (Blyth et al., 2013). The condition becomes more severe when the person is co-infected with other respiratory viruses, such as respiratory syncytial virus, especially in patients with impaired immune systems (Goka et al., 2013).

10.8 Complications of Influenza

Cardiovascular: cerebrovascular problems, ischemic heart disease, and myocarditis (Warren-Gash et al., 2012)

Hematologic: hemolytic uremic disorder, hemophagocytic disorder, and thrombocytopenia purpura (Kosugi et al., 2010).

Musculoskeletal: myositis and rhabdomyolysis (Ayala et al., 2009).

Neurologic: acute disseminated encephalomyelitis, encephalopathy, post-influenza encephalopathy, and transverse myelitis (Goenka et al., 2014)

Ocular: Optic neuritis, retinopathy, uveal effusion syndrome, and conjunctivitis are the most common complications of influenza and encephalitis (Mansour et al., 2012; Fukami et al., 2005).

Pulmonary: diffuse alveolar hemorrhage, acute respiratory bruises syndrome, hypoxic respiratory syndrome, prioritized viral pneumonia, and secondary bacterial pneumonia (Perez-Padilla et al., 2009).

Renal: multi-organ dysfunction, acute kidney injury (Fearnley et al., 2011; Ru et al., 2011).

10.9 Diagnosis of Influenza virus

The influenza virus A (H1N1) pandemic in 2009 caused health problems and obtained consideration from global researchers for diagnosis, prevention, and better treatment. It is essential among adults and children, causing severe respiratory problems (Dawood et al., 2009; Jeon et al., 2010; Monto & Kioumehr, 1975; Flasche et al., 2011). It also involves other complications like encephalopathy, myocardial symptoms, myositis, and pneumonia (Wright et al., 2007). It is more common in developing countries due to the lack of proper diagnosis tools, treatments, poverty, and overpopulation. Antigenic changes in the influenza virus have diverse effects on diagnosis and treatment strategies (Webster et al., 1992). Rapid detection tools like laboratory methods, symptoms of influenza, reverse transcriptase-polymerase chain reaction (RT-PCR), viral genome amplification technique, virus culture, serological testing, and immunofluorescence tests can be used for influenza detection for early treatment to avoid its complications.

10.10 Laboratory Diagnosis

A sample of nasal aspirates or nasopharyngeal swabs detect the influenza virus (Wright et al., 2007). First, sterile saline is used to sanitize the nose. A swab is inserted about 1-2 inches in the nose, rotated, and removed for adult sample collection, and the nose is washed, or aspirates are collected for children's influenza detection. Then, the sample is placed in a sterile container with specific preservatives for delivery to the laboratory (Online lab tests are available). The patient's blood is collected for antibody detection, and PCR techniques are used (Online lab tests).

10.11 Viral culture Approach

It is an essential technique for influenza virus diagnosis that has been used for many years since the 1940s but is also very time-consuming (Treanor, 2005). In this method, the influenza virus is detected by preparation of cell lines like Madin Darby canine kidney (MDCK), rhesus monkey kidney tissues or cells (RhMK), African green monkey kidney cells (AGMK), mink lung epithelial cell lines (Mv1Lu). The virus is inoculated in cell lines and monitored for 7-10 days to examine its effects on cells and further isolation. The last confirmation is achieved using stained antibodies and immunofluorescence microscopy to diagnose H1N1 09 (Higgins et al., 2010). Respiratory epithelial cells of humans encourage the growth of the influenza virus (Zhirnov et al., 2002). It awaits further processing, like antiviral treatment and other methods to control infection (Cox & Subbarao, 1999; Lopez Rao et al., 2011; Mahony, 2008). Nowadays, the shell viral culture (SVC) technique is used, which is considered more specific and sensitive (Matthey et al., 1992). Using this technique, the virus is inoculated in mammalian cells grown in shell vials. Then, the virus is stained with virus-specific fluorescent monoclonal antibodies. This technique has a turnaround time of about 24-48 hours (Microlab for detecting Respiratory viruses).

10.12 Immunofluorescence Technique

Direct fluorescence antibody assay (DFA), also called immunofluorescent antibody test (IFA), is a cheap and fast method to detect antigens (influenza viruses) in nasopharyngeal swabs or nasal aspirates. Many studies and surveillances have been performed on DFA (Sadeghi et al., 2011). This method involves staining influenza cells obtained from samples followed by conjugation of antibodies produced in patients after influenza infection against the virus with a fluorescent dye and observation under a fluorescent microscope. This easy and quick diagnostic method takes 2 to 4 hours (Centers for Disease Control and Prevention CDC 2010; Pollock et al., 2009). This method has 60-80% sensitivity for seasonal influenza virus diagnosis. This test is better for children below ten but less sensitive for people above 30 (Lee et al., 2011). It requires experts for fluorescence microscopic analysis and is less sensitive than RT-PCR, which is used for confirmatory test of H1N1 09 (Ganzenmueller et al., 2010).

10.13 Serological Assays

Antibodies produced after influenza infection can be detected by complement fixation test, virus neutralization test (VN), hemagglutination inhibition test (HI), and enzyme-linked immunosorbent assay (ELISA) (Cox & Subbarao, 1999).

Haemagglutination inhibition assay is a simple, inexpensive, and sensitive method to detect avian influenza A virus. This method uses specific haemagglutinin (HA) antibodies that prevent the combination of influenza virus with blood cells. This method is used to prohibit vaccinations that serve during the next outbreak of influenza virus (Chen et al., 2010; Allwinn et al., 2010). This method is ineffective and time-consuming because it requires two serum samples simultaneously (Cox & Subbarao, 1999). *Virus neutralization assay* neutralizes the viruses and inhibits their infection-causing ability. It is more sensitive than HA assay but not used in BSL 2 and BSL3 laboratories (Stephenson et al., 2009).

The complement fixation test is an immunodiffusion-based method that quantifies internal antibodies' response to the influenza virus and is used for vaccination (Haaheim, 1977). It has low sensitivity.

Enzyme-linked immunosorbent assay (ELISA) diagnoses various bacterial and viral infections. However, it is also a less sensitive method. Microtiter plates and paper strips are available for ELISA tests. It is replaced with nanoparticles-based immunoassay (ENIA), a rapid method that uses monoclonal antibodies directed to detect influenza viruses A and B. It is also an effective method to detect 29 strains of Influenza A and 10 different variants of the Influenza B virus (Zhand et al., 2014).

10.14 Rapid Influenza Diagnostic Tests (RIDTs)

This rapid method is used for clinical decision-making in samples with rich titer, such as children, rather than adults (Landry, 2011). This is more common in developing countries that do not have PCR availability. This method uses commercially available monoclonal antibodies that target the viral nucleoproteins and are employed in immunochromatographic techniques. It is completed in 30 minutes and can be detected by eye (due to a color change). However, it is not sensitive compared to viral culture assay and PCR methods.

10.15 Nucleic Acid-Based Test (NAT)

Kary B Mullis developed the technique of PCR in 1983, which has now evolved into NAT and can be used to detect viral DNA or RNA rather than antibodies against viruses or viral antigens. It is more sensitive than other antibody-based tests.

10.16 RT-PCR

It is mainly used in the conventional NAT method to identify influenza. This method is used in laboratories all over the world. In this method, RNA is extracted from viral samples, reverse transcription of RNA generates complementary DNA (cDNA) by using reverse transcriptase enzyme, and then amplification occurs by using fluorescently labeled PCR products (Wang & Taubenberger, 2010).

10.17 Microarray-Based Approach

It is a very convenient technique for diagnosis and sub-typing of the influenza virus; for instance, the FluChip microarray is a DNA-based microarray that is used to determine the H5N1, H1N1, and H3N2 variants of influenza very quickly (Dawson et al., 2007; Moore et al., 2006; Townsend et al., 2006). Combi Matrix Corporation of USA has developed a semiconductor- influenza A microarray that detects all subtypes in less than 5 hours (Liu et al., 2006; Lodes et al., 2006). The nano microarray technique can produce images of strains using purified PCR fragments of H5N1 viral RNA. Thus, the result can be seen by intensity patterns produced by the naked eye. However, it is a labor-effective method to diagnose seasonal epidemics or pandemics of influenza in laboratories (Zhao et al., 2015).

10.18 Sequencing of Nucleic Acid

Sanger Sequencing

Sanger sequencing is a DNA sequencing chain resistance method developed by Fredrick Sanger in late 1970. This method uses chain-terminating dideoxy nucleotides (di-ddNTPs), DNA primers, DNA polymerase, and dNTPs that are not labeled. Di-ddNTPs are incorporated in newly synthesized DNA and inhibit the further elongation of DNA strands by restricting the addition of dNTPs. This method is widely used to produce genomic sequences of viruses like influenza. It is also used to detect antiviral resistance to the circulating influenza virus.

10.19 Drug Resistance Detection of Influenza Virus

Molecular techniques are used to check genetic-based resistance to drugs by identifying mutations in genes that cause the development of resistance. Oseltamivir is the most widely used drug for influenza virus, against which the virus has developed resistance due to histidine to tyrosine substitution in the neuraminidase active site H275Y (Bao et al., 2011). Real-time PCR (RT-PCR) pyrosequencing and neuraminidase inhibition assay are the techniques that are used to detect drug resistance (Bao et al., 2011), but they are all time-consuming. Oseltamivir resistance can also be monitored using the cycling probe RT-PCR method, which results in 3 hours of seasonal influenza and the H1N1 09 virus (Suzuki et al., 2011).

10.20 Preventive Measures

Vaccination

To prevent influenza infection, vaccination is the best approach (Paules & Fauci, 2019). The relation between vaccine antigens and circulating influenza virus variants is the parameter for protection from influenza virus for populations and individuals. According to some studies, vaccination has 60% efficiency in the fight against the influenza virus (Recombinant, attenuated, and inactivated influenza vaccines are currently available) (Noor & Gradidge, 2018).

10.21 Treatment

Early detection of influenza infection is the major challenge in its treatment because antiviral medications effectively initiate it. Respiratory viral infection symptoms detection, ELISA test, and PCR-based test are recommended for early diagnosis to process for further treatment. However, World Health Organization (WHO) guidelines recommend providing treatment before the laboratory tests against influenza infection (WHO, 2009).

Body fluid loss during fever can be reduced by hydration. WHO guidelines recommend antiviral drugs and neuraminidase (NA) inhibitors for all influenza A and B cases (including immunocompromised patients suffering from chronic lung disease, obesity, pregnant women, and respiratory problems). Aspirin, ibuprofen, naproxen, and diclofenac sodium are anti-inflammatory drugs that reduce fever, headache, and muscle aches (Eyers et al., 2010). Oseltamivir is orally administered and can diminish the symptoms in children and adults having acute influenza. It is also recommended as a chemoprophylaxis agent in adults and 1-year-old children (McLean et al., 2015). Flufirvitide is used to inhibit peptide binding and, thus, block viral HA. Nitazoxanide belongs to phase III vaccines that block the viral HA maturation.

It has been observed that NA inhibitors restrict symptoms and risks of pneumonia (Dobson et al., 2015). In the future, it is thought that different antiviral drugs will be combined to create different modes of action (Dunning et al., 2014). NA inhibitors are mostly recommended because they are effective even if administered 48 hours after symptom onset. It is applied by intravenous injection in critical condition patients (Louie et al., 2012). In combination, antibiotics, and antiviral drugs are suggested in patients with hospitalized influenza virus and lung infiltrates chest radiography by Jain et al. (Jain et al., 2012).

10.22 Therapeutic Strategies

New revolutionary strategies are established to overcome the growing prevalence of AIV drug resistance that limits the replication and outcomes of the influenza virus. Classical drugs target the viral proteins and affect their functions. Like, favipiravir has evolved in phase III, inhibiting the viral polymerase complex activity (Furuta et al., 2009; Furuta et al., 2002). Neutralizing antibodies (Ab) are developed against HA and M2 and effectively treat various influenza strains (Fleury et al., 1999; Wei et al., 2011). Cyanovir-N 56 is effective against HA, which binds to its glycosylation sites and prohibits cell adsorption (Smee et al., 2008). Small RNA inhibitors (Lin et al., 2012; Tompkins et al., 2004) and smaller inhibitory molecules directly targeting viral components are developed. Alveolar epithelium repairing after viral clearance is mediated by CD-mediated antiviral immunity by GM-CSF, which mediates macrophages (Sever-Chroneos et al., 2011; Cakarova et al., 2009; Huang et al., 2011). Patients with pneumonia can be treated by administration of GM-CSF, which also improves oxygenation and outcomes of IAV-induced ARDS (Herold et al., 2014). Respiratory problems induced by influenza can be treated by host-modulating therapies, and further research is thought to be conducted with antiviral agents in the future (Dunning et al., 2014).

10.23 Future Aspects

Influenza virus infections are constant life threats that include the knowledge gap, which has to be addressed in future experiences. For instance, a lack of memory phase exposure experiments, the impact of influenza and its vaccines on adults and infants, and immune memory mechanisms in influenza virus experiments should be addressed. Investigations have shown fast recovery in 14-28 days after secondary exposure to the influenza virus (Linderman & Hensley, 2016; Kosikova et al., 2018; Cao et al., 2015). However, the actual host immune mechanism in this regard has not been adequately understood. The duration between infection and vaccination bot properly reflects the host's immune response to seasonal viral challenges (Lipstich & Viboud, 2009). Most studies have focused on viral HA and host humeral responses, but it will be more important to pay attention to alternative immune response pathways in the future. Adaptive memory cells like CD8 + T cells, CD4 T cells, and natural killer cells (NK) can be more effective, but the challenge is the lack of immune signaling components for their response and function. Plasma blasts can be caused by vaccinating memory B cells, as suggested by studies by Andrews and colleagues (Andrews et al., 2015). Immune response drivers against dynamic infections investigation may suggest improved vaccine production. RNA polymerase subunits' (PB1 & PB2) catalytic and endonuclease activity could also be targeted to produce inhibitors. To produce other therapeutic strategies, it is essential to understand the three-dimensional structure of every single protein and the complex proteins of the host and virus. Higher protein complexes like M1, NS2, and RNP may include the interacting domains of membrane proteins (HA, NA, M2). So, it is essential to map the functional genome of influenza to understand its pathogenesis and replication, which will, as a result, help to diagnose and treat influenza infection.

10.24 Conclusion

Influenza is a contagious, zoonotic disease that directly or indirectly spreads in the human community by respiratory droplet transmission, which causes severe pneumonia and multiorgan failure. Mutation at a specific point in the viral genome causes genetic drift, developing novel strains that invade the host defense mechanism and move to the previous influenza variants to cause annual influenza epidemics. The genetic diversity in man, avian, and swine influenza viruses having HA and neuraminidase (NA) protein genes has established the emergence of influenza, especially in humans that lack immunity against them. Proper time diagnosis and treatment can bruise the influenza virus infection and help to restrict its outbreak. Treatment therapies are highly cost-effective, particularly in regions with less practical resources. It is complicated to discriminate influenza disease from another respiratory syndrome, leading to treatment delays. It causes many complications, but influenza vaccination effectively prevents it and its complications. However, more effective strategies, in addition to antiviral agents, are needed for older people and immunocompromised patients. Moreover, this chapter also concludes that increasing resistance in avian-derived H5N1 and H7N9 influenza A viruses against NA inhibitors lightens the requirement for more molecular surveillance and experiments of imputed virulence factors and antiviral resistance markers for treatment and diagnosis to improve public health during seasonal outbursts and epidemics.

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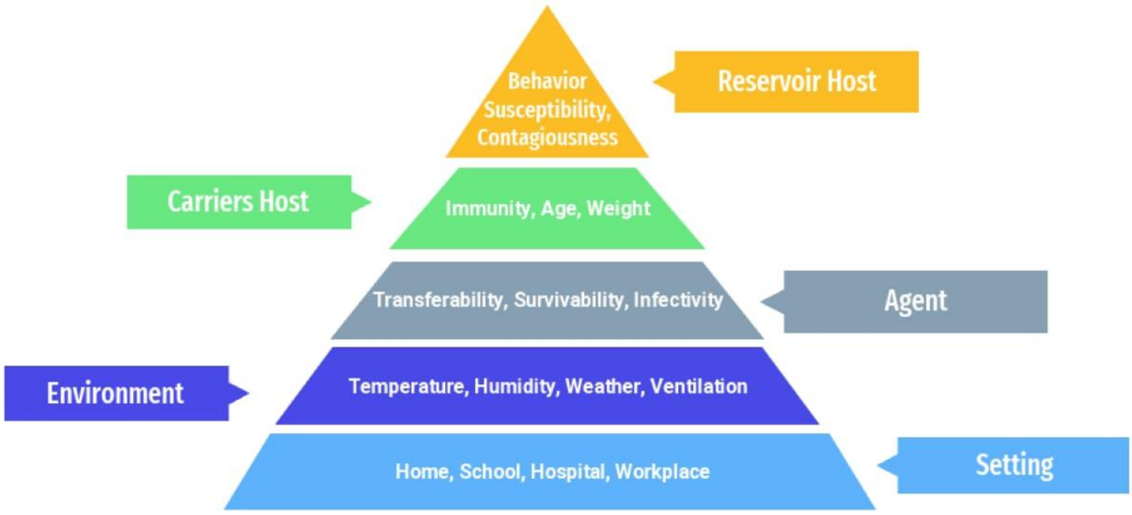

Figure 6: Factors causing transmission of Avian Influenza

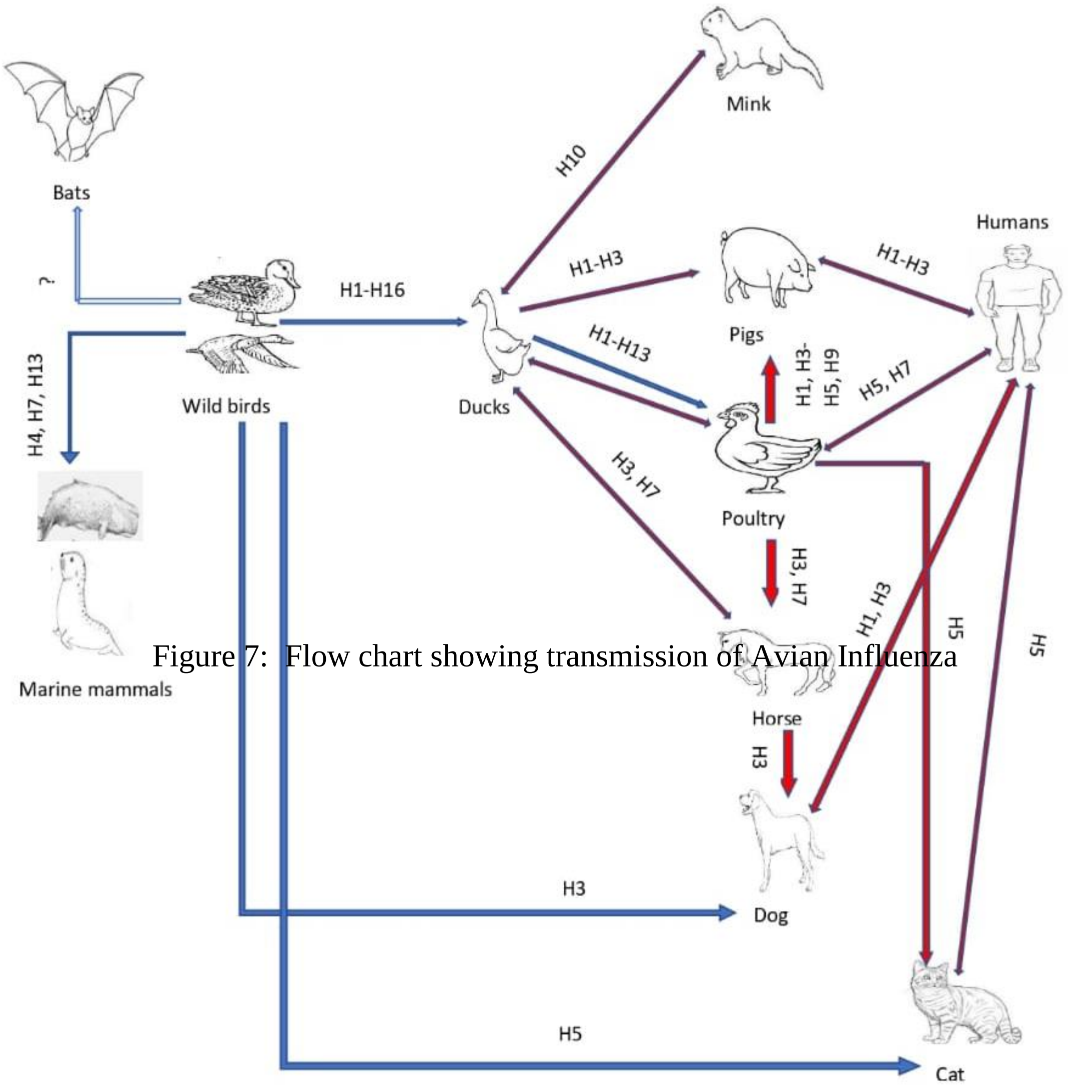

Figure 7: Flow chart showing transmission of Avian Influenza

Chapter 11 : Nutrition And Livestock Health

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Abstract

Nutrition is a foundational aspect for growth, production, reproduction, health and overall all wellbeing of livestock, and plays a vital role in ensuring sustainable agriculture production and food security. The relationship between nutrition and animal health is intricate, where quality of feed and nutrient composition directly influence the efficiency of metabolism, physiological functions, immune responses and disease prevention. Livestock require sufficient and balanced amounts of macro-nutrients like fiber, non-fiber carbohydrates, proteins and lipids as well as micro-nutrients like minerals and vitamins. All nutrients function in an integrated manner to influence growth, development and health of livestock and poultry. Nutrients in the feed are translated into products that human consume and therefore, any hazard in the animal feed negatively affects human health too. This chapter presents an overview on the impact of nutrition on metabolism, physiology, ecology of gastro intestinal tract, immunity and reproduction in animals, in relation to health sustainability and wellbeing. Nutritional imbalances and deficiencies can lead to lower production efficiency, reduce fertility, poor health outcomes and increased susceptibility to infections. To ensure a healthy and balanced nutrition for livestock, a careful formulation of diet is essential which requires a sound knowledge of characteristics and limitations of ingredients, nutrient requirements according to physiological functions of animal and manufacturing processes to make finished feed. Different in-practice nutritional strategies which promote animal health are also discussed in this chapter including the use of various additives and dietary supplements specially the probiotics, regular monitoring of livestock health indicators such as body condition score, efficient management of grazing livestock in relation of parasitic infestation, nutritional aspects for stress management, role of water and forage in sustaining health, feed safety and preventing mycotoxicosis in feed manufacturing, nutritional intervention to improve gut health and nutritional manipulation to reduce methane gas emission. The modern livestock production system with the objective of producing organic food, nutrition should be considered as 'preventive medicine'. As the global demand for animal-based products continues to grow, a

holistic approach combining the health aspects and nutrition management is essential for promoting sustainable livestock farming and ensuring food safety.

11.1 INTRODUCTION

Nutrition is a process used by an organism to acquire and assimilate food (or feed) in order to be used for growth and replacement of damaged tissues. The nutrients (Learning) are essential for an organism to carry out its various physiological functions (Izquierdo et al., 2021; Kohlmeier, 2015). In animal nutrition, priority has always been given to the welfare and health of the animals (Salobir et al., 2012). Whereas, understanding of metabolism is learning about life as ultimate goal of metabolism is to provide energy to the cells through a series of reactions (metabolic pathways) to sustain life of the organisms (Judge & Dodd, 2020).

Nutrition, metabolism, and livestock health are closely interrelated concepts in animal agriculture. Proper nutrition and metabolism are foundation for animal growth, development, reproduction and health of livestock (Wu, 2022). Proper management of animal health involves delivery of nutritious feed (composition of diet with ample necessary nutrients), provision of clean water, clean and comfortable environment (living) as well as reliable veterinary services can enhance productivity (as animals are less morbid with reduced mortality), and safeguard public health by preventing contact with zoonotic diseases (Sudda et al., 2017). In this chapter, we will provide an overview of the nutrition (and metabolism) on health management of livestock especially ruminants.

11.1.1 Importance of Nutrition and Livestock Health in the Context of Livestock Production

Livestock being one of the major subsectors of agriculture witnesses rapid growth due to augmented demand for livestock products. Despite the importance of livestock sector in both developed and developing counties; more livestock growth can be predicted in developing countries in the years to come (Abubakar et al., 2020). In order to maintain satisfactory performance of newborn, growing, finishing and breeding livestock, it is utmost necessary to meet nutritional requirements of animals through quality feeds. Quality feeds not only supports the productivity of livestock, it is also of substantial value to healthy living of the animals. Quality feed thus readily enhance key metabolic functions of the livestock for immunity, improved fertility and reproductive efficiency, response of animals to vaccines, survival of newborn, growth, feed use efficiency, and meat quality (Lawal-Adebowale, 2020) *(refer to Fig. 1 for 'feeds and feeding impact on livestock operation').*

Unbalance feed (under or over nutrition) has direct adverse effect on livestock health, and puts animals in more risk of capturing infectious diseases. Further, treatment of diseased animals in the form of medicines is not only costly but may be less effective in certain cases. Vaccination during malnutrition may not also properly immunize animals. Balanced nutrition thus can lessen the risk of infective diseases by improving cell-tissue integrity and immune system (defense mechanisms) (FAO, 2012b). Proper nutrition also enhances blood immuno-globulins suggesting improved immunity (FAO, 2012a). Supplementation with antioxidants, minerals and amino acids (AAs, e.g., methionine) play a role in immune stimulation (Celi et al., 2014; Jankowski et al., 2014). Good nutrition is also a biosecurity measure for control of infective and zoonotic diseases (Makkar, 2016).

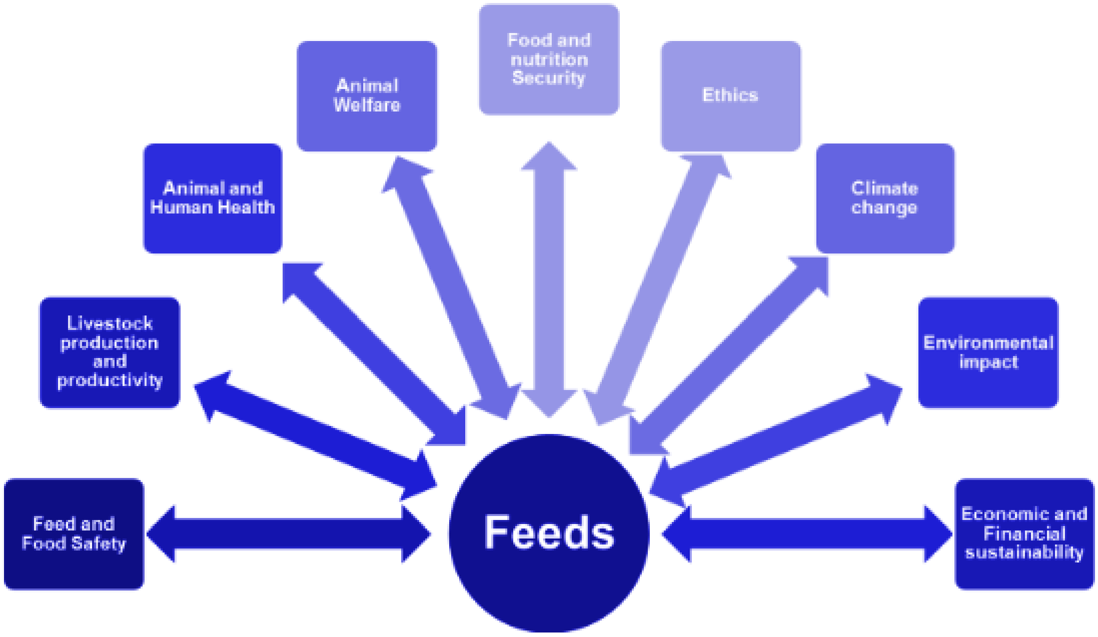


Figure 8 Feeds and feeding impact on livestock operation. Source adapted form (Makkar, 2016).

11.1.2 Link between nutrition, livestock health and human health

In the face of growing worldly population of human and animals, and with rapid changes in environment; the link between human, animals, and health of environment are becoming more evident (Rabinowitz & Conti, 2013). One of the key point of the profitable livestock industry (with a special focus on ruminants in this chapter) is to feed animals with low-cost, readily available (sustainable as well) and high-quality ingredients (Tedeschi et al., 2017). Further, livestock feed represents the initial point of food safety in the farm-to-table supply chains e.g., in case of ruminants (Fig. 2). Thus, provision of safe feed to livestock is central to human food safety, since, feeds might get toxic or contaminated with physical, chemical or biological hazards (e.g., pesticide residues, mycotoxins due to fungal infections or heavy metals in industrial areas especially lead, arsenic, chromium, copper, cadmium) during the harvest of raw ingredients, manufacturing, processing, packaging, storage, or transport of finished product (Patra, 2022). Eventually, foods of animal origin containing high concentrations of these heavy metals become concern of public health (Kar & Patra, 2021).

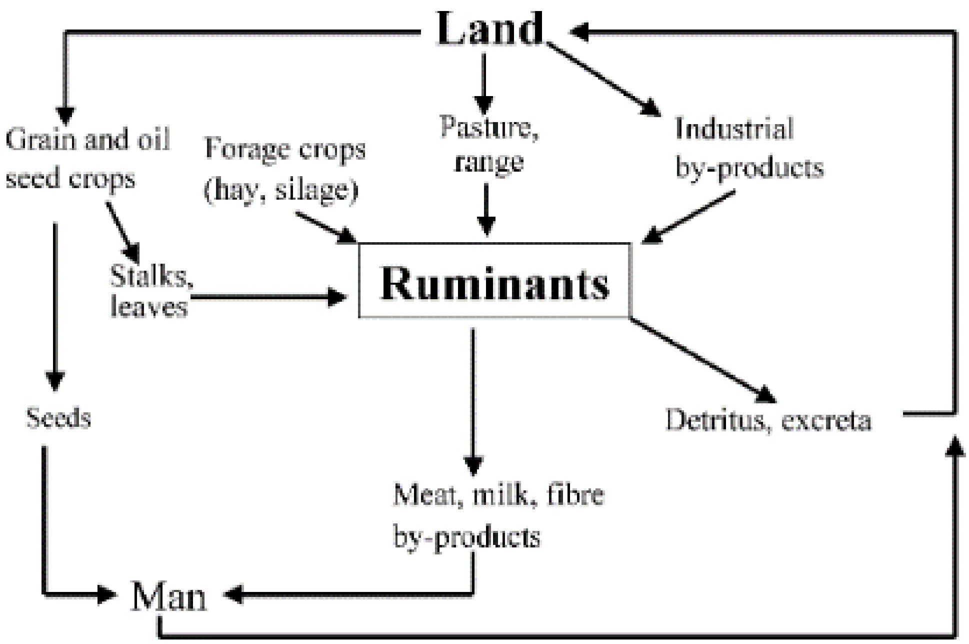


Figure 9 Role of ruminants in human nutrition. Source: (DeRamus, 2004)

11.1.3 Overview of the chapter's objectives and structure

Chapter’s objective is to elucidate of the importance of quality nutrition in maintaining proper metabolism and hence the health of livestock. Thus, the healthy livestock products have ultimate connection with the human health. When it comes to the health of livestock, veterinary medicine (VM) comes into the mind. The importance of VM in maintaining livestock health is crucial but little attention has been given on the concept of ‘healthy feed for healthy livestock to healthy food’. Although the contents covered in this chapter are exhaustive in nature and each and every aspect may need a book chapter to be described properly; yet an attempt has been made to at least clarify the main concepts. The readers must pay attention that description of livestock in general should be considered as ruminants with much focus on dairy animals as much of our knowledge on ruminants comes from dairy research; and similar is the case with monogastric species which denotes the poultry birds especially chickens and pigs in some instances as well.

11.2 UNDERSTANDING LIVESTOCK NUTRITION FOR VETERINARY HEALTH

11.2.1 Essential nutrients for livestock and their functions

In 18th and 19th centuries, animal nutrition science identified essential nutrients such as carbohydrates, lipids and proteins. It took however, more than 100 years (around latter half of the twentieth century) of research in nutritional sciences, more nutrients like vitamins, amino acids, fatty acids and minerals (inorganic compounds of food/feed) were recognized in details. Moreover, during that era, the nutrient requirements of animals at various physiological stages (i.e., growth, pregnancy and lactation) were defined (Pond et al., 2004; Stanton & LeValley, 2006). From a practical perspective, feeding program should warrant sufficient intakes of proteins (supply of essential as well non-essential AAs), carbohydrates, lipids, vitamins and minerals (Greene, 2016; NRC, 2002; Wu, 2017) through a supplementation program to correct deficiencies in basal diets (such as maize and soybean meal-based (SBM-based) diets for monogastric, milk replacer for kids, lambs and calves; and forage for ruminants) of livestock (Dept of Anim Sci. T. A. M. U, 2024). Likewise, non-ruminants (e.g., pigs and chickens) fed typical maize and SBM-based and not having supplementation with deficient AAs, high productive performance in case of meat and eggs can't be reached (He et al., 2021; Zhang et al., 2021). Growth, development and health of the livestock is influenced by the interaction of all feed nutrients with one-another, gut microbes, and with environment (Wu, 2022). Although nutrients deficiency may not be problem in confined animals (intensive productive system; IPS) when diet are carefully balanced but grazing ruminants (extensive production system; EPS) show suboptimal performance i.e., reduce growth and lactation if not supplemented with protein or minerals (Bergen, 2021; Greene, 2016) to cover deficiencies.

As stated above, typical maize and soybean diets need supplementation to cover amino acids deficiency. For all animals as well as humans, there exist the dietary requirement of proteinogenic AAs (for AAs synthesized de novo i.e., proline, glycine, glutamate and glutamine and AAs having no de novo synthesis i.e., leucine and lysine (Wu, 2021). AAs are not only important in protein synthesis inside animal body but these also regulate cell function and are vital for maintaining growth and health of animals (Wu, 2010) (Fig. 3).

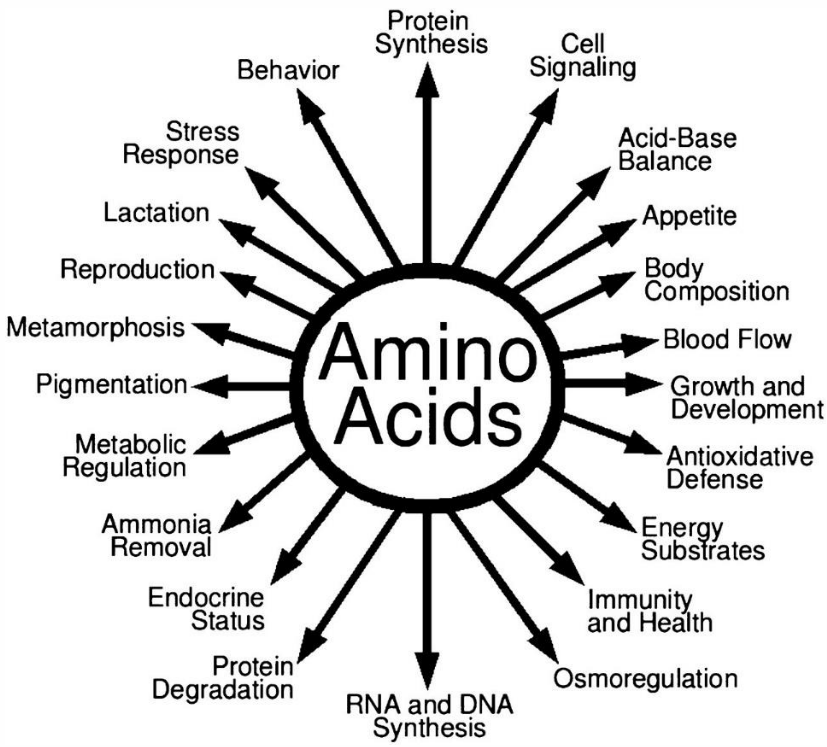


Figure 10 Role of AAs in nutrition and homeostasis of whole-body. Source: (Wu, 2010).

Some functional AAs are responsible for metabolic regulation, health improvement, growth and development as well as reproduction and lactation of animals. These relatively new concepts in nutrition are shaping feeding practices for livestock (poultry and aquaculture as well) around the globe (Chalvon-Demersay et al., 2021; Rodrigues et al., 2021; Rossi Jr et al., 2021).

11.2.2 Digestive physiology of different livestock

Farm livestock (including poultry) are broadly classified as either ruminants (pre-gastric fermenters) or non-ruminants (post-gastric fermenters) based on their diets and stomach type i.e., herbivores (ruminants such as sheep, goat, cattle and buffalo etc.) have complex stomach to efficiently digest plant materials mainly fibrous stuff, whereas omnivores (non-ruminants such as swine and poultry) have simple stomach due to relative ease for digestion of food stuff. Ruminant's stomach is known as forestomach (because most of digestion process takes place in front portion of gastro-intestinal tract; GIT), which is composed of four chambers – (1) the rumen, (2) reticulum, (3) omasum and (4) abomasum ('true stomach' equivalent to single stomach of monogastric) (Fig. 4). The rumen is large fermentation vat with complex and dynamic ecosystem consisting of strictly anaerobic bacteria and fungi, archaea, protozoa, and bacteriophages. These microbes work their own way and interact with each other to establish a symbiotic relationship with host in a way to provide it basic nutrients (mainly energy) from the breakdown of fibrous parts of plant feeding material. The microbial digestion of plant material in ruminants enables them to convert human inedible feeds into edible nutritious food of human use (i.e., milk and meat) and to thrive on meagre lands unfit for cultivation of food crops (Huws et al., 2024).

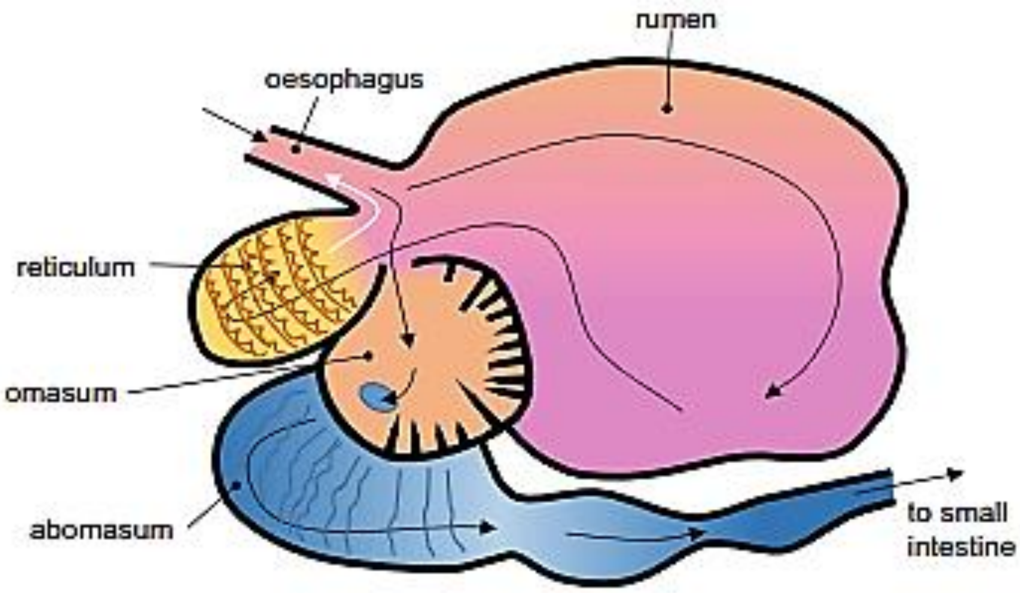
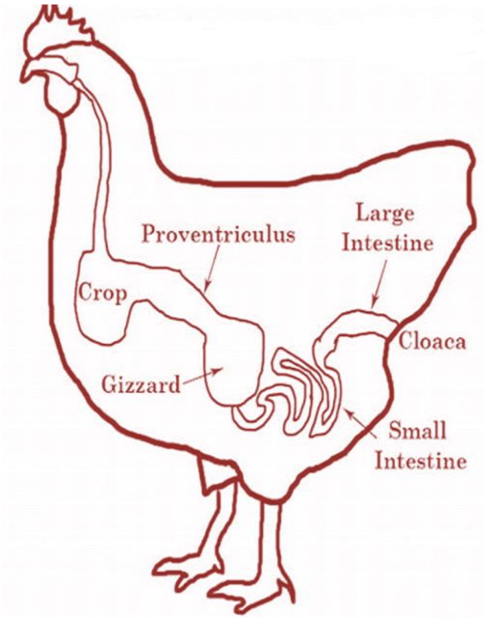


Figure 11 Basic morphology of the ruminant stomach. The flow of feed is indicated with black arrows white arrow indicates feed regurgitation for re-mastication (rumination) Source: adapted from (García-Yuste & Pérez-Barbería, 2020).

Non-ruminant animals are single stomach animals (rabbits, cats, dogs, pigs, horses and birds) with in the digestive tract hence called monogastric (Fig. 5 & 6). The diet of non-ruminants is either meat (called carnivores) or diverse diet with mixture of plant material and meat (omnivores). The only difference between ruminants and non-ruminants is the presence of compound stomach or single stomach in these animals respectively, while other organs of the digestive tract remain same such as mouth, esophagus, small intestine (rabbits, however, have bigger small intestine and cecum, portion of large intestine to allow more time to digest plant material). The poultry birds have two chambered stomachs: (1) the proventriculus (secretory organ of gastric juice) and (2) the gizzard (organ of storage, soaking and mechanical grounding feed).



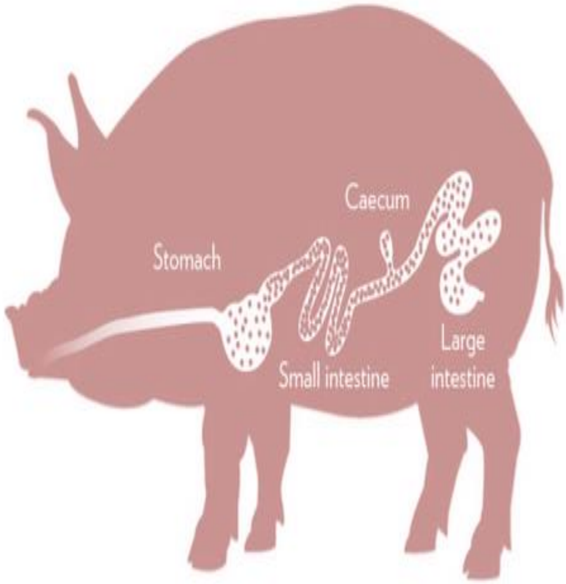


Figure 12 Non-ruminant stomach (swine)

Figure 13 Non-ruminant stomach (bird)

11.2.3 Dietary strategies to influence the gastro- intestinal ecology and to improve intestinal health

Feeds and feed components contribute to animal wellbeing (physical as well as mental). Nutrition through a well-balanced diet contributes to optimal intestinal and microflora. Active ingestion of functional foods, such as dietary fiber (DtF), oligosaccharides (prebiotics), and probiotics encourage colonization of useful bacteria (while suppressing pathogenic bacteria). Recent research advancement in intestinal flora have provided background for the appearance of functional foods (Mitsuoka, 2000).

The productive performance of animals and their impact on environmental are chiefly the result of biochemical process occurring in the rumen after entry of feed and start of digestion process by rumen microbes (Huws et al., 2018; Mizrahi et al., 2021; Ungerfeld, 2020). Basically, carbohydrates in the diet of ruminants after their break down by the rumen microbes and going through biochemical processes that culminate in the production of short chain fatty acids (SCFAs) which ultimate provide energy source to the host (Fig. 6 and 7). This process results in the production of hydrogen (H_2), and it is used by methanogens (ruminal archaea) mainly to convert CO_2 into methane (CH_4) (Fig. 6).

Robert Hungate, an American scientist developed culture techniques (still used trough out the word) for anerobic bacteria (Clarke & Hungate, 1966; Sterilization, 1969). That is the reason that he is widely considered to be the "father of rumen microbiology". Using culture techniques, it has been demonstrated that bacteria are the diverse and most abundant group with a multitude of functions in ruminal ecology. Based on their multitude of functions, ruminal bacteria can be described as amylolytic (converting starch into sugar), cellulolytic (converting cellulose to short chain fatty acids), proteolytic (bacterial protease enzymes that break peptide bonds in protein molecules) or lipolytic (break lipids due to extracellular lipases which hydrolyze triglycerides), with many bacteria described as generalists or specialists i.e., having broad range of functionality or specific to certain tasks only respectively (Fig. 7). Despite of technological advancement and more understanding of the ruminal microorganism, the functional role of many rumen bacteria and their interactions among themselves and with the host are still largely unknown. This knowledge gap has relatively constrained attempts to beneficially modulate rumen microbiome for improved production and reduced environmental impact (*refer to 'balancing the diet for a healthy rumen ecology' for details*).

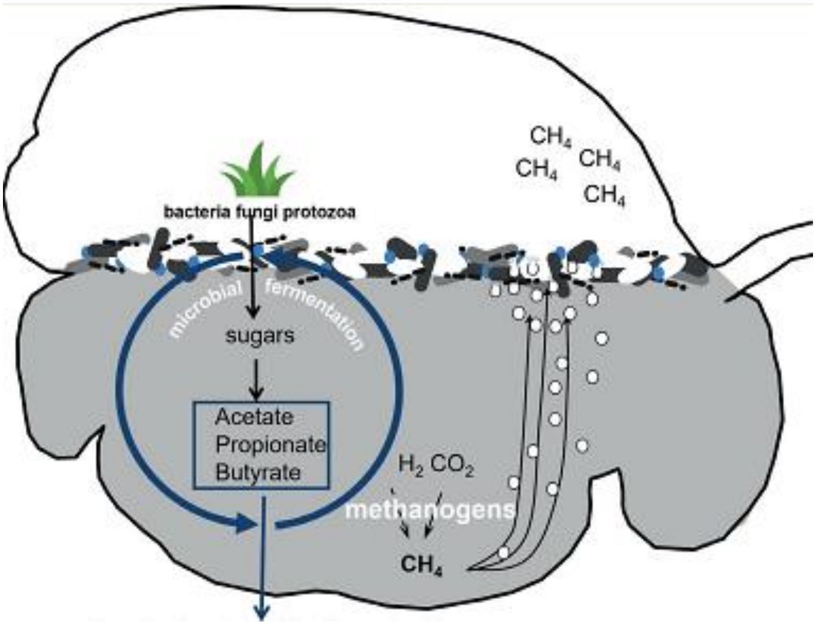


Figure 14 Stomach of ruminants. Source: Adapted form (Huws et al., 2024).

Apart from energy source, SCFAs trigger epithelial immune response of rumen, hence contributing to ruminants' health (Zhan et al., 2019). Production of certain SCFAs i.e., propionate uses more H_2 than others (acetate and butyrate), and can therefore redirect H_2 away from methanogenesis (CH_4 generation) in rumen. This H_2 redirection away from methane generation can be a reason that ruminants having improved parameters of production emit less methane naturally (Huws et al., 2024). (*Refer to 'nutritional strategies to mitigate methane emission' for details).*

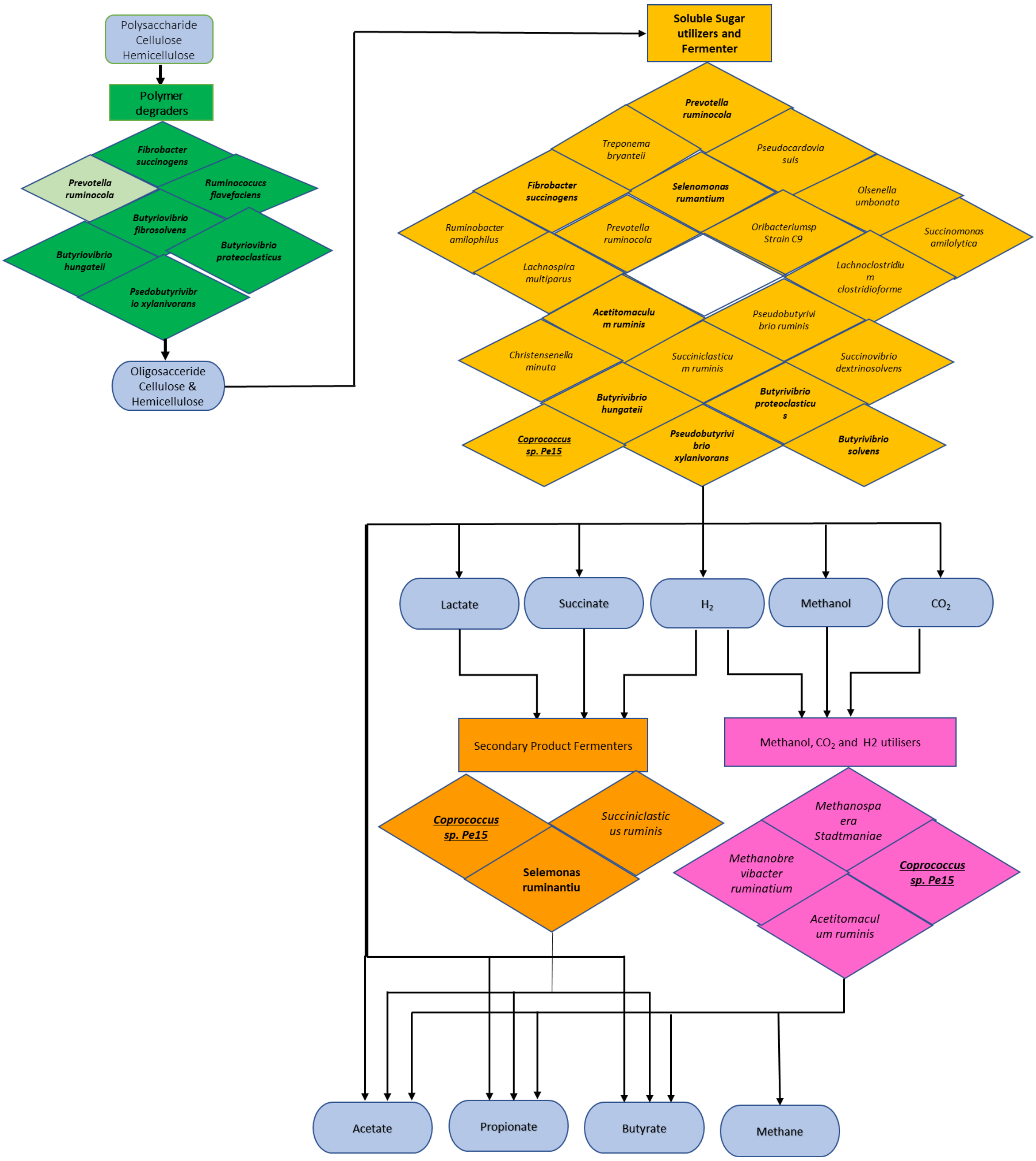


Figure 15 Outline/overview of the stages and examples of bacteria involved in plant material degradation in rumen ecosystem. Source: Adapted from (Huws et al., 2024)

Note: The bacterial species shown in the above figure act as polymer degraders, utilizers of soluble sugar and fermenters, fermenters of secondary product, and methanol, utilizers of CO_2 and H_2 in the rumen. Many in specific stages of this process are specialist, but some of these bacteria (names highlighted in bold) are identified as being active in two phases, or even bacterial activity (highlighted names in bold and underlined) in three phases. Polymer degraders consist of primary colonizers (light green), secondary colonizers (dark green) and those with no early-phase temporal pattern (white).

11.2.4 Metabolism of Nutrients

Metabolism is consisted of total chemical reactions in cells necessary for life and involve either break down of complex macromolecules into simpler ones e.g., CO_2, H_2O and NH_3 (catabolism) or synthesis of complex macromolecules e.g., nucleic acids, lipids, proteins and polysaccharides (Fig. 8). Metabolism provides energy (in the form of ATP) to organisms continuously for maintenance of cellular and whole-body function, for mechanical work and active transport of ions such as Na^+, K^+, Ca^{2+} and Mg^{2+} (Judge & Dodd, 2020).

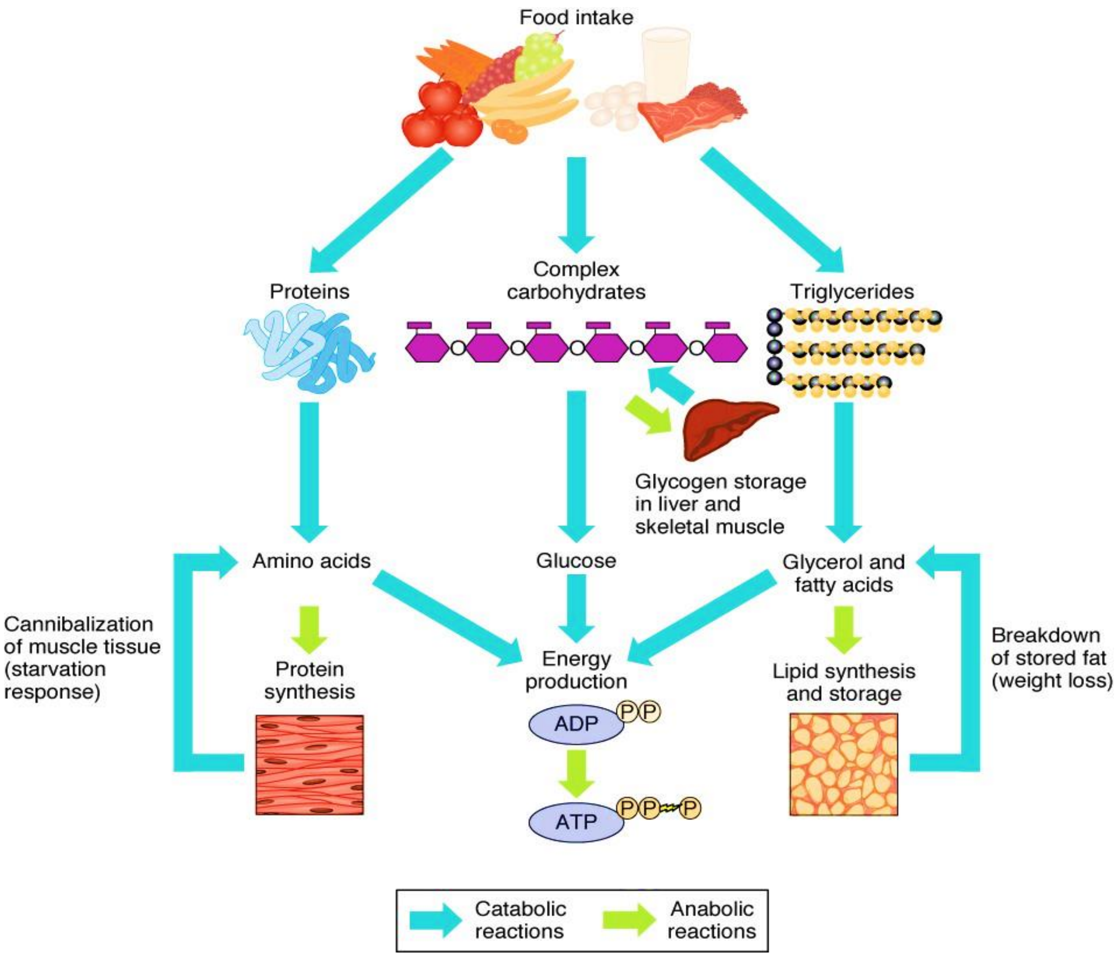


Figure 16 Connection of catabolic and anabolic processes during metabolism. Source adapted from: (Learning, 2023).

Catabolism results in release of energy when macromolecules are broken down into basic building block (proteins into amino acids, fats to fatty acids and polysaccharides to monosaccharides), whereas, catabolism consumes energy to form macromolecules and biomolecular polymers inside cells from available building block after catabolism.

11.2.5 Role of Feed Composition in Meeting Nutritional Needs

Fulfilling need of required nutrients though feed is one of the basics to maintain animal health (Salobir et al., 2012). Hence, the ultimate goal of feed composition (feed analysis) is to predict the productive response (also known as biological response) of animals when given a 'feed' in question with known composition. This response is function of feed chemical composition and the animal's ability to fetch useful nutrients (digestibility for availability of a nutrient in the feed for absorption into animal body and its ultimate efficiency of use for productivity and health) from the feed (Stanton & LeValley, 2006).

To support the productivity of the livestock; provision of quality feeds adequately supplying the energy requirement for maintenance (animals with higher body weight will have higher energy requirements for maintenance) and production (high producing animals will have high energy demands) of animals are needed. Using information of nutrients required (from nutrient requirement tables for various livestock species), rations can be formulated in a manner that animals remain healthy, efficient and productive. Till now, > 40 nutrients have been identified and deemed necessary to be included in diets of various livestock species. Manufacturing of nutrients for inclusion as feed additives and greater knowledge related to biochemistry, physiology of digestion and metabolism has helped in improved diet formulation and to adequately feed our livestock according to different physiological states (growth, pregnancy & lactation), leading to obvious benefits of animal welfare and health (Pond et al., 2004).

11.2.6 Metabolic Disorders of Macro and Micro Nutrients

Poor quality forages or lack of supplemental feeding on EPS are probably most frequent cause of energy and/or protein deficiency (malnutrition due to macronutrients). Although vitamin deficiency is not a problem on grazing yet mineral deficiency can be a problem (most often minerals supplementation is recommended in grazing conditions) due to low soil availability of minerals or spoilage of mature grasses (a possibility of excess intake of minerals due to soil conditions cannot be ruled out). However, under confinement situation (IPS), there is usually no deficiency or excess of macro or micro (vitamins and minerals) nutrients when diets are formulated exactly according to the requirements of high-yielding animals. However, unsuitable feeding or unbalanced feeding may lead to nutrient deficiency during very early lactation with increased risk of metabolic (ketosis) or infectious (e.g., mastitis) diseases (Drackley, 1999).

Malnutrition can occur in several ways, deficiency of single macro or micro nutrient, or in combination, such as energy-protein malnutrition and micronutrients deficiency such as vitamin and minerals (Calder & Jackson, 2000) (Fig. 9).

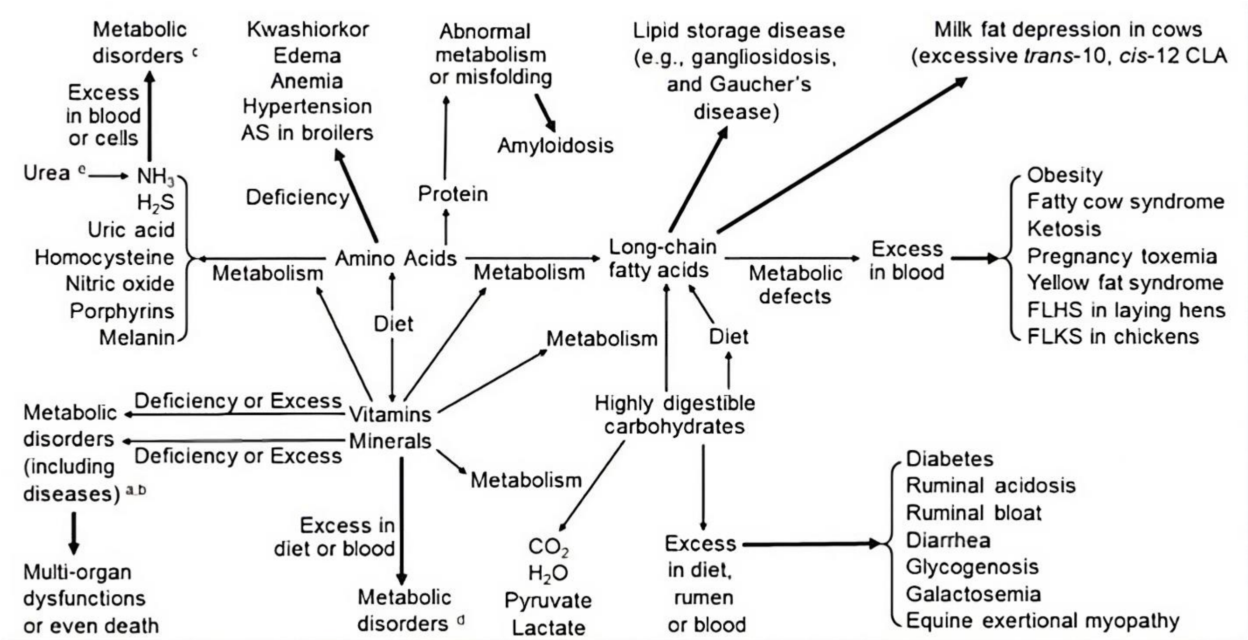


Figure 17 Overview of metabolic disorders (including metabolic diseases as well) in ruminant and nonruminant livestock species. Source: Adapted with slight modification from (Wu, 2022).

Livestock (including poultry) can suffer from dysfunctions of multiple organs, disease conditions or even mortality due to abnormal metabolism which stems from abnormal biochemical pathways resulting from deficient enzymes, co-enzymes and co-factors. [a]Multiple disorders or diseases can occur due to deficiency of vitamin(s) in ruminants and other species such as polioencephalomalacia (also known as cerebrocortical necrosis, a nutritional disease characterized by neurological signs in ruminants) and beriberi (nutritional disorder manifested by nerves impairment and heart, feeling of numbness and weakness in limbs and extremities, digestive irregularities and generalized symptoms of appetite and overall lassitude due to thiamin/vitamin B1 deficiency, pellagra (characterized by diarrhea, dermatitis, dementia, and death) due to niacin/vitamin B3 deficiency, burning-foot syndrome (uncomfortably hot and painful feet) due to pantothenic acid/vitamin B5 deficiency, neural tube defects (birth defects of the brain, spine or spinal cord) due to folate/vitamin B9, scurvy (weakness, fatigue, spontaneous bleeding, sore arms and legs and sometimes ulceration of the gums as well as loss of teeth) due to ascorbic acid/vitamin C deficiency, photophobia (extreme sensitivity to light causing pain and discomfort in eyes due to riboflavin/vitamin B2 deficiency, xerophthalmia (abnormal dryness of conjunctiva and cornea of eyes with inflammation) and keratomalacia (softening of cornea of eyes, is a conditions grouped under xerophthalmia) due to vitamin A deficiency, rickets (bone deformity in kids due to defective mineralization of growth plate resulting into soft and week bones giving rise to bone pain and poor growth) and osteomalacia (week of soft bones in adults result from defective mineralization of the preformed osteoid) due to vitamin D deficiency, liver steatosis (build-up of excess fat in the liver cells) due to choline (a nutrient similar to B vitamins) deficiency, myopathy (generalized term to indicate disease affecting muscles which control voluntary movement in body) and hepatic necrosis (death of liver cells) due to vitamin E (fat-soluble vitamin that include four tocopherols and four tocotrienols) deficiency, hemorrhage (loss of blood from a damaged blood vessel) due to vitamin K (vitamins K1 (phytonadione) and K2 (menaquinone) are commonly available as supplements.) deficiency, and infertility (vitamins A & E). [b]Disorders (including diseases) caused due to mineral deficiency include parturient paresis in dairy animals (milk fever with subnormal body temperature is a metabolic disorder occur around calving due to insufficient calcium mobilization), rickets, and osteomalacia due to Ca, grass tetany (a nutritional or metabolic disorder occurs more frequently in cattle eating lush green immature pasture, but sometimes winter tetany may also occur to feeding dry forages) due to Mg deficiency, anemia (condition in which the body does not have enough healthy red blood cells) and hemorrhage due to Fe deficiency, ammonia toxicity due to Mn, Keshan's disease (a highly lethal but treatable cardiac disease first reported in 1935 in Keshan country of Northeast, characterized by congestive heart failure, acute heart failure, and cardiac arrhythmia) due to Se, goiter (any enlarged thyroid gland) due to I/iodine, dental caries (holes, or areas of tooth decay) due to F, Menke's (disease is caused by mutations in blood Cu metabolism regulating ATP7A gene) and Wilson's diseases (due to genetic disorder, there is problem in removing extra Cu leading to build up of Cu in tissues e.g., in eyes, brain, liver and other organs) due to Cu, and infertility due to P. [c]Disorders (including diseases) due to excessive levels of amino acid metabolites are: hyperhomocysteinemia (condition result when blood homocysteine level is >15 micromol/L), hyperammonemia (raised levels of ammonia, a nitrogen-containing compound and is toxic at high level), gout (a painful form of arthritis, a disease inflammation and stiffness of the joints), melanosis (a form of hyperpigmentation associated with increased melanin), and porphyria (buildup of natural chemicals 'porphyrins' in the body which affects skin, gastro-intestinal system and nervous system). [d]Disorders (including diseases) caused by hypervitaminosis (excessive vitamin) of lipid soluble vitamin i.e., vitamin A, D, E, and K, whereas diseases due to excessive minerals in ruminants include polioencephalomalacia (common neurologic disease with signs of incoordination, circling, wandering, cortical blindness, head pressing, recumbency and seizure activity) due to sulfate, hypertension due to Na, hyperkalemic periodic paralysis (a disorder that sometimes results in more than normal K blood levels and with occasional episodes of muscle weakness) in horses due to K, Cu toxicity, and selenosis (poisoning due to accidental or intentional excessive intake) due to Se. [e]This reaction in rumen fluid (of ruminants) and intestines (of all animals) is catalyzed by an enzyme urease.

Abbreviations: AS, ascites syndrome; CLA, conjugated linoleic acid; FLHS, fatty liver hemorrhagic syndrome; FLKS, fatty liver and kidney syndrome

However, malnutrition effects are mainly manifested on tissue damage, digestive disorders and metabolic diseases; all of these cause release of cytokine leading to inflammation and impairment of animal welfare (Fig. 10) (Bertoni et al., 2016).

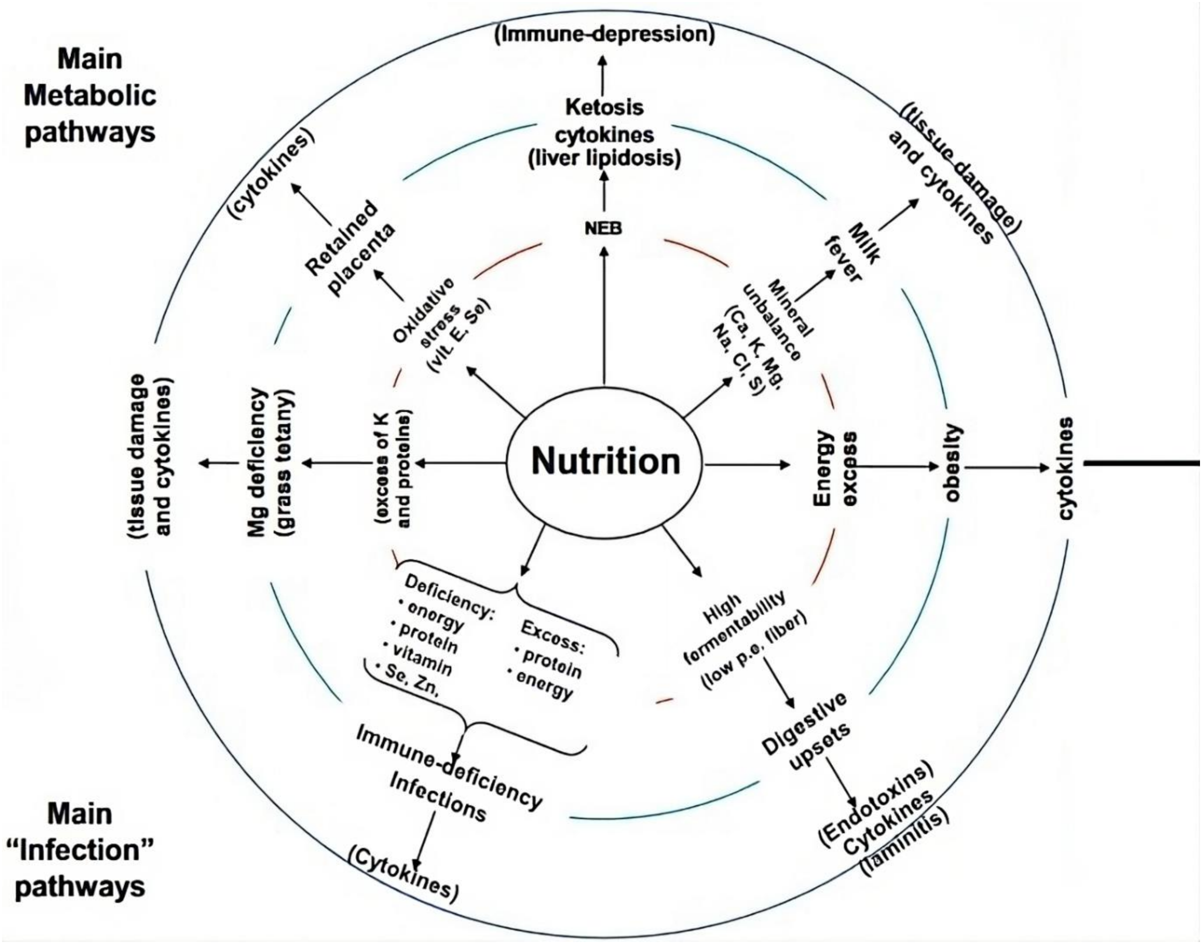


Figure 18 Different pathways from nutrition to health disorders. Source: Adapted from (Bertoni et al., 2016).

In EPS, energy deficiency due to lack of good forages or concentrates can cause a specific metabolic disease in ewes and goats known as pregnancy toxemia. This disease specifically hits animals carrying twins or triplets (due to non-fulfillment of high energy demands of fetuses) at the end of pregnancy period (LeValley, 2010). In goats, hyperketonemia in late pregnancy is followed by inflammation (Trevisi et al., 2005), suggesting induction of cytokines (inflammation) even without clinical signs contribute to energy shortage and thus lead to metabolic disease (Bertoni et al., 2016). Intoxicants i.e., phytoestrogens with *Medicago spp*. and *Trifolium spp* (subterranean clover) (D'mello, 2000); mycotoxicants from *Claviceps spp.* in fungi-infected grasses, especially fescues; or toxic weed intake (in certain cases of grass shortage) are also a nutritionally related problem under grazing management system (*refer to 'health management of grazing animals' for details)* (Bertoni et al., 2016). A number of mycotoxins adversely affect digestive morphology and rumen function (D'mello, 2000).

11.2.7 Nutritional management of reproductive performance

Since long, dairy farmers, animal nutritionists, veterinary professionals, feed dealers, and extension service providers have been interested in connection between reproduction and animal nutrition (Amin, 2014; Hess et al., 2005). Nutritional deficiencies cause various reproductive problems. Malnutrition of macro-nutrients i.e., energy and protein (or even excessive protein), and micro-nutrients deficiency i.e., trace minerals, and vitamins are stated as contributory factors to infertility and thus poor reproductive performance. Poor nutrition cause both the cessation of estrus along with suppression of ovulation or may cause ovulation without estrus. Under-nutrition also results in greater number of services per conception (Amin, 2014).

The nutritional status of animal determines the live weight (LW) and body condition score (BCS; *refer to 'monitoring body condition score (BCS) for livestock health' for details*) throughout lifetime period; and both were declared more than half a century ago to underpin fertility in pubertal heifers and postpartum cows (Michael et al., 2019). There has a strong correlation between BCS of a cow and her productive and reproductive performance (Felix et al., 2024). BCS in *Ovine* can reflect lamb birth and growth rate, ovulation, lambing rates and earlier returns to breeding (Fisher, 2020). In dairy cows, BCS at calving are related to first estrus after calving (cows in greater condition soon resume normal estrous cycles) and ultimately result in shorter calving intervals. Around calving, cows with greater BCS deliver calves having better average daily gain and heavier weaning weights. Avoiding a decrease in BCS is must during pregnancy as it may affect fetal programming. Resultantly, poor nutritional of dam during pregnancy may have long-term negative impacts on the offspring.

The importance of proper nutrition in success of dairy cattle reproduction has been recognized by the professionals of livestock industry since long ago (Hess et al., 2005). Direction of nutritional inputs towards anabolic process is utmost necessary to stimulate key events for reproductive success. Reproduction is greatly influenced by hypothalamo-hypophyseal-ovarian axis in female cows. Nutritional inputs likely impinge on the hypothalamo-hypophyseal-ovarian axis to influence reproduction. In this regard, energy balance for example in beef cows before parturition (pre-partum) and after parturition (postpartum) has been described to affect duration of the postpartum interval to first estrus. Nutrition of beef cows with dietary fats for approximately 60 days before calving can improve pregnancy rates in the upcoming breeding season. In a meta-analysis, it has been indicated that fats feeding can improve fertility (overall 27% improvement in pregnancy to service and decrease in calving to pregnancy interval) in dairy cows (Rodney et al., 2015). commonly available sources of lipids to increase energy density may include cotton seed, soybeans, rice bran, safflower, sunflower, animal tallow, fishmeal, and calcium salts of fatty acids. However, supplementation of lipid high in in linoleic acid to postpartum diet can impede reproductive performance (Hess et al., 2005).

Starting from prepubertal period, low plane of nutrition delays the onset of puberty through inhibition of the endocrine reproductive system development (Izquierdo et al., 2021). Whereas, puberty in animals comprises a complex biological event involving maturation of the reproductive- neuroendocrine-axis and subsequent initiation of high-frequency, episodic release of reproductive hormones i.e., Gonadotropic releasing hormone (GnRH) and Luteinizing hormone (LH). High plane of nutrition (in HFxKankrej crossbred heifers) from early age shortened (2-3 months) the age at puberty and sexual maturity than routine farm fed heifers (Dhami et al., 2019). Increasing proteins levels in the diet of Zebu cattle has also been associated with improved reproductive performance (Oyedipe et al., 1982). Hence, nutrition is very critical for neuroendocrine control of puberty (Cardoso et al., 2020).

Although, the effects of poor nutrition especially energy is critical to reproduction in dairy cows, yet surplus energy during late lactation and after drying off (dry period) can cause “fat cow” that can reduce reproductive efficiency in subsequent lactation. Heifers with insufficient energy intake will have delayed sexual maturity (Amin, 2014). In this regard, dietary polyunsaturated fatty acids (PUFA) have been described to energy balance and thus to positive modulate reproductive function with the stimulation of follicular growth. PUFA of the ω-6 and ω-3 were found to greatly enhance the cattle reproductive responses if fed (especially ω-6 fatty acids) during late gestation and early lactation. Likewise, during lactation period, feeding ω-3 fatty acids helped maintain pregnancy and improved embryo quality (Santos et al., 2008). Some non-conventional feed such as seaweed or macroalgae are rich source of ω3 fatty acids, bioactive compounds, and colorants (Patra, 2022). The best recommendation for reproductive health at present is to devise a feeding program for livestock which meets the nutrients requirements and contains balanced amount of all nutrients (on a long term and continuing to obtain success).

11.3 NUTRITIONAL STRATEGIES FOR OPTIMAL HEALTH

11.3.1 Balancing the diet for a healthy rumen ecology

The concept of “gut health” is gaining substantial interest amongst nutritionists, veterinarians and scientists over the past few years (Celi et al., 2019). The rumen is a big fermentation chamber with microbial ecosystem consisting of huge populations of bacteria, protozoa, fungi, yeast, archaea (methanogens), and bacteriophages living symbiotically with the host (Soltan & Patra, 2021). Firstly, ruminants chew and regurgitate feed to physically break down the feed into smaller particles and thereby increasing surface area for microbial attachment on feed particles (dietary substrates). Microbes secrete enzymes for degradation and digestion of dietary substrates. Strong rumen movement to mix digesta provides more opportunity for microbes to attach with substrates and mix with these feed particles; and ultimately production of fermentation products mainly volatile fatty acids (VFAs; precisely described as SCFAs, useful for host), H_2, CO_2 and NH_3 (unusable by host). Further, rumen provide optimal ruminal environment (e.g., pH) by utilizing SCFAs (and to incorporate into meat and milk) to maintain growth and microbial protein synthesis (Belanche et al., 2021). (Fig. 11).

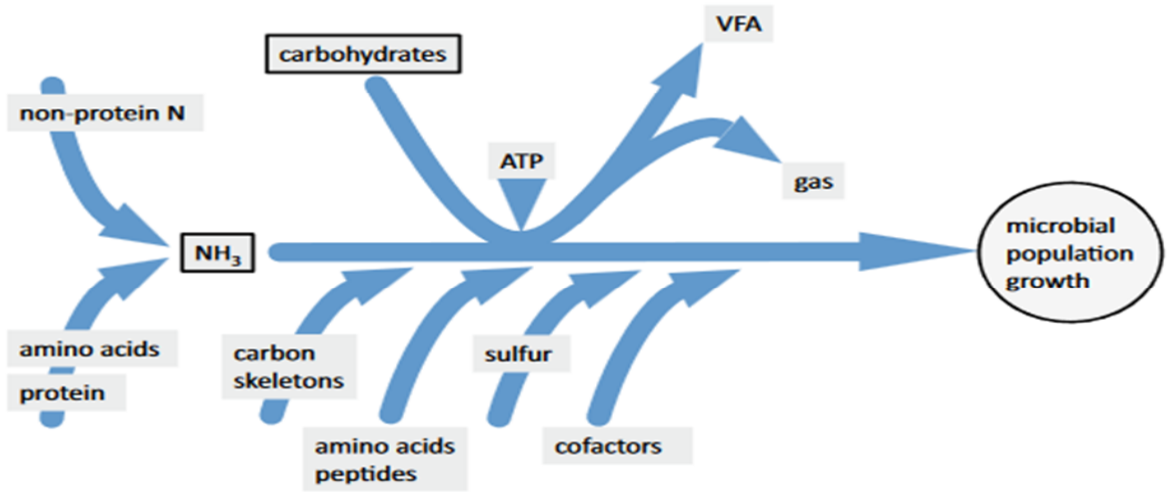


Figure 19 Flow diagram of nitrogen (protein and non-protein sources) and energy use for ruminal microbial protein synthesis (microbial population growth). Source: adapted from (García-Yuste & Pérez-Barbería, 2020).

Rumen ecosystem is consisted of a variety of micro-organisms living in a symbiotic relationship with host. The fermentation activity of the rumen can be modulated to a more useful output of microbial products by the nutritional strategy. Use of enzymes for degrading non-starch polysaccharides (NSP) improve fiber degradability and result into change ratio of acetate and propionate will lead to lower methane emission Some species of yeast stimulate acetogenesis and thus dispose H_2 away, which may otherwise be used for methanogenesis. Also adding dicarboxylic acids (e.g., fumarate) for the conversion to propionate utilizes H_2 and thus reduces the possibility of methane formation (Salobir et al., 2012). In this regard, propionate formation competes with methanogenesis for metabolic H_2 in rumen; and a rumen microecosystem favoring propionate formation can mitigate methane emission (Wang et al., 2023) (r*efer to 'nutritional manipulation to mitigate methane emission' for details*).

There exist an interaction among all dietary nutrients, with gastro-intestine (GIT) microbiota, and environment which in turn influence growth, development and health of animals (Wu, 2022). (Fig. 12). The composition (substrate type) and intake of diet are main determinants of not only the GIT microbiota (rumen microbiol ecology as well), but also productive outcomes in ruminants (Belanche et al., 2021). It has been indicated that cows fed high forage-diets had higher bacterial diversity with abundance of *Bacteroides, Fibrobacter and Ruminococcus* (plant cell polysaccharides degraders) when compared to cows fed high concentrate diet (Wang et al., 2019). Another metagenomics study on dairy goats fed high rumen degradable starch (HRDS) diet as compared to low indicated that goats fed HRDS had higher abundance of microbial genes encoding for amylases leading to higher propionate levels in rumen and with lower levels of genes encoding for cellulose degrading enzymes (Shen et al., 2020). Since, microbes play an important role in digestion of substrates in an interactive ecology of rumen, the manipulation of the ruminal fermentation pathways seems most effective approach to improve ruminant health and production efficiency without exaggerated increase in nutrient supply.

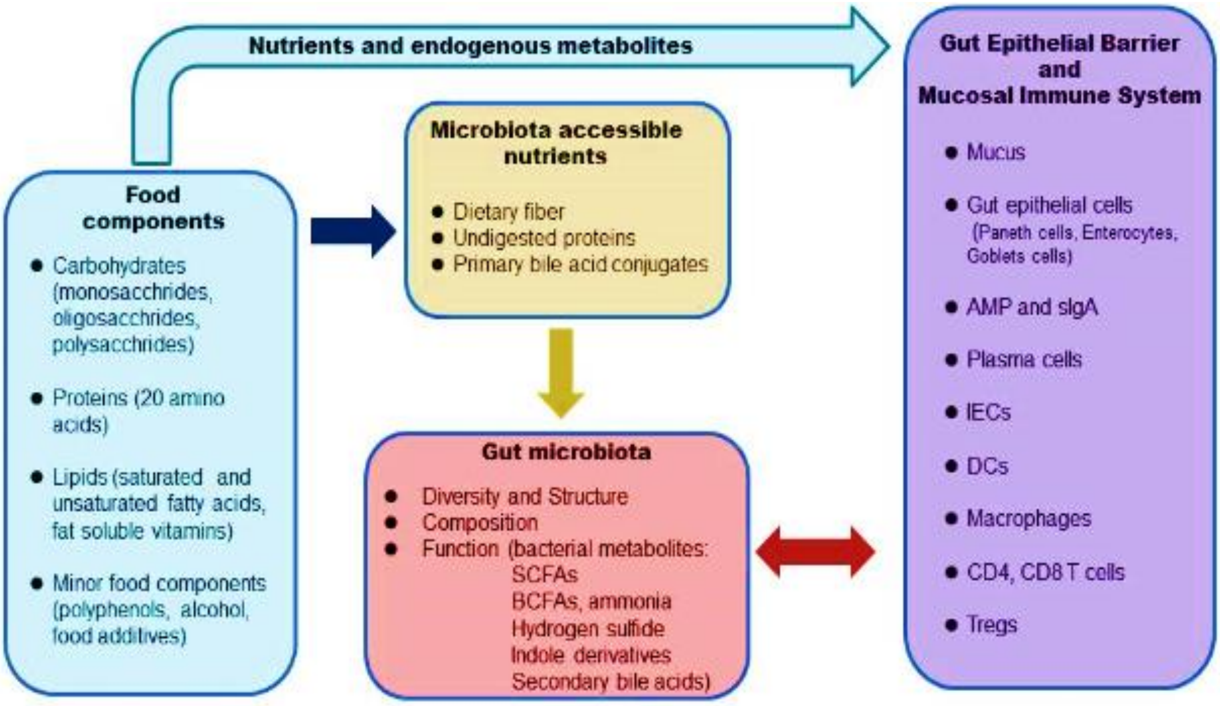


Figure 20 Foods and nutrients impact on the microbiota-host interactions in gut. Source: adapted from (Zhang, 2022)

Regulatory pathways are indicated by arrows. Food nutrients (carbs, protein, lipids, polyphenols and additives etc) and endogenous metabolites of nutrient metabolism directly modulate gut epithelial barrier function and mucosal immune response. Gut microbial ecology is also essentially determined by diet which regulates microbiota-accessible nutrients. In this way, interaction of gut microbes with the host epithelium and mucosal immune system shapes intestinal homeostasis.

Abbreviations: IEC, intraepithelial lymphocytes; AMP, antimicrobial peptides; sIgA, secretory mmunoglobulin A; DCs, dendritic cells; SCFAs, short-chain fatty acids; BCFAs, branched-chain fatty acid.

The diet of ruminants needs to be changed gradually to adjust microbiota to diet change. Otherwise, sudden diet change may challenge the microbiota and result in poor feed efficiency and rumen health. In such an experiment to check adaptability of beef calves to feedlots (with the assumption that nutrient portioning was the sole responsibility of the rumen microbiota), it was showed that beef calves previously fed concentrate diets for at least two weeks in comparison with those fed hay only (control) or restricted quantity of hay (low quality roughage diet), seem to adapt better to high concentrate diets which resulted in higher DMI, increased propionate concentration to control and lower species diversity to restricted animals during adaptation phase (Pinto et al., 2020); a phenomenon that have been described previously with higher feed efficiency in dairy cows (Shabat et al., 2016). It was concluded that that animal who had previously been exposed to concentrate diets were better adapted to high-concentrate diets when compared animals on restricted diets (Pinto et al., 2020).

The recent research focus has centered on the use of natural feed additives (*refer to 'health benefits of feed additives and dietary supplements'*) such as plant secondary metabolites (PSM) to improve ruminal fermentation, production and health of ruminants while minimizing the environmental impact (Singh et al., 2021; Singla et al., 2021) (Fig. 13). However, recently there has been a quest for substitution for chemical feed additives with natural feed stuffs and bioactive phytochemicals, with potential to modulate rumen fermentation and reduce CH_4 production (Dhanasekaran et al., 2020). In this regard, medium chain fatty acids (MCFA) as feed supplements in livestock for quick absorption mostly in stomach and start of small intestine (Dierick et al., 2002; Messens et al., 2010). MCFA are exciting source of energy and inhibit the colonization of bacterial pathogens. They also act as antimicrobials because lipid membranes of microorganisms are permeable for the MCFA and cell lysis occurs due to a decrease in intracellular pH.

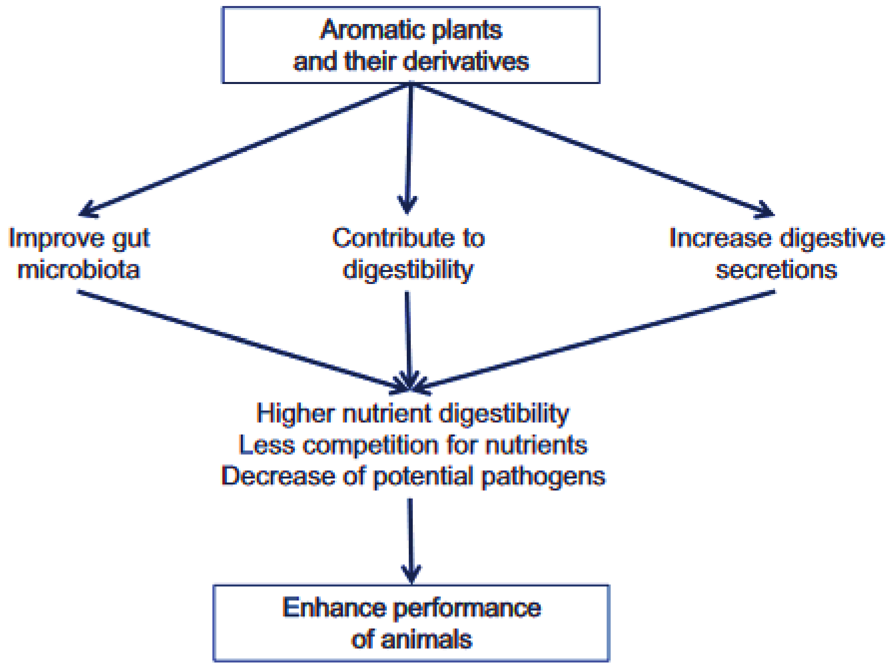


Figure 21 Possible modes of action of aromatic plants as growth promoters. Source: (Christaki et al., 2020).

These act through improvement in gut microbes, digestibility and digestive juices, reducing the potentially pathogenic bacterial loads in GIT and improved antioxidant and anti-inflammatory activities.

Use of metabolic modifiers ins dairy cows can change, augment or interfere with normal metabolic processes of ruminant digestive tract, or alter post absorption partitioning of nutrients among body tissues (McGuffey, 2017). Supplementations of various rumen modifiers such as dietary fats (polyunsaturated fats or medium chain) up to 6% of dry matter (Patra, 2013), some bacterial (*Lactobacillus*, *Bifidobacterium, Enterococcus, Streptococcus, Bacillus, Propionibacterium, Megasphaera elsdenii and Prevotellabryantii)* and fungal species (of yeast such as *Saccharomyces* and *Aspergillus),* fungal microorganisms as a direct-fed microbial

(Elghandour et al., 2015), Chitosan (Jiménez-Ocampo et al., 2019; Zanferari et al., 2018), phytochemicals and essential oils as substitutes for chemical feed additives in ruminants, PSM [terpenoids, tannins, saponins, and flavonoids] (Agarwal et al., 2009; Goel & Makkar, 2012; Ku-Vera et al., 2020; Matloup et al., 2017; McSweeney et al., 2001; Newbold et al., 2004; Patra & Saxena, 2010; Patra & Yu, 2015), bee propolis extracts (Morsy et al., 2015), nitrates and fumarates (Pal et al., 2015; Patra & Yu, 2015), Mimosine-a non-protein amino acid found in species of *Leucaena leucocephala*} (Soltan et al., 2017), *Moringa oleifera* root bark (Soltan et al., 2018), unconventional oil seeds such as safflower, poppy, hemp and camelina (Wang et al., 2017) have been used. For an in depth insight, refer to a comprehensive review of the 100 years (1917-2017) use of modifiers in dairy industry (McGuffey, 2017).

The specific gut microbes (*Megasphaera elsdenii*, and *Coprococcus catu*s; both may use acrylate pathway to utilize lactate for propionate production, are enriched microbes of efficient cows. Although, in lower diversity and richness, these bacteria have been suggested to be have a large impact on microbial community composition and ecosystem functioning yet these are very energy efficient and help harvest energy for efficient ruminant animals in rumen (Shabat et al., 2016) and in distal gut from dietary polysaccharides in obese human beings (Le Chatelier et al., 2013; Turnbaugh et al., 2009). Further understanding of these findings could shed light on underlying ecological mechanisms that describe interaction of complex microbial communities and their interaction with host for sustainable production of food resources for humans.

11.3.2 Balancing the diet for a physiological stage

Metabolic products of nutrition which are absorbed form GIT include glucose, SCFAs, vitamins and minerals. Nutrient requirements can be classified into two groups: Maintenance requirements (minimum needs of animals to only continue living without any type of production) and production requirements (required above maintenance to support animals to produce meat, milk, young ones and work, etc). Total feed requirement are calculated by adding the maintenance and production needs (Izquierdo et al., 2021).

Shortage of quality feed may result in suboptimal supply of nutrient required for a physiological stage and hence optimum potential performances of animals may not be expressed (Patra, 2022). Nutrient requirements of animals must be calculated taking into account the physiological state of animals i.e., dry cows (from drying off to three weeks before parturition). Dry cows are supplied with high fiber and modest energy (little or almost no concentrate supplementation). Prepartum cows (three weeks before calf delivery) are supplied with energy and protein dense (because eating capacity of rumen drops down due to calf growth in late pregnancy) diet (usually through concentrate and/or fat supplementation). Prepartum cows should be provided with low potassium forages to prevent milk fever (parturient paresis) around the calving.

Cows in the physiological state of the three weeks after parturition are called as "fresh" cows. Such cows have reduced dry matter intake (DMI) due to reduced consumption capacity.

So, these cows are supplied with huge quantities of grains at lactation peak to fulfil their nutrient demands. However, cow's rumen must be adapted before larger quantities of cereal grain could be fed. As far as the physiological status of reproduction is concerned, the nutritional needs of animals with respect to reproduction are critical (*refer to 'nutritional management of reproductive health' for further details*) (Izquierdo et al., 2021).

Feeding of the cows (dairy and beef) should be done with the objective of meeting all its needs through provision of nutrients in a best possible manner. After a lot of research, nutrient requirements of various farm animals (during various physiological stages) have been elaborated in tables of the National Research Council (NRC). Based on these tables of nutrient requirements, the nutrient needs for maintenance, pregnancy, and lactation are calculated (Izquierdo et al., 2021).

11.3.3 Water provision

Clean water provision is crucial for the nutrition program of animals. Livestock producers need to maintain good water supply both qualitatively and quantitatively, because insufficient water intake reduce feed (roughages and concentrates) consumption and producer will lose profit due to production drop (Dept of Anim Sci. T. A. M. U, 2024).Water of sub substandard quality may give rise to health problems in livestock. The need for water consumption takes precedence over food consumption (Dept of Anim Sci. T. A. M. U, 2024; Wiseman, 2002), e.g., without food a healthy human adult can live for several weeks, but survival without water is mere a matter of few days (or in hot weather and exercise, this duration may be shortened to 24 hours only). Since, water reserves of the body are very small, and if dehydration occurs then water deprivation of bodily tissues including brain soon produces collapse and death (Wiseman, 2002).

Almost 25% of water in body present is in the blood and in extracellular spaces (fluids outside cells), while 75% water present inside the cells. Also, male (e.g., humans) have higher contents in body due to high musculature. While aging (reduced muscles) and increasing fat contents decrease water contents of body (Wiseman, 2002). Every day, the water becomes available to the body through ingestion of liquids, the water in feed material and the metabolic (water produced when the food is metabolized). Both thirst and kidneys controls the water intake (when the amount taken is near the lower end of the range) water excretion (when taken in excess of need) (Wiseman, 2002).

Carbohydrates are composed of C, H and O. Water (H_2O) is also the component of carbohydrates $(CH_2O)_n$ in its HOH form. Up to six carbon structure of carbohydrates i.e., hexoses ($C_6H_{12}O_6$), hydrogen and oxygen are in 2:1 ratio. Water acts as nutrient transporter from the soil to plants and from feeds into animal body. Use of polluted water near factories or townships or irrigation with polluted water contaminates herbages, and accumulation of harmful factors (chemicals, metals and pathogens) occurs inside plant tissues such as Pb, Hg, As, F, Cu, Se and Mo etc. Many plant species have been found to excessively accumulate harmful substance due to their affinity with those substance. Resultantly animals consuming rice plant which selectively absorbs Se suffer from degnala disease and plants ingestion with high F can give rise to skeletal problems in animals. (Saha et al., 2021)

11.3.4 Forage quality and quantity

Forage quality is a complicated topic. It crosses academic disciplines, so the agronomist, who may understand little about animals, and the animal nutritionist, who may not understand forage plants well, have different perspectives. Forage quality is perhaps the most important factor influencing ruminant productivity, whether grazing or feeding in a lot (Van Soest, 1994). Intensive production systems (IPS, animals in confinement) allow superior control of the feed quality and quantity as compared to extensive production system (EPS, grazing animals). Whereas IPS ensues balanced diets with continuous supplies resulting in stable production systems However, IPS involves selective animals with high genetic merit for production are chosen) (Gueddari & Vázquez, 2020).

Forage quality has close connection to that of DtF (bulk of plant material) which is needed in coarse form to be processed by the digestive tract as a source of energy (although lignified portion of fiber is indigestible, yet it is necessary to elicit adequate rumination) for rumen microbes and thus is important in promoting rumen function. Thus, forage quality is indicative of several contrasting factors: the supply of plant cell wall, its optimal digestibility, and the rate of digestion. The rate of digestion is important because it sets the amount of food energy available per unit of time. Forages of poor quality i.e., having too much total fiber contents may have fermentation rates in adequate for maintenance requirements of rumen bacteria, thus setting severe limits on rumen out-put for the animal to use.

Generally speaking, nutritive value of mature plants usually declines due to increased lignification and a decreased ratio of leaves to stems. However, not all leaves are more digestible than stems. Also, not all the leaves decline in nutritive value with maturity (maize, do not decline in nutritive value with age although the stover may decline). Parts of some plants may not change in quality with age; alfalfa leaves, for example, which have little structural function. In grasses the structural function of the leaves or the function of the stem as a storage organ can give stems a higher nutritive quality than leaves. This situation normally occurs in immature temperate grasses, although sugarcane is a tropical grass that falls into this category.

Generally speaking, cereals and seed crops at harvesting have reached ultimate maturity. Consequently, hulls, husks, bran, straw, and so on may be regarded as mature structures that have declined to their lowest quality. Nevertheless, there are important exceptions; soybean hulls and com bran are relatively devoid of lignin and are highly digestible. Individual forages vary in the degree to which they decline in value with age, the effect being in part related to maturity and in part to the environment in which the plant is growing (Van Soest, 1994).

Season change also affects the quality and quantity (including protein, mineral, and fiber content, of pasture species. However, nutrient requirements of stock on pasture depend on their growth and physiological stage and not on the season change, affecting animal performance. Thus, gazing animals during grazing season may suffer from imbalance of nutrients due to discordant balance of nutritional supply and demand between the plants and the animals (Nakajima & Yayota, 2019). During any season, tree foliage are very useful fodder resources for small ruminants which can supplement low-quality forages with crude proteins and micronutrients (Patra, 2010). Exploitation of tree leaves to reduce GHG emission is also suggested (Huang et al., 2021).

11.3.5 Health benefits of feed additives and dietary supplements

Feed additives are products used in animal nutrition to improve the quality of feed (for animals) and food (for humans from animal origin), and to improve the health and performance of animals (Hashemi & Davoodi, 2011). To improve health status of the animals, feed supplements are used frequently in livestock industry. Certain nutrients such as Arginine, Glutamine, Zinc, and Conjugated Linoleic Acid when given as dietary supplementation can provide benefits of improved fertility, neonatal survival, immune function, feed efficiency, growth, and better-quality meat by regulating gene expression and key metabolic pathways (Dept of Anim Sci. T. A. M. U, 2024). Feed additives e.g., in pigs have been evaluated for the purposes such as (1) immunity enhancers such as immunoglobulin; ω-3 fatty acids, yeast derived ß-glucans), (2) additives lessening pathogenic load in GIT such as high levels of prebiotics, bacteriophages, zinc oxide, essential oils, organic and inorganic acids, herbs and spices, some types of anti-microbial peptides, (3) additives to establish gut friendly microbes such as probiotics and certain prebiotics), and (4) additives such as nucleotide, glutamine, threonine, cysteine, lactic acid, gluconic acid and butyric acid which stimulate digestive function (De Lange et al., 2010).

Antibiotics use in the past were most effective feed supplements. However, EU put a ban on the use of antibiotics in 2004. As an alternate to antibiotics, farmers are now using organic acids, probiotics, prebiotics and symbiotics (combination of pro- and pre-biotics). The action of aforementioned molecules is on the principle of lowering pH, regulation of gut microbiota, gut stimulation and immune system development and function (Salobir et al., 2012).

There is active research going on in the field of natural feed supplements (containing bioactive molecules). Increasing awareness for the use of strategic nutritional supplements has far reaching consequences viz. increased DMI, growth, milk, meat, and wool (quality as well), reproductive efficiency, parasitic control , reduction in oxidative stress (OS; oxidation of biological molecules leading to damage and dysfunction of tissues and organs), enhancement of immunity and diseases resistance, microbial population and health status of the intestine (Frankic et al., 2008). In an EU project called 'Rumen up' that evaluate 500 plants and their extracts invitro for their effect on fermentation in one of the comprehensive studies of its kind, identified at least 25 with potential value as feed additives (https://www.abdn.ac.uk).

Recent years have witnessed a great trend toward use of feed additives of natural origin versus synthetic additives (potentially harmful). For this reason, keen interest has been shown for innovative use of aromatic plants and herbs, since they are considered as an untapped reservoir of valuable substances and their research is an ongoing discipline (Christaki et al., 2020) (Fig. 14). The recent applications of aromatic plants as source of growth promoters (through increased secretions of endogenous digestive enzymes, saliva, bile and mucus), antimicrobials (as they can reduce potential pathogenic bacteria), immunostimulators, antioxidants (may improve gut morphology), flavorings, pigments, and as preservatives, in animal nutrition, especially those that can satisfy the increased consumers' demands for functional foods and natural products in relation with human health (Christaki et al., 2020).

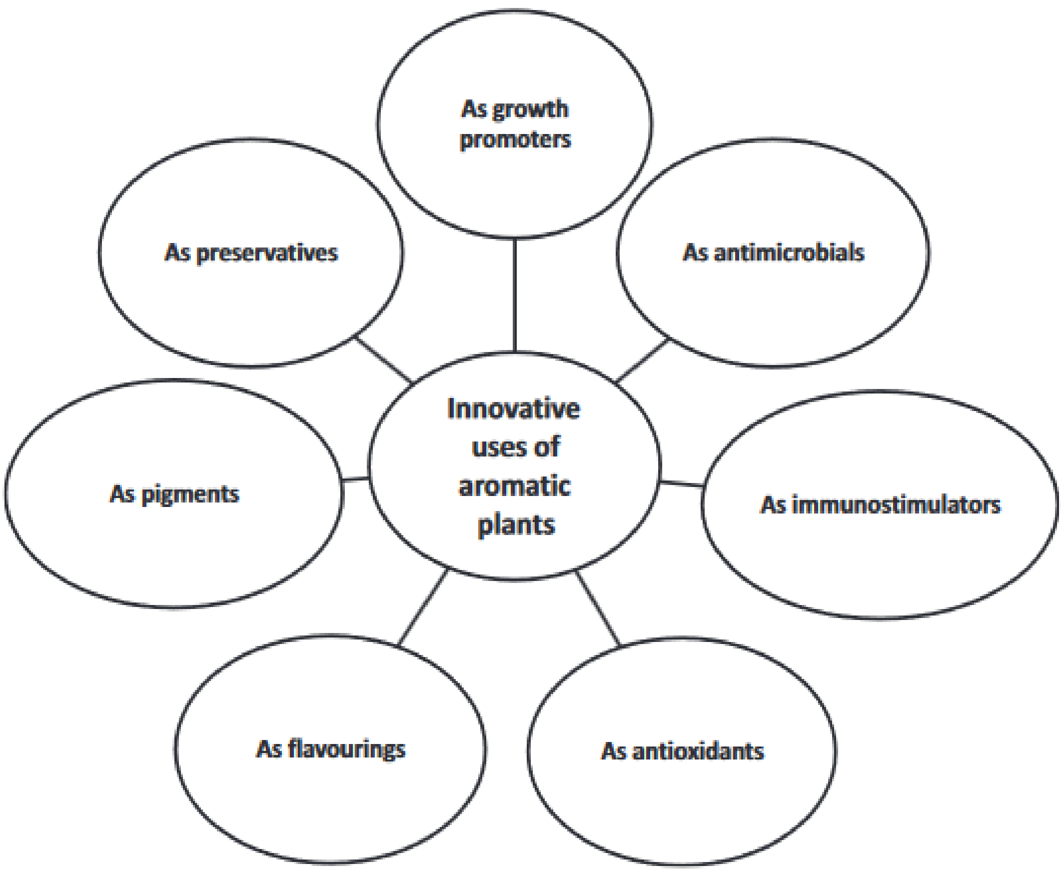


Figure 22. Innovative uses of aromatic plants as natural supplements in nutrition. Source: adapted from (Christaki et al., 2020)

11.3.6 Nutrition and Stress management

Stress is an aberrant response of an animal to an external environmental stimulus. Stress occurs when homeostasis is threatened or perceived to be so. Homeostasis is re-established by various physiological and behavioral adaptive responses (Chrousos, 2009). Therefore, stress describes the condition when natural regulatory capacity of an animal in an unpredictable and uncontrollable situation can't cope with environmental demand (Koolhaas et al., 2011).

Various stressors such as overcrowding, overproduction, heat stress, cold stress and transportation are not only welfare issue but also induce protective reactions in the animal body, showing obvious physiological metabolic abnormalities (stress induces a series of metabolic disorders such as reduced feed intake, disruption of physiological processes of nutrient metabolism, digestion, and metabolic disorders, which in turn lead to nutrient deficiencies) and non-specific immune dysfunction, reduced gut health, and immune defense (increase vulnerability to disease), and ultimately leading to a decrease in production performance (along with deteriorated product quality) and feed conversion rate (Patra, 2022). Good nutrition during periods of stress provides ways to relief stress symptoms. Use of medicinal plants, gut microbiota-modulating agents, and antioxidant minerals and vitamins can result in good productivity of livestock along with improved antioxidant status, gut health, and immunity (Patra, 2022).

Prolonged stress of any physical or physiological origin has devastating impact on digestion and nutritional needs. As stress generates greater physiological demands such as for energy, oxygen, circulation, and therefore requirement is enhanced for metabolic cofactors (e.g., vitamins and minerals). Role of specific nutrient in stress such as dietary AA-tryptophan results in synthesis of Serotonin (the mood-boosting hormone that promotes feelings of calmness), whereas depletion of the serotonin precursor i.e., tryptophan has been found to increased depressive mood (Yuly Bersudsky et al., 2010). A short description of various nutrients and their role in coping with stress is mentioned below.

Nutrients like tryptophan (to synthesize mood-boosting serotonin),(*refer to 'improving immune function through nutrition' for details)* found in Banana and Walnut (Singh, 2016), whereas whole grains (allow Tryptophan to get into brain; whole, unprocessed grains complex carbohydrates, such as oat grains, quinoa and brown rice provide a steady release of energy and stimulate the production of serotonin) (Banham, 2022); Phenylalanine and tyrosine (Singh, 2016); L-Theanine (Haskell et al., 2008; Kimura et al., 2007); ω-3 fatty acids (anti-inflammatory and antioxidant properties reduce cortisol levels) (Alagawany et al., 2022; Thakur et al., 2012) found in eggs, flax seeds, spinach, walnut. flaxseed, chia seeds, fatty fish, such as salmon and tuna; vitamin C (supports the adrenal glands, lower cortisol levels and boost the immune system) (Acute, 2024; Brody et al., 2002) found in oranges, blue berries, Banana, tomatoes, peppers, leafy greens, broccoli and spinach; vitamin B (Nelson, 2009) found in eggs (B2, B5, B12), brocoli (B6), walnut and banana (B6); Se and Mg found in spinach, chocolate, nuts, seeds, beans, avocados, bananas, and dark leafy greens (Banham, 2022; Singh, 2016); and Probiotics (*Lactobacillus acidophilus* and *Lactobaillus rhamnosus*) have been proven beneficial to tackle stress.

Nutritional deficiencies or imbalances in farm animals can cause OS and metabolic disorders. Whereas, high production performance puts these genetically superior animals on high nutrient demand, which may enhance the development of free radicals (leading to oxidative stress), if not fed according to production level (Ponnampalam et al., 2022). Moreover, OS is result of discordant balance between oxidants and antioxidants, which in turn is indicated by a continuous increase in ROS production (Ponnampalam et al., 2022). Similarly, many other diseases such as mastitis, steatosis and coccidiosis as well as thermal stress also have connection to OS.

Nutrition provides health benefits under oxidative stress (Lykkesfeldt & Svendsen, 2007); such that supplementation with antioxidants (antioxidant vitamins, whole plants or parts or their extracts) balance the oxidant-antioxidant environment. Some interesting plant extracts were also used in swine and chickens, examples include black currant, sweet chest-nut tannins, mixture of spices and calendula extracts etc. (Frankič et al., 2010; Frankič & Salobir, 2011; Frankič et al., 2009). For humans, various other natural antioxidants such as Vitamin E, coenzyme Q10, melatonin, polyphenols, curcumin, and selenium have shown promising results in treating Alzheimer Disease; whereas, saffron, vitamin E, curcumin, Zn and Se found effective in depression treatment (Juszczyk et al., 2021). Fig. 15 presents an overview of how the dietary supplements help to ameliorate oxidative damage caused by stress conditions mainly through activation of antioxidation system, protection of lipid membrane oxidation and upregulation of cell protective mechanism through heat shock proteins.

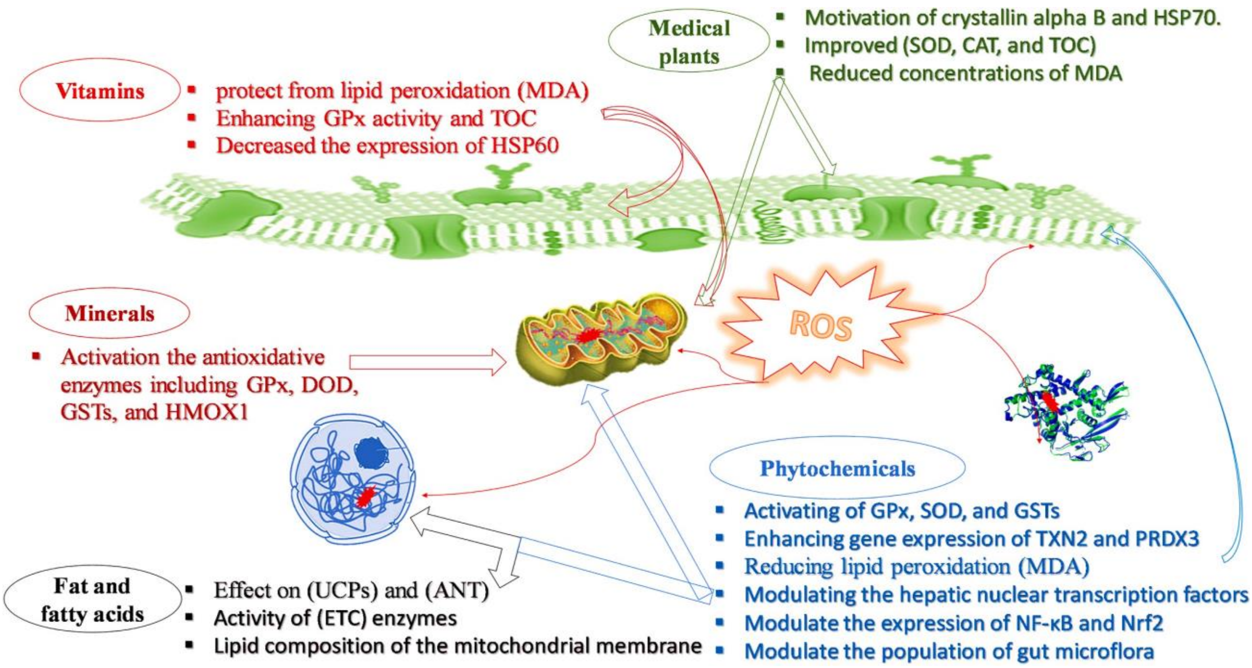


Figure 23 Use of dietary supplements and their mechanism for mitigation of oxidative damage and motivation of immunity. Source: (Abdel-Moneim et al., 2021)

11.3.7 Monitoring body condition score for livestock health

Body condition score (BCS) is of significant value to optimize health and productivity of flock and/or flock (Fisher, 2020). BCS is just a graded scoring system of an animal's ability to accumulate fat reserves (Fig. 16) on its body and are developed as a homeostatic mechanism of animal. Fat reserves thus developed used to overcome nutritional changes when energy demand is greater than availability from feed e.g., in winter, during pregnancy, and lactation, or other times of the year with infrequent forage supply. For dairy animals a body condition score of 1-5 is used (Fig. 16). The BCS of one means severely under conditioned, score of two means under conditioned, score of three means good condition, score of four means over-conditioned and score of five means severely over-conditioned (Pasture.io, 2020). For animals with poor genetics, with poor nutrition or health, it is hard to put on weight and accumulate fat reserves (Pasture.io, 2020).

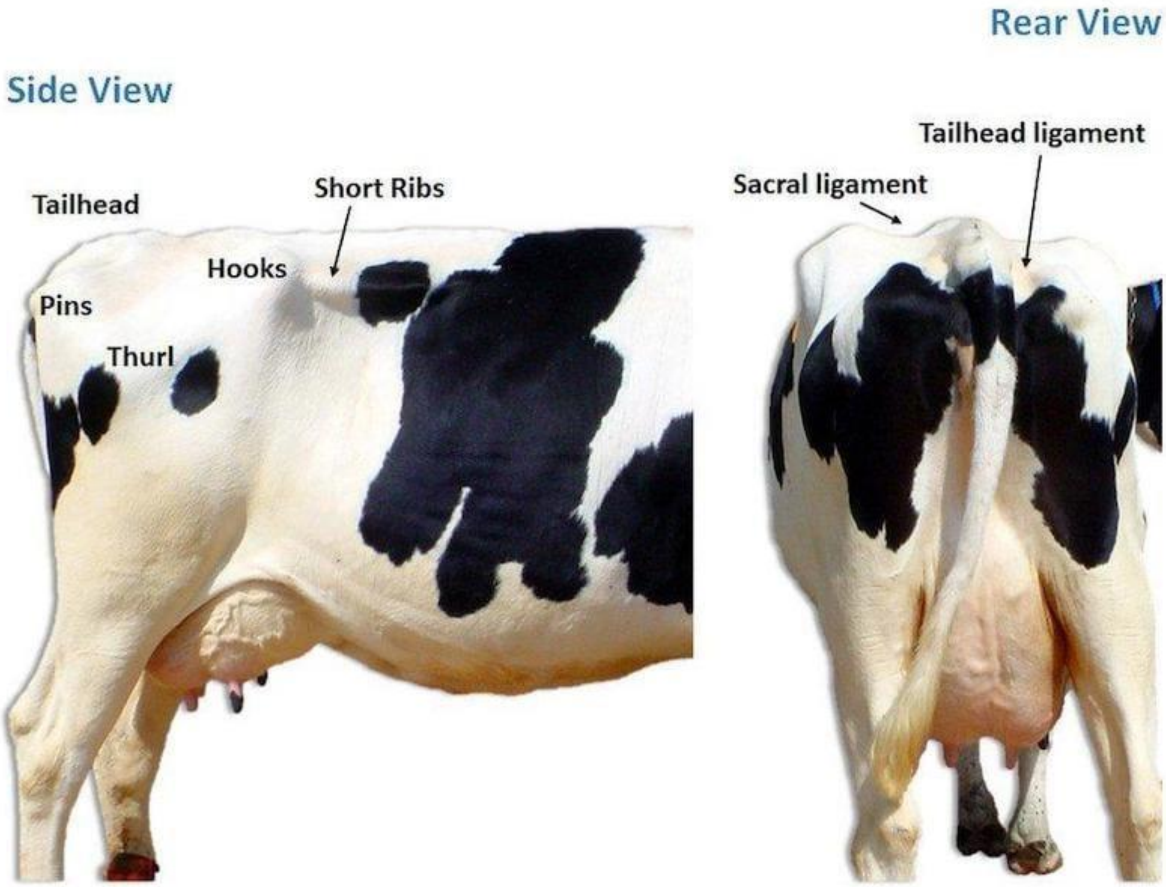


Figure 24 In dairy cows, for BCS measurement, fat reserves in the backbone, near hip and tail are accessed. Source: Penn State University Extension & (Pasture.io, 2020)

BCS is not only an accepted measure of energy reserves (reflecting a better access to nutrients) of an animal, a good score helps individuals in better survival, growth, lactation, reproduction, and health (Fisher, 2020). As stated earlier, a body condition score of 1-5 is used for dairy animals (Fig.17). The BCS of one means severely under conditioned, score of two means under conditioned, score of three means good condition, score of four means over-conditioned and score of five means severely over-conditioned. BCS is an easy management tool and could be used both in management and in breeding (reproductive) programs as an indirect measure for infertility. Since high yielding cows (of high genetic merit) having lower BCS than average cows at early lactation; also lose more BCS in early lactation (than BCS change from 1st to 10th week) is unfavorably related to reproductive performance (Pryce et al., 2001). Hence cows with high genetic merit for milk production that lose more BCS in early lactation, consequently, are more susceptible to health disorders after calving. Moreover, if animal has excessively low BCS at calving; the greater BCS loss after calving can increase the risk of metabolic disorders like milk fever, ketosis and fatty liver disease. However, experiments during the transition period point out benefits from restricting energy intake pre-calving (i.e., high-dietary roughage) was associated with reduced incidence of ketosis, fatty liver and milk fever. Collectively, it can be stated that at calving body condition score of 3.0 for adult cows and 3.25 for heifers on diets with modest energy restriction pre-calving will minimize the risk of BCS-related health disorders around calving (Roche et al., 2013).

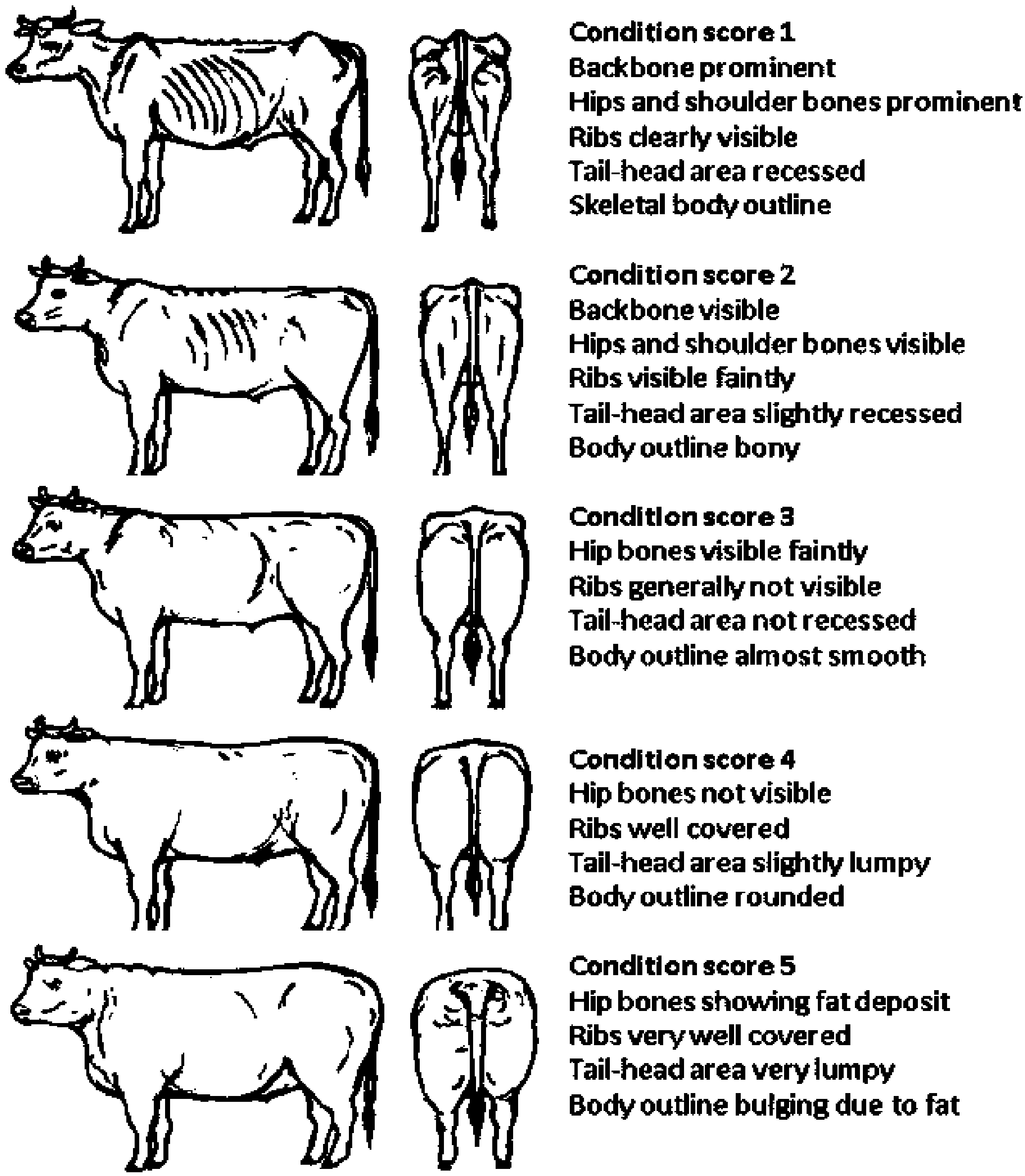


Figure 25 Cattle body condition scoring system (Anon,1994). Body condition scores of 1-5

Since, BCS has a significant association with dry matter intake (DMI) in high-yielding dairy cows; animals with obese condition (BCS > 4) have less DMI (about 1.68% of live weight). However, cows with normal condition (3 < BCS < 4) and poor condition (BCS < 3) had DMI equal to 1.84% of their live weight (Atalay, 2019). This is the reason that accessing fat reserves can serve as easy, fast and trustworthy marker to indicate animal health condition. Condition of animal also provide clues about the animal's nutrient intake, productivity and fertility; thus, prediction of general and reproductive health can made through use of BCS.

Although body condition scoring is a simple technique yet it can contribute significantly to good husbandry (possibility of formulation of diets according to the needs of the cow and calf), welfare (reduce calving difficulties and calf losses) and management (good performance) of beef cows. For beef cows, a nine-point BCS (scale 1-9 with one extremely thin and weak while nine extremely obese) can be used to manage beef herd (Selk, 2004) (Fig. 18). Beef cows through their homeostatic mechanism store excess (from requirement) dietary energy in the form fat to use at some future point in the cow's life at need. Fat reserved not only provide energy during seasonal changes in feeds availability but also insulate the cow against extreme cold conditions by not allowing heat loss. For beef cows, a body condition score of 1-9 can be used to manage the cow herd. The BCS of cattle can be evaluated at any time, but it is recommended at weaning, sixty to ninety days before calving and around calving (Farney et al., 2016).

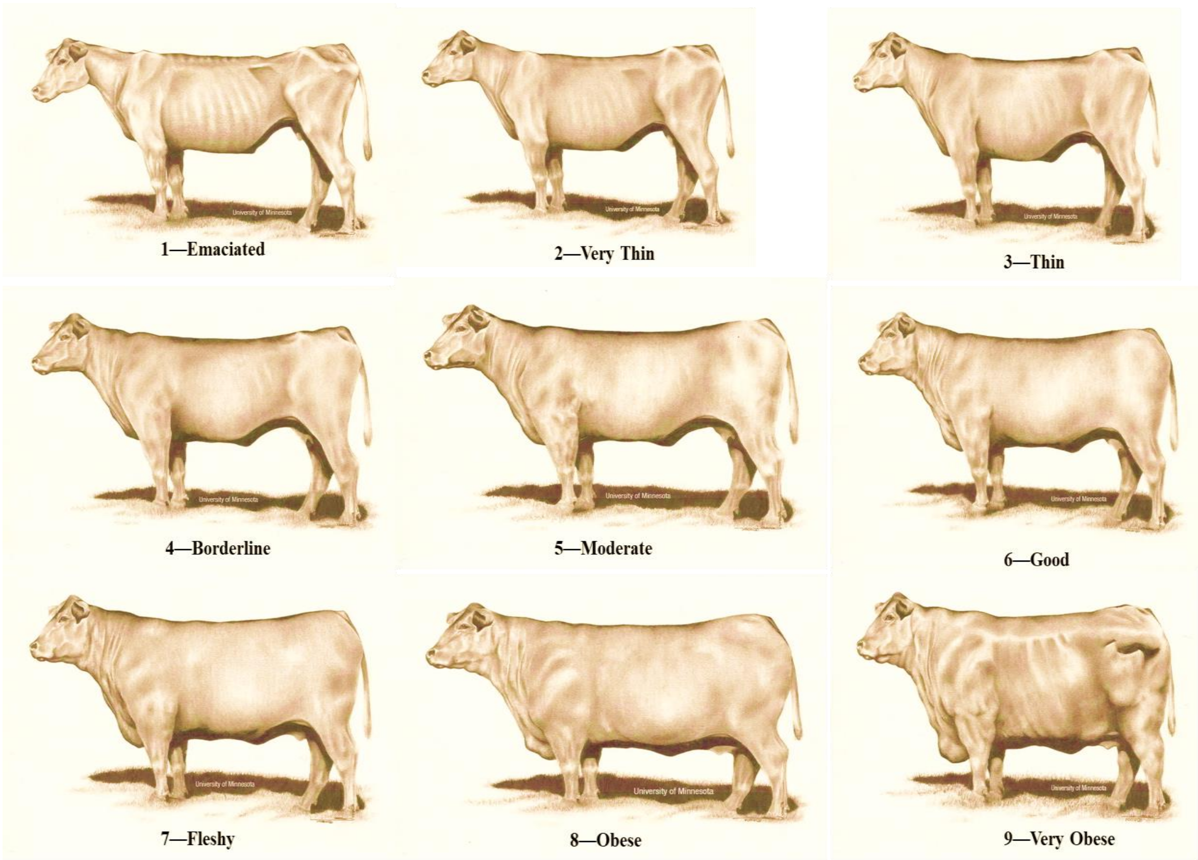


Figure 26 Nine-point scale of body condition scoring system for beef cows, according to the

Nutrient Requirements of Beef Cattle (NRC, 2016) are as follows Source: (Felix et al., 2024).

Indication of condition loss due to low score can also be used to depict an underlying disease or parasitic infestation or as a result of neglect in care of animals (sometimes due to peoples' difficult situation such as financial or personal situations such as in drought conditions, diseased state and challenging relationship).

11.3.8 Managing health problems in grazing animals

Animal welfare highly influence the productivity and health of animal and is considered as integral aspect for a sustainable livestock production systems (Rivero & Lee, 2022). Grazing in pasture or open areas is reported to influence positively on the behavior and comfort of the animal (Herlin et al., 2021; Nakajima & Yayota, 2019). However, grazing involves various factors such as vegetation, weather, temperature, humidity and carrying capacity (Nakajima & Yayota, 2019). Although grazing contributes to animals' welfare, yet there are potential drawbacks with respect to animal health and welfare which include negative energy balance (NEB) due to poor forage condition, much higher risk of internal parasites, deficiency of micronutrients, delayed activity of estrus after parturition, and in some situations, welfare concern due to exposure with unpredictable and extreme weather conditions (Rivero & Lee, 2022).

The question arises which form of grazing whether traditional continuous grazing (TCG) or intensively managed rotational grazing system (IMRG) is better for animal health? In an effort to determine the effect of grazing management systems on bulk tank milk quality (BTM) and udder health; Goldberg et al., 1992 compared IMRG, TCG and confinement housing. They concluded that adoption of IMRG can reduce exposure to environmental pathogens and may be a practical and profitable alternative to enhance milk quality and mastitis control (Goldberg et al., 1992). Comparably, a review found that a regenerative grazing systems with nitrogen cycling, biodiversity, carbon storage is more closed to wild grazing ecosystems which is considered as more environment friendly and sustainable as compared to conventional grazing system. (Kleppel & Frank, 2022).

Grazing has positive effect on immunity in livestock; however, the underlying mechanism through which grazing affect immune system in cattle is largely unclear (Nakajima & Yayota, 2019). One of the mechanisms through which grazing may improve immunity may be forage selection of native plant species which contain plant secondary metabolites (PSM). These PSM can beneficially modulate animal health (Provenza & Villalba, 2010) by protecting animals from health deteriorating microorganism (Lozano, 1998). Thus, grazing livestock may change their forage selection (given that provision of diverse vegetation is there) based on their physiology and to ensure immune status. Likewise, parasite infested sheep like medicinal plant species; whereas plant such as *Lotus corniculatus* are reported to boost immunity (Egea et al., 2014; Min et al., 2002). A diverse diet (ryegrass, chicory, alfalfa, plantain, and red clover) in comparison to ryegrass only can improve the total antioxidant capacity of ewes at lambing, ewes gave birth to heavier lambs as well as reduction in some markers of oxidative and metabolic stress (Garrett et al., 2022). Moreover, diverse vegetation has also been shown to be beneficial for improving mineral intake and balance of the grazing animals (Ogura et al., 2017; Ohlson & Staaland, 2001). However, under grazing conditions, cattle with inappropriate nutritional conditions can suffer from immunological disturbances (Butler, 2014; Hammon et al., 2006; Martinez et al., 2012). Also grazing occasionally can induce negative energy balance (NEB) in livestock. Negative energy balance coupled with protein deficiency in the feed or lower protein digestibility results in the decline in number of circulating lymphocyte (Lewicki et al., 2014). Furthermore, an increase in β-hydroxybutyrate (BHB and plasma non-esterified fatty acids (NEFA) under NEB conditions can result in neutrophil dysfunction and uterine disorders in cows (Hammon et al., 2006). Such disorders can suppress the body immune system and therefore supplementation of macro- (energy and protein) and micro-nutrients is necessary in the grazing system with inadequate supply of nutrients.

It is reported that growth and lactation performance was reduced in the grazing ruminants which were not supplemented with protein and minerals. (Bergen, 2021; Cao et al., 2021; Gilbreath et al., 2021).

Animals can now receive better nutrition. Thanks to the introduction of feed additives with improved nutrient availability, which also promote better health. One of the important feed additives is the use of organic sources of microminerals, which not only give animals more nutrients due to their higher bioavaiability but are also less toxic. Higher digestibility of such sources means that small amount of the ingredient can fullfile the requirement and therefore pose less burden on overall ecosystem (Brennan et al., 2011).

One of the important and often neglected issue of livestock which negatively impacts on health, production and reproduction performance is the presence parasite in animals. Internal parasite infections are often reported as a major constraint to the productivity of small ruminants affecting their health and reproduction (Khan et al., 2023). An effective control of parasitism to mitigate overall detrimental impact of parasites on host, and further improving the host resistance to parasitic infestation, can be achieved primarily through nutritional management (Sahoo et al., 2011). Most prevalent parasitic infestation are caused by nematodes that include *Strongyloids, Hemonchus, Ascaris, Oesphagostomum, Ostertagia, Cooperia, Enterobius, Chabertia, Gunagylonema, Trichuris, Trichostrongyliods, Dictyocaulus and Trichnellia* which are distributed globally and can affect all types of livestock. The gastrointestinal nematodes are detrimental to the animals' health. Nematodes in the gastrointestinal tract primarily hinder digestibility and utilization of feed, and decrease feed efficiency. A severe type of infestation can lead to various ailments and death of animal. This segment presents an overview to summarize the current knowledge on the role of nutrition in host resistance to parasites, growth, and reproductive efficiency of small ruminants, and to develop future strategies for better animal health and disease resistance.

It is important to understand the host-parasite interactions. The host-parasite interactions are complex and are influenced by a various factors including the host (breed, age, nutritional and health status of the animal), the parasite (species, strain and stage of development of the parasite), and the environment (climate, pasture management and the presence of other animals) (Sahoo et al., 2011). The host-parasite interactions can be divided into two main categories which can be termed as direct effect and the indirect effect. The direct effects of the parasite on the host animal means a physical damage caused by the parasite, the loss of blood and nutrients and reduced immunity of the host animal. The indirect effects of the parasite on the host means the changes in the host's behavior (behavioral changes in feeding, reproduction and social activity in herd), the changes in the host's metabolism (energy, protein and mineral metabolism), and the changes in the host's reproductive performance (fertility, fecundity and hormonal balance of the host) (Sahoo et al., 2011).

In terms of prevalence and burden, it is reported that pastures contain about 95% of the parasitic burdens while host animals contain about 5% of the parasitic burden (Bowman, 2014). Therefore, managing and optimizing pasture and grazing areas is of utmost importance to minimize parasitic infestation in the animals. Key strategies in pasture management include controlling stocking density, pasture rotation (moving animals to next pasture after 4 days in warmer season and 7 days in cooler season), resting pasture (keeping animals away from pasture for 2-3 months), using mixed species of livestock along with the use of synthetic pesticides in the pasture (AHDC, 2024; Bath, 2014; Kelemework et al., 2016; Taylor, 2012). Another study suggests that a rotation period of 3-6 months for a pasture can effectively reduce the overall magnitude of parasitic infestation (Niranjan Kumar et al., 2013). However, some nematodes are resilient enough that their eggs can survive for one year under a non-extreme environmental conditions (Bowman, 2014). Lessening the duration of grazing period decreases the chance of any high accumulation of infectious parasitic larvae and thus ensures an efficient pasture utilization (Bath, 2014). In addition to the prolonged grazing period, keeping the number of animals higher than the stocking density of a pasture or paddock area and unhygienic conditions also results in buildup of infectious larvae (Bath, 2014).

Supplementation of metabolizable protein and micro minerals specially in a time of protein scarcity results in low parasite-egg count in cattle and sheep and the animals demonstrated higher resistance to parasitic infestation (Greer, 2008; Hughes & Kelly, 2006; Ibrahim et al., 2008). High protein diets promote immune response by the animals against parasitic infection, providing a satisfactory performance of susceptible breeds (Arsenos et al., 2007). An immunogliobulin response in the sheep was analysis when diets with difference potein levels was fed and it was reported that low protein diets resulted in low immunoglobin production and such animals were more susceptible to parasitic infestation (Niranjan Kumar et al., 2013). Protein sources like casein, ovalbumin, sunflower meal, cotton seed meal, soybean meal, dry distiller grains with solubles as well as urea are important protein ingredients which contribute to promote immunity in the livestock (Bertoni et al., 2016).

Another approach which is gaining popularity around the globe is the use of bioactive plants and phytogens that possess or produce certain compounds and metabolites like tannins (contained e.g., in *Calluna vulgaris*) which are reported to possess anthelmintic effect for different species of ruminants (Beauchemin et al., 2020; Hoste et al., 2006). In organic farming systems, the use of plant-based anthelmintics is gaining importance. The use of tannin-rich feed ingredients (containing 4-8% condensed tannins) to control gastrointestinal nematodes in small ruminants is one of the most promising alternatives to chemical anthelmintics (Hoste et al., 2006, 2008). Some forage species have shown higher anthelmintic effect in the ruminant animals, like sainfoin (*Onobrychis viciifolia*), trefoil (*Lotus pedunculatus*), sainfoin (*Onobrychis viciifolia*) sulla (*Hedysarum coronarium*) and birdsfoot trefoil (*Lotus corniculatus*). The anthelmintic effect of these plants is attributed to their condensed tannin content. Condensed tannins are polyphenolic compounds that bind to proteins and other macromolecules, forming complexes that are not degraded in the rumen. This reduces the availability of nutrients to the parasites and can lead to their death. In addition to their anthelmintic properties, condensed tannins in the feed are also reported to mitigate the methane emission from ruminant livestock and contribute reducing environmental pollution (Patra, 2022). When using tannin-containing ingredients, it must be kept in consideration that tannin can act as antinutritional factor when used levels higher than recommendation.

There is a link between nutrition, gastro-intestinal parasites and reproductive status of animals. Nutritional insufficiency can give rise to gastrointestinal parasites (Sahoo et al., 2011). In free-ranging population of Mediterranean sheep, reproductive females had 2.6 times high gastrointestinal strongyles (GIS, a parasitic spp.) fecal egg values than non-reproductive females. It is recommended that young animals, reproductive females and animals with weak body condition should be given special attention in parasite management for herd health because such animals highly susceptible to parasitic infestation and source of contamination for rest of the herd (Bourgoin et al., 2021).

Likewise the inclusion of nutraceutical plants (the plant which contain chemical compounds that may prevent development of disease and promote better health) in the ruminant feed and their health benefits for livestock has been well documented (Villalba et al., 2017). Some plant substances with nutraceuticals properties are onions, garlic (containing nutraceutical substance as 'Allyl sulfur compounds' and Adenosine), soybeans, other legumes and apios (Isoflavones), onion, red grapes, citrus fruits, broccoli (Quercetin), pepper fruit (Capsaicinoids), Tomatoes (Lycopene), Cruciferous vegetables (Isothiocyanates), Oat bran (α-Glucan), Grapes skin (Resveratrol), Citrus fruits, carrots, pumpkin (α-Carotene), rosemary (Carnosol), teas and berries (Catechins), cabbage, broccoli, cauliflower, kale, brussels sprouts (Indoles), turmeric (curcumin), grapes, strawberries and raspberries (Ellagic acid), celery (3-n-butyl phthalide) and most plant (components of cells wall, cellulose) (Wildman et al., 2016).

The idea of the development of healthscape maps in a grazing management system for ruminants can help the grazers to identify nutraceutical plants with respect to spatial distribution. This is a new method which combines spatial and nutritional analyses of plants to create healthscape maps. Such a holistic approach to manage the pastures and livestock production systems is very effective as the plants are analyzed as a source of medicine in addition to the source of nutrients (Pereira & Gregorini, 2022). Such information is helpful to manage and distribute the enclosures of a pasture according the need of animals. For instance, it is suggested that to attenuate the parasitic burden in the animals during peripartum period, which is considered to be a sensitive period of internal parasite attacks, the animals shall be moved to the enclosures containing herbage rich in anthelmintic properties. To combat stress during weaning transition period in animals, they could graze the area with higher antioxidant properties. Likewise, animals with bacterial infections could be separated from the flock and allowed to graze in the areas containing plants with antibacterial properties (Pereira &

Gregorini, 2022). A recently introduced concept of designer livestock products can also benefit from such healthscape maping. For instance, to increase the certain types of fatty acids (e.g long chain polyunsaturated fatty acids, omega-3) or phytogenic compound in the meat of animal finishing animals may be allowed to graze the area containing plant which are rich in those particular compounds. So, healthscape maping has multidimentional benefits encompassing quality of the product, production efficiency, better health, lesser expenses on drugs/treatment, lesser impact on environment, overall welfare of animal and provision of organic food products for human consumption (Pereira & Gregorini, 2022).

Regular evaluating grazing animals through BCS (*refer to body condition score for livestock health for details*) is utmost necessary for maintaining animals in good condition. However, under grazing conditions, monitoring the animals manually can be challenging. In this regard, animal welfare surveillance can be increased under large pastures through use of modern equipment such as cameras, drones, digital sensors/precision livestock farming (for direct or indirect detection of abnormal behavior or physiological change i.e., disease/fever, thermal stress or imminent calving), GPS (positioning equipment mounted on head collar) for detecting movement of animals over large area, can track their location and somewhat the indicators of welfare and health. Whereas as electronic positioning such as radio frequency identification can record animals at fixed points. These tools can give information that helps in evaluating and managing the free-ranging animals efficiently (Herlin et al., 2021).

11.3.9 Mycotoxin and intoxicants in animal nutrition and health

Presence of mycotoxins in feed is very detrimental for the health of livestock and overall farm production system (Pitt et al., 2012). Appearance of oral lesions (distract them from eating, and deteriorate growth performance) are painful in chickens and have often relevance to mycotoxins. Oral lesions may have other causes i.e., very fine feed particle size, deficiency of Vitamins A, E, B6 and Biotin, excessive levels of copper sulphate and some parasite infections. However, to confirm the case of illness, mycotoxin analysis must be done. The most common ill-health effects of mycotoxins include disorders of neurological and respiratory systems, nephrotoxicity, hepatoxicity and cancer. Growth of fungi and yeasts on plant materials give rise to toxic molecules, which are known as mycotoxins (Becker-Algeri et al., 2016). Mycotoxins are heat resistance and highly toxic and when ingested by animals though contaminated feeds are not only detrimental to animal health but are also harmful for humans as these are passed on to animal products (Baydan et al., 2017). Thus, mycotoxins in feed materials are important with respect to public health concerns and one-health concept. Among hundreds types of mycotoxins, the most prevalent and concerning are aflatoxins, ochratoxins, fumonisins, deoxynivalenol, putalin and zearalenone (Wu et al., 2014).

Intoxication problems in extensive system can be common which can be caused by ingestion of certain toxic weeds (when pasture grasses are limited and animals have no choice but to feed on weeds) or with certain forage grasses like fescues which are more susceptible to toxic fungus like Claviceps spp. Consumption of such forage grasses can cause depression in intake of dry matter and milk production, suppression of immune system and increase in respiration rate (Bertoni et al., 2016). Toxic chemicals which are produced due to consuming feeds containing ergot alkaloids which in turn are produced by fugus *Claviceps purpurea* or grasses, especially fescue and rye grass, contaminated by fungi (like *Neotyphodium* spp. and *Epichloë* spp) developed as a result of endophytic symbiotic relationship, were found responsible for various ailments in farm animals. Different endophytes produce ergot alkaloids which are responsible for low milk production, growth rate and reproductive performance in cattle and fever (due to peripheral vasoconstriction), neurological disorders and reproductive issues in horses. Sorghum-based feeds may also cause outbreaks of ergot poisoning in pigs and cattle. Consumption of rye or barley grains contaminated with ergots cause lameness (due to peripheral vasoconstriction) and necrosis of feet, tail and ear in cattle (Pitt et al., 2012).

To prevent and control mycotoxicosis in livestock, certain strategies have been proposed. One of the strategies is the use of probiotic-like bacteria. It is demonstrated in vitro that certain strains of normal gut microbiota when supplemented with probiotic such as *Bifidobacteria* bind 25-60% of the ingested mycotoxins and render those unavailable for the absorption at intestinal level (Oatley et al., 2000). The dietary control of mycotoxins also includes inclusion of bentonite clay minerals which aid in binding and elimination of aflatoxin from the GIT (José Mendes et al., 2024)

11.4 ADDRESSING SPECIFIC HEALTH CHALLENGES

11.4.1 Improving immune function through nutrition

An enormous amount of literature is available revealing the effects of nutrition on host-resistance through boosting immunity. Malnutrition and nutrient deficiency suppress immune functions. This immune suppression may give rise to virulence of pathogens in the animal thus, compromises the health and welfare of herd through negatively influencing the dry matter intake, digestion, absorption, cellular metabolism and hormonal regulation (Bertoni et al., 2016).

Role of nutrition in immune response may be directly through nutrients or indirectly by secondary metabolites of protein metabolism or through metabolites of gut microflora (Kamareddine, 2017). Physiological imbalance of the nutrients can occur in lactating cows which are struggling to adapt to enhanced nutrient needs for lactation demands. Improper metabolic status and immune interaction may increase disease risk during early lactation. (Ingvartsen & Moyes, 2013). Diet affects the immunity in different ways, such that dietary amino acids modulate immunity by activating lymphocytes and macrophages, and through excretion of hormones (Stephen and Avenell 2006; Osorio et al. 2014). Dietary supplementation of branched chain amino acids like leucine and valine results in improved immune response by increasing blood cytokines and decreaing interleukin-4 (Bassit et al. 2002).

Microbiota-derived products of dietary molecules (e.g., tryptophan) has the potential for co-metabolism between the microbiota and the host. Free tryptophan in GIT has metabolic fate in three ways; Enterochromaffin cell metabolism via the serotonin production pathway and gut increase gut motility, Immune and epithelial cell metabolism via the kynurenine pathway, and microbial metabolism, which generates amines and bioactive molecules (Diether & Willing, 2022).

Microbial fermentation of tryptophan via tryptophanase enzymes can produce indole-containing compounds (are now well characterized as AhR ligands) (Fig. 19). Indole is an important signaling molecule in host physiology and also to other microbes in community. These AhR ligands are important component in immune competency in the GIT. Whereas microbe may also help (although they are not direct enzymatic contributor) in conversion of tryptophan to kynurine in epithelial cells through stimulation to produce enzyme (indoleamine 2,3-dioxygenase 1) responsible for this conversion (Diether & Willing, 2022).

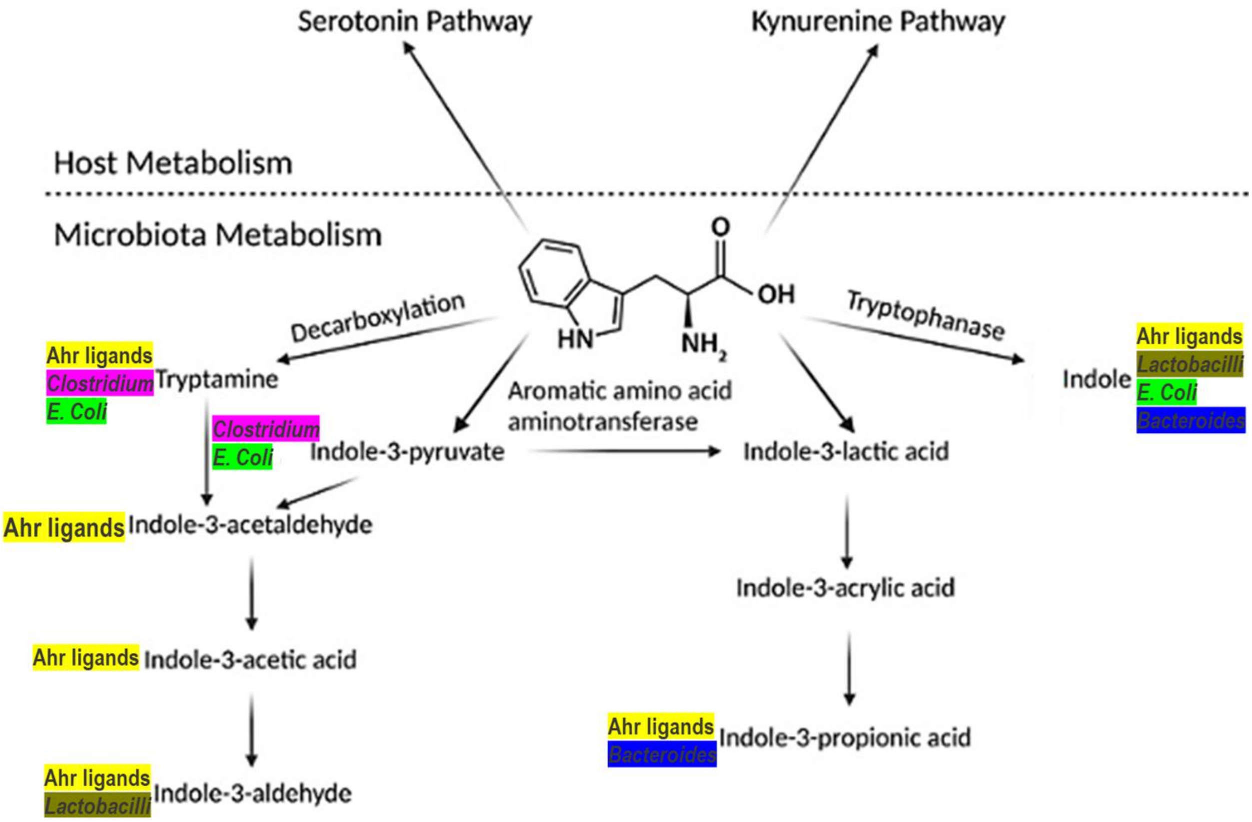


Figure 27. Host and microbial metabolites of tryptophan. Whereas, host metabolism give rise to serotonin and kynurenic pathways. Microbial metabolism can give rise to various products with important effects on host health. AhR ligands are denoted with yellow color. Microbial species known to produce each metabolite are denoted using other colors. Source: adapted and modified from (Diether & Willing, 2022).

Fatty acids nutrition for improved immunity is a field that needs to be researched. Polyunsaturated fatty acids (PUFA) such as omega-3 and omega-6 needs to supplemented for proper immune function. Supplementing omega-3 PUFA to sows increased concentration of immunoglobulin G (IgG) in the colostrum and thus supported the immune system of suckling piglets (Leonard et al., 2010).

Not only immune system plays an important role in fighting infections and neoplasms, it also benefits systemic metabolic homeostasis. Several metabolic processes are reported to be engaged in the maintenance of immune homeostasis (Kamareddine, 2017). This cross-talk between metabolic and immune system is essential in promoting 'metabolic health' and helps organism to adapt to changing environmental conditions and available nutrition throughout its life (Zmora et al., 2017) and is regulated by host genetics, diet, gut microbiota and epigenetic (Zmora et al., 2017).

As stated earlier, nutrition impacts immune system directly or indirectly. Figure 20 below indicates that diet components typically impact directly the immunity through interaction with immune cells via receptor-mediated signaling, or through indirect interaction via gut microbiota by modulating the metabolites, which in turn regulate the metabolic homeostasis of the host (Zmora et al., 2017). Currently, various phytogenic-plant extracts possessing immunomodulatory functions are being used in the diet of animals. However, investigations on alternate and more effective immunomodulators as feed additive are underway. A promising weed called laminarin has shown to be an efficient source of β-(1-3) and (1-6) glucans. Feeding them to monogastric animals improves the phagocyte activity of suckling animals just before weaning and improves microflora and absorbability of intestines (Leonard et al., 2010).

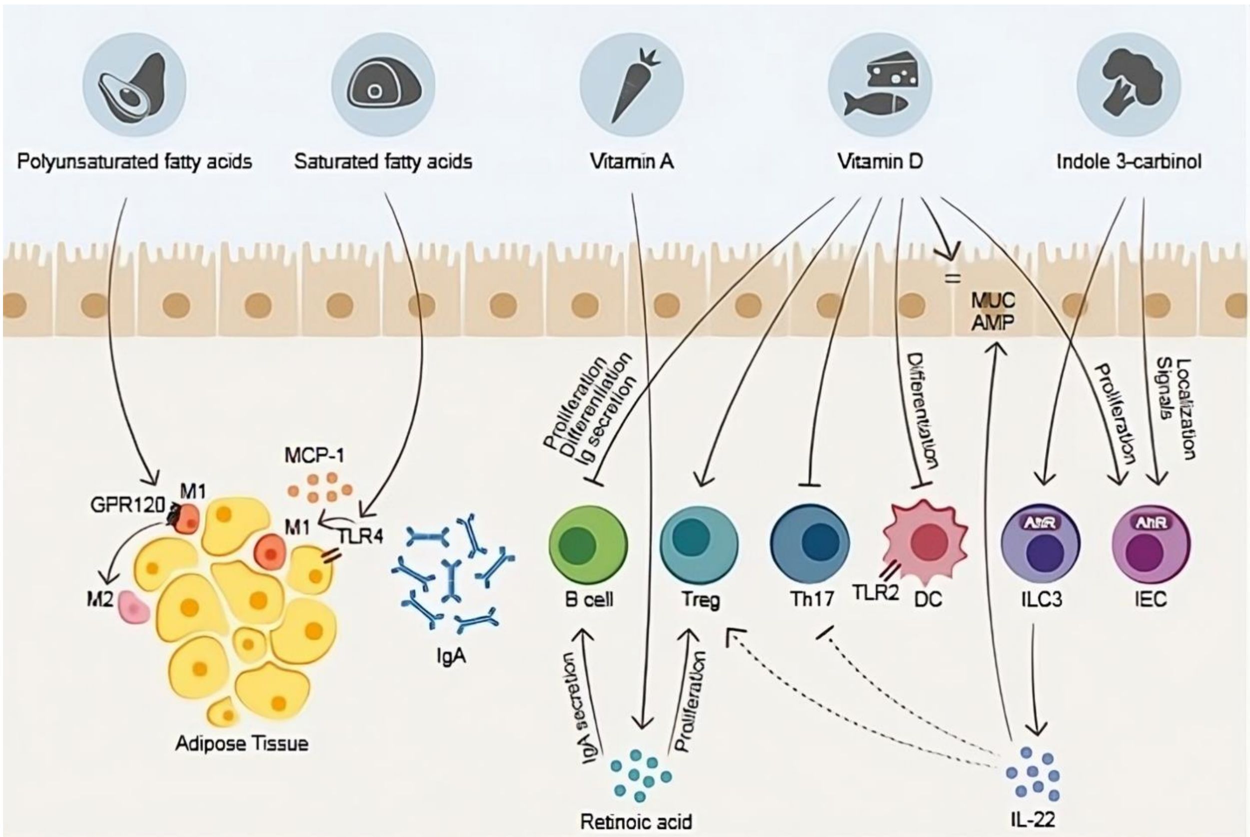


Figure 28. Nutrients and their immune and metabolic effects Source: adapted from (Zmora et al., 2017).

In monogastric animals, supplementation of vit A and D improve the integrity of gut mucosa and activate internal hematopoietic cells which in turn modulate metabolism of immune system of the body. regulate metabolism of immune system of the body. Likewise, supplementation of saturated and polyunsaturated fatty acids interacts with the immune system of adipose tissues.

11.4.2 Preventing metabolic disorders

High producing dairy cows suffer more of the metabolic disorders due to physiological imbalances (reduced nutrient intake through diet and higher nutrients outflow in milk). Metabolic disorders occur when cow is unable to cope with enhanced metabolic demands the related metabolic processes in the body are disturbed. Metabolic disorders and intoxication are the major concerns when the cow is in transition period and such problems can also serve as predisposing factors for many other health disorders (Sundrum, 2015). So, care and dietary management of the cow during the transition period, which last 3 weeks prepartum to 3 weeks postpartum, is of utmost importance for production performance, health and reproductive success (Grone et al., 2024). Dairy cows with high BCS (> 3.5) at the time of calving are more susceptible to metabolic disorders like milk fever and ketosis, while those with lower BCS (< 2.5) are more likely to have low conception rate. A BCS of 3.0 at the time of drying off the cow, 3.0-3.5 at calving, 2-2.5 at first service and 2.5-3.0 in mid-lactation will optimize the overall lactation cycle with less incidence of metabolic disorders. To improve the chances of conception at first service after calving, the cow should be gaining weight at earliest after losing weight in early lactation (Kennedy, 2007).

Metabolic disorders also known as production diseases such as milk fever, mastitis, ketosis, metritis and lameness are closely related to a suboptimal nutritional management. Production diseases although can occur at any time in the life of a dairy cow, yet more pronounced occurrence of disorders happen during the transition period (shift from dry state to high yield of milk). Thus, a great deal of adaptation and metabolic regulation is required. In high-producing cows grain feeding is very common to fulfill the great energy demands which may result in subacute ruminal acidosis leading to reduces milk yield, rumen health, and barrier function. Practical solution to prevention of metabolic disorder is that quality feed is available to all animals without any restriction to fulfill their nutrient requirements. In case of elevated levels of NEFA or BHB, various dietary administration such propylene glycol administration and supplementation with insulin (prebiotics) may be beneficial.

11.4.3 Managing gastrointestinal tract health

The nutritional and overall health status of monogastric and ruminant species is reflected by its gut health (Muneeb et al., 2024; Steele et al., 2015). It is now clearly evident that health of GIT has relevance in animal nutrition from starting from mood to immune function and productive performance of the animals. Gut health is an increasingly important topic in animal nutrition. A new concept of “gut health” through healthy functionality of GIT has been introduced by giving an overview of the features involving gastrointestinal functionality and animal health (Fig. 21) (Pietro Celi et al., 2017).

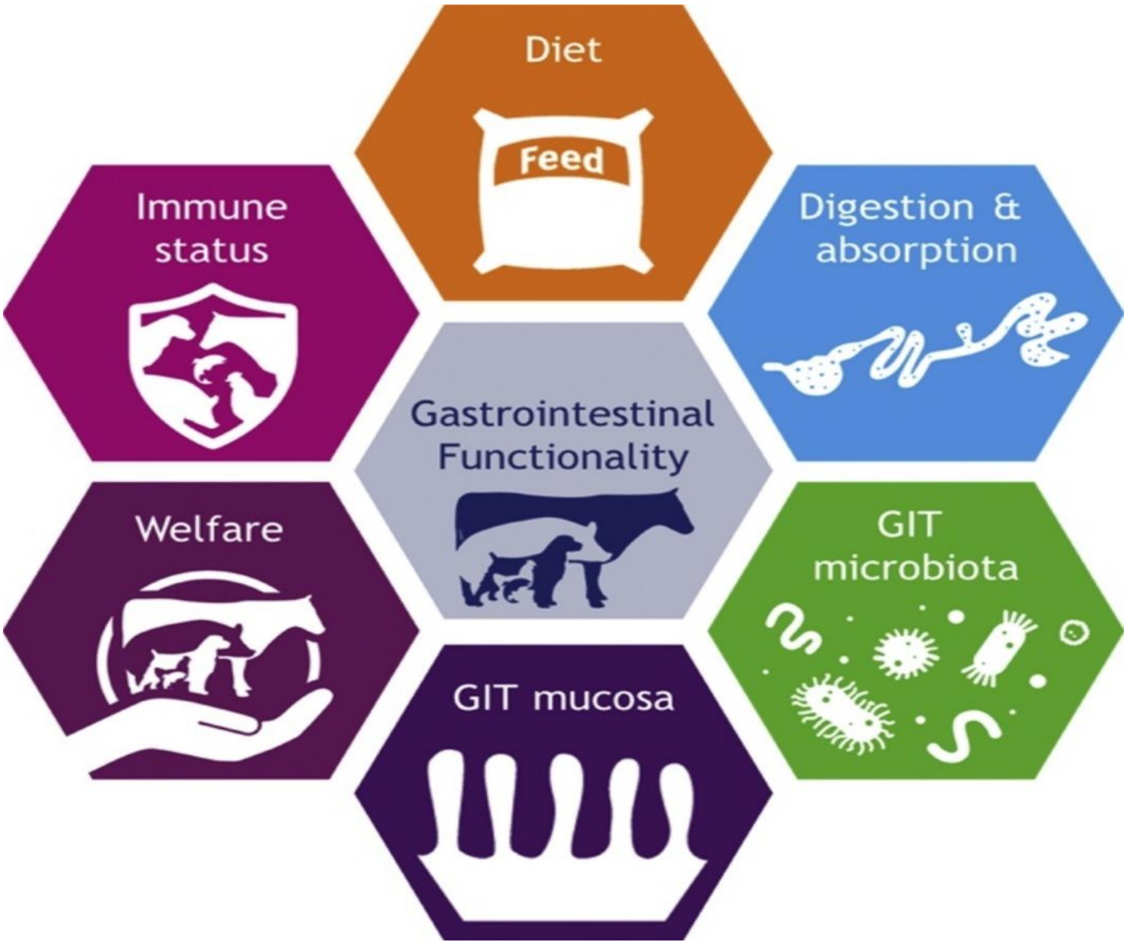


Figure 29. Gastrointestinal functionality and related factors

Feed efficiency, production performance and overall health are heavily dependent on gut health and microflora (Jha et al., 2019). Optimum functionality of GIT and its health are necessary for sustainable production performance in terms of growth, milk yield, egg and meat quality in livestock and poultry setup. The gut is the chief organ for the digestion and absorption of nutrient and largest immunological organ in the body (Kraehenbuhl & Neutra, 1992). Furthermore, it also serves as a protective mechanism to exogenous pathogenic organisms which can enter and colonize the host cells and tissues (Mathew, 2001). Thus, a healthier gut promises a healthier animal by supporting proper immune response and through impact on physiological processes such as efficient metabolism, digestion and utilization of nutrients (Yegani & Korver, 2008). However, physiology and biology of gut is highly complex and involves macro- and micro-structural integrity of the gut, type, concentration and balance of the microflora and the status of the immune system (Choct, 2009).

In chickens, not only the type of feedstuffs, but also particle size and form of feed influence gut health and function (Jha & Kim, 2021). Like other animals, chicken gut health comprises the immune system, the balance of the gut microbial population and the overall structural integrity of the gut at both macro and micro levels. Consideration of gut health in feed formulation of monogastric animals has becoming a trend in animal feed industries (Choct, 2009). During last couple of decades, instead of using antibiotic growth promoters, an encouraging trend was seen for use of prebiotics to modulate gut ecosystem through modifying the intestinal microflora, improving the digestibility and health of epithelium and stimulating the metabolism of the immune system (Teng & Kim, 2018). In ruminants, use of probiotics, the powder form of living beneficial microorganisms such as *Bifidobacterium or Lactobacillus,* as a feed additive has been reported to maintain the intestinal microflora and gut health and to improve production performance (Mahesh et al., 2021; Soccol et al., 2010).

11.4.4 Role of dietary fiber in gut health

Our knowledge on fiber fermentation in intestine of ruminants and its effect on health (due to interaction of fiber with microbes) has expended in last decade. Ruminants are able to digest fiber i.e., structural carbohydrates that are not consumed by other mammals (Izquierdo et al., 2021). As a food for microorganisms, the microbial products (*refer to 'dietary strategies to influence the gastro-intestinal ecology and to improve intestinal health' for details*) thus produced are absorbed through ruminal or intestinal walls to enter into metabolic process. A good source of fiber not only have impact on the microbial community but also have potential to reduce colonization with pathogenic bacteria (Metzler-Zebeli et al., 2010).

Dietary fiber (DtF) found only in plants is a group of complex substance consisting of celluloses, hemicelluloses, pectin and lignin. DtF is also sometimes called as non-starch polysaccharides (NSP) or unavailable carbohydrates due to presence of cellulose, hemicellulose and pectic substance (carbohydrates). DtF may vary from plant-to-plant species and variation may cause different effects of fibrous diet. Although humans don't have any mechanism to digest DtF in stomach but microbiota of colon can break it down. Fibers are necessary to good functioning of stomach and intestine due to longer stay in stomach with slow carbohydrates absorption and steadily flows of serotonin (mood boosting hormone) (Wiseman, 2002). DtF can contribute to gut development in monogastric animal species due to provision of physical stimulus to gut by hard and solid particles of fiber. DtF has been known to not only reduce chronic disease risk but are energy source to gut microbiota as fermentable substrate. The gut microbes in turn can influence host health by production of beneficial microbial metabolites and modulation of microbial profile (Wong et al., 2017).

Hindgut fermentation in ruminants contributes 5-10% of total dietary energy and affects production and health of the animal (Gressley et al., 2011). However, certain conditions such as ruminal acidosis (indicative of failure of healthy rumen function) can lead to excessive carbohydrate fermentation in the hindgut resulting in hindgut acidosis. Hindgut acidosis (HGA) occurs in high producing dairy animals with higher intake of fermentable carbohydrates which results in higher propionate to acetate ratio, decreasing the pH of digesta and dramatic alterations in microbial populations (Gressley et al., 2011).

11.4.5 Use of antioxidants for livestock health

Adequate dietary sources of antioxidants can contribute to protection of livestock against harmful effects of reactive oxygen species (ROS; free radicals). Natural antioxidants are consisted of natural molecules such as tocopherols (vitamin E), ascorbic acid (vitamin C), carotenoids (i.e., xanthins, β-carotene, luthein and lycopene), flavonoids (i.e., morin, quercetin, catechins, rutin and epigallocatechins) and non-flavonic phenols. Minerals such as Mn, Se, Cu and Zn may act as antioxidants (Abdel-Moneim et al., 2021). Antioxidants supplied through diet activate bodily (endogenous) antioxidant defenses system which protects the cells from oxidative damage. This endogenous antioxidant system in tissues can safeguard cells under stress condition by regulating the amount of ROS through numerous enzymes and lipid peroxidation detoxifying molecules. These enzymes include superoxide dismutase (SOD), glutathione peroxidase (GSH-Px) and catalase (CAT) whereas, lipid peroxidation detoxifying molecules include glutathione s transferases, ascorbate peroxidase and phospholipid-hydroperoxide (Abdel-Moneim et al., 2021).

V. Sustainable and Ethical Considerations

11.4.6 Nutrition for animal welfare

In recent years, animal welfare and comfort has been in focus due to economic and ethical reason in animal production systems. Animals maintained under good welfare conditions have higher profitability due to higher productions, whereas poor care conditions can result in low productivity and ill health in animals including sheep, goats and buffaloes (Makkar, 2016).

Nutrients delivered to animals are extremely important as fundamental inputs in life processes since animals cannot survive without appropriate nutrients (Phillips, 2016). Ruminants of superior genetics in order to maintain high production can suffer from metabolic disorders causing welfare issues such as acidosis and lameness due to an excess or deficiency of nutrients. Breeding animals may suffer from welfare issues of chronic hunger when they are restrict-fed to optimize production and health. A poor quality and unsafe feed are one of the factors responsible for welfare issues in ruminants. So, a well-balanced diet which is free from contaminants, along with supply of clean water according to the requirement may prevent the animals from physical and physiological suffering of hunger and thirst. Without any doubt correct nutrition is vital to sustain optimal productive performance and fitness of animals (Makkar, 2016).

As list above, metabolic disorder due to excess or deficiency of nutrients, restricted or poor-quality feed and contaminated water (or unavailability) causing welfare issues of hunger and thirst; may have direct or indirect link with release of pro-inflammatory cytokines. These cytokines are indication of inflammation which may 'sickness behavior' leading to physical and mental discomfort. Inflammation can ultimately lead to digestive problem of ruminal and/or intestinal nature, may cause bacterial translocation through mucosal barriers (barrier function disorder) leading to endotoxins along with welfare problems (appetite issue and resultant negative energy balance and metabolic disorder risk). Moreover, nutrient deficiency during pregnancy could cause fetal deficiency of nutrients and variation in disease frequency later in life. (Bertoni et al., 2009)

11.4.7 Sustainable sourcing of feed ingredients

For most of the livestock and poultry farms, feed costs up to 70% of recurring production costs. For a sustainable livestock production system, a sustainable feed inputs is crucial (FAO, 2014). Increasing human population and decline in fodder cultivation land has exerted immense pressure on food and feed production. Food security directly connects the food-feed competition and the biologists are undertaking researches with the objective to minimize this competition. In this regard, efficient utilization of feed sources which are either waste or in excess for human consumption, in animal feed is regarded as one of the top strategies. In this regards, ruminant animals are efficient nutrient recycler, which can use the feeds like crop residues, agro-industrial by products, lignocellulosic biomass and other high fibrous feeds (which are waste for human consumption) and convert them into high quality human foods like milk and meat (Sufyan et al., 2022) Grains represent 14% of the food consumed by farm animals, which represents about a third of the world production of cereals, therefore such ingredients create a competition with human food (Mottet, 2024). However, ever increasing cost of grains and energy nutrients, has put a pressure on economics of farmers and a paradigm shift is required from costly to low-cost crop residues, wastes and coproducts from food processing industries and forages for grazing (Benoit & Mottet, 2023). On the other hand, cereal straw, stover, corn cob, oil cakes and meals and cereal brans are not edible for human and balancing the feed using such ingredients is highly applaudable for a sustainable agriculture production system. As most of the crop residues and by-products are high in fiber and/or lignin which impede the digestibility of ingredient, it is recommended to treat such feedstuff in such a way to improve the availability of nutrients in ruminant animals. For that, various physical, chemical and biological treatments have been suggested to improve the nutritional value and digestibility of feed (Sarnklong et al., 2010).

Phytochemical extracted from fruit and food industrial wastes contain medicinal value could be used as feed additive to improve livestock production health. In addition to improve rumen fermentation characteristics, metabolism, productive performance and health of the animals, use of such phytochemicals also supports minimizing the environmental burdens of greenhouse gases emitted from livestock (Singh et al., 2021; Singla et al., 2021). Future research on food, feed and fuel production must focus to ensure food security and nutrition for all.Use of alternate and non-conventional protein rich sources can also play important role in lessening the burden on conventional protein sources. Use of insect meal which is not only good protein source but also contain immunomodulatory and antimicrobial compounds (Veldkamp et al., 2023), microalgae, like spirulina (still much costly than traditional protein sources), yeast proteins, mycoproteins, and co-products extraction from crop industry like potatoes (Mottet, 2024) are gaining interest of the stakeholders relating to animal feed industry.

11.4.8 Nutritional manipulation to mitigate methane emission

Methane is regarded as greenhouse gas (GHG) having 28-fold warming potential of globe to that of carbon dioxide (CO_2) (Pachauri et al., 2014), and ruminal anaerobic fermentation is major (50–60%) contributor to this gas (Karekar & Ahring, 2023). Methanogenesis is a normal process of ruminal fermentation as it captures metabolic H_2 which is produced during the anaerobic fermentation of sugars in the rumen and, thereby, methanogenesis represent a pathway for the disposal of metabolic H_2 of microbial metabolism in order to balance the fermentation (Karekar & Ahring, 2023; McAllister & Newbold, 2008). Then question arises why to disrupt normal fermentation process and mitigate CH_4? The answer is to reduce environmental consequences of enteric CH_4 and increase ruminants' production efficiency.

The animals having certain archaea (*Methanobrevibacter gottschalkii*) usually produce more CH_4 in rumen, since it is the principal source of enteric methane emissions. Whereas, ciliate protozoa produce major substrate of methane formation in rumen, the H_2. Moreover, bacteria such as low *Proteobacteria* abundance and differences in certain *Bacteroidetes* and anaerobic fungi (due to high H_2 and format production) as well seem to be associated with greater methane emissions. Production and utilization of formate by ruminal microbiota is not properly understood and hence may be a source of variability between animals with respect to methane generation (Tapio et al., 2017).

We will discuss the nutritional strategies to mitigate enteric methane (Fig. 22). A range of nutritional strategies, such as phytochemicals (secondary plant compounds such as essential oils, tannins, saponins, flavonoids and organosulphur compounds with anti-methanogenic properties), lipid nutrition with addition of high concentrations of medium-chain fatty acids or polyunsaturated fatty acids, (Grainger & Beauchemin, 2011; Patra, 2013), increasing the cereal grains/concentrates (Hristov et al., 2013), improved forage quality (forages with higher ratio of non-fiber carbohydrates to NDF and containing less lignified NDF, which also promote organic matter(OM) break down in rumen (Hristov et al., 2013) and use of probiotics as well. Several new potential technologies, such as the use of plant secondary metabolites, predominantly polyphenols, essential oils (Cobellis et al., 2016), tannins (reduced CH_4 production when expressed relative to digestible OM (established in meta-analysis of in vitro and in vivo experiments) (Jayanegara et al., 2012), saponins, and alkaloids, have been explored to modulate ruminal microbial fermentation and decrease methane production because of their antimicrobial and antimethanogenic properties (Patra, 2022; Samal & Dash, 2022). Since certain animals have lower tendency for CH_4 emission, hence, genetic selection for low emission is another strategy in addition to nutritional intervention. Seaweeds (containing plant secondary compounds) are not only potential feed for ruminants, but can also be used as dietary supplement due to antimethanogenic effects (by modifying rumen fermentation and inhibiting methanogenesis pathways), which may also be further beneficial environmentally (Min et al., 2021).

As stated earlier, additives such as essential oils have potential to reduce methane production invitro and in vivo (with thyme showing promising results). In this regard, essential oils have shown following reduction in methane production invitro (except when mentioned in vivo): thyme 30% (Günal et al., 2017), 21% in dairy cattle (Laabouri et al., 2017), and enhanced propionic acid production in rumen of Holstein Friesen calves (Vakili et al., 2013), lavender 60% (Yadeghari et al., 2015) and 45% (Beyzi, 2020), peppermint > 25% (Patra & Yu, 2012), eucalyptus up to >17% (Patra & Yu, 2012), 85% (Sallam et al., 2009) and no effect in sheep (Wang et al., 2018), rosemary > 20% (Roy et al., 2014) and 9% (Cobellis et al., 2015), garlic 42% (Patra & Yu, 2012), 73% (Busquet et al., 2005) and 94% (Soliva et al., 2011), Clove 34%

(Patra & Yu, 2012), and no effect in dairy cattle, oregano 11% (Zhou et al., 2020) and 87% (Patra & Yu, 2012).

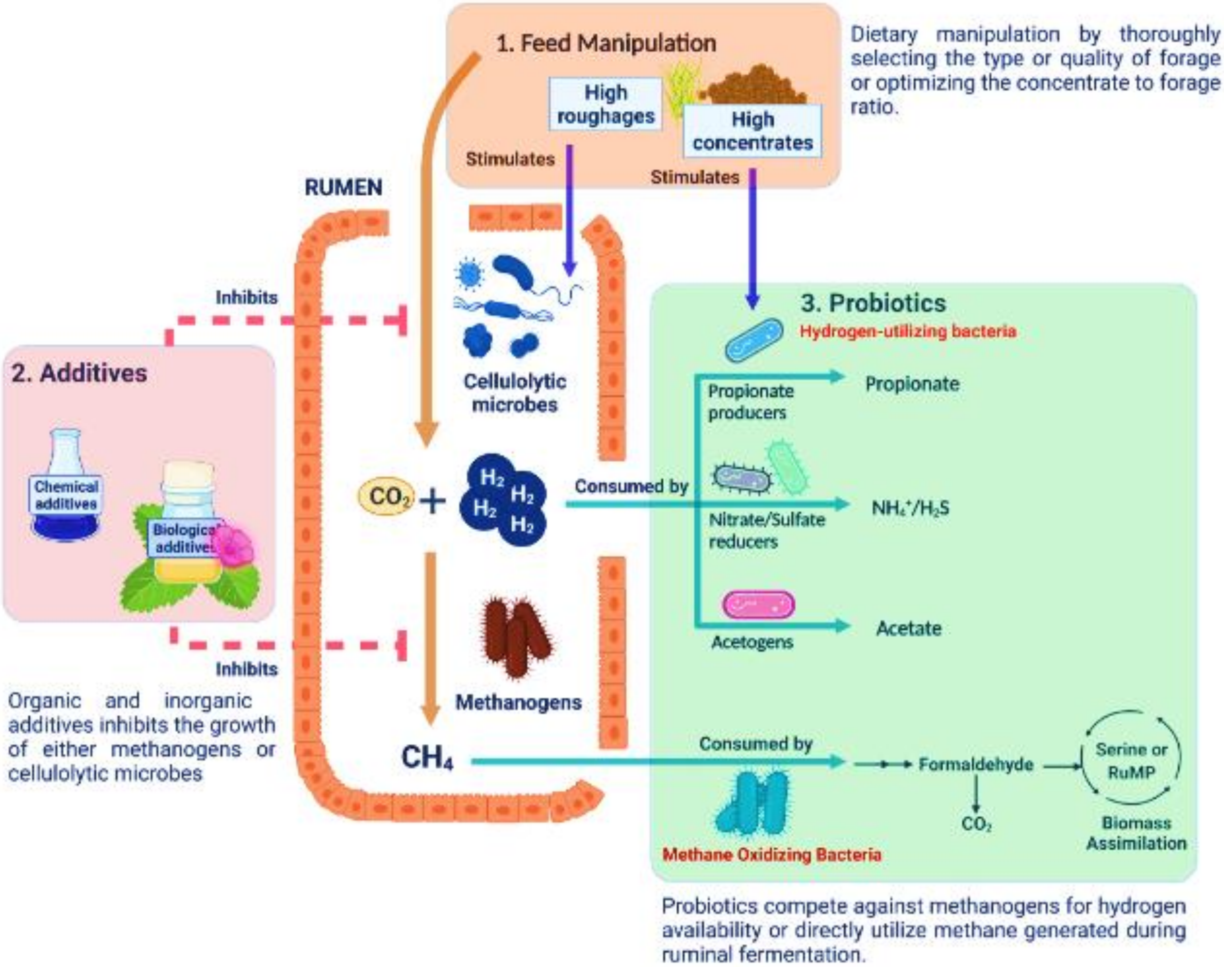


Figure 30. Nutritional strategies for mitigation of methane from ruminants. Source: (Tseten et al., 2022).

Technique used are feed manipulation, supplementation of probiotics and prebiotics as additives. Light brown indicate line flow of rumen fermentation, pink line indicate inhibition, purple line indicate stimulation, and green line indicate consumption.

Dicarboxylic acids (organic acids such as α-ketoglutaric acid/oxaloglutarate, pyruvate, fumarate, tartate, citrate, aspartate, lactate, succinate etc.), when enter into rumen can be converted into propionic acid and consequently restrict the routing of hydrogen towards the formation of methane gas (Fig. 23). Under invitro conditions, organic acids such as acrylic acid and fumaric acid were reported most prominant in reducing methane formation (Newbold et al., 2005). It has been shown that fumarate @3.5 g/L in continuous fermenters using pasture as a substrate resulted in reduction in methane output by 38% (Kolver et al., 2004). Supplementing diet with multiple propionate precursors (containing two or more dicarboxylic acids) exerted additive effect in lowering the methane gas production in rumen (McAllister & Newbold, 2008). In sheep, a higher dose of fumaric acid (encapsulated in fat for slow ruminal release) @10% of diet resulted in methane emission by 40-75% in sheep (Wallace et al., 2006). In contrast, a non-significant effect of supplementation of fumaric acid on the emission of methane was observed in cattle (Beauchemin & McGinn, 2006).

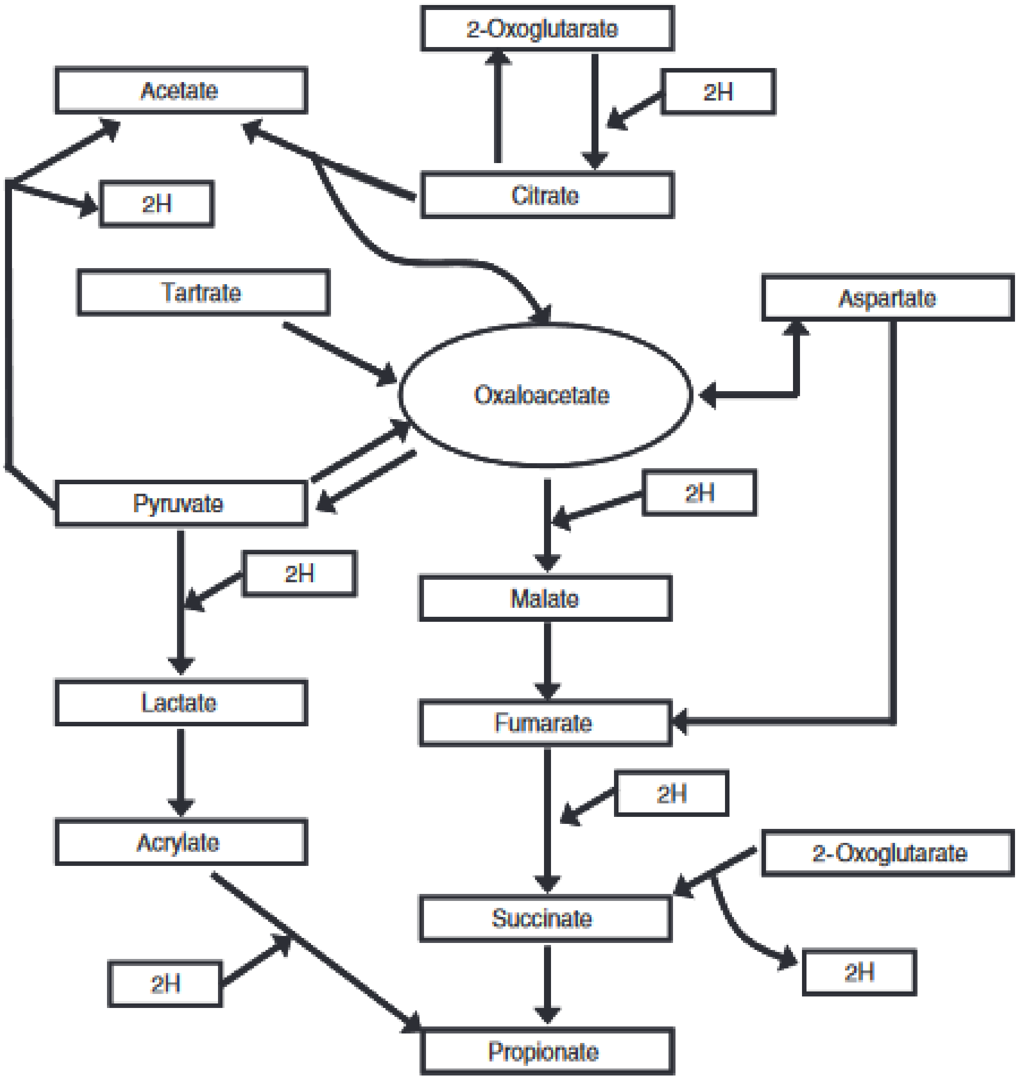


Figure 31 Metabolic pathways for different precursors of propionic acid in rumen. Source: (Newbold et al., 2005).

11.5 Emerging trends in Animal Nutrition and health

11.5.1 Feeding for designer animal products

In recent years, a higher trend has been observed for the demand of human food products which are safe and contain particular nutritional compound which promote health or are deficient in human food items, globally. Production of such animal products, usually termed as ‘designer food products’, is a challenge for animal nutritionist and requires extensive research. Animal products like meat (source of essential AAs, folate, vitamin A, D and B vitamins (especially B12), Fe, Zn and Se) and eggs (source of ω-3 and ω-6 long chain PUFA) are extremely important as they are nutrient rich. Using breeding and nutritional strategies, certain nutrients of interest which promote health (like critical amino acids, PFA, omega-3, long chain eicosapentaenoic (EPA) and docosahexaenoic (DHA) and linoleic acid which contain anticarcinogenic, anti-atherogenic, and immunomodulatory effects) can be enriched and other nutrients which are supposed to pose risk to the human health (tans and saturated fats, mycotoxins and antinutritional factors) can be reduced in milk, meat and egg. Likewise, certain micro-nutrients like mineral, vitamins and bioactive compounds of natural origin can also be incorporated into designer animal products. In this regard, it should be kept in mind that type of animal, breed and strain vary to respond the higher level of certain nutrient in the feed. For instance, animal products supply some essential minerals like Ca, K, Fe, P, Zn, Se and S, however the response of certain minerals such as addition of I and Se in the feed affect the concentration of I and Se in milk, meat and eggs more prominently as compared to others. Likewise, is the response of certain vitamins when added in the feed in the upper range, e.g., vitamin A / beta-carotene, E and B complex. However, such capability and flexibility of body metabolism to respond the higher-level nutrition in the diet can be improved by developing special breeds or strains using breeding techniques.

11.5.2 Nutrigenomics in animal nutrition and health

Nutrigenomics involves the study of effects of nutrients on genome functioning, protein synthesis and epigenetic mechanism (Nowacka-Woszuk, 2020). Studies on Nutrigenomics at underlying level will not only help in determining the requirement of nutrients needed for certain physiological function, but also for gathering a more reliable data on how various nutrients impact at gene level to product health and productive functions in the animal. Different techniques in the field of nutrigenomics, such as metabolomics and transcriptomics offer insights into intricate regulatory relationships, including the interactions among diet, nutrients, metabolites, genes and gene functions. Such techniques can effectively be used to study the processes occurring at molecular level of genome when it receives a particular nutritional-signal and responds to them by engaging in specific metabolic activities within the organism (Zduńczyk & Pareek, 2009). Metabolic signals turn on or off the gens after nucleus in cell receives signals from internal (hormonal) or external (nutrients) factors being most influential of the environmental stimuli. To understand the underlying mechanism involved in relationships between nutrition and health, biologists are working on various biomarkers and their effect on body functions, disease response and overall well-being (Picó et al., 2019).

11.6 ANTIBIOTIC ALTERNATIVES FOR LIVESTOCK HEALTH

11.6.1 Nutritional interventions to reduce antibiotic usage

Antimicrobials commonly used in the animal industry either as growth promoters or treating infectious diseases, but use of antibiotics can pose a serious risk of development of antimicrobial-resistant (AMR) in human when consume such animal products (Patra, 2022), when humans consume antibiotics residue containing foods of animal origin. Antibiotic resistance in livestock represents an iceberg that the world is facing in both veterinary and human medicine due to frequent use of antibiotics as growth promoters and therapeutic agent (Ayukekbong et al., 2017; Saettone et al., 2020).

Considering the harm or complete ban on antibiotics for livestock use, scientific community has tried organic acids (OAs), probiotics, prebiotics, postbiotics, phytogenics, enzymes, antimicrobial peptides, clay mineral as most promising products that are capable of preventing or treating enteric/gastrointestinal diseases (Lillehoj et al., 2018; Muneeb et al., 2024); along with integral approach of taking into account the composition of the diet and effective husbandry practices (yielding highly promising outcomes). This segment will look for innovative dietary strategies as viable alternatives/practical alternatives to antibiotics in monogastric as well as in ruminant livestock species.

11.6.2 Health benefits of organic acids as acidifiers in animal feed industry

Organic acids (OAs) possess anti-microbial nature due to the capability to decrease the pH in digestive tract and therefore, can serve as potential alternative to antibiotics (Du et al., 2024). However, the poultry and pork industry has seen conspicuous preference to acidifier as growth promoters. Various acidifiers such as formic, fumaric, citric, acetic acid, propionic acid, butyric acid, sorbic acid, tartaric acid, and lactic acids are produced during metabolism of animals; and hence due to these qualities of nutrition (energy source) and preservation, OAs are used in livestock feed industry to acidify the feed as a prophylactic measure to counter microorganisms like bacteria, fungus and molds in the feed industry. Various OAs and their salts have been described to promote health and performance of animals besides their role in pathogens intake in feed, digestion of feed and digestibility of nutrients and correction of gut microbiota dysbiosis.

Thus OAs in animal nutrition represent an economical option for performance enhancement which exert their effect on animals through feed, intestine and metabolism (Pearlin et al., 2020). In addition to having antimicrobial activity, OAs can also perform functions as antifungal (can increase shelf-life and storage capability of feed ingredients), anticoccidial and antiprotozoal agents for their therapeutic effects on pathological disorders. Moreover, OAs improve immunity, morphology and functioning of GIT (Du et al., 2024), as illustrated (Fig. 24).

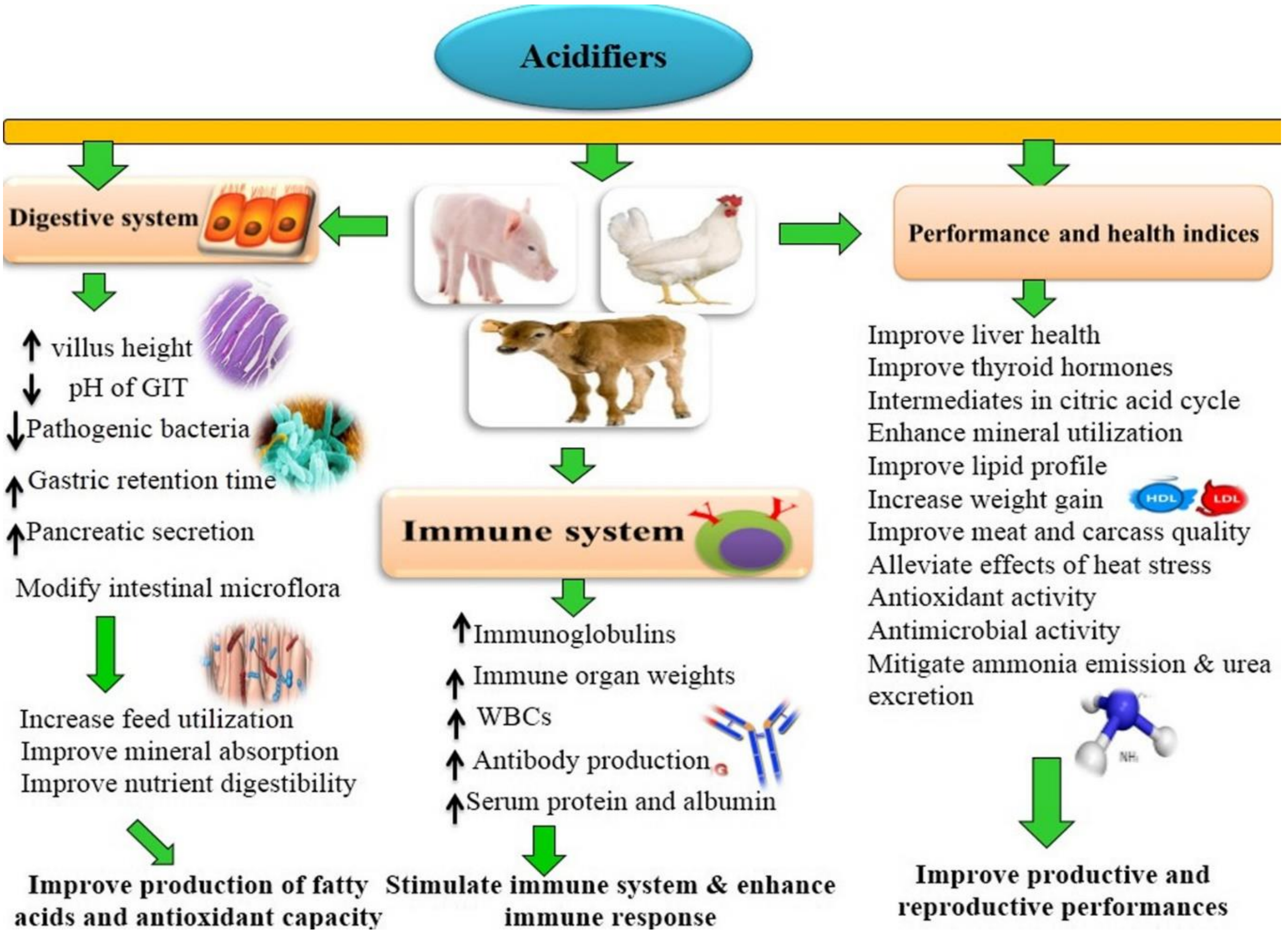


Figure 32 Effect of acidifiers on various parameter related to production, digestive health and immunity Source: (Pearlin et al., 2020)

11.6.3 Probiotics

Probiotics are live microorganisms (falling under specific criterion) that promote physiology and heath of the animals when given in adequate amounts (Hill et al., 2014; WHO, 2002). Mode of administration of Probiotics can be through spray, drinking water, dietary supplements or functional feed as pure or mixed culture (Cassani et al., 2022; Krysiak et al., 2021). Various microbes have been identified that can benefit the host animal when used as additive in the feed of animals such as lactic acid producing bacteria (e.g., *Streptococcus*, *Lactococcus Lactobacillus* and *Bifidobacterium*), non-lactic-acid bacteria (e.g., *Acetobacter and Propionibacterium),* yeasts (e.g. *Saccharomyces*, *Issatchenkia* and *Kluyveromyces*) and spore forming bacteria (e.g. *Bacillus*, *Sporolactobacillus, Clostridium* and *Brevibacillus*) (Cassani et al., 2022).

In the context of natural and antibiotic free livestock production, animal agriculture around globe has immensely tried probiotics due to the myriad of production and health benefits. Animal nutrition programs have widely evaluated probiotics for improvement in beneficial gut microbiota (eubiosis) and for elimination of pathogenic gut microbes (dysbiosis), resulting in advantages i.e., enhanced GIT functionality, improvement in immunity at gut as well as whole-body (systemic) level with better health status of both ruminant and non-ruminant species (Fig. 25) (Mahesh et al., 2021).

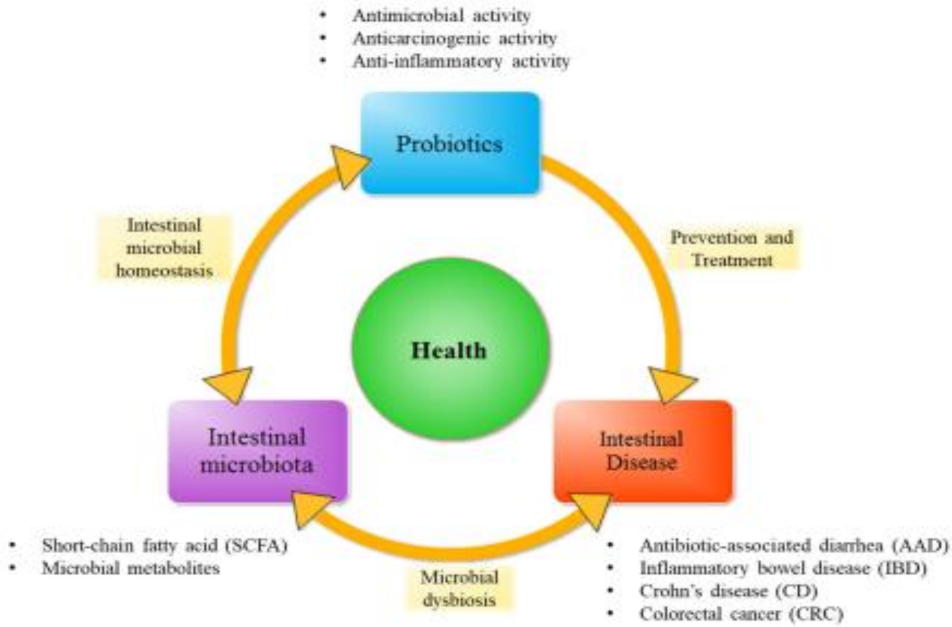


Figure 33. Role of probiotics relevant to intestinal diseases via recovery of dysbiosis in the intestinal microbiota Source: (Kim et al., 2019)

Various intestinal diseases such as diarrhea, inflammation and even colon cancer are closely related to dysbiosis of the intestinal microflora. Administration of probiotics has been proposed as a therapeutic approach to prevent intestinal disease. Probiotics can have anti-inflammatory, antimicrobial and anti-carcinogenic effects, so they can help restore an imbalanced gut microflora. This recovery can also relieve the symptoms of these intestinal diseases.

Probiotics for example in poultry help in synthesis of an array of metabolites such as H_2O_2, diacetyl, bacteriocins/antimicrobial peptide and SCFAs that impede proliferation of pathogens and thus benefit in modulation of the intestinal microbial profile (Ben Lagha et al., 2017; Kulkarni et al., 2022; Muneeb et al., 2024). Probiotics in livestock or humans have been shown to exert their effect on intestinal health by producing antibacterial (bacteriocins & bacteriocin-like inhibitory substances) and other compounds which inhibit adhesion of pathogenic microbiota to intestinal wall; boosted immune system of host with raised serum concentrations of immunoglobulins (especially IgG/ IgY and IgA), interaction with toll-like receptors, along with decrease in bacterial enzyme activity, lower NH_3 production, enhanced digestive enzyme activity, increased lactic and other volatile acids, production of primary metabolites (hydrogen peroxide, carbon dioxide, and diacetyl), siderophores, biosurfactants, competitive exclusion of pathogenic microbes (through bacteriostatic effect and/or direct killing of pathogens and provide advantage to probiotic bacteria to compete with pathogens), and neutralization of toxins are some of the distinguishing characteristics associated with using probiotics (Cassani et al., 2022; Hill et al., 2014; Krysiak et al., 2021; Patel & Katole, 2019).

11.6.4 Prebiotics

A substrate of compound which support and improve the efficiency of microorganisms of the host animal and cause production and health benefits (Gibson et al., 2017). Prebiotics as natural products (mainly carbohydrates based such as fructooligosaccharides, inulin, lactulose, galactooligosaccharides, and β-glucans) when act as fermentable substrate will stimulate growth of beneficial bacterial and metabolic processes associated to various health problem (Voss et al., 2022). Prebiotics now may also include peptides, polyphenols and PUFAs (Gibson et al., 2017). The presence of prebiotics in the diet may lead to various health benefits through positive effects on the gastrointestinal tract (Fig. 26). Prebiotics provide a substrate to beneficial intestinal microorganisms for proliferation, modify gut microbial profile, boost immune responses, inhibit the invasion of pathogens, lower cholesterol, improve gut health through the modulation of intestinal microbiota equilibrium, facilitate enzymatic processes, mitigate the formation of phenol and NH_3 byproducts, and hence improve the production efficiency and has been described to have the potential to serve as a viable substitute for antibiotic growth promoters through their ability to modify intestinal bacteria and immune system. This modification aims to reduce the colonization of pathogens, enhance the utilization of nutrients such as amino acids and proteins, improve gut health and ultimately enhance overall performance (Muneeb et al., 2024).

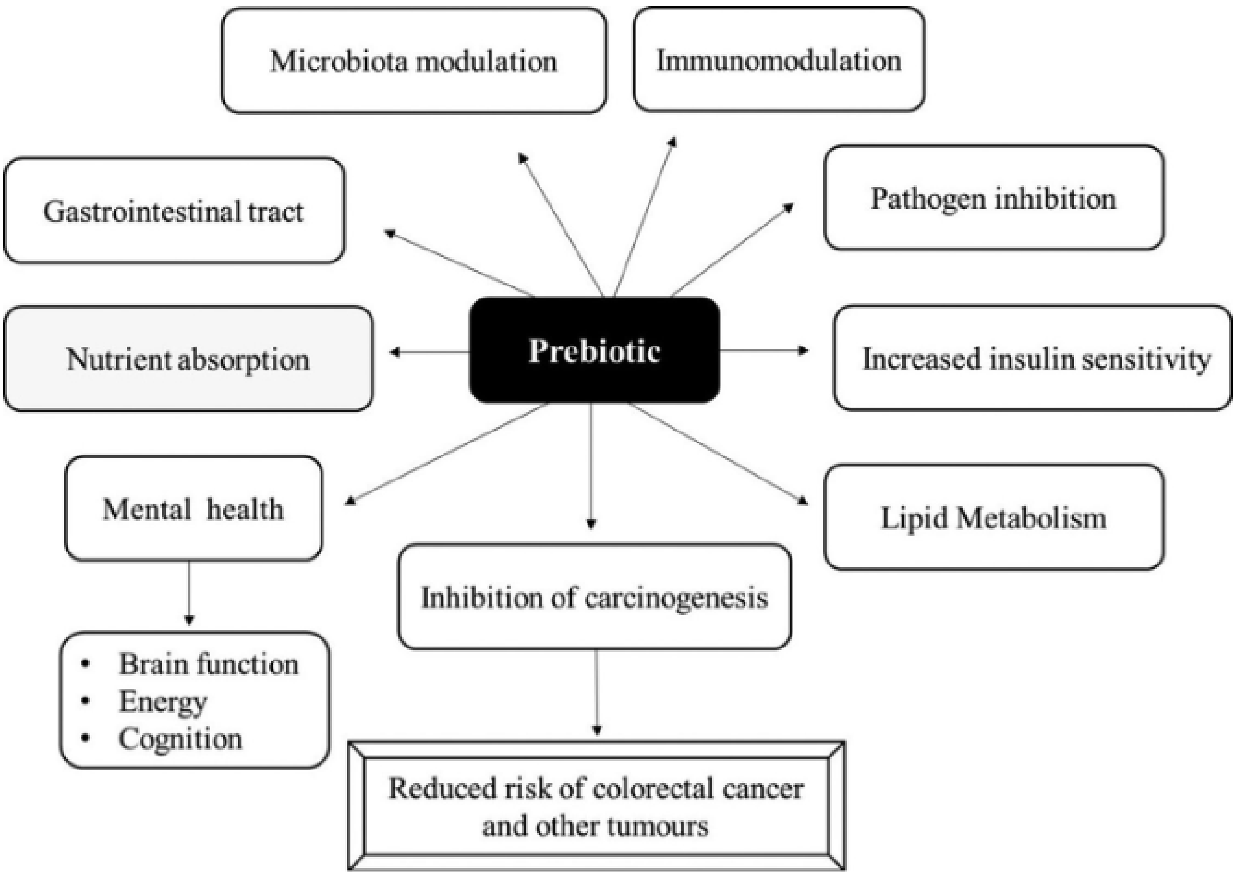


Figure 34. Different physiological outcome and health benefits of adding prebiotics in animal feed. Source: (Voss et al., 2022).

11.7 CONCLUSION

Occurrence of diseases in the farm animals results in huge economical losses. Nutrition, in addition to providing essential nutrients for maintenance, growth, production and reproduction, also greatly influence the immunity, health and overall well-being of livestock. Providing nutrients in adequate amount and balance ensures quality animal product and food security which is one of the major concerns of human. To have a sustainable agriculture, efficient use of industrial by-products, crop residues and lignin/fiber rich ingredients in the animal feed along with applying stratigies which promote nutrient utilization, productivity and health of the animal are imperative. Professionals related to livestock and poultry production must have good understanding of nutrients, digestive physiology, metabolism and how they effect on growth, production and health of various classes of animals. In any animal production system, the best strategy is ‘prevention is better than the cure’ and in this regard nutrition shall be dealt as ‘preventive medicine’.

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Chapter 12 : Bacterial Plagues: Understanding Major Diseases in Livestock

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Abstract

Livestock is one of major sector of Agricultural. It plays an important role in economy of any country. It also plays an important role in GDP and source of income for many families. There are many major diseases of animals but major diseases include Listeriosis, Theileriosis,

Tetanus, Brucellosis, Hemorrhagic Septicemia, Enterotoxemia, Foot and Mouth Disease (FMD), Rabies, Paratuberculosis (Johne's Disease), Botulism, Blackleg Disease, Lumpy Skin Disease (LSD). Listeriosis, is a rare but serious illness caused by Listeria monocytogenes with a high fatality rate, mainly transmitted through contaminated food. Theileriosis, affects domestic animals caused by various species of Theileria, characterized by fever, anorexia, and anemia. Control may include tick control, supportive care, vaccination, and selective breeding for resistance.Tetanus, is one of important bacterial disease caused by Clostridium tetani, show muscle spasms and neurological symptoms. Brucellosis, caused by brucella abortus and transmitted in animals by various routes, affects many animal species with clinical signs such as abortion and orchitis and cause abortion in 3rd trimester. And also, a zoonotic disease,Pasteurella multicide, is causative agent of Hemorrhagic Septicemia, and it mainly affecting water buffalo and cattle.Enterotoxemia, is also major disease caused by Clostridium perfringens type D, results in sudden death and GIT problems in cattle.Footand mouth disease (FMD), a highly contagious viral disease, results in fever and lesions in cloven-hoofed animals. It is Control by proper vaccination of animals and movement restrictions.Overall, effective disease management include a proper vaccination of animals, sanitation, control of vector, and proper veterinary care to reduce the impact of these diseases on livestock population and public health.

Key words

Livestock, GDP, ECONOMY, Diseases and Prevention

12.1 Introduction

Pakistan is an agriculturally important country. Raised in an agricultural environment, livestockare domesticated animals that are raised to supply labor and a variety of consumable goods, including meat, milk, eggs, wool, and leather.

Livestock Animals:

Cattle, Buffaloes, Sheep, Goats, Horses, Camels,

Estimated Livestock Population

Species	2022-23 (in Million)
Cattle	55.5
Buffalo	45.0
Goat	84.7
Sheep	32.3
Camel	1.1
Horses	0.4

12.1.1 Role Of Livestock in National Economy

Livestock is vital to the national economy of many nations worldwide. Some salient features ofthe input are given below:

Food Security: A major amount of food security in the world helps provide food security. Income Generation: Particularly in rural areas, millions of people rely on raising livestock as a source of income. Along the entire value chain, from farming and animal husbandry to processing, shipping, and marketing, it offers job opportunities.

Livestock Farming contributes to agricultural diversification by lowering dependency on a single crop. This diversification aids in reducing the risk of crop loss caused by pests, illnesses, or unfavorable weather.

GDP Contribution: The livestock industry significantly increases the gross domestic product (GDP) of several countries. Animal husbandry, processing, trading, and other related businesses,all of which support the country's economy, are included.

Export Revenue: Meat, dairy, and leather products are examples of livestock products that are significant export goods for many nations. By exporting these goods, the strength of the economy and trade balance can be increased by earning foreign exchange.

Jobs: Throughout the value chain, livestock husbandry generates jobs, especially in rural areas where there are no other reliable sources of protein or vital nutrients from livestock. Animal products such as meat, milk, and eggs are staple sustenances for billions of people worldwide. Because livestock farming guarantees a steady supply of items derived from animals,

There may be a shortage of jobs. Livestock production helps maintain rural communities and provides a means of subsistence for both large commercial enterprises and small family farms. **Contribution to Rural Development**: By creating jobs, boosting local economies, and enhancing infrastructure, livestock farming can help rural communities flourish. It promotes sustainable development and keeps the social fabric of rural communities intact.

The use of By-products: Crop leftovers and agro-industrial waste are examples of by-products from other industries that can be used in livestock production as animal feed. This reduces wastage and improves the effective use of agricultural resources.

Livestock contributes to employment, income generation, food security, and rural development among other aspects of the national economy. Because of its significance, sustainable practices that strike a balance between the economic, social, and environmental factors.

12.2 LISTERIOSIS

1. History:

- Listeria monocytogenes causes listeriosis, and depending on the area and region, there are 0.1 to10 instances per 1 million people annually, making it a relatively uncommon illness.

- Although listeriosis is extremely rare, its high fatality rate makes it a serious public healthconcern.
- Pandemics have impacted all nations worldwide.

2. Causative Agent:

- L. monocytogenes is found in large quantities in nature. It is present in vegetation, water, soil, and animal wastes. Contaminated food is the main source of infections.
- L. monocytogenes can thrive and proliferate in low temperatures, such as those found in refrigerators, in contrast to many other hazardous bacteria.

3. Clinical Signs:

- The non-invasive variant, known as familial listerial gastroenteritis, affects people who are otherwise healthy, but have mild symptoms.
- Fever, chills, headache, upset stomach, diarrhea, nausea, vomiting, muscle aches, and confusion are some of these symptoms.
- Invasive form: It has the potential to spread to the brain or the circulation.
- Pregnant women, fetuses in development, the elderly, and people with compromised immune systems are among high-risk populations.

4. Treatment:

- Treatment May involve the use of antibiotics.
    - Tetracycline
    - Oxytetracycline
    - Inj.Cephur

1. Control

Prevention:

- Carefully prepare and wash food.
- Steer clear of high-risk foods such as cold-smoked seafood products, soft cheeses, and deli meat.
- Use good hygiene when handling food. Vector and tick controls. (José A et al., 2001).

12.3 Theileriosis

2. History:

- Haemoprotozoa of the genus Theileria cause tick-borne disease theileriosis, which affects both domestic and wild Bovidae (cattle, buffalo, and other related species) globally.

- Different Theileria species are responsible for different manifestations of illness.

3. Causative Agent:

Theileria species are parasitic protozoans that are obligate intracellular members of the phylum Apicomplexa. Although they are most closely linked to Babesia, they are distinct in that they first infect leukocytes and then spread to erythrocytes.

- Among the notable species are

- T. parva: In cattle, causes *East Coast fever (ECF)*.

- T. annulata: Responsible for Mediterranean or Tropical theileriosis (TT).

- T. orientalis: Theileria-associated bovine anemia (TABA) or *Oriental theileriosis (OT)* are caused by this bacterium.

- Small ruminants were affected by T. lestoquardi, T. uilenbergi, and T. luwenshuni.

4. Clinical Signs:

The symptoms of acute diseases include the following:

- Elevated temperature - Swollen lymph nodes - Anorexia - Severe illness loss Symptoms of conjunctivitis include pale mucous membranes.
- Hemolytic anemia is caused by the immune system and/or non-regenerative anemia.

5. Treatment:

(i). The most effective medication is dipervaquone, and 2.5 mg/kg BW is the recommended dosefor sheep, goats, and cattle (ii). Successful treatment has been achieved with the use of broad- spectrum antibiotics such as oxytetracycline, chlortetracycline, and tetracycline (iii). 1.2 mg/kg of halofuginone lactate b. Oral information regarding recuperation (iv). Bereline has also been successfully used.

6. Control:

The primary goals of management are: Tick control: Avoid tick infestation.

- Supportive care: blood transfusions, fluid treatment, and antipyretics.
- Tick vectors are the focus of vector control.
- Vaccination: Certain species are eligible for certain immunizations.
- Selective breeding: Creating cattle breeds with resistance.
- Movement limitations and quarantine: stopping the spread.
- Surveillance and monitoring: Early identification and management. (Mohsin, M et al., 2022.

12.4 Tetanus

(Lock Jaw,Wooden Horse disease)

1. **History:**

- The neurotoxin tetanospasmin causes tetanus. Numerous livestock animals are affected; however, certain species are more vulnerable than others are.
- According to historical accounts, Hippocrates first described tetanus in the fifth-century BC. Antonio Carle and Giorgio Rattone of the University of Turin identified the cause of tetanus in 1884.

2. **Causitive Agent:**

- Gram-positive bacteria called Clostridium Tetani

3. **Clinical Signs:**

- Between 103 and 105°F Hyperatresia
- Seizures
- Locked mouth, salivary drooling, prolapse of the third eyelid, elevated tail, erected ear, cramping sound, constipation, and maturation. (Bikhane et al.,1998)

4. **Diagnosis:**

- Drum Stick Visual
- Does not result in lesions
- ELISA
- Clinical indicators and history; • Radio Immuno Assay (RIA)

5. **Treatment:**

- 20,000–40,000 IU/Kg of pencillin by (I/M)
- 3 lakh IU/kg of Anti Tetanus Serum (ATS) given every 12 hours via (I/M, I/V, S/C)
- Chlorpromazine 0.88 mg/kg (I/V) or 0.22 mg/kg (I/M) • 30–50g chloral hydrate
- Multivitamins
- Phenvil injection; • Flunixin meglomine injection; • Diazepam

6. **Prevention:**

- Tetanus Toxicoid 5–10 ml (I/M) • Adequate Vaccination (vaccine developed in 1924)

- Wound Surveillance
- Cleanliness of the environment; •Awareness and education; •ATS 1500–3000 IU/kg prior to surgery performed on the animal. (Thwaites et al., 2020).

12.5 Brucellosis

- Many species of Brucella cause bacterial brucellosis, which mostly affects cattle, sheep, horses,pigs, and other animals.

1. **Etiology**

- Brucella melitensis and Brucella abortus

2. **Transmission**

- Colostrum, Milk; Placenta; Littering of the aborted fetus, contaminated feed, contaminated water

3. **Clinical Signs**

- Orchitis and infection of male accessory sex glands; abortion; stillbirth; retained placenta;fatality

4. **Diagnosis**

- History of abortions; PCR; ELISA, etc

5. **Treatment**

- Just detection and prevention are carried out; there is no particular treatment for camels

6.**Control**

- Control measures included vaccination, separating sick animals from healthy ones, culling sickanimals, and providing decontaminated water and feed for animals. (Corbel M. et al., 1997).

12.6 Hemorrhagic Septicemia (HS)

- Water buffalo, cattle, and bison are the primary hosts for hemorrhagic septicemia, an acute andextremely lethal type of pasteurellosis.

1. **History:**

- From 1974 to 1986, infectious infections that killed buffaloes and cattle, most frequently in India, were caused by HS. 2.17 billion Pakistani rupees were lost financially as a result of HS inPakistan in 1996.

2. **Causative agent:**

- P.multocida

3. Clinical signs:

- High fever; Severe nasal discharge and salivation; Severe throat oedema; Breathing difficulties; and Animal death within 1-2 days.
- Weak and fast pulse; pale mucous membrane and mucosal hemorrhage.(Abenayake et al.,1993).

4. Treatment:

IV oxytetracycline injection (10 mg/kg BW) for three to five days

- Ampicillin 10 mg/kg BW IM for five days
- Ceftiofure 10 mg/kg BW IM for 5 days; • Dexamethasone 1 mg/5 kg; • Enrofloxacin 2 mg/kg BW IM for 5 days.

5. Prevention:

- Keep sick animals apart from healthy ones; • Prevent crowding, particularly in the rainy seasons.
- Vaccinate every animal that is six months of age or older once a year.www.dairyknowledge.in/dkp/article/Haemorrhagic – septicemia

12.7 Enterotoxemia

- Caused by the bacteria Clostridium perfringens type D, enterotoxemia, also referred to as "overeating disease" or "pulpy kidney disease," is a dangerous and frequently fatal ailment thataffects cattle. Here's a thorough explanation:

1. Causative Agent:

- The bacteria Clostridium perfringens type D, which is frequently present in the intestines of healthy animals, is the main cause of enterotoxaemia. Certain situations, including abrupt dietarychanges or overindulging in high-carb meals, might cause the bacteria to multiply quickly and create toxic amounts of their products.

2. Mode of Transmission:

- Soil and excrement are common places to find Clostridium perfringens type D spores.

3. Risk Factors:

Several factors are important that cause enterotoxaemia in cattle, including:

- Sudden diet change: Suddenly changing feed from a high-forage to a high-concentrate diet allow Clostridium perfringens to proliferate by disrupt the bacterial balance in the intestines,

- Overeating: Eating fermentable carbohydrates in large quantities of, such as grain or lush pasture, can provide an ideal environment for Clostridium perfringens to live.
- Stress: Stress of transportation, weaning, or adverse weather conditions play an important role in weaken the immune system and increase chances of enterotoxaemia occurrence. (Niilo, L et al.,2003)

4. Clinical Signs

In cattle the clinical signs of enterotoxaemia vary, they mostly depend on the severity of the infection, but most important are:

- Sudden Death: In acute cases, affected animal may suddenly die without showing any clinical signs of disease.
- Depression and Lethargy: Animal effected with enterotoxaemia show signs of depression, lethargy, and weakness.
- Abdominal Pain and Distension: Animal may show signs of abdominal discomfort, including restlessness, groaning and stretching (Hussain, R., et al., (2022).
- Diarrhea: Diarrhea may range from mild to severe is a common clinical feature of enterotoxaemia and.
- Muscle Tremors or Convulsions: In some severe cases of this disease, affected animals may show signs of muscle tremors, twitching, or convulsions, especially if nervous system is affected by the toxin.
- Coma: In severe cases, Coma is important sign of this disease that lead to death of animal.

5. Treatment

Treatment of enterotoxaemia in animal typically include supportive care and administration of specific antibiotics therapies to counteract the effects of the bacterial toxins.

1. Fluid Therapy: To correct dehydration and electrolyte imbalances resulting from diarrhea andfluid loss fluid therapy is important.
2. Electrolyte Supplementation: Electrolyte supplementation along with fluid therapy, may beimportant to replace lost electrolytes such as sodium, chloride and potassium.
3. Antitoxin Administration: Antitoxins, can be given to neutralize circulating Clostridium perfringens type D toxins in the bloodstream which are antibodies against the toxins produced,by Clostridium perfringens type D
4. Antibiotic Therapy: Antibiotics may be given to control bacterial multiplication and to avoid secondary bacterial infections. However, antibiotics alone are not important to treat enterotoxaemia and should be used in combination with other supportive therapies.

5. Pain Management: To reduce abdominal pain and discomfort associated with this disease, Analgesics or pain-relieving medications may be given.
6. Isolation and Rest: To prevent further spread of the disease affected animal should be isolatedfrom the rest of the animals. Providing a quiet and stress-free environment can facilitate recovery.
7. Monitoring and Follow-Up: Close monitoring of an animal's condition is essential throughoutthe treatment process. Follow-up examinations by a veterinarian may be necessary to assess the response to treatment and to make any necessary adjustments.

Prevention through vaccination and good management practices are key to reducing the incidence of enterotoxaemia in cattle populations. (Hussain, R., et al., (2022).

12.8 FOOT And MOUTH DISEASE

History ◦

- FMD most likely started in 1514 when Fracastorius wrote about a related cattle illness in Italy(159).
- Loeffler and Frosch (271) proved that a filterable substance induced FMD nearly 400 years later,in 1897.

Clinical Signs

1. High fever and mouth saliva leakage
2. Tongue sores that resemble blisters
3. A lip sore that is present
4. Sores located on the hoof's coronary band
5. Production Losses (Aslam, M et al., 2023).

12.9 Etiology

An aptho virus belonging to the Picornaviridae family causes FMD. Seven strains of this virus have been identified. A "O" C Worldwide endemic are Sat 1, Sat 2, Sat 3, and Asia 1. Each strainrequires a different vaccine, and the majority of affected animals have clovens.

Transmission

- By direct touch ◦ By secretions from saliva ◦ By secretions from the nose
- Infections obtained from meat and milk as well of animals that are afflicted

Treatment

No particular FMD treatment

- It is possible to manage secondary bacterial infections using antibiotic therapy. Mouth lesions were cleaned with a 1% KMNO4 solution.

Control

1. Keeping sick animals apart
2. Cattle that get the adjuvant vaccination
3. Give sick animals soft food and fresh water. (Sáiz, M et al.,2002).

12.10 Rabies

1. **1.Common Names**

(Hydrophobia, Lyssa, Tollwat, Lerage, Mad Dog, Madness, Lytta, Habboo, Rabere&Jalatanka.)

2. **Clinical Signs**

- Abnormal Behavior
- Aggressions
- Excessive Slalivation
- Self-mutilation
- Paralysis
- Seizures
- Difficulty in swallowing
- Cerebral dysfunction
- Cranial nerves dysfunction
- Lethargy
- Fever (Kumar, H et al.,2023).

3. **History**

- It is one of the oldest diseases in history, with cases dating back 4000 years ago.
- According to the first written record, death occurred in dogs and nam occurred in Mosaic Esmua.
- Today, it is found worldwide.

4. **Causative agent**

- Rabies virus in the genus Lyssa virus in family Rhabdovaridea

5. Treatment

- Wash the wound with 20percent soft soap solution.
- Wash bite site using benzalkonium chloride.
- Use Anti rabies serum.

6. Control

Vaccination of wild animals

- Stray dogs' control
- Registration of dogs and cats. (Jackson, A. C. et al .,2014).

12.11 Paratuberculosis (Johne's Disease)

1. History:

Animals can contract paratuberculosis (PTB), also known as Johne's disease (JD), from Mycobacterium avium subsp. paratuberculosis (MAP). It is a chronic, infectious disease. The illness was initially identified in sheep in Bosnia in 1908 and was first described by Johne andFrothington in 1895.

2. Causitive Agent:

- The bacterial parasite that causes paratuberculosis is Mycobacterium avium subspeciesparatuberculosis.

3. Clinical Signs:

- loss of weight and diarrhea
- Under the jaw, soft swelling known as "bottle jaw" or intermandibular edema may develop.

4. Diagnosis:

- The presence of MAP in the feces is demonstrated by microscopy, culture, or the polymerasechain reaction (PCR) with DNA probes, confirming the clinical diagnosis of paratuberculosis.

5. Treatment:

- The animal will receive oral isoniazid at a dose of 20 mg/kg every 24 hours for the remainder ofits life

6. Prevention

- There is no known effective treatment. Good management and sanitation techniques that minimize the organism's exposure to young animals are necessary for control. Calves, youngsters, or lambs should be born in manure-free locations, taken out of the dam as soon as possible if they are dairy cattle, bottle-fed pasteurized colostrum or derived from dams that test negative, and raised as far away from adults and other animals as feasible Unless the waste milkhas been pasteurized, using milk replacer is advised in place of wasted milk. (Aitken JM et al., 2024).

1. **Botulism 1.Clinical Signs:**

- Difficulty in swallowing
- Weakness of muscles
- Dry mouth
- Drooping eyelids
- Respiratory failure
- Blurred vision
- Paralysis

2. **Causative agent:**

Clostridium botulinum bacterium, which produces botulinum toxin.

3. **History:**

- Botulism was first recognized in the late 18th century when it was linked to consumption of spoiled sausage. It has since been identified in various forms, including foodborne, wound, andinfant botulism.

4. **Treatment:**

- Supportive therapy
- Administration of antitoxin
- Mechanical ventilation for respiratory support

5. **Control**

- Prompt wound care and management
- Proper food preservation and hygiene practices
- Avoiding consumption of improperly processed foods. (Sobel J et al., 2005). "

Blackleg Disease

- Also known as black quarter, quarter ill, symptomatic anthrax, Emphysematous gangrene is ahighly fatal bacterial infection primarily affecting cattle and occasionally other ruminants likesheep and goats.

Causative Agent:

- Clostridium chauvoei is the bacterium responsible for causing blackleg. It is a spore-forming, anaerobic, Gram-positive bacterium commonly found in the soil and gastrointestinal tract of animals.

Clinical Signs:

- Blackleg typically manifests as sudden onset lameness, severe pain, and swelling in affected limbs (usually hind limbs). Other clinical signs may include fever, depression, loss of appetite,and rapid breathing.Swelling often feels crackly due to gas production in tissues, giving rise tothe name "gas gangrene."

Pathogenesis:

- The disease usually occurs when C. chauvoei spores are introduced into muscle tissue through wounds or punctures. These spores then germinate in the low-oxygen environment of damaged muscle, producing toxins that cause tissue necrosis and gas accumulation.Rapid multiplication of bacteria and gas production lead to tissue destruction and the characteristic swelling seen in affected limbs

Differential Diagnosis:

- Must be differentiated from anthrax , malignant edema and bacillary hemoglobinuria

Treatment:

- Pencillin Large dose 40,000IU/Kg BW
- Surgical debridement of the lesion including fasciotomy Newer antibiotic Cephaloridine(kefelong) may be tried

Control:

- It is best to keep the young animals away from such areas. Either burn the corpse or bury it.
- It is not appropriate to let the deceased body to skin
- It is not advisable to let the sheep and calf graze on endemic pasture. Every animal living in an endemic zone has to receive the appropriate

Vaccination:

- Black quarter vaccine polyvalent Dose: Cattle and buffalo 5ml

- Sheep and goat 2-3ml
- Black quarter vaccine (BAIF)
- Combined HS and Black quarter vaccine

Lumpy Skin Disease (LSD)History

- In 1929, an epidemic of lumpy skin disease was first observed in Zambia. It was formerly believed to be the consequence of poisoning or an extreme allergy to bug stings. Between 1943and 1945, there were more cases in South Africa, Zimbabwe, and Botswana.

Causative Agent

- The lumpy skin disease virus (LSDV), a member of the Poxviridae family and genus Capripoxvirus, is the causative agent of lumpy skin disease (LSD).

Clinical Signs

- The disease causes fever, depression, and the distinctive skin nodules in animals that areaffected.
- Clinical indicators consist of:
- Firm, elevated skin nodules that can measure up to 50 mm in diameter appear on the limbs, neck, genitalia, and head. Nodules can develop on the body in any location.
- Large holes that could get infected are left behind when the scabs that form in the center of the nodules fall off.
- Potential for genital, brisket, and limb swelling Reluctance to move and consumeocular and nasal secretions enlarged lymph nodes on the surface Reduction in milk yield Abortion. (Liang et al., 2022).

Diagnosis

- Fever, lacrimation, hypersalivation, and distinctive skin eruptions are examples of clinicalsymptoms. The diagnosis can be made by PCR, viral isolation, or histology.

Treatment

- The best method of control is vaccination as prevention as there is currently no therapy for thevirus.
- Antibiotics (topical +/- injectable) and non-steroidal anti-inflammatory drugs (NSAIDs) can beused to treat secondary skin infections when necessary.

Control

- The most effective defense is a well-planned, early preventive immunization program for alllivestock in high-risk locations.
- Cattle movements within the nation and across borders ought to be tightly regulated or outrightprohibited. A veterinary certificate that includes all information regarding the animals' provenance and assurances of their health should be presented with any livestock movements that are authorized.
- Cattle herds in impacted villages should, if at all feasible, be maintained apart from other herdsby refraining from communal grazing in order to protect animal welfare. But occasionally, the entire hamlet becomes a single epidemiological unit, in which case it becomes necessary to assess whether separation is feasible on a case-by-case basis.
- Animals that have received vaccinations may be permitted to travel within a country's restricted areas once it has been determined that A vaccine with demonstrated effectiveness has given fullimmunity (28 days after inoculation).
- Insect repellents should be applied to cattle on a regular basis to reduce the possibility of diseasevector transmission. Although it can't completely stop transmission, this approach can lessen thedanger. (Akther M et al., 2023).

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Chapter 13 :Malpractices of Veterinary Medicine in south Punjab-Pakistan

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Abstract

Malpractice in veterinary medicine refers to misdiagnosis, surgical mistakes, Medication errors, inadequate patient monitoring, failure to follow established protocols, poor communication with clients and Inadequate record-keeping posing serious threats to domestic animals at farm and local level resulting in harm to an animal patient. Non registered veterinarian, quacks, self-medication by animal lovers, false believes are major cause of malpractice. Failure to act in accordance with the standards of veterinary ethics is considered as professional negligence. Ethical values are imposed upon veterinarians by administrative government bodies that regulate various aspects of veterinary practice. Prevention is the best defense against malpractice. The physician should care for every patient with scrupulous attention to the requirements of good medical practice. The Purpose the study is to make aware the farmers, veterinarians, students and general public about the consequences of these malpractices. So that major loss can be prevented for the welfare of animal as well as to improve the socio-economic status in south province of Punjab-Pakistan.

Key words: Malpractice, veterinary, ethics, Medication, Animal

13.1 Introduction

Malpractice in veterinary medicine refers to negligence, error, or deviation from accepted standards of care by a veterinarian, resulting in harm to an animal patient. Examples of malpractice in veterinary medicine include Diagnostic errors, Surgical mistakes, Medication errors, Inadequate patient monitoring, Failure to follow established protocols, Poor communication with clients, Inadequate record-keeping, Failure to refer to specialists when necessary, Unnecessary procedures or tests, and failure to obtain informed consent. Proper client communication is a key aspect of veterinary practice, enhancing understanding and compliance with treatment plans (Pugliese et at., 2019). To help veterinarians fulfill their moral and professional responsibilities in their interactions with patients, clients, coworkers, and society at large, veterinary ethics is a collection of moral precepts derived from both professional and animal ethics. The protection of animal health, alleviation of animal suffering, preservation of animal resources, and advancement of public health are the cornerstones of veterinary ethics, as stated in the veterinary oath (Tannenbaum et al., 1993).Veterinary ethics is the application of moral principles, values, and judgments to the practice of veterinary medicine. History, philosophy, theology, sociology, and real-world application in veterinary clinics are all included in veterinary ethics (Vettical , 2018). Veterinarian ethics is a combination of professional veterinary ethics and animal ethics. It could be seen as a key illustration of the kind of care that veterinary professionals need to give in order to meet their duties to both people and animals. Many moral conundrums arise in the business unit of a veterinary practice. These kinds of issues can always give rise to moral dilemmas, and one must always make appropriate moral decisions only after giving them considerable thought. In the business unit's ethical decision-making process, veterinary professionals and paraprofessionals use ethical theories, principles, and rules to resolve ethical issues. Resolving ethical conundrums requires knowledge of and comprehension of ethical theories. Animal welfare and ethical concerns need to be carefully considered in a good ethical veterinary practice (Bernstein et al., 2013). The ethics and key steps involved in the ethical decision-making process in a veterinary practice business unit are reviewed and discussed in this article when faced with moral conundrums in a veterinary practice, moral stress levels can be reduced by a strong commitment to the role, the organization's commitment to ethics, and staff support. It's possible that applying ethical frameworks won't affect the choices that veterinary practitioners ultimately make. But maintaining a positive public image for oneself and the veterinary practice, as well as for client communication and job satisfaction, requires a solid grasp of ethical issues (Vettical , 2018). It also emphasizes how crucial moral judgment is to upholding a high standard of professionalism and how it affects animal welfare. In the southern part of Punjab province-Pakistan, along with the modernization of the veterinary profession, Veterinary malpractices are deteriorating this noble profession in terms of unethical actions by veterinarians, veterinary technicians, veterinary quacks, or veterinary clinics that result in harm or injury to animals. In addition to this misdiagnosis, Improper and unnecessary treatment, Medication errors, Surgical errors, Negligence in patient care, malpractice by owners, Failure to obtain informed consent, false beliefs, Breach of standard of care, unsanitary conditions, Overcharging or fraudulent billing, Self-medication and Failure to maintain accurate records are those factors which enhancing the malpractices and playing a negative role in the socio-economic status of that country. The livestock sector has a major share of the national GDP. As key stakeholders in the veterinary field, a crucial role in preventing malpractice is required. By staying current with continuing education, Following the established protocols and guidelines, communicating effectively with clients, maintaining accurate records, practicing evidence-based medicine, referring to specialists when necessary, and obtaining informed consent, you can prioritize patient care and welfare and contribute significantly to preventing malpractice (Schneider, 2009). The consequences of malpractice in veterinary medicine are severe and far-reaching. They include Animal suffering or death, Legal liability for the veterinarian, financial losses for the client, Damage to the veterinarian's reputation, Loss of trust in the veterinary profession, Regulatory disciplinary action, Increased insurance costs, and Emotional distress for the client and veterinarian. The study aims to make farmers, veterinarians, students, and the general public aware of these consequences. By doing so, we aim to prevent major losses for the welfare of animals and to improve the socio-economic status (Krauss et al., 2021).

13.2 Malpractice by veterinarians

13.2.1 Fail to diagnose

Due to advances in preventive and specialized veterinary medicine, as well as increased client awareness of the advantages of the human-animal bond, veterinarians are more vulnerable to malpractice claims today (Quartarone et al., 2011). Veterinarians frequently lead the way in ethical issues striking a balance between the needs of society, employers, clients, animal welfare and animal health. A primary reason behind a physician's inability to identify a set of symptoms associated with a specific disease or serious condition is the absence of diagnostic tools, such as X-ray machines, ultrasound machines, diagnostic test laboratories, and chemical reagents meant to run the tests. At the hospital and district levels, these instruments are not working in veterinary hospitals or diagnostic labs. Furthermore, the results of these tests go beyond the means of a local farmer.

13.2.2 Wrong medicine

A physician may prescribe the wrong medicine for an animal due to a wrong diagnosis. This medicine may prove fatal for the affected animal and cause antibiotic resistance (Quartarone et al., 2011). Furthermore, antibiotic residues appear in milk and meat used for human consumption. These residues because serious complications in the human body, especially newborn babies, mostly nourished on powdered and packed milk.

13.2.3 Surgical errors

As mentioned earlier, no diagnostic tools are available for ultrasound, x-rays, endoscopes, etc. In veterinary practice as compared to the medical field. Veterinary surgeons may go through wrong surgical interventions. For example, in the case of repeated ruminal tympani, the physician may suspect a foreign body. While performing the laminotomy, the surgeon found nothing inside the rumen. So, veterinarians observe such surgical and technical faults. Mishandling in surgery animals by veterinary doctors can have serious consequences, including Surgical site infections, Internal injuries or organ damage, Anesthesia complications, postoperative pain or discomfort, Incomplete or failed procedures, Unnecessary amputations, Surgical instruments left behind in the animal's body, Incorrect surgical technique, Failure to close surgical sites properly (Quartarone et al., 2011).

Inadequate post-operative care. Mishandling in surgery include poor surgical techniques, Insufficient technical skills, Negligence to follow up, running through procedures, failing to monitor instituted protocols, ignoring safety guidelines, using outdated or poorly maintained equipment, Insufficient patient preparation, failing to observe anesthesia properly, Ignoring patient vital signs during surgery. Such mishandling can lead to Animal suffering and death, Legal consequences for the veterinarian, financial losses for the owner, Damage to the veterinarian's reputation and Loss of faith and trust in the veterinary career are major setbacks Veterinary doctors need to follow best practices, stay updated on the latest techniques, and prioritize patient care to prevent mishandling in surgery (Quartarone et al., 2011)

13.2.4 Negligence

Failure to act in harmony and accordance with the principles of veterinary ethics is considered as professional negligence. There may be one or more perfectly proper standards, and if he confirms with one of these proper standards, then he is not negligent. For example veterinary doctor at civil veterinary dispensary or hospital like to rely on their veterinary assistant. He did not bother to attend the sick animal.in this way, they show professional negligence as veterinary assistant have zero knowledge about the standards.Veterinary negligence can encompass a failure to diagnose, incorrect treatment, and failure to caution a patient of notorious risks. In cases of relayed responsibility or direct corporate negligence, privileges may also be brought against hospitals, clinics, managed care organizations or medical Corporations for the mistakes of their employees (Quartarone et al., 2011).Professional slackness and negligence is the prevalent theory of liability regarding allegations of veterinary malpractice. A person who alleges negligent medical malpractice must prove all four elements of the tort of negligence; a physician/veterinarian-patient/animal owner relationship existed, so a duty of care was owed by the physician/ veterinarian, the physician/veterinarian violated the applicable standard of care, giving proof of negligence, generally confirmed by expert testimony or understandable errors, the physician's negligence caused the injury; and the person/animal suffered a compensable injury, meaning that the injury led to precise damages. The burden of proving these origins is on the plaintiff; the healthcare worker is the defendant (Meschievitz, 1991)

13.2.5 No follow up

Follow-up care involves regular medical checkups, including a physical exam, blood tests, and imaging tests. However, the veterinary profession has many concerns because no patient-admit facility is available.

13.2.6 Lack of knowledge

Lack of awareness among veterinarians on well-judged antibiotics and their use and lack of antibiotic safekeeping are the significant factors that need attention to combat the rising AMR in the veterinary sector in India It's essential for veterinarians to follow to ethical standards and unsurpassed practices to prevent malpractice and ensure the well-being of animals in their care. If you suspect veterinary malpractice, report it to the relevant regulatory establishments (Geleris et al., 2011)

13.3 Malpractice by quacks

13.3.1 None registered

Due to poor administration, many quacks are working in the field in south Punjab. Concerned government departments failed to stop these quacks from practicing. A lot of animals are affected by their wrong practice (Bernstein et al., 2013). They are using high potency and broad-spectrum antibiotics from the start. Due to this, antibiotic resistance is produced in animals. This further leads to the death of animals and the financial loss of farmers. In addition to this, quacks badly damage the local breeds of cattle and buffalo by using local and unregistered semen. This is a great economic loss for animal care countries in terms of quality milk and meat (Abdu et at., 2006).

Malpractice by non-registered veterinarians is a serious concern in the animal healthcare industry. Non-registered veterinarians, also known as unlicensed or unqualified veterinarians, may not have the necessary education, training, or experience to provide adequate animal care. This can lead to misdiagnosis, improper treatment, and even harm or death to animals. Some common examples of malpractice by non-registered veterinarians include improper administration of medications or vaccinations, inadequate surgical procedures, Failure to diagnose or misdiagnose diseases, Neglect or abandonment of animals, and Unhygienic or unsanitary conditions in clinics or facilities (Abdu PA et at., 2006). Animal owners must confirm their veterinarian is enumerated and licensed to practice in their domain and jurisdiction. It is essential to check with your area's relevant veterinary regulatory association to verify a veterinarian's registration and essential credentials. If you suspect malpractice by a non-registered veterinarian, you should report it immediately to the proper authorities, such as the local veterinary governing body or animal welfare association.

13.3.2 False tags

Moreover, many veterinary workers, especially veterinary assistants and Artificial insemination technicians, are adding fuel to the fire with false tags as full veterinary doctors. They are also a bone of contention in animal welfare and pose serious threats to animals and farmers. Malpractices by quacks in the veterinary field can have serious consequences for animals and their owners. Quacks in the veterinary field claim to be veterinarians but do not have the necessary qualifications, training, or experience (Lesperance et al., 1993). Here are some common malpractices by quacks in the veterinary field: Fake vaccinations and certificates, Unsterilized equipment and poor hygiene, Misdiagnosis and improper treatment, Administration of fake or expired medications, Unqualified surgery and dental procedure, Lack of anesthesia or improper use of anesthesia, Ignoring animal welfare and neglecting animal care, Overcharging for services not rendered, using unapproved and untested treatments, Misrepresenting qualifications and experience. These malpractices further lead to Animal suffering and death, Spread of diseases, financial losses for animal owners, and Erosion of trust in the veterinary profession (Centner et al., 2011).

13.3.3 Malpractice by owners.

Malpractice by owners is also a burning issue in south Punjab. The majority of farmers have two or three animals and consider themselves doctors. They start to treat and self-medicate their animals without the advice of registered veterinarians. Social media or YouTube channels play a critical role in this regard. Owners make misuse of social media. Over/underdoes of drugs, wrong injection site, and wrong medicine are common issues that arise from malpractice by the owner (Huss RJ, 2003).

Malpractice by owners in the veterinary field refers to negligence or wrongdoing by animal owners that can harm their animals or compromise their care (Centner et al., 2011). Here are some examples of malpractice by owners: Neglect or abandonment, Failure to provide satisfactory food, water, or shelter, Ignoring medical conditions or failing to seek veterinary care, Managing medications or treatments without veterinary guidance, Improper restraint or handling, leading to injury or stress, Breeding animals without proper knowledge or care, Keeping animals in unsanitary or inhumane conditions, Lacking to provide necessary vaccinations or preventatives like heartworm medication, over-feeding or underfeeding, leading to obesity or malnutrition, Denying animals necessary surgery or medical procedures. Such malpractices can result in Animal suffering and death, Legal consequences for the owner, financial losses for veterinary care or legal fees, and emotional distress for the owner and others involved (Lesperance RJ et al., 1993). In addition, false beliefs are another major drawback of veterinary practice adopted by the owner. For example, they do not offer colostrum to the newborn calf till the calf starts sucking itself from the dam and till the expulsion of the placenta from the dam. They avoid the correct medication with the view that this medicine is highly metabolic (gram/hot) and may harm their animal or cause abortion in their animals. So, they spend much money on local herbal medicine, mustered oil, and desi ghee compared to cost-effective medicine Furthermore, prolonged anestrus period of up to one and half years due to ovarian cystic disorders is another drastic issue and has caused severe productive loss in the milking animal and dairy industry as well. Farmers at the local and farm levels tend to use costly local herbal substances rather than the proper drug of choice (Krauss et al., 2021).

13.4 Malpractice reasons

13.4.1 Non-implication of govt laws

Numerous factors support the practice, chief among them being that the veterinary profession defines the veterinarian's duty. Based on the most recent veterinary knowledge and clinical experience, the veterinarian should behave as a knowledgeable and conscientious member of the veterinary profession. The veterinary profession, not any one veterinarian, sets the medical professional standard (Pugliese et at., 2019).

13.4.2 Lack of awareness

Awareness about the veterinary profession is negligible in the southern region of Punjab. The south area is very underdeveloped and administratively poor. Most animal lovers and farmers rely on local quacks instead of competitive practitioners. Farmers, as well as educated groups, remain unaware of the veterinary profession. They are even unaware of animal surgeries, ultrasonography, stomach tube passing, and Intravenous infusions.

13.4.3 Mis-use of Telemedicine & social media

The trend of telemedicine is increasing in the southern region and around the globe. The people of the southern region, as well as the whole of Pakistan, misuse telemedicine and social media. The majority of veterinarians post prescriptions and treatment protocols on social media and explain via some telephonic source. Animal owners and quacks misuse the prescription. In this way, animal and pet lovers do not call for a doctor, which leads to complications in their animals.

13.4.4 Actions to stop malpractice

It is primarily the duty of veterinarians to alleviate the suffering and anguish that animals endure from illnesses, disabilities, mistreatment, starvation, violations of their rights as animals, etc. To ensure the health and welfare of their patients, as well as the public's safety, veterinarians must provide veterinary services that are both adequate and appropriate. Veterinarians who provide veterinary services or care must adhere to recognized professional standards (Geleris et al., 2011). During the provision of services, they should not allow personal gain or aggrandizement to cloud their judgment; instead, they should be guided by the needs of the patients as well as the public's health and safety (Abdu et al., 2006).

13.4.5 Role of Local Administration

Administrative government bodies also impose ethical values on veterinarians that regulate various aspects of veterinary practice. Such ethical standards are especially important because they have the force of law. If they are disobeyed, punishment or some deprivation of or restriction upon one's ability to practice can result. The state veterinary licensing boards, along with the practice acts and regulations that these boards oversee, are the primary sources of administrative veterinary ethical standards. Virtually every state has on its books practice act provisions or regulations requiring various kinds of ethical behavior toward patients, clients, the general public, and other veterinarians (Geleris et al., 2011). Because they are the only medical professionals tasked with safeguarding both human and animal health veterinarians are in a unique position. In addition to providing for the medical requirements of various animal species, they also have environmental protection obligations to fulfill. (Meschievitz, 1991).

13.4.6 Role of State Laws

The liability for veterinary malpractice is determined by state law, and in most states, recovery for veterinary malpractice is limited to the animal's fair market value. This approach fails to acknowledge the emotional value animals have for their owners. Awareness creation through workshops and seminars is needed to educate animal owners about cooperating with veterinarians in providing care for their animals. Owners should accept veterinarians' opinions in managing their animals' medical care, regardless of emotional attachment or potential economic gains (Meschievitz, 1991).

13.4.7 Role of Veterinarians and Healthcare Workers

The best defense against malpractice is prevention. Every patient should receive the utmost care from doctors, who should also be aware of their legal obligations to the patient and follow the guidelines of good medical practice. They have to abstain from disparaging and unethically criticizing the work of other medical professionals (Pugliese et at., 2019). Every case should have "ideal" medical records that could be presented in court, clearly documenting actions taken and when they were completed, proving that nothing was overlooked, and proving that the treatment received complied with legal requirements.

The record should indicate any early treatment terminations or noncompliance with instructions, such as by attaching a hard copy of the letter cautioning against the unwise course of action (Hell, 2011). Any statement that might be taken as an admission of guilt should be avoided by doctors. An agent or physician employee acting in the course and scope of their employment may make such an admission, which could be used against the doctor. It could also be made to the patient and to a third party prior to the trial. For the purpose of preventing and controlling various zoonotic diseases, there should be a strong connection between human medical professionals and veterinarians to report disease patterns and trends to state public health and regulatory agencies (Tannenbaum et al., 1993). They also support the objectives and results of public health by offering guidance on animal health matters to regional health boards and commissions. The contributions made by veterinarians to public health are divided into six main areas. Veterinary devices, feeds, and medications for animals are governed by the Center for Veterinary Medicine. Biologics and animal vaccinations are regulated by the USDA; animal flea and tick products are regulated by the FDA; and other products are regulated by the EPA.

13.4.8 Role of Pakistan Veterinary Medical Council (PVMC)

A veterinarian should carefully treat the animals without any negligence. He should be capable of treating the animals with skills, experience, and attainments. If a veterinarian does not follow the standard of pharmacological principles, shall be considered to have committed a willful act of negligence. If a veterinarian is deficient in moral character, education, and skills shall not be allowed to treat the animals. A veterinarian should enlighten the public about animal disease control, zoonoses, toxicity, and various forms of diseases. A veterinarian should focus on good veterinary practice, awareness, and public engagement. They must refrain from negative, destructive, and unprofessional criticism of other veterinarians. A complete medical record of cases should be kept for vetro legal cases. If a patient does not follow the case treatment protocols, the record should reflect this. A veterinarian should make statements very carefully to prevent them from any inconvenience in future.

Veterinarians should report disease events and outbreaks to public health agencies to prevent and control zoonotic diseases. They should also engage local public health boards to prevent and control disease outbreaks. Veterinary public health protocols are divided into certain domains; The Center for Veterinary Medicine (CVM) regulates animal drugs, feeds and veterinary devices, The United States Drug Authority (USDA) regulates animal vaccines and biologics, The Food and Drug Authority (FDA) regulates certain flea and tick products for animals and The EPA regulates various other protocols. A veterinarian shall treat cases requiring his advice or services. He should make decisions on the basis of complete case history, clinical and laboratory examination, and recommended medications (PVMC Act, 1996).

13.4.9 Use of Telemedicine and role of clients

Telemedicine"-the utilization of technological devices such as telephones, facsimile machines, and the internet as tools to create a professional support network-is another advancement that has contributed to the increased standard of care now being demanded of the veterinary medical profession. (MJ et al.1990). Trans telephonic electrocardiography has been available for more than two decades, with IDEXX Cardio Pet being the innovator in helping veterinarians manage the many new developments in cardiology. Recently, competitors such as the Veterinary Heart Institute have offered, in addition to trans-telephonic electrocardiography, advanced services such as color Doppler echocardiography, radiograph interpretation, and specialist consultation in other areas. The advantages telemedicine offers to the practicing veterinarian are clear. First, the income from diagnostic studies remains with the attending veterinarian instead of going to a specialist. Second, telemedicine offers the convenience of support from a board-certified specialist, thereby enhancing the quality of care for both veterinarians and clients. As a result, Clients should Research and choose a reputable veterinarian, ask questions and understand treatment plans, monitor their animal's condition and report concerns, request a second opinion if necessary, and Report suspected malpractice to regulatory authorities. Medical malpractice occurs when a patient suffers harm, injury, loss or damage to function by a physician, veterinarian, dentist, pharmacist, therapist or any other medical care provider who fails to competently perform his or her medical duties by providing improper, unskilled (Quartarone et al., 2011).

13.5 Conclusion

Malpractice is a real threat to all healing professions, with doctors of medicine being the primary targets. Ethical and moral issues are common in veterinary practice. Education in ethics during veterinary training can help improve ethical decision-making and compassion among veterinary professionals. Establishing good communication in the veterinary-client relationship is crucial when dealing with serious welfare matters. The ethical decision-making process requires thoughtful consideration of facts, alternative perspectives, consequences, and ethical principles. Commitment to ethical values, organizational support, and strong veterinary law knowledge can help lower moral stress when facing ethical dilemmas. Further research on veterinary practice-related ethical issues can enhance veterinary education. Understanding professional ethics is essential for practitioners to comply with legal requirements. Veterinary jurisprudence reflects ethical values central to the veterinary profession. This book chapter argues that paying serious attention to legal obligations is the most effective way for practitioners to gain a general understanding of their professional responsibilities. Professional ethics as Veterinary ethics describes the search for "correct" norms or professional behaviors and attitudes in veterinary practice.

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Chapter: 14 Insects as a Sustainable Protein Source for Poultry: A Comprehensive Guide

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Abstract

The use of alternative protein sources has become an emerging interest including insect meal used by poultry nutritionists to ensure poultry protein requirements are met given the growing concern over sustainable protein sources in poultry such as fish and soybean meal. Common insects used for the production of insect meal include Mealworms, house crickets, and black soldier flies. The nutrients included in the meal vary based on the type of insect the substrate it is raised on and the method of production. An in-depth description of insect meal has become a novel type of concentrated protein for poultry diets is given in this chapter along with information on its benefits drawbacks and potential applications. Insect meal is a beneficial feed item especially for chicken production as it is demonstrated to be a rich protein source, minerals (calcium, phosphorus, zinc), and amino acids (lysine, methionine). It also has a high rate of digestion. Insect meals could also lower manufacturing costs and the industry's environmental effects when used in poultry feed. Moreover, the development performance and meat quality of poultry species may be enhanced by the usage of insect meal. However several issues need to be resolved regarding consumer acceptability, legal regulatory frameworks, and large-scale insect production. Subsequent investigations and advancements may aid in surmounting these obstacles and augmenting the insect's acceptance as a prospective protein source in poultry feed.

14.1 Introduction

The human population has increased rapidly over the years and by the end of the current century it might have reached 12.3 Billion (Randive et al., 2021). According to Roser's (2023) report, the population of the world is expected to be more than 10 Billion people by 2050 (Roser, 2023). This emphasizes how vital the food production industry will be to ensuring the security of food and nutrition, especially in the production of animal protein. It takes a significant amount of high-quality food to meet the needs of such a huge population. Meat and eggs from poultry may meet people's demands for protein and they are reasonably priced food sources with a quick turnaround time. As a result, there will always be a need for enough poultry feed to meet human food requirements. Poultry meat has become a more tasty source of animal protein and is easily accessible to thousands of people around the world (Asghar et al., 2022).

Over the years, the use of insect meal in poultry diets has become an increasing trend as one of the sources of protein. Here for the first time, the researchers have tried out sustainable poultry production by use of fresh insects and it is thought to increase the profitability of poultry production as feed costs can be minimized (Khan et al., 2013). Given they are a source of nutrients which is almost similar to other conventional animal protein sources and have a positive environmental footprint compared to conventional animal protein sources insects prove more suitable for human consumption than the issues they present (Van Huis, 2022).

Nowadays, poultry has a critical role in the economics of many developing nations and provides a living for a large number of people particularly in rural regions (Demeke, 2004). Humans obtain animal protein mostly from poultry products consumption like eggs and meat (Wu et al., 2022). Insect meal is thought to present a viable substitute protein source for feed. Additionally, certain challenges including consumer acceptability and regulatory restrictions must be solved. Nonetheless, several nations, such as the European Union and Canada have already approved the production of sustainable animal feed through the use of insect culture (Valdes et al., 2022).

Since chicken is currently the most popular white meat it is expected that utilization of poultry meat in particular will continue to rise over the next ten years (Henchion et al., 2021). Poultry farming has a comparatively low environmental impact due to its lower feed conversion ratio (FCR) and lack of enteric fermentation compared to other meat production methods (Gerber et al., 2007). Raising poultry has advantages such as its rapid growth rate and economical use of resources, but production costs are rising. There are several reasons for the high expenses associated with raising chickens, but first and foremost is significant contribution to the total cost of production is the price of feed (Asghar et al., 2021). The cost of soybean production and processing as well as its worldwide supply and demand are the main causes of the high price of chicken feed (Adeniji, 2007).

Another area that depicts the problem of acquiring feed ingredients has been the interruption of readily available and cheap ingredients like cereals, soybean, and fish meals due to competition between the food, feed, and fuel industries not to mention the effects of climatic changes. As affirmed by (Mugwanya et al., 2022) this has in effect, reduced accessibility while also subjecting feed resource costs to a high level of fluctuation. The availability of its traditional feed resources has been stretched over the past 10 years and hence high feed prices consequently the demand for finding animal protein substitutes for chickens. The insects used in feeding farmed animals are promising because of their nutritional significance and the benefit of environmental sustainability connected with this formula of farming (Van Huis et al., 2020). Therefore, for the insect meal to produce desirable qualities in the feed formulated, the quality of by-products and the substrate used as an insect meal are rarely defined.

14.2 Insect Meal is a Sustainable Source of Nutrition in Poultry Diets

The current trend for intensive poultry production has high requirements for new feed components that can support the feed needs in the future years due to high projections in the consumers' demand for poultry products (Lähteenmäki-Uutela et al., 2018). Insects can be consumed as protein sources as they are nutrient-dense particularly, fat and some essential amino acids. Also worthy of notice is that, despite coming from the same species of insects the assembling of protein contents in insect meals can significantly differ by 40–60% (De Marco et al., 2015). Even among closely related insect taxa, there can be differences in the nutritional profiles due to factors like developmental stage, environmental conditions, and dietary habits which include the particular foods eaten in their ecological niche (ideally natural eating patterns and how these factors impact the insects' total nutritional profiles and nutrient levels) (Salomone et al., 2017).

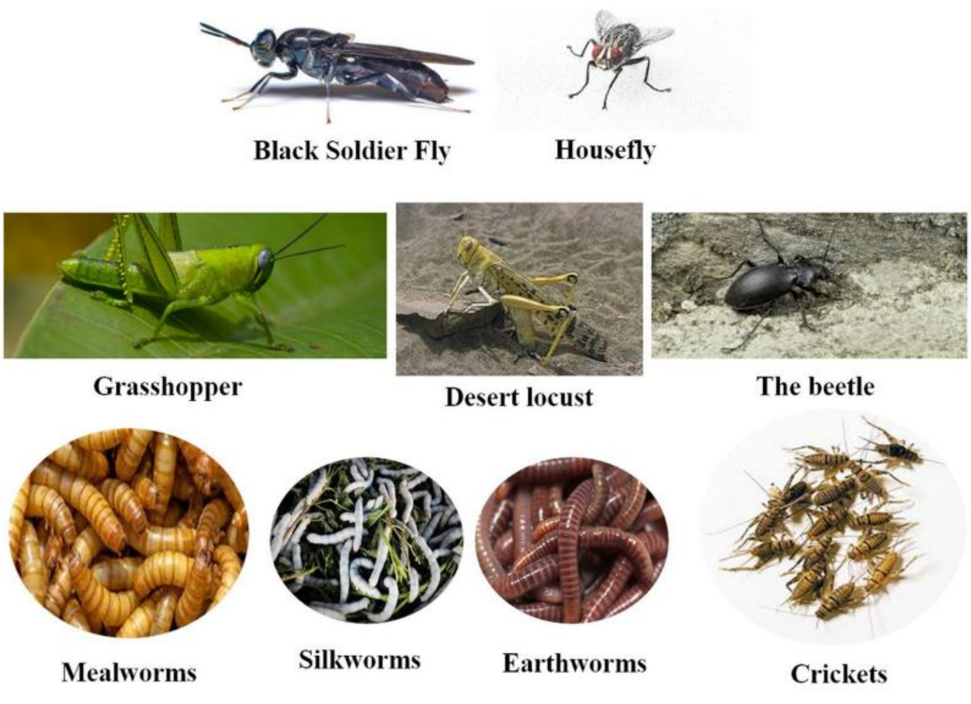

Figure 35: Insects Used in Poultry Feeding.

14.2.1 Black soldier fly (BSF)

The *Stratiomyidae* flies family includes the widespread and common black soldier fly (BSF). BSF meal is an innately healthy, high-protein, high-calcium food. The crude fat (CF) and content protein (CP) content of BSF meal vary from 35 to 61% and 7 to 42%, respectively. There have also been reports of high LA and PA contents (Moula et al., 2018). The concentration of the essential amino acids; lysine, methionine, valine, and threonine were observed to be between 0.34 to 3.30%, 0.08 to 0.90%, 0.33 to 3.38% and 0.22 to 2.26% respectively (Mwaniki, Kimani & Wambui, 2018). These nutrients should have concentrations of 0.34 to 3.38% (Neumann et al., 2018). The percentage of nutrients varies while the contents of calcium and phosphorus range from 0.74 (Liu et al., 2021) 0.95% (Mwaniki et al., 2018),

and 1.21% to 4.39% (Chu et al., 2020). The high substrate, the maturity of harvest, and the processing technique are key factors that determine what nutrient value is in the BSF meal. The fat content may decrease as a result of the pressing and defatting processes from 0.33 to 3.38%, 37% (original) to 27.36% (steamed), 17.95% (normal pressing) or 13.05% with hot pressing, or 17.05% (methanol extracted) (Astuti et al., 2018).

Figure 36 Black Soldier Fly (BSF)

Table 17 Black soldier fly larvae supplementation's effect on poultry diets (as a percentage of total feed).

Broilers	0, 5, 10, 15, and 20%	A 10% addition can add more performance traits that could increase the performance level.	(De Souza-Vilela et al., 2019)
Broilers	75 and 100%	There are no special variations in the performance traits.	(Benzertiha et al., 2019)
Broiler (Cobb)	5, 7.5, and 10%	Enhanced performance traits and traits of meat	(Dahiru et al., 2016)
Ross 308 broiler	20%	Improved traits of meat quality	(Vilela et al., 2021)

Ross 708 broiler	5%	Cecal microbiota and enhanced performance qualities	(Biasato et al., 2020)
Laying hens (Hy-line brown)		Enhanced blood biochemistry characteristics and performance	(Chu et al., 2020)
Laying hens (White Leghorn)	5 and 7.5%	Enhanced functionality and qualities of egg quality	(Mawaniki et al., 2018)
Growing quails	10–15%	No variations in carcass features or performance were found.	(Jayanegara et al., 2015

14.2.2 Housefly (HF)

All countries are home to houseflies (HF), which are raised on food waste and animal dung. Significant levels of crude fiber and crude protein range from (2.5 to 28% and 40 to 64%) respectively present in HF meal (Table 2.1). HF larvae age, the content of CP drops while the content of CF increases (Aniebo et al., 2008). The two most restricting amino acids for poultry nutrition are methionine and lysine. High concentrations of these two amino acids can be found in HF meal (Hall et al., 2018). It's interesting to note that FM and HF meals have similar amino acid profiles. Like BSF food, the insect's processing technique affects the meal's nutritional content (Aniebo & Owen, 2010).

Figure 37 House Fly

Table 18 Effects of adding housefly meal to poultry diets.

Bird Line	Rate of replacement (%)	Findings	Sources
Anak broilers	20 and 40% of fish meal	Improved performance traits and meat quality	(Okah & Onuujiariri, 2012)
Broilers	60% from soybean meal	Improved performance and carcass traits	(Khan et al.,2018)
Ross 308 male broilers	4% from from total feed	Improved performance traits	(Elahi et al., 2020)
Ross 308 male broilers	10% from from total feed	Improved growth performance, carcass, and meat quality traits	(Pieterse et al., 2014)

14.2.3 Mealworms (MW)

MW is a beetle that is found in any place where there exist warm and moist areas that are hiding. The MW contained a CP quantity from 27 to 54% and the contents of CF from 4 to 34% and therefore classified as a healthy food fat and protein source. Migiro et al. (2016) noted that the energy, ash, and crude fibers content of MW in larvae ranged from 1378 to 4029.63 Kcal/Kg DM (De Marco et al., 2015), 3.0 to 4.5%,(Hall et al., 2018) and 5.0 to 8.8% (Hall et al., 2018) respectively. MW larvae had higher Crude protein, Crude Fibre, and ADF than SBM larvae (Bovera et al., 2015). In comparison to the control, SBMs had a significantly higher content of (methionine+cysteine, arginine, valine, lysine, leucine, isoleucine, histidine, and threonine) although MWs had significantly higher tryptophan (Bovert et al., 2015). Further, MW larvae contained high K and P but low Ca; the concentration values in hundred milligrams per kilogram were Ca: 434.59 mg/kg, K: 9479.73 mg/kg, and P: 7060.70mg/kg. Hence it is suggested to apply calcium since the Ca/P ratio is not recommended for poultry production, especially hens (Makkar et al., 2014). In addition to these minerals, there are also enrolled probe concentrations of Cu, Zn, and Fe in MW (13.27, 104.28, and 66.87 mg/kg) respectively (Ravzanaadii et al., 2012).

Figure 38 Mealworms

Table 19 Effects of Supplementing Poultry Diets with Mealworm

Broilers (Ross 308 male)	4% from from total feed	Improved performance traits at starter phase	(Elahi et al., 2022)
Broilers (Hubbard hybrid free-range)	7.5% from from total feed	Improved fat profile of meat	(Dabbou et al.,2020)
Broilers (Ross 708 male)	15% from from total feed	Improved performance traits	(Biasato et al., 2018)
Broilers	100% from soybean oil	Improved breast meat's fat composition	(Benzertiha et al., 2020)

14.2.4 Crickets and grasshoppers

As for poultry, people can eat grasshoppers and crickets which are also rich in proteins. It stated from Table 1 that the range of percent for CP and CF is from 48% to 65% and 3% to 21%, respectively. Of course, the contents varied between Chinese grasshoppers (*Acrida cinerea*), African grasshoppers (*Acanthacris ruficornis*), wild edible grasshoppers (*Ruspolia nitidual*), desert locusts (*Schistocerca gregaria*), and short-horned grasshoppers (*Oxya hyla hyla*) (Amobi et al., 2020). The crude fiber content has a variation of 1.06 to 9.21%. Sun and his colleagues ascertained that ME values varied between 3923 and 4018 Kcal/Kg (Sun et al., 2012) while GE values were 1618 and 1917 Kcal/Kg (Ojewola et al., 2005). Concerning the amino acid (AA) content, it was found to be very close to that of FM's. The quantitative ratios of cysteine, methionine, and lysine were determined at (0.69, 1.70, and 3.79%) of DM, respectively (Wang et al., 2007).

Figure 39 Grasshoppers and Crickets

14.2.5 Earthworms (EW)

When it comes to calories, protein, and amino acids, earthworms (EW) and crickets are excellent sources (Janković et al., 2020). The meal obtained from EW has a wide range of crude fiber content ranging from 3.5 to 18% and crude protein content ranging from 41 to 66%. Methionine and lysine levels were reported to be significant in EWs (Finke, 2002). It is crucial to note that the freshness and dryness of the EWs affect these values.

14.3 Insect-Based Feeds and their Digestibility in Poultry Diets

Although the usability of insect protein in poultry feeds has been growing in popularity, little is known regarding the safety and ease of assimilation of these compounds in the diets of birds. There is a knowledge gap about the digestibility of certain nutrients, especially in insects used in livestock production. Insect meals are known to enhance the digestible amino acid content in broiler diets as well as the apparent metabolizable energy source (De Marco et al., 2015). In the pig study, the estimated fecal digestibility of crude protein in diets with larvae meal and soybean meal some difference but were close (76.0% and 77.2%) respectively (Newton et al., 1977). However, the high crude fat digestibility of the larval meal diet was higher 83.6% than the soybean meal diet of 73.0%.

The analysis shows that nearly 95% of crude fats are ingested by the pigs when fed on insect-based diets, particularly those containing black soldier fly larvae. According to the studies by (Gasco et al., 2019) various qualities like the insect's type, the extent of the inclusion, and the processing methods that can facilitate how insects can be processed into meals for animals will also influence its ease in breaking down into meals for animal feed. In addition, the outer layer of the insect is hard and stiff and is called the exoskeleton and it has chitin, which may hinder the digestive processes of nutrients. It is possible to use black soldier fly larval meal as a replacement for meat meal and maintain fecal quality and nutrient digestibility for dogs without compromising nutrient quality (Abd El-Hack et al., 2020). It is for this reason that insect meal may go a long way in helping to replace some ingredients in the making of animal feeds. As an alternative to current ingredients in canine and feline diets, insect-based protein sources could improve overall health and nutrient digestibility (Reilly et al., 2022).

Feeding insect powder into quails mopane worms (Gonimbrasia belina) has shown hope in the conversion of traditional animal protein and reducing reliance on fish and soya meal, which form conventional protein sources. Is a highly nutritious feed ingredient Mopane worm is mostly rich in protein, which may averagely hit 55% balance and a correct proportion of the amino acids (Mnisi et al., 2022). In an experiment, the supplementation of broiler live weight, total feed consumption, and relative growth rate with house fly larval meal was significantly enhanced (Pretroius et al., 2011).

Studies on the portion of house fly larval meal have indicated that it can serve as a substitute for other protein sources and enhance broiler performance in the same way as other feeds without adversely affecting the carcass characteristics. Another study looked at adding house crickets and mulberry silkworm pupae to adult mixed-breed dogs' diets as a poultry meal supplement. Therefore, supplementing the meals of pets with insects can add a good replacement for protein sources that are conventional in their diet (Areerat et al., 2021). Several studies conducted in the recent past have shown that insect meals are well suited for efficient nutritional digestion in many animal species as is evident even though the existing evidence and literature on chicken on the effect of replacing insect-based feeds in chicken diets is still scarce. Thus, insect meals can be seen as promising substitute components in formulas for animal feed kategor-American Institute of Biological Sciences (El-Sabrout et al., 2023).

Table 20 Apparent digestibility of certain insect diets across the whole tract (%).

Substrate	**Digestibility (%)**						**Sources**
	OM	**DM**	**N**	**CP**	**EE**	**GE**	
Insect meal							
BSF Larvae	84.3	53	89.7	51	88	69	(De Marco et al., 2015)
Mealworm	66–91.5	60	91.3	60	99	64	(Bosch et al., 2014)
HF Larvae	-	81	-	69–98	94	77	(Pretorius, 2011)
HF Pupae	-	83	-	79	98	-	(Pretorius, 2011)
House	88	-	91.7	-	-	-	(Bosch et al.,

cricket							2014)
Reference							
SBM	80.6	-	94.7	98	-	-	(Bosch et al., 2014)

OM, organic matter; DM, dry matter; N, nitrogen; CP, crude protein; EE, ether extract; GE, Goss energy; BSF, black soldier fly; HF, housefly; SBM, soybean meal

14.4 Detailed Guidelines and Safety Standards for Using Insects as Animal Feed

The safety of novel feed sources must be guaranteed to protect animal health and performance, which in turn protects human health. We will talk about several issues that have been identified in earlier research that have an impact on using insects as food in this section. To ensure a risk-free or significantly reduced supply of edible insects to consumers throughout the entire production and distribution process, all governments, developed and developing alike, must give top priority to enacting laws and regulations about edible insects (Imathiu, 2020). Consumption patterns have changed significantly as people become more aware of the benefits and rights associated with eating healthful foods; this is the case with edible insects and the products derived from them (Lähteenmäki-Uutela et al., 2021).

14.4.1 Safety Concerns for Using Insects as Feed

The feasibility of using insects as a food source has drawn criticism. Controlling for chemical issues, potential allergies, and microbiological danger is necessary to increase the nutritional assessment of insects as protein sources (Van der Fels-Klerx et al., 2018).

14.4.2 Microbiological Safety

In overcrowding farms the viruses, bacteria, and yeasts grow very fast, and insects are genetically produced. These extensions regarding the use of insects and the microbiological risks implicated with feeding insects to animals have more recently given rise to numerous research alarms. A detailed study on the microbiological quality of the freeze-dried insects found that more than half of the insects especially mealworms and locusts had a total of aerobic bacteria count over 6log colony-forming units (CFU) per gram and Enterobacteriaceae at over 3 log CFU/g. This report establishes that different categories of bacteria such as *Clostridium* species, *Staphylococcus* species, and *Bacillus* (cereus group) are among the several bacteria that can be isolated from insects (Vandeweyer et al., 2021).

Mammalian pathogenicity is not a general trait of fungi, but a characteristic exhibited by certain compounds called mycotoxins which fungal species are capable of infecting mammals with. Innumerable varieties of fungi namely *Aspergillus* species, *Fusarium* species, and *Penicillium* species release mycotoxins. Further research is needed in this area because feeding studies did not yield any conclusive evidence of mycotoxin build-up in insects (Govorushko, 2019).

14.4.3 Chemical Safety

Chemical pollution affects every stage of insect development as well as their habitat. Chemical pollutants can be things like pesticides, heavy metals, and some veterinary drugs (Pousga et al., 2019). Insects used in the experiment could contain chemical pollutants either due to their production of toxins or the contaminants from the substrate on which they were raised. Some of the common heavy metals that can form and accumulate within insect bodies are lead, arsenic, mercury, and cadmium; the authors found that there is a direct correlation between the amount of the same in the substrate and the concentration of heavy metals in the internal organs of the insect (Diener et al., 2015).

An illustration of heavy metal found in insects and substrates was described (Van derFels-Klerx et al., 2016). They found that the amount of cadmium peaked in *T. molitor* larvae at (2 mg/kg dw) as the substrate increased (0.13mg/kg).

14.4.4 Allergens

While an allergy is one form of hypersensitivity, it is triggered by specific immune pathways that can be easily identified. The identified potential insects that can cause a severe allergic reaction and include silkworm pupae, grasshoppers, bee larvae, locusts, cicadas, and pupae (Van der Fels-Klerx et al., 2018). However, before their extensive use in animal feeds, more studies on the allergens of insects are needed. People may have allergic reactions to chitin, an aminopolysaccharide that is virtually always found in insect cuticles. The good news, however, is that the finer size of chitin particles can reduce the inflammatory reaction (DiGiacomo & Leury, 2019). It should also be noted that some people who have allergies may develop an allergic reaction if they consume insects because they may cross-react with other allergens. Cross-reactive allergic reactions occur when the allergic reaction impacts those allergic to different proteins; (Premrov-Bajuk et al., 2021).

14.4.5 Anti-nutrients

It is imperative to take into account the existence of trace amounts of anti-nutritional chemicals (less than 1%) in certain bug species, including tannins, oxalates, and alkaloids, as they may have an impact on the digestion of proteins and the absorption of minerals (Ojha et al., 2021).

14.5 Legal Framework for Use of Insects as Animal Feed

Since the use of insects as animal feed brings about several safety issues, certain factors must be considered. There is a broad space in the laws regulating the feeding of insects because every state has its laws and practices based on its history (Sogari et al., 2019). This section is dedicated to the analysis of the feed legislation in various countries because it is the most significant and influential component influencing commercial insects. Unfortunately, the vast majority of countries in the Western world have very strict prohibitions against utilizing insects for human consumption or as animal feed. Now available on shelves are insect items under the definition of 'novel food' as defined by the European Food Safety Authority for human consumption. These products will need to be proposed for registration as novel food by the year 2024.

In response to this need, many of the EU member states have passed their laws in this regard (Lähteenmäki-Uutela et al., 2017). Similarly, there are general rules governing innovative food in North America. One must note that the Association of American Feed Control Officers, the Canadian Food Inspection Agency, the US Food and Drug Administration, and Health Canada demand authoritative authorization for the Feed Ingredients Definition Committee (DiGiacomo & Leury, 2019).

14.6 Advantages of Using Insects

14.6.1 Composition of Nutrients

Prey consumed by many domesticated animals are insects yet the nutritional composition of insects differs greatly depending on the type of insect and the medium in which the insects are farmed (Koutsos et al., 2019). Although studies have been conducted to make insects a new source of protein for the formulation of animal feeds, it is important to note that the nutritional quality of insects depends on the species, rearing technologies as well as a substratum (Danieli et al., 2019). Insects have a somewhat complete amino acid composition and a high protein content of 30–68% (Koustas et al., 2019). In addition, they exhibit potential as a source of fat and energy (Biasato et al., 2018). This is because they contain a significant amount of lipids (between 10% and 30%), even if the makeup of their fatty acids varies greatly. Insects also contain considerable levels of bioavailable minerals particularly iron and zinc and vitamins particularly B12 (Finke, 2019).

14.6.2 Bioactive Compounds

There is the possibility of using insects as supplement feed where emphasis is pulled away from crop-based feeds such as fishmeal and soybean meal. Moreover, they contain bioactive molecules that can elicit more positive changes in the health and efficiency of chickens (Biasato et al., 2019). Among the claimed benefits of insect-derived compounds including antibacterial, chitin, and antioxidant peptides identified for insect-consuming animals are improved animal welfare (Wu et al., 2018), alteration of the gut microbiome of the animal (Antonopoulou et al., 2019) and boosted the immune response of the animal body (Henry et al., 2018).

14.7 Limitations

14.7.1 Market Price

That's why one can meet rather big problems in finding information concerning market pricing since demand is extremely high while supply is still poor. However, it cannot be as flexible as necessary so that the parties of a business have to make particular agreements if their customers require certain conditions and the required amount of acquisitions. However, it is equally expensive between 2.0 and 10.0 e/kg when compared to other countries of the world and even more expensive when compared to the EU member countries (Mancuso et al., 2019). This is particularly due to regulatory hindrances and a relatively small industrial base. Even more, cheap insect products could be produced by developing more extensive manufacturing in the EU and employing various floor materials for insects' breeding (Amro et al., 2023). Cultivate studies suggest that the cost of protein from insects can be comparable to that of fishmeal by 2023 (Arru et al., 2019).

14.7.2 Polyunsaturated Fatty Acids and Minerals

While there are insect species that are rich in polyunsaturated fatty acids in their bodies, like grasshoppers, there are others that contain a moderate amount of this ingredient, for instance, black army flies. This is determined by the species of the insect and the conditions in which it is raised. Terrestrial insects may pose a challenge to animal nutrition and the quality of meals made from them because of their low amount of polyunsaturated fatty acids (Dalle Zotte et al., 2019). The same applies to minerals since some species like locusts and grasshoppers have extremely low calcium or phosphorus contents. However, with the right nutritional intervention, this feature in insects might be improved (Pinotti et al., 2019).

14.7.3 Customer's Acceptability

Hitherto, the limited literature review finds that acceptability is not a constraint to the insect protein's growth for the feed market. Nevertheless, one would like to know, if from this point of view, the overall acceptability will be regarded as superior concerning traditional commodities and if consumers willing to pay for fed insects, it results in an increase in animal proteins and production becoming a highly sustainable feed component. Recently, the question of whether people in the West will happily take insects on their plates has become a popular subject of research. Several variables have been researched about insect consumption, such as neophobia (Pliner & Hobden, 1992), disgust (Verbeke, 2015), experience (having consumed insects before), and between processed insects and raw insects (Menozzi et al., 2017).

14.8 Insect farming and its nutritional interaction value

The use of insects in poultry diets is strongly encouraged by the food security arguments for doing so particularly because scientific studies have verified insect meals as sustainable as well as valuable protein sources that can fulfill these birds' production needs. However, from the stage of raising insects to the phase of manufacturing feed, the nutritional content of insects fluctuates.

14.8.1 Farming

The settings under which insects are raised have a critical impact on the larvae's nutritional value. In actuality, the larvae's body composition is determined by their density as well as the amount and caliber of their food. The impact of food nutrient content and larval rearing density on the chemical composition of BSF and growth performance was examined by (Barragan-Fonseca et al., 2018). Three nutritional concentrations (low, medium, and high) and four (04) rearing densities (400, 200, 100, or 50 larvae per container) were examined. Despite the larval density, the authors found that in all groups that received high nutritional concentration, both individual larval weight and overall larval output rose.

Groups receiving high nutritional concentrations and low rearing density had increased larval crude fat content. Groups with low rearing density and low nutrition concentration had increased larval crude protein. This study showed that the interplay between rearing density and nutrition concentration affects the CP content of larvae. This study revealed that the regulation of larval CP content occurs within specific bounds and that the quantity and density of nutrients determine the amount of crude fat in larvae. Indeed, it was reported that larval crude fat content increased with high-fat and high-carbohydrate diets (Zheng et al., 2012). Another study found that larval crude fat content decreased with low-fat and/or high-fiber meals (Nguyen et al., 2015). These results emphasize how crucial insect diets and rearing techniques are to the development of larvae with valuable nutritional profiles.

14.8.2 Processing

The insects are usually offered to animal feed in the form of a meal. It entails the consumption of whole insects that are either roasted or dehydrated before being ground into a fine powder (Licaega, 2021). Some processing methods include steaming, boiling, frying, smoking, drying, and toasting among others (Mutungi et al., 2019). The thermal treatments can reduce a very high level of microbial risks but at the same time, it reduces the insect's nutritive value. It was noted that all heat treatments which include boiling, toasting, oven drying, and the combined treatments eliminated the number of bacteria and the presence of yeast and mold in the prepupae of BSF (Nyangena et al.,2020).

Meanwhile, toasting, boiling, oven-drying, and solar-drying treatments lowered the crude fat content down to 14%.3-28.2g/Kg DM. They also improved the available carbohydrates and increased the CP level to 37 – 41. 3g/Kg DM in the same order as previously described. The effect of heat treatment and lyophilization on the proximate composition and amino acids of the black cricket *Gryllus bimaculatus* at 45 and 120 °C was studied (Dobermann et al., 2019). It was evident from the results that the protein and calcium content could be preserved by drying at 45°C. Moreover, when compared to drying at 120°C, freeze-drying provided better protection to long-chain polyunsaturated fatty acids. Therefore, during heat processing, one needs to be more cautious when preserving the nutrient value of insects.

14.9 Future Potential of Insect Meal in Poultry Nutrition as a Concentrate Protein

For any number of reasons, a meal of insects still has promising prospects for the future as a protein source concentrate for poultry feeding. Thus, it is suggested that by increasing the technological efficiency of insect production and more effectively adapting technologies that can be used for insectary production of insects, insect meals can be made cheaper than conventional proteins (Olarotimi & Adu, 2017). Furthermore, improvement of the regulations and awareness of the consumers to the insect-based diets may create new exports for insect-based feeds in the poultry supply chain (Sogari et al., 2019). Even though insects contain a relatively high content of micronutrients, have high energy and protein calories, and have the potential for use in feeding and as protein sources, still more data is required to specify the utility of insects for human consumption, discover pathogenetic effects of insects, whether there exist dangerous components in the edible insects, ways to eliminate them, and storage conditions (Thirumalaisamy et al., 2016).

Furthermore, meals of insects can be combined with other non-animal protein sources such as proteins of plant origin to produce diets for chickens that are nutritionally balanced. This tactic can lessen the need for conventional protein sources while also lessening the environmental impact of chicken farming (Park et al., 2022). All things considered, insect meal as q protein concentrate for chicken diets has bright prospects. More studies and advancements in this field will increase the likelihood of meeting the poultry industry's increasing need for affordable protein sources.

Further research should be done on the levels of insect product integration in feeds for pigs and poultry. Research conducted in Asia and Africa provides the bulk of the published data on animal performance. Studies conducted in diverse areas using varying pig and poultry husbandry systems are required to evaluate the impact of insect components on animal product performance and explore their potential. Further investigation is needed to ascertain the viability of insect products and any other advantages they might offer as protein sources (Banday et al., 2023).

14.10 Conclusions

The nutritional profile of insect meals along with the cost-effectiveness makes insect meals a good candidate for a substitute protein source in poultry diets. The current source of proteins seems to be under pressure as the worldwide call for proteins increases. One sustainable solution to these challenges is to use substitute sources of protein in feed including plant protein and insect proteins. They have their limitations and challenges as the utilization entails massive insect production, legal endorsement, and customer acceptance. Insect meals could be used more in feeding for the chickens if there many more research and developments done to overcome these challenges.

More studies should be carried out to establish the impact of these diets on immunology, the gastrointestinal tract, and the performance of chickens in subsequent stages of growth. Various types of nutrient-rich products are derived from the rearing of insects, which also has a positive impact on the health and nutritional value of meats. They have a relatively rich content of protein and energy, and they also contain antimicrobial peptides, chitosan, and chitin. Future research in the utilization of bug meat for cattle feed may include establishing the optimum levels of inclusion over the long term for laying hens and breeders.

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Chapter : 15 Sex Semen: The Next Frontier

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Abstract

Sexed semen technology has the potential to expand animal breeding, providing control over the sex of offspring and advanced transforming genetics. Embracing innovative technologies of sex semen like flow cytometry and microfluidics by farmers helps in separating sperm cells based on sex chromosomes, enabling the preferential production of male or female calves. Sex semen technologies provide multiple advantages like improved herd management and precise control over offspring, dystocia may have a very low level of occurrence, and it also increases the well-being and reproduction of Cows and buffalos which is the main demand for them to expand their livestock. Hence, it is proved that sex semen plays an important role in veterinarians and animal breeders, especially for achieving specific breeding objectives and incorporating sustainable agricultural practices. Therefore, the study will provide a comprehensive understanding of sorting accuracy, productivity, and efficiency in the breeding of cows and buffaloes. Moreover, the combined results of sexed semen with assisted reproductive technologies (ART) mainly in vitro fertilization proved effective at a high level.Similarly, innovative technology has potential applications in prioritizing male and female calves with promising implications. Although reporting a lot of benefits, sex semen still faces potential hurdles and challenges such as higher costs and ethical considerations highly associated with sex selection. Future directions in sexed semen technology focus on refining techniques and expanding its applicability across species, contributing to the overall goal of improved animal health, well-being, and sustainable livestock production of cows and buffalos.

In short, the book has presented the transformative impact of sexed semen technology and underscores its potential to revolutionize animal breeding and conservation efforts.

15.1 Introduction

Large healthy animals like cows and buffalo are in the medical veterinary because of their paramount importance (Roth, 2011). In trade, agriculture economies worldwide and contribution significantly to food protection and livelihood all these animals play a vital role. Their welfare and health are important to guarantee because it is not only important for ethical reasons but also for the sustainability of farming operations and maximum production. The aim of veterinary medicine is for large animals and encompasses a wide range of practices to prevent the diagnosis and for the treatment of diseases as well as to optimal living Conditions promotions. All the large and healthy animals are less prone to diseases more productive, and more capable of contributing to the agricultural economies. Cornerstone is a reproduction of animals' health, directly influencing the protection and sustainability of the animal population (Perry, et al., 2020).

In Cow and buffalo livestock is important for the successful reproduction of the maintenance of herd size and genetic diversity, milk products for other animals and meats, etc. The problem of reproductive health in the animals not only effect affects the ability of animals to give birth but also affects the overall vitality and resilience to the animal's diseases. For instance, the efficient cycle of reproduction in the cattle of dairy leads to more milk production and healthier calves while the production of milk is guaranteed to steady the supply of animals for the market. So reproductive health to the economic viability of farming operations is integrated with the welfare of the animals themselves.

For semen sexing numerous techniques are used. The most widely used technique for semen sexing is cytometry (Sharpe, 2010). This method though efficient, sorting can be laborious and costly, and the repetitive motions during the process could at times affect sperm viability, resulting in somewhat lower rates of conception than with normal semen.

The equipment of sexed semen has many advantages (Seidel, Update on sexed semen technology in cattle, 2014). For instance, producers generally favor female calves in the dairy business because they produce more milk, but in the beef business, male calves are frequently chosen owing to their faster growth with higher meat yield. Farmers can raise the percentage of desired offspring by utilizing sexed semen, which improves herd management and productivity. Additionally, by enabling producers to choose female calves, who are often smaller at conception than male calves, sexed sperm helps lessen calving issues.

The technology of gendered embryos has many advantages. For instance, producers usually favor female calves in the dairy business because they generate greater quantities of milk, but in the beef business, male calves are frequently chosen because of their faster growth and higher meat yield. Producers can raise the percentage of desired progeny by utilizing sexed sperm, which improves herd administration and productivity. Additionally, by enabling farmers to choose female calves, who are often smaller at childbirth than male calves, sexed semen assists in preventing calving issues (Weigel, 2004).

15.2 What is Sexed Semen?

The process of thawing semen can reduce its viability, low conception rate is leading. Artificial insemination with frozen semen, while offering more control over the time and genetics of offspring can also have its drawbacks (Seidel & DeJarnette, 2022). Additionally, the need for special equipment and expertise and the cost of cryopreservation can also be prohibited for the farmers.

Figure 40 Sex Semen

15.2.1 Technology Behind Sexed Semen Production:

In reproductive technology recent advances like sexual semen aim to address these to improve the efficiency and effort of the program of breeding. The sorting of sperm to produce offspring of a desired sex is involved in the sexual semen. This is partially beneficial to the industry of livestock where one sex is more valuable than the other (Amit & Birthal, 2023).

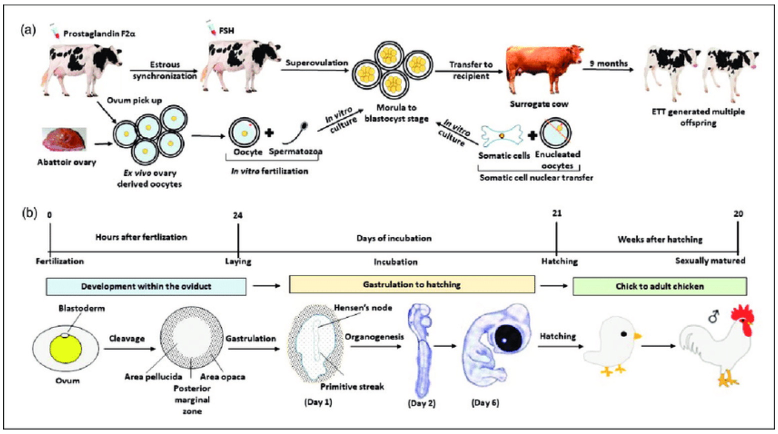

Figure 41 Technology Behind Sexed Semen Production

15.2.2 Difference Between Conventional Semen and Sexed Semen

The term "conventional semen" describes the type of semen that is traditionally extracted from bulls and transferred into cows. Using an artificial vagina or an electroejaculation technique, a bull's ejaculate is used to harvest conventional semen. A concentrated suspension of sperm cells in a nutritional medium is what remains after the collected semen has been treated to eliminate seminal plasma and any other debris. In contrast to conventional semen, sexed semen has more potential to produce offspring. Sexed semen has a 90% probability of producing male and female rate of offspring. A dairy bull calf is not worth much to the dairy farmer because they don't make milk and don't have the right genes to make a powerful beef beast. For this reason, sexed semen is a valuable asset since dairy farmers would prefer to have solely female kids in the herd (ABSGlobal, 2024).

Traditional breeding methods which involved the mating natural and artificial insemination with frozen semen, have decades with wide usage. Moreover, for all these limitations, several methods are introduced. So, in the natural motion both the presence of male and female animals is necessary. So, this ratio leads to logistics challenges and also increases the risk of transmission of many diseases. Moreover, the motion schedule is more difficult, especially in large herds. The maintenance of motion is more and more difficult.

15.2.3 Techniques for Sexing Semen

For semen sexing numerous techniques are used. The most widely used technique for semen sexing is cytometry. This method though efficient, sorting can be laborious and costly, and the repetitive motions during the process could at times affect sperm viability, resulting in somewhat lower rates of conception than with normal semen.

15.2.4 Microfluidics:

Microfluidics, which organizes sperm according to its shape and size using the fundamental principles of fluid motion, is another recently invented method for sexual activity semen. This technique might give you a more effective and non-intrusive selection procedure. The new emergence of micro involves sperm through microscopic procedures. These tubes' are laid out to separate X and Y sperm according to their physical traits. There is still working on more and more research on the effectiveness and affordability of the sorting process (Fernandez, 2023). Furthermore, additional strategies are being investigated to bring more pragmatic results. To enable targeted separation, researchers are using particular markers and antibodies that link to either X or Y sperm of animals (Kumar & Yata, 2021).

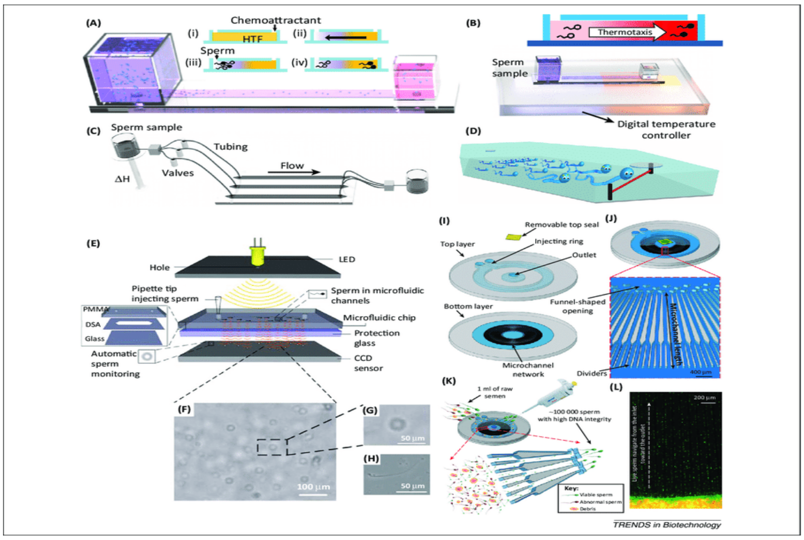

Figure 42 Microfluidics

15.2.5 Flow Cytometry

Cell and molecular biology offer several techniques for accurately detecting and identifying the percentage of spermatozoa bearing X and Y, including PCR and adding and differentiating sperm samples sorted by flow cytometry for DNA content taken from the Cows and Buffaloes. Compared to single-labeled FISH and quinacrine staining, double-labeled FISH involves more strong and more effective spermatozoa isolation. The main and fundamental technique for separating sperm is flow cytometry. Cytotoxicity always has a chance to occur, mutagenesis effects from DNA stains and the ultra-laser beam, that eventually lower the rate of reproduction, and very harsh effects on the rate of the blastocyst (Parth, Saini, Saharan, Bisla, & Yadav, 2020).

15.2.6 Magnetic-activated Cell Sorting (MACS)

Magnetic microspheres are used in the process of magnetic-activated cell sorting (MACS), which divides several cell fractions (Schmitz, 2012). With a 73.8% recovery rate, it has been demonstrated to successfully differentiate dead and apoptotic sperm. Additionally, high-quality sperm samples from bovine sperm have been produced using MACS. By utilizing MACS in conjunction with scFv antibodies, this study seeks to assess the effectiveness of sexing bovine sperm, with particular attention paid to sperm motility, kinematic factors, sperm quality, and the X/Y-sperm ratio (Sringarm, et al., 2022).

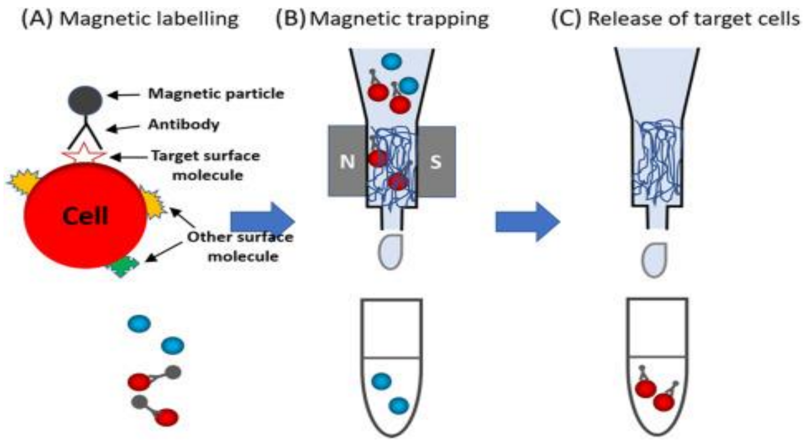

Figure 43 Magnetic-activated Cell Sorting (MACS)

15.2.7 Density Gradient Centrifugation

Sperm are separated employing the previous method based on their relative concentration. It is more affordable and easier to use than flow cytometry, but not being as exact. Nevertheless, in comparison to more sophisticated approaches, its efficacy in sexing testosterone is restricted (Yekti, Rahayu, Ciptadi, & Susilawati, 2023).

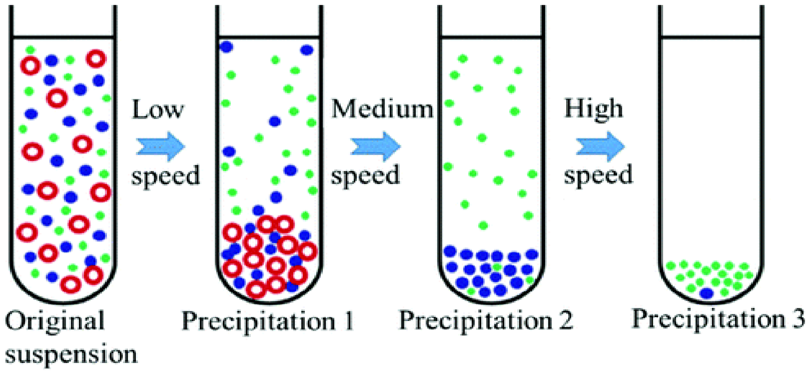

Figure 44 Density Gradient Centrifugation

Sexed semen technologies are a huge move forward in breeding animals and have several advantages for animal welfare, well-being, and production. Sperm sorting methods like circulation cytometry and microfluidics remain to be researched and improved, and this will hopefully lead to increased accessibility and efficiency for farmers around the world. Studies will probably lead to the establishment of our having to more economical and successful methods for sexing semen, which will increase the relationship of technology's influence on the cattle sector (Seidel, Update on sexed semen technology in cattle, 2014).

15.3 Benefits of Sexed Semen

15.3.1 Improved herd management and production efficiency

- To generate a greater number of offspring

In this case, sexed semen that employs gender-specific sperm has a far higher chance of producing offspring of the target sex, which improves the reproduction rate and helps meet certain animal semen objectives.

- With male births, calving difficulty was associated

  Since larger male calves have a harder time calving, sexed semen frequently leads to a higher percentage of male births, which calls for cautious methods for handling.

- In breeding programs faster generation turnover present

  Using innovative technologies that reproduce sexed semen allows for faster generation turnover, accelerates genetic development, and improves herd efficiency over a shorter period (Quelhas, 2023).

- For farmers, there are potential economic benefits

  Even though using sexed semen is more expensive than using unsexed semen, it is typically economically justified. A primary benefit is the possibility of using this technology to breeding farms that offer replacement females or males, or to markets where one gender may be preferred, like dairy production or breeders of specific horse and dog breeds.

15.3.2 Animal Health and Welfare

- **Better mother health due to a decreased requirement for dystocia interventions**

To address shoulder dystocia, rescue techniques involve removing the posterior shoulder, and reduced need for dystocia therapies leads to better maternal health, which in turn leads to fewer difficulties and increased general well-being for breeding females. using an axillary sling and attempting to break the fetal clavicle. In the event of a failure, symphysiotomy, and abdominal rescue techniques are the last options.

- **Associated with male newborns; a possible decrease in unintended pregnancies and abortions;**

There is a correlation between a woman's age, where she lives, amount of education, expected family size, and media exposure with her chance of becoming pregnant against her will. Five percent of all heterosexually active men said they had become pregnant unintentionally in the previous five years with a partner. Twenty percent of the recent pregnancies that men reported were considered unplanned, and forty-five percent of those resulted in induced abortion (Elul, 2021).

- **Enhanced biosecurity by reducing bull contact with females:**

Any effort made to reduce the possibility of disease agents entering and spreading throughout the farm and, consequently, to maintain the health of the animals and the farm is referred to as biosecurity on a cow farm. Through the implementation of biosecurity protocols and effective management practices, farm animals are safeguarded against endemic and pandemic diseases (Subasinghe, 2022).

Figure 45 Biosecurity by reducing bull contact with females

15.3.3 Ethical Considerations

The possibility of enhancing the welfare of male animals reared for slaughter by lowering the number of such animals. The use of sexed sperm raises ethical questions about animal welfare, genetic diversity, undesirable offspring welfare, and the broader consequences of tampering with normal reproductive procedures.

15.4 Challenges and Limitations of Sexed Semen

The introduction of sexed semen technology has transformed the breeding of livestock like cows, and buffalo by allowing farmers to choose the sex of their offspring, and increase productivity. Still, many challenges and restrictions hinder its widespread adoption and effectiveness.

15.4.1 Lower Pregnancy Rates Compared to Conventional Semen

After comparing sexed semen with the traditional Semen process, it is identified that currently produced have lower pregnancy rates in the cows and buffaloes. The method of sorting can harm sperm, lowering their ability and motility by decreasing the life span of the sperm. Research has demonstrated that reproduction rates with sexed semen are lower in about 10-20%. Therefore, it is noted that this can be a big problem for farmers who depend on conception rates to keep their livestock productivity by sexed semen resolve with effective and careful management and inseminations to get the desired production results.

15.4.2 Higher Cost of Sexed Semen Compared to Conventional Semen

The study also identified that the cost of sexed semen is higher than that of conventional semen, due to the intricate and labor-intensive sperm sorting methods. Besides, the sperm sorting methods, other techniques are expensive operations. Similarly, the cost of producing sexed semen can be two to three times higher than that of conventional semen which eventually makes it difficult for the researchers and farmers to increase their breeding of livestock. Higher costs must be weighed against the financial benefits of producing offspring of a desired sex and ease must be ensured for the farmers (Galina, 2023).

15.4.3 Availability of Sexed Semen for All Species and Breeds

An additional constraint is the availability of sexed semen among species and breeds, especially dairy breeds, and its significant application in livestock species like cows, buffalos, and horses. The process of separating sperm offers effective biological and technological difficulties for sexed semen within cattle, which restricts the benefits of technology to particular areas of the livestock business (Dhangada, 2024).

15.4.4 Ethical Concerns Surrounding Sex Selection Technology

There are several ethical issues regarding sex selection technology as it is against natural procedures. Deliberately altering an animal's sex can interfere with the natural reproduction process. Researchers questioned argue sex selection of cows and buffaloes to increase their animal welfare by farmers. Meanwhile, altering the genetic impacts of breeding for one sex leads to a reduction in genetic diversity as well as it can be prone to illness.

12.4.5 Societal and Ethical Implications of Manipulating Animal Populations

In addition to ethical concerns, there are severe ramifications of manipulating animal populations by the farmers to increase productivity and also maintain the imbalance in animal populations by preferential breeding of females in dairy herds may produce an excess of male calves. Moreover, many calves are put down soon after birth, raising animal welfare challenges and issues. Therefore, the study also noted that these techniques promote unethical practices.

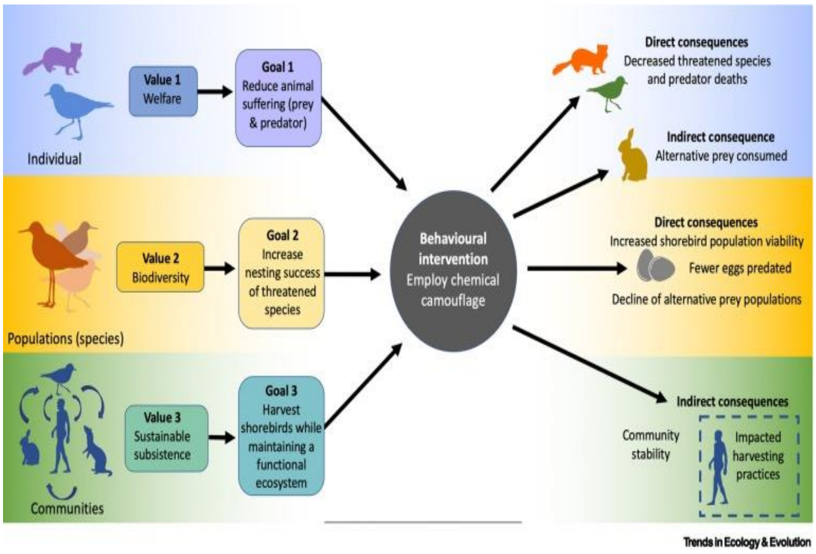

Figure 46 Ethical Implications of Manipulating Animal

12.5 The Future of Sexed Semen Technology

Sexed semen technology has revolutionized livestock breeding, enhanced productivity, and yielded gains by adopting the selection of offspring sex. As this technology develops, productive advancements and new applications are expected, with potentially even greater effects on animal breeding and welfare.

12.5.1 Advancements in Sorting Techniques

At the top of the discussion, the fundamental issue regarding sexed semen technology has been the efficiency and accuracy of sperm sorting which needs more productive procedures. Flow cytometry the modern way of reproduction is much more effective but is reported too costly which can even harm sperm and lower production rates of calves. Despite severe sperm damage, a great level of advancement is ongoing. These great technologies can be seen in the form of new and innovative techniques such as microfluidics and magnetic-activated cell sorting (MACS) to improve sorting accuracy and efficiency (Garg, 2021). Microfluidics leverages the precise manipulation of fluids at the microscopic level to sort sperm based on the physical properties of animals like cows and buffalos. Moreover, the technique of (MACS) is less harmful to sperm and could potentially expand the reproduction rates. This method uses magnetic fields to separate sperm tagged with specific antibodies, offering a gentler sorting technique, which ensures-sexed semen technology is more accessible and effective, while, broadening its use in livestock breeding and well-being.

12.5.2 Application of Sexed Semen Technology in Large Tame-Like Cows and Buffalos Conservation

Sexed semen technology has robust applications in large tame animals like Cows and buffalos' conservation. The new and effective techniques of sex semen are very useful for managing the sex ratio of offspring and can be crucial for increasing the population mostly preferable sex calves by the farmers to expand the level of livestock by depending on the needs of the species.

This case can be comprehensively understood with examples like in species where females are critical for population growth mainly cows and buffalos, sexed semen could help produce more females to increase reproduction rates. On the other hand, for species where the male sex is at a lower rate, the technology could help balance it, supporting healthier and more sustainable populations.

12.5.3 Integration with Other Assisted Reproductive Technologies

Furthermore, the study also demonstrated that the integration of sexed semen technology with other assisted reproductive technologies (ART) like in vitro fertilization (IVF) is also a very crucial and effective technique. Therefore, these integrating methods of technologies could help to improve as well as increase the efficiency of breeding programs, particularly in large tame animals with less natural fertility rates.

Figure 47 Assisted Reproductive Technologies

Using sexed semen in IVF methods incorporates the pre-selection of embryo sex before implantation, highly promising the chances of successful pregnancies and desired offspring whether male or female. This integration is particularly beneficial for large animals like Cows and buffalos for those that produce few offspring per breeding cycle.

12.5.4 Innovations in Semen-Based Therapies

Along with animal breeding and well-being, sexed semen technology is innovating day-by-day in animal medicine, particularly in regenerative and personalized or prioritized sex medicine.

- **Regenerative Medicine**: Semen-based therapies are revolutionized and innovated for their high level of potential in regenerative medicine which provides a lot of benefits to the farmers. Sperm cells, rich in stem cell properties are used for the treatments for several significant diseases and injuries. However further research is being conducted to develop more and moreproductive and effective regenerative of these cells for repairing tissues and treating infertility. The most important thing about this innovative technology is that it is even developing new organs for animals to treat them in a timely.
- **Animals Prioritized and Personalized Medicine**: In animal medicine, the unique genetic material in sperm can be used to tailor treatments and care for the species. This is observed that analyzing sperm DNA can provide insights into genetic predispositions to certain diseases, enabling early and immediate interventions and customized treatment regimens that prevent the spread of these illnesses among more animals (Rehman, 2021).
- **Enhancing Semen's Role in Reproductive Health:** The future of sexed semen technology will also play a bigger part in reproductive health through cutting-edge genetic engineering and advanced reproductive technologies.

- **Genetic Modifications and Enhancements**: Similarly, this research also finds that CRISPR and other modifying gene technologies can enhance and improve the genetic quality of sperm. These developments result in the eradication of genetic illness, improvements in the reproduction process, and the creation of offspring with desirable traits. This aspect of fertility health ensures to significant impact on animal breeding and fertility treatments, offering new solutions to longstanding genetic disorders (Richardson, 2023).
- **Advanced Reproductive Technologies**: Innovation in artificial gametes and cryopreservation procedures are being developed to further improve the productivity of semen in the reproductive health of animals. Artificial gametes made up of stem cells open innovative doors for fertility treatments, particularly for unfertile animals with viable sperm. Improved cryopreservation methods and techniques increase the longevity and viability of stored semen, making it more effective for long-term use in breeding programs and cycles.
- **Expanding the Scope of Semen Research**

  Semen research has directly challenged the conventional methods of breeding, exploring the environmental as well as lifestyle impacts on semen quality. Furthermore, semen study also promotes cross-disciplinary research opportunities.

- **Environmental and Lifestyle Impacts on Semen Quality**:

  Research highly emphasizes improving the semen quality to benefit environmental factors and lifestyle it can increase the breeding for the farmers bringing a huge amount of profit for them. Factors like pollution, diet, and exposure to toxins are being studied for their impact on the sperm health of cows and buffaloes. Therefore, the farmers should understand these influences to better management practices and interventions to improve reproductive health in animals.

- **Cross-Disciplinary Research Opportunities**: The intersection of semen research with other scientific disciplines is providing new and innovative techniques and methods. Therefore, effective collaborations between reproductive biologists, geneticists, and biomedical engineers are helping in comprehensively understanding sperm biology and developing new technologies and procedures. This cross-disciplinary approach is very crucial for finding novel applications for sexed semen technology.

12.6 Emerging Technologies in Semen Research

There are many significant emerging technologies in sex semen research such as:

12.6.1 Section 1: Advanced Semen Analysis Techniques

Advanced semen analysis techniques, especially proteomics and metabolomics, have revolutionized the study of semen.

1. **Proteomics**: Proteomics is a study in which functions and structure of proteins are studied and experienced with significant practices. Scientists and veterinary researchers

detect and measure the proteins of sperm cells and seminal plasma for the discovery of more and more insights. The study has demonstrated essential details associated with proteins in sperm function. It underscores the causes of male reproduction by experimenting with several acrosome reactions that are developed, providing valuable and productive insights into the mechanisms processes, and methods. Moreover, it is examined that by understanding the protein composition of semen, researchers can develop targeted interventions in the reproduction process.

2. **Metabolomics:** Metabolites are the tiny molecules created during the metabolism process. Metabolomic profiling of semen depicts a complete image of the metabolic state of sperm cells and seminal plasma for a better understanding of the process (Jia, 2021). This analysis can identify metabolites regarding energy production, and other critical and complex cellular procedures. By comparing the metabolomic profiles of fertile and infertile semen samples, researchers can improve metabolic efficiencies or may contribute to infertility.

12.6.2 Section 2: Semen Cryopreservation and Storage

To increase the effectiveness of breeding operations and distribute high-quality germplasm through artificial insemination, semen must be cryopreserved. Sperm functions deteriorate during semen cryopreservation because of significant osmotic and thermal stress, physical damage from intracellular ice crystal formation, and oxidative stress from excessive ROS production.

Because of the large concentration of polyunsaturated fatty acids in the plasma membrane and their weak intrinsic antioxidant defense system, which gets worse as semen gets diluted, sperm are vulnerable to oxidative stress (Tirapelle, 2021).

Since a sperm subpopulation with active mitochondria has a high proportion of motile, morphologically normal sperm that can undergo an acrosome response, mitochondrial functionality is necessary for sperm fertilizing competence. By propagating the genetics of exceptional bulls, artificial insemination has the potential to increase animal productivity. This highlights the need to maintain semen quality during cryopreservation. This way, oxidative damage can be lessened by adding traditional antioxidants to extended semen to maintain the quality of chilled, cryopreserved semen, which has been extensively studied in a variety of species. Nevertheless, taking into account the crucial correlation between mitochondrial activity and sperm functional competency.

Nanoparticles (NPs) are pliable, microscopic particles composed of polymers such as proteins, polysaccharides, and metals. Their stable, soluble in water, and biologically efficient qualities have led to their widespread use in medicine. The use of NPs in medication delivery for lipophilic or destabilizing hydrophilic compounds in medicinal formulations is growing. By preventing drug digestion, extending the drug's half-life, getting beyond blood-tissue barriers, delivering the medication to certain tissues, lowering the effective dose, and minimizing side effects, nanotechnology can improve the efficiency of pharmaceuticals. On the other hand, additional study is required to fully understand the effects of nano compounds on cells and tissues, especially in the reproductive system. Concerns are also been voiced regarding their systemic dispersion and biocompatibility.

High-quality gametes and embryos must be chosen for modern breeding systems, and several techniques have been devised to get around restrictions on male reproduction. Sorting and purifying sperm are useful techniques for removing trash, contaminated cells, and subpopulations of the best spermatozoa. To this end, methods such as density gradient centrifugation, swim-up test, filter through columns, and simple layer centrifuged have been developed. On the other hand, these processes may result in lower insemination doses, longer infusion times, and higher expenses. Targeting particular negative biomarkers of sperm quality, such as ubiquitin on sperm surfaces or abnormalities in the cytoplasm and acrosome membrane, is a unique strategy for semen selection. Bulls and humans have both had success using magnetic beads covered with probes that bind to these indicators to remove faulty spermatozoa by exposure to a magnetic field (MACS) (Falchi, Khalil, Hassan, & Marei, 2018).

Contemporary cryopreservation technologies and sophisticated semen analysis tools, in especially, are revolutionizing the comprehension and control of fertility in males in the field of semen research. Personalized fertility treatments make made possible to learn about the biochemistry and metabolism traits of semen. At the same time, developments in cryogenic preservation and biobanking technology improve sperm accessibility and affordability for researchers and farmers. These technologies possess the potential to desired demand for the calves like males and females are preferred for the growth of livestock (Gordon, 1, 2021).

12.7 Conclusion

In conclusion, this chapter is about sex semen technologies. The technology of sexed semen offers exact management, improving genetic development in animals like cows and buffaloes. This type of equipment holds the power to breeding operations, enhancing the overall well-being of animals. The technology of Sexed semen separates sperm cells according to their sex chromosomes employing complex technologies like flow cytology and nanotechnology to produce after preferring male or female offspring. The capacity to choose the sex of progeny has an important effect on productivity and genetic diversity.

Additionally, by decreasing requirements for invasive businesses and the probability of dystocia in cows and buffaloes mainly facing difficulties in pregnancies with male offspring sexed semen. By lowering challenges regarding breeding practices, this advancement in controlling reproduction increases growth in the fertility of female calves (Gordon, Reproductive technologies in farm animals, 2021).

12.7.1 Significance as a Tool for Veterinarians and Animal Breeders

Sex-based semen technology is very crucial for veterinarians and researchers to enhance the selection and reproduction of female calves that increase the livestock and therefore, remain the target of farmers. Veterinarians can help ranchers accomplish certain breeding goals more effectively by providing carcass quality in beef cattle or milk production in dairy cows and buffaloes. Cattle populations can become more efficient and healthier over several generations as genetic advancement becomes more insightful and productive.However, it raises multiple ethical issues and corresponds with customer expectations for sustainable farming procedures and techniques by diminishing the proportion of male animals reared exclusively for slaughter.

12.7.2 Ongoing Research and Future Directions

The goal of ongoing research is to optimize the sex semen sorting process and techniques to ensure the effectiveness of breeding cows, buffaloes, and horses. It is anticipated that innovation and developments in genetic markers, artificial intelligence, and microfluidics will improve sexing methods (Iyer, 2022).

In the future, sexed semen technology by merged with other assisted reproductive technologies, like embryo transfer and in vitro fertilization which will ultimately enhance the reproduction and well-being of these animals. By integrating sex selection with preimplantation evaluations of embryo quality, this integration may provide successful pregnancies and offspring with desired features. In conservation biology, alternately sexed semen may help maintain genetic variety and regulate the gender balance in populations of endangered species by bringing progress in semen analysis and cryopreservation technology eventually customized reproductive therapies and regenerative health applications.

In conclusion, embryo of sexed semen innovations represent a breakthrough advancement in animal breeding which gives unparalleled authority over the gender and genetic variety of descendants. It enhances reproductive effectiveness and reduces risks associated with traditional breeding methods, which has a significant positive influence on the wellness the welfare of cattle. As more research is done to improve and expand the potential uses using sexed semen technology, veterinarians, animal producers, and conservationists will likely find that it is a valuable tool. By embracing these improvements ethically, stakeholders can take full advantage of sexed semen technology to advance animal welfare, increase the sustainability of farming, and advance the availability of nutritious food worldwide.

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Chapter 16 : Animal Physiotherapy: The Ignored Miracle

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SUMMARY The authors assessed the perception of pet physiotherapy by pet owners and veterinarians in China. The results show that 94% of pet owners have heard of pet physiotherapy, and most would use the treatment if needed, typically once a week, for RMB 6,000/occasion. 28% of veterinarians work in practices where such a service is already provided, and another 24% plan to introduce it. Most veterinarians consider continuing physiotherapy training important. They expect RMB 7,000 for a treatment, and almost half would be willing to invest more than half a million RMB in physiotherapy equipment, demonstrating their commitment and dedication to the field.

Abstract

Background: Physiotherapy has become a widely used supplementary treatment in China's small-animal veterinary clinics.

Objectives: The study aimed to survey the attitudes of veterinarians and pet owners towards small animal physiotherapy in China.

Materials and Methods: Two questionnaires were completed - one for pet owners (available in both printed and online forms) and one for veterinarians (only available online). They could be filled out between June and September 2023. We received 200 responses from pet owners and 100 from veterinarians, and the collected data were processed using the Microsoft Excel™ program.

Results and Discussion: A significant 94% of pet owner respondents were aware of small animal physiotherapy, with an overwhelming 99% willing to employ it if necessary. The main sources of pet owners' information about physiotherapy were the therapist (47.3%) and the Internet (47.3%), with the veterinarian also playing an important role (39.9%). An impressive 81.5% of pet owner respondents were willing to pay a maximum of 18.8 euros per treatment, and 61.5% would take their animals to a therapist a maximum of once a week, with others even

more often. When choosing a therapist, the most common factor was the recommendation of other dog owners (46.5%), but 37.9% decided based on the vet's advice. A commanding 88% of veterinary respondents would recommend physiotherapy to their clients if they see it justified. In these cases, an assertive 66% of them would require a consultation with the therapist. A noteworthy 28% of the vet respondents work at a clinic where physiotherapy is provided, and 24% of the surveyed practices plan to introduce it in the short or medium term. Most veterinarians (an impressive 67.7%) expected an income of up to 21.9 RMB per treatment. Furthermore, an impactful 47.1% of the vet respondents are willing to invest more than 1,563 RMB into physiotherapy equipment, with a strong 76% of them encouraging vets and/or assistants to take part in physiotherapy training, and 64.5% of the vets showing great support by offering financial assistance to complete the course.

Keywords: physiotherapy, pets, therapist, rehabilitation, veterinarians

16.1 Assessment of Small Animal Physiotherapy in China

Going back to the Greek origin of the word, physiotherapy can be defined as healing with natural energies [1]. However, it is incorrect to classify it as an alternative medicine, as it is a complementary therapy supported by research results [2]. Its goal is optimal fitness and ensuring the best possible quality of life in connection with free movement and health by maintaining, restoring, and developing bodily functions [3]. Physiotherapy treatment of animals has a relatively short history. While the spread of human physiotherapy can be roughly linked to the First World War, the treatment of animals only began to gain ground in the 1980s [4, 5]. In Great Britain, the Association of Chartered Physiotherapists in Animal Therapy (ACPAT) was founded in 1985 and consists of human physiotherapists with professional qualifications in animal therapy. This was necessary because even then, too many people without reliable knowledge were involved in the physiotherapy care of animals. In Great Britain, in the mid-1990s, animal physiotherapy did not yet offer an independent livelihood; 97% of the respondents also worked in the humane field and most often treated only 5 or fewer animals a week. At that time, the patients were typically more valuable animals from an economic point of view, such as racehorses, dogs, and sometimes farm animals with a significant breeding value [6]. The initial significance of animal physiotherapy in the USA can be attributed to greyhound racing, where the need for the complete musculoskeletal rehabilitation of dogs after injuries first appeared [5].

The research aimed to assess the perception of pet physiotherapy among pet owners and veterinarians in China.

16.2 Material And Method

SURVEY

The survey was conducted between June and September 2023 using separate questionnaires for veterinarians and animal keepers. Pet owners could fill out the questionnaire in printed form in the waiting room of the doctor's office and online in several pet owners' groups on social media. Veterinarians could answer the survey questions online in closed social media groups created for veterinarians. The online questionnaires were created using the Google Forms™ program. The pet owner's questionnaire contained 18 questions, in which - after the socio-demographic and animal husbandry data - we asked the pet owners about their prior knowledge of pet physiotherapy, their judgment, experience, willingness to pay, and criteria for choosing a therapist. The questionnaire prepared for veterinarians contained 16 questions, during which the veterinarians answered socio-demographic questions related to their own animal husbandry and veterinary practice and pet physiotherapy (general knowledge, perception, service options, owner's willingness to pay). Most of the questions were simple-choice, to a lesser extent multiple-choice, and open questions requiring short answers. Both questionnaires took approximately 10–15 minutes to complete. The questionnaire prepared for pet owners was closed after 200 questionnaires and 100 questionnaires for veterinarians were completed in an evaluable manner. The collected data were processed using the Microsoft Excel™ program.

16.3 Socio-Demographic Data And Animal Keeping Characteristics Of Respondent Animal Keepers

91.5% of the responding pet owners were women, and 8.5% were men, probably because most women take care of their pets, including taking them to the vet and filling in the questionnaires, which also shows a greater willingness than men. 0.5% of the 200 responding pet owners are under 21 years old, 28% are between 21 and 30 years old, 28.5% are between 31 and 40 years old, 34.5% are between 41 and 50 years old, 8, and 5% belonged to the over 50 age group. More than half (52%) of those who completed the livestock questionnaire were residents of Shanghai, as the questionnaire was also available here in printed form; one-third (33%) lived in rural towns and 15% in villages. Despite the large proportion of respondents from the capital city, 54.5% of pet owners lived in family houses, and 67.5% had a garden or garden connection. In comparison, 32.5% kept their pet in an apartment without a direct garden connection.

94.5% of the responding pet owners kept dogs, which is welcome from the point of view of our research; since dogs are the main target group of pet physiotherapy, they are by far the most common patients. The number of cats kept as pets was also significant (32%), but 5.5% kept rabbits, 3.5% kept small rodents, 0.5% kept ferrets, and 11% kept other animals, i.e., many animal owners kept several species of animals. The living conditions of the animals resonate with the features of the living environment: 48.5% of the respondents kept their pet animals exclusively in the apartment, another 48.5% in an outdoor-indoor system, and only 3% exclusively in the garden. 84.5% of farmers handled their animals several times a day, 7% once a day, and 8.5% several times a week. Nobody marked the answer option once a week. 96.5% of pet owners see their pet as a family member or companion. The former classic role of dogs as house guards has now significantly decreased; 9% of the responding pet owners marked this answer as the role played by their pet, but more than four-fifths of them (7.5% of all pet owners) also said that he sees his animal as a housekeeper, he also considers it a member of the family (Figure 1).

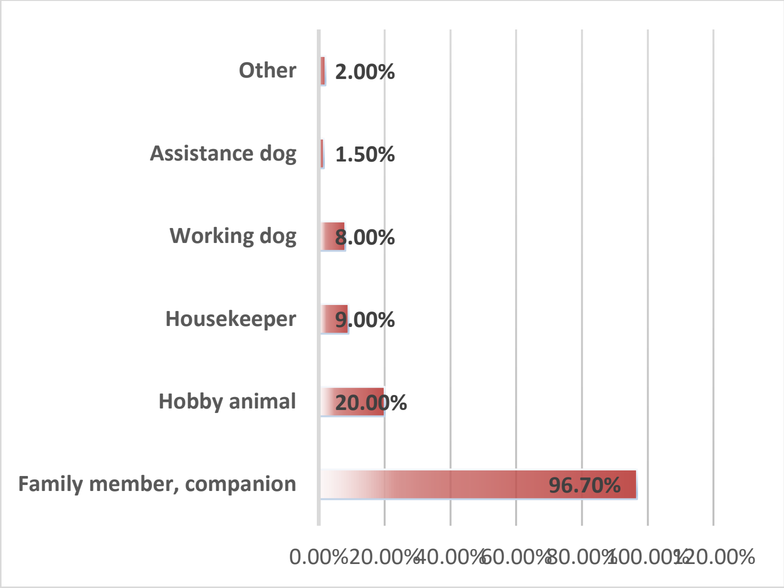

Figure 48 The role of small animals in the households (n = 200)

16.4 Socio-Demographic Data And Animal-Keeping Characteristics Of Responding Veterinarians

Of the 100 responding veterinarians, 70% were female, 55% were between 21 and 30, 29% were between 31 and 40, 9% were between 41 and 50, and 7% were over 50. belonged to the age group. The questionnaire was mostly filled out by women and members of the younger age group, which could be for several reasons. On the one hand, it is a trend of about 3 decades that proportionally more and more women choose this profession, which is also related to the shift of practices in the direction of small animal medicine since this field is often more suitable for a woman than farm animal medicine [7, 8]. On the other hand, the higher proportion of the young veterinarian age group's willingness to answer may be due to the connection to the fact that they are more active on the Internet and social media sites than older people. By the age distribution, most veterinarians who completed the questionnaire graduated within 5 years (60%), 18% graduated 6–10 years ago, 10% graduated 11–20 years ago, and only 12% graduated more than 20 years ago. Most responding veterinarians (91%) worked in small animal practice, and two-thirds were active exclusively in this field. Veterinarians dealing with exotic animals were also represented in a significant number (13%), but 10% of the respondents worked in farm animal practices, 8% in public administration, 2–2% in veterinary pharmacies and horse practices, and 1–1% also did teaching, research, laboratory or non-veterinary work.

88% of the answering veterinarians kept their own animals at home, and 90.9% of the veterinarians who kept animals considered them family members. Only 6.8% considered them exclusively pets, and one veterinarian (1.1–1.1%) considered them working dogs or for a housekeeper. The distribution of the veterinarians' role played by their pets in their own household was very similar to that of the surveyed pet owners.

Perceptions Of Pet Physiotherapy Among Pet Owners

Our survey results show that pet physiotherapy is already present in the public consciousness, as 94% of pet owners have heard of it (two-thirds of those who have not heard of it were not residents of Shanghai). To the question, “If so, where did you get information about this field?” Pet owners mainly received information from the pet physiotherapist and the internet (47.3–47.3%) and also heard about this treatment option from their veterinarian (39.9%) (Figure 2).

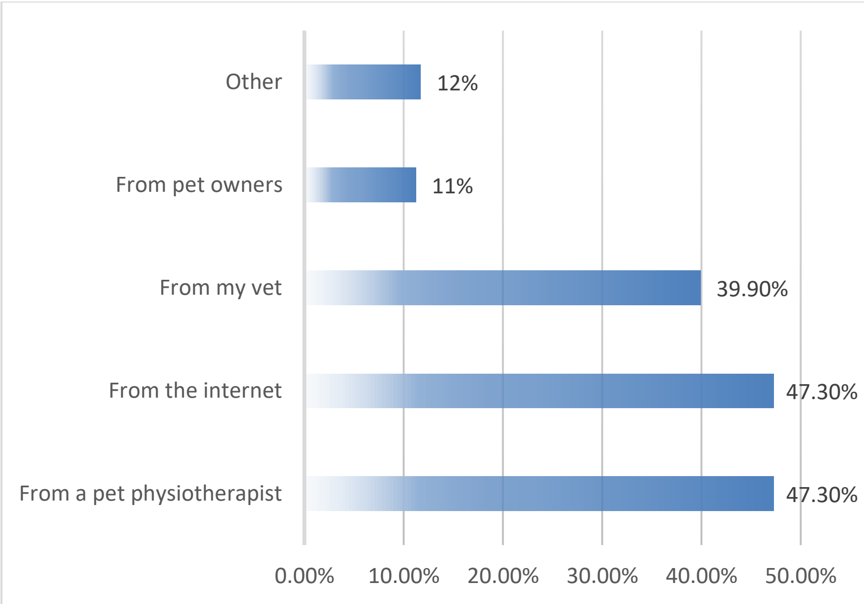

Figure 49. The pet owners’ information sources about physiotherapy (n = 196)

People are open to this therapy method; 64.5% have already visited a pet physiotherapist, and 34.5% have not yet needed one. However, according to their claim, they would visit a therapist if it was justified. Out of 200 respondents, only 2 people (1%) stated that they did not consider physiotherapy important. Most pet owners (61.5%) first consider physiotherapy for its role in rehabilitation after orthopedic surgery, followed by helping to recover from neurological injuries (51.5%). Still, almost half of them (48.5%) would also use it for prevention if some locomotor disorder is revealed, and more than a third (35.5%) would visit the therapist to treat lameness. It is gratifying to see that a quarter (25.5%) of the responding pet owners would already include this treatment in the training program for sports dogs (Figure 3).

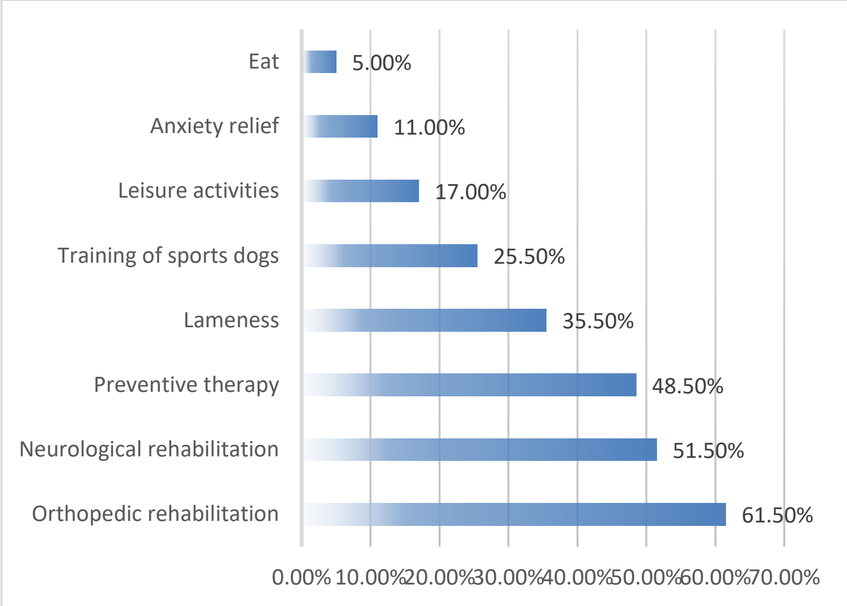

Figure 50. The pet owners' motivations for physiotherapy treatments (n = 199)

Physiotherapy can be used in many areas of veterinary medicine, and orthopedics is one of the most important. Postoperative rehabilitation significantly speeds up and, in many cases, makes recovery more complete in cases such as surgical solutions for anterior cruciate ligament tears, limb amputations, femoral head resection, other hip joint surgeries, and surgical repair of tendon and ligament injuries. However, it is also useful in treating orthopedic problems that do not require a surgical solution, such as arthrosis, soft tissue injuries, or conservatively treated hip or elbow dysplasias. It can also play a significant role in pain relief management since the amount of pain-relieving drugs used can be significantly reduced by using the appropriate physiotherapy devices, which can be of critical importance, especially in the case of high-risk patients [2, 9].

The other main rehabilitation target group is neurological patients. Rehabilitation after surgery is, if possible, even more important here than in orthopedics. Most often, these interventions are aimed at decompressing the spinal cord. However, it has an important role in the rehabilitation of central or peripheral nerve injuries, as well as in the management of chronic diseases such as degenerative myelopathy [2, 10, 11]. A growing branch of physiotherapy is the preparation of animals, especially dogs, for various sports. This includes learning the correct warm-up, developing important skills in the given sport, strengthening important muscle groups, and rehabilitation after sports injuries [12]. In addition, physiotherapy can also play a role in regulating body weight and maintaining the quality of life of elderly animals; it can be used for stress relief, but it also has the means to facilitate the emptying of phlegm that accumulates in the lungs [13]. Examining the willingness to pay, more than a third of pet owners (34.5%) would only accept a price below RMB 5,000, almost half (47.5%) would accept a price between RMB 5– 6,000, and 10.5% considered a price of RMB 6–7,000 acceptable for a physiotherapy treatment. Only 8% of the respondents would gladly pay more than this for one session (Figure 4).

Examining the willingness to pay of pet owners every month, most of them (35.5%) could allocate less than RMB 20,000 for the physiotherapy treatment of their pet, 31.0% of them would spend RMB 20–25,000, and 7.0% of them 25 - RMB 30,000. It is encouraging, however, that more than a quarter of pet owners (26.5%) would spend more than RMB 30,000 per month for their pet's physiotherapy (n=200). Almost half of the pet owners, 48%, could take their pet for treatment once a week, which in reality is the most typical practice, but in justified cases, 22% of the pet owners would accept the therapy more often than 3 times a week (Figure 5). To the question "Would prefer to go to a treatment center designed for this purpose or would prefer treatment at home," the pet owners gave the following answers: two-thirds (66%) of them would choose a physiotherapist over treatment at home, 17% would always choose home therapy, even if there is a separate exit fee, also 17% on the other hand, they only want to use treatment at home without a disembarkation surcharge (n= 200). The answers to the question about the form of treatment reveal that the majority of pet owners would like to be an active part of the therapy process in some form; only 4% of them stated that they would leave their pet completely to the therapist for the duration of the treatment and would pick it up at the end of the treatment (Figure 6). Choosing a physiotherapist is always a matter of trust, and almost half of the owners (46.5%) listen to the experience of their friends with dogs. However, the veterinarian's word is also important here; 37.9% of people would make a decision primarily based on their veterinarian's recommendation. 9.1% would choose a therapist based on internet information, and 6.6% based on other criteria (n=198).

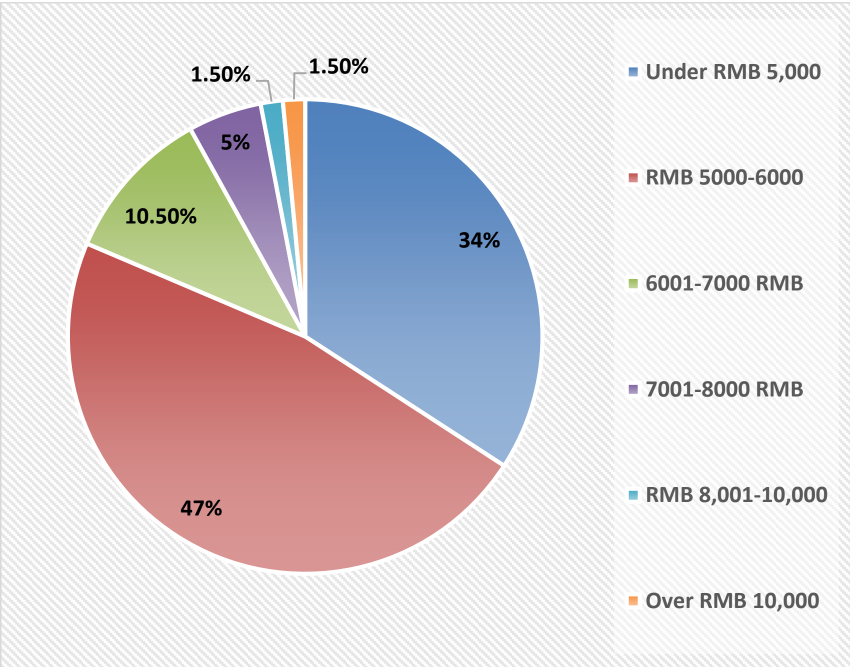

Figure 51. The amount per physiotherapy treatment that pet owners are willing to pay (n = 200)

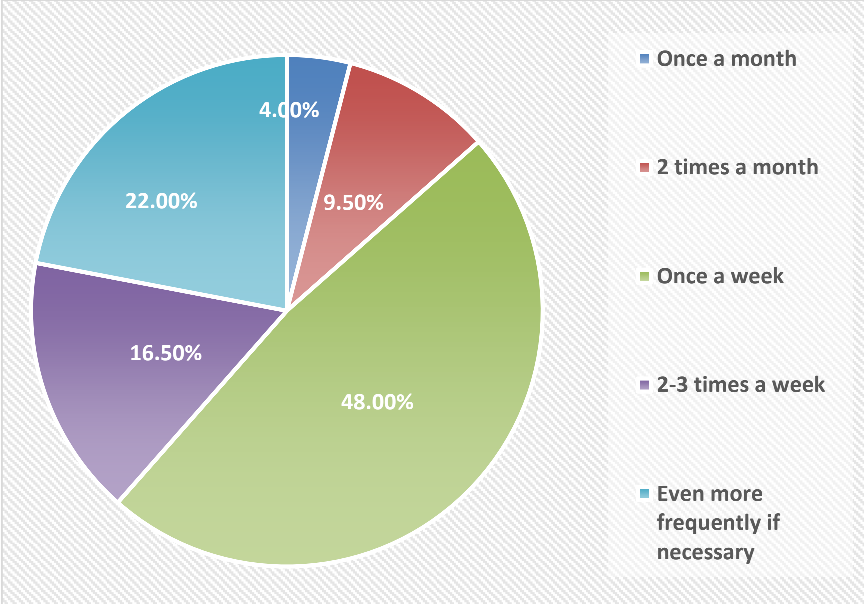

Figure 52. How often are the pet owners able to take their pets to physiotherapy treatment (n = 200)

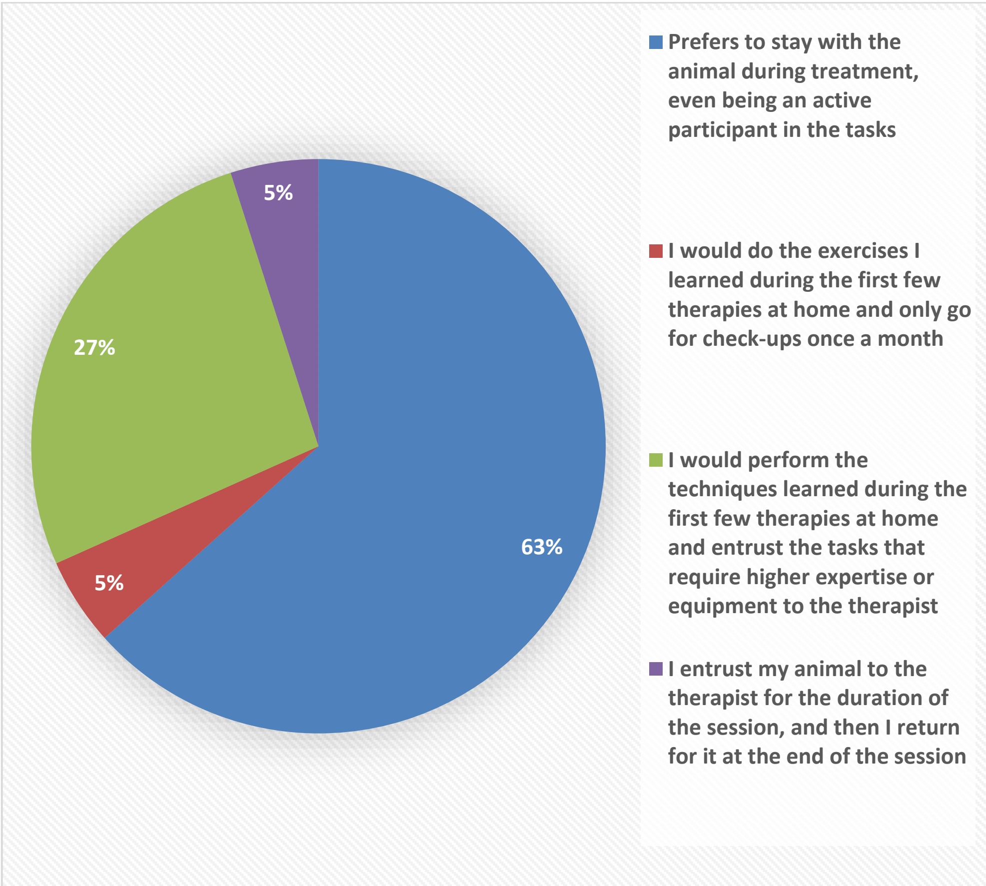

Figure 53. The pet owners' preferred form of physiotherapy (n = 200)

16.5 Perceptions Of Small Animal Physiotherapy Among Veterinary Doctors

Most respondents obtained information about physiotherapy from veterinary colleagues (67.0%), so professional discussion between colleagues is essential in ensuring that veterinarians are up-to-date on this complementary therapy (Figure 7). The second most significant source of information was the pet physiotherapist himself (55.0%). This is not surprising since physiotherapists often visit veterinarians at the beginning of their careers to inform them about the services that can benefit animals. In addition, if they are lucky, many physiotherapists can agree on one or a few veterinarians with whom they can cooperate to refer patients and learn from each other mutually. The third most significant knowledge base was university courses (37.0%). This is certainly an encouraging trend for the future, as it means that newly graduated colleagues enter the practice with a basic idea of the benefits of pet physiotherapy.

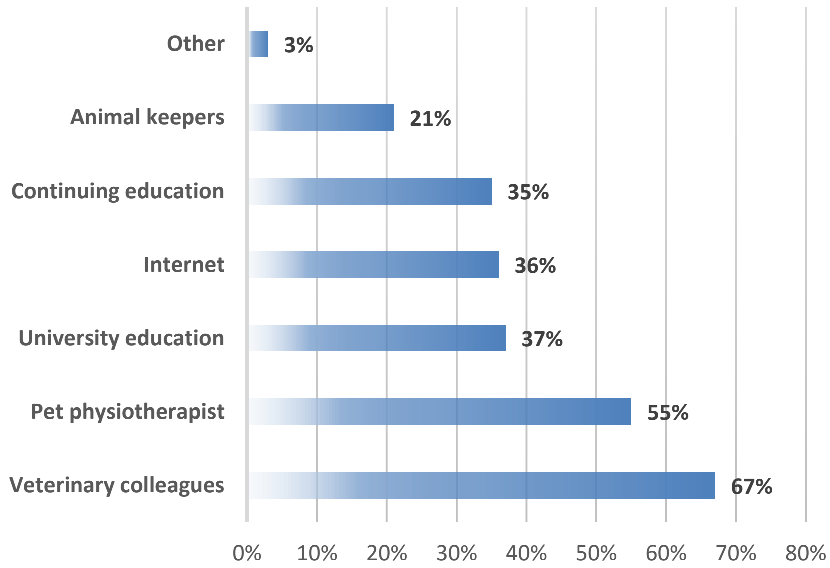

Figure 54. The veterinarians' information sources about physiotherapy (n = 100)

Fourth- and fifth-year students at the Sichuan University Faculty of Veterinary Medicine were asked about their knowledge and opinions about animal physiotherapy. Most of the students knew about this branch of therapy, but their knowledge typically came from informal sources. In their opinion, the most important benefits of physiotherapy are maintaining joint movement paths, increasing muscle strength, and reducing pain. Even though only 5 respondents (5.4%) had personal work experience with a physiotherapist, they demonstrated a high awareness of the methods used. It was generally agreed that physiotherapy was helpful, but it was debated whether it should be done by a veterinarian or a licensed physiotherapist. However, they all supported the regulation that only patients referred by veterinarians for physiotherapy could be treated [14]. The regulation of animal physiotherapy in China is also in line with this. According to the professional position of the China Veterinary Chamber issued in 2016, physiotherapy is a complementary and supporting veterinary activity, not an independent healing activity, which can be performed if the veterinarian deems it so and is subject to the veterinarian's professional supervision. According to this, animal physiotherapists can only collaborate with a veterinarian or should have a veterinary qualification themselves [15]. The survey results show that the internet, further training, and animal keepers also played a significant role in expanding the knowledge of the responding veterinarians. (Figure 7). Some highlighted their experiences with their own animals or got information from acquaintances, and some had a physical therapist qualification themselves. In other words, most domestic veterinarians learned about pet physiotherapy from several sources, which is very positive, considering that dog physiotherapist training has only been available in China since 2010. The rate of development of small animal physiotherapy is clearly shown by the fact that even in the early 1990s, even in Great Britain, less than half of the veterinarians had heard of animal physiotherapy, even though there was already an association of certified physiotherapists experienced in the treatment of animals. (ACPAT). However, their general attitude was considered positive, and there was a clear need for more information on the subject [16]. In 2003, 79% of veterinarians had heard of animal physiotherapy in Ireland. Most of their knowledge came from colleagues, farmers, and professional magazines. However, very few referred patients for physiotherapy; just over a quarter (26%) of the veterinarians knew about this complementary therapy, but none of them had a bad experience with treating the referred animals; in fact, the vast majority of them were particularly satisfied. The general opinion was that this therapeutic option is primarily for sporting dogs and horses. Most were aware of physiotherapy use in locomotor complaints but heard less about other treatable conditions. It was also a general conclusion that veterinarians needed more knowledge, and close cooperation and communication between the two professions were also considered important [17]. The main sources of information for domestic veterinarians are similar to foreign practice because, in both Great Britain and Ireland, the most important source of information was information from colleagues, in addition to specialist articles, physiotherapists, and farmers [16, 17]. 88% of the interviewed veterinarians recommend physiotherapy to pet owners, almost all of them for rehabilitation after orthopedic surgery (95.5%), but more than two-thirds also recommend it for neurological complaints (78.4%). Joint problems and developmental disorders (70.5%) and arthrosis (63.2%) are also common indications. (Figure 8).

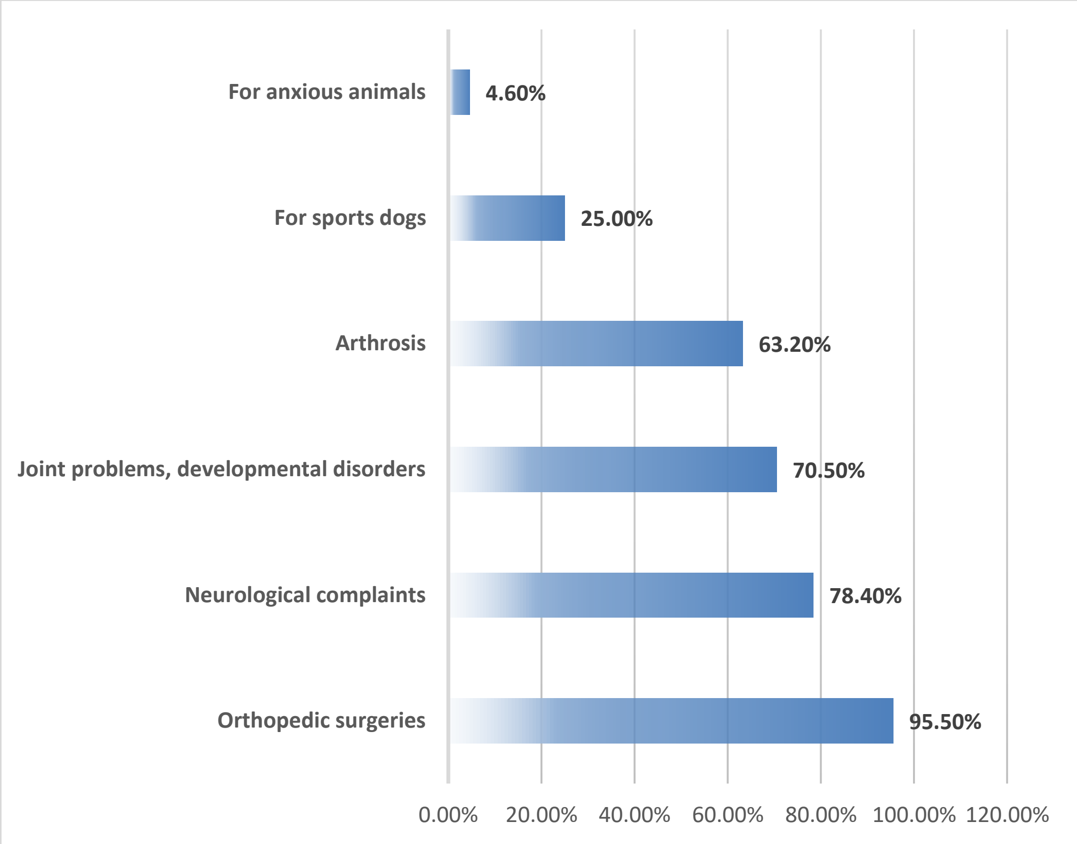

Figure 55. The cases in which the veterinarians would recommend physiotherapy (n = 88)

The "How often do you meet pets with locomotor complaints?" answers to the question, it was revealed that, not surprisingly, the veterinarians who least often recommend physiotherapy to their patients are those who meet animals with musculoskeletal complaints less often than monthly (7% of the respondents), and only 57.1% of them recommend it. this additional therapeutic option for his patients. Most often, veterinarians who meet several such animals daily (39% of respondents) recommend this additional treatment and only one of them does not usually recommend this treatment (2.6%) (n=100). The increase in the popularity of physiotherapy is also clear here because, at the beginning of 1990, only 57% of veterinarians in Great Britain who were aware of pet physiotherapy recommended this therapy to their clients [16]. The openness and interest of veterinarians in physiotherapy are shown by the fact that almost two-thirds of the responding veterinarians (66.0%) would request to see the progress of their cases sent to physiotherapy and to be consulted about the therapy by the physiotherapist and only 14% of them said that does not require consultation (n= 100). Another 20% of responding veterinarians only want to contact the treating physiotherapist regarding complicated cases. These colleagues presumably already have more experience in animal physiotherapy, so they have more confidence in the physiotherapist's decisions, or they probably work with a larger caseload. Among the veterinarians working in equestrian practice, those who have already encountered some additional therapy in connection with their patients, e.g., with physiotherapy, were mostly very satisfied with it; however, about half of them also reported that they had to correct the pet owners in cases where they received incorrect information from the therapist [18]. The "Given the growing demand for physiotherapy, are you planning to introduce this service within the practice?" question, it was revealed that 28% (!) of the responding veterinarians worked in clinics where physiotherapy services were already provided, and 24% of the surveyed clinics had short- or medium-term plans to introduce the service. The other half of the respondents (48%) do not plan such developments, but only 8% of the interviewed veterinarians said that they do not consider this additional therapy important (Figure 9).

We also asked the veterinarians, "What income would you expect from a physiotherapy treatment?"(Figure 10). Two-thirds of the 90 responding veterinarians (66.7%) expected a maximum amount of RMB 7,000 for physiotherapy treatment, which also meets the price indicated by most pet owners. Most (33.3%) have 5,000 and 6,000 RMB, and only 16.7% would expect an amount greater than RMB 8,000. Based on the willingness of pet owners to pay, people still need to accept the introduction of higher prices. However, proper management, a wider range of physiotherapy tools, and a professional attitude can also provide this opportunity. In Great Britain, the price of physiotherapy treatment has also caused mixed opinions among veterinarians. Those who regularly sent animals to a physiotherapist did not consider the average price of the treatment to be high. In contrast, those who did not continue to send animals indicated the size of treatment costs as one of the main risk factors for using the service [16].

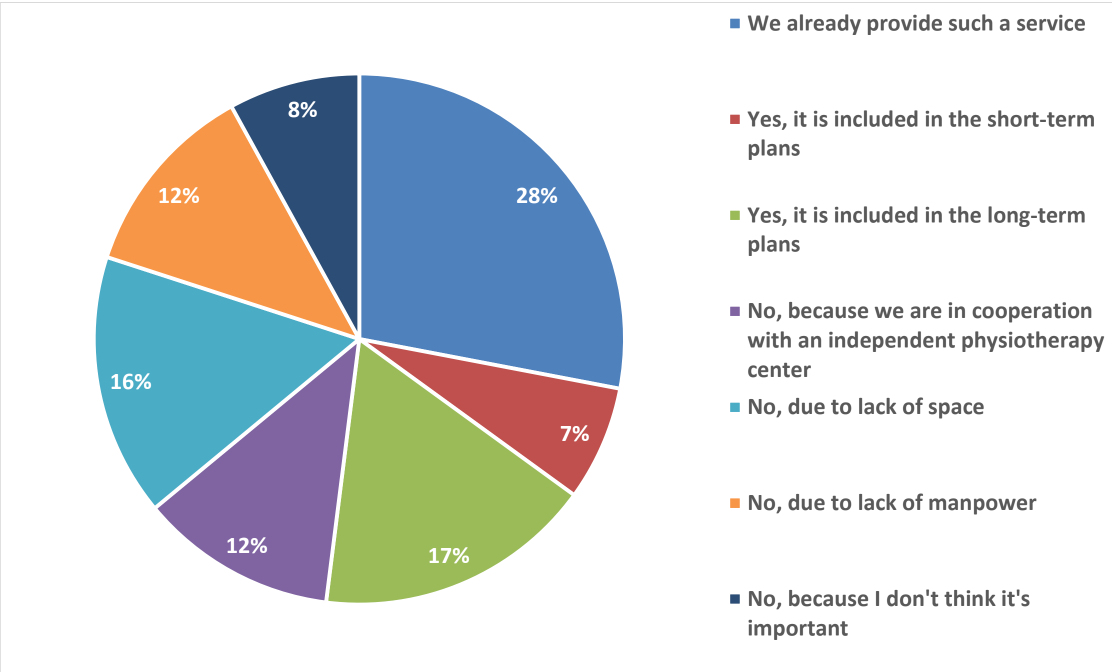

Figure 56. Availability of physiotherapy service in veterinary clinics (n = 100)

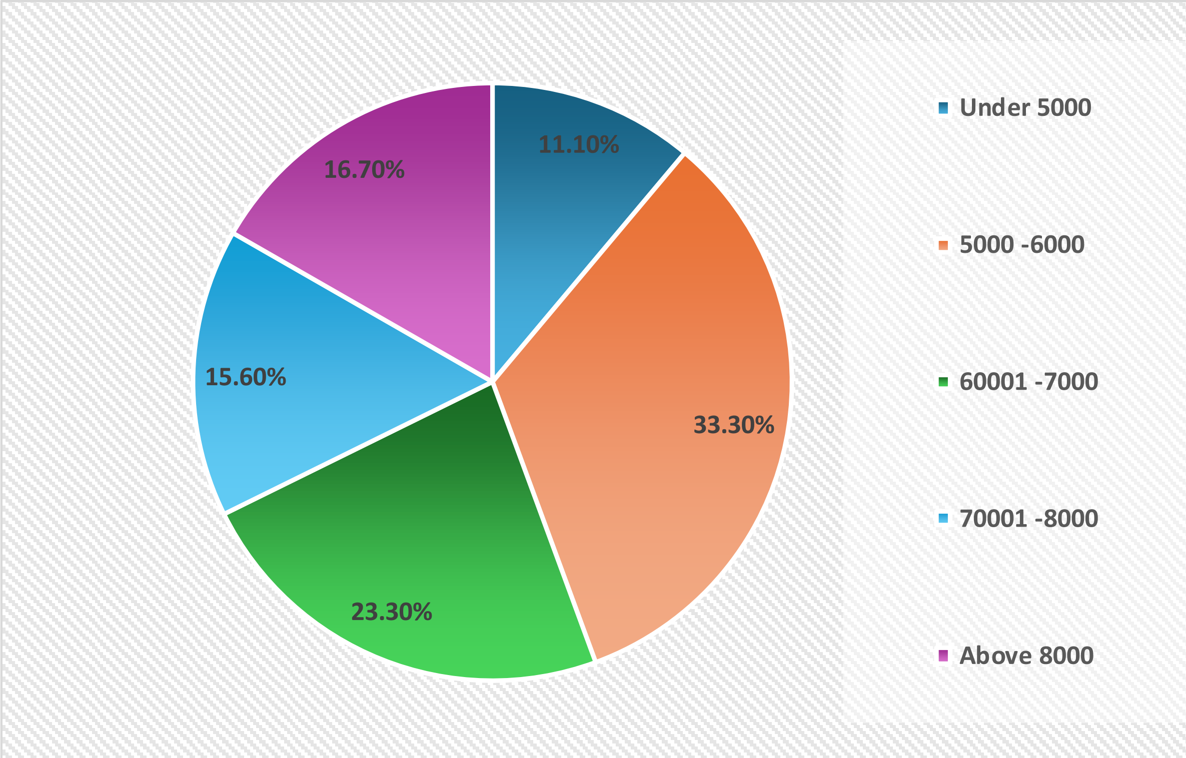

Figure 57. The expected amount per physiotherapy treatment by the veterinarians (n = 90)

We also asked the veterinarians “how much they would invest in physiotherapy equipment" (Figure 11). Overall, it can be concluded that there is a strong willingness to invest among veterinarians, as nearly a third of the 85 respondents (32.9%) would be willing to invest between RMB 101,000 and RMB 500,000, and 47.1% of them would be willing to invest more than half a million HUF in physiotherapy equipment translate. (It should be noted that 3.5% of them would invest more than ten million RMB!)

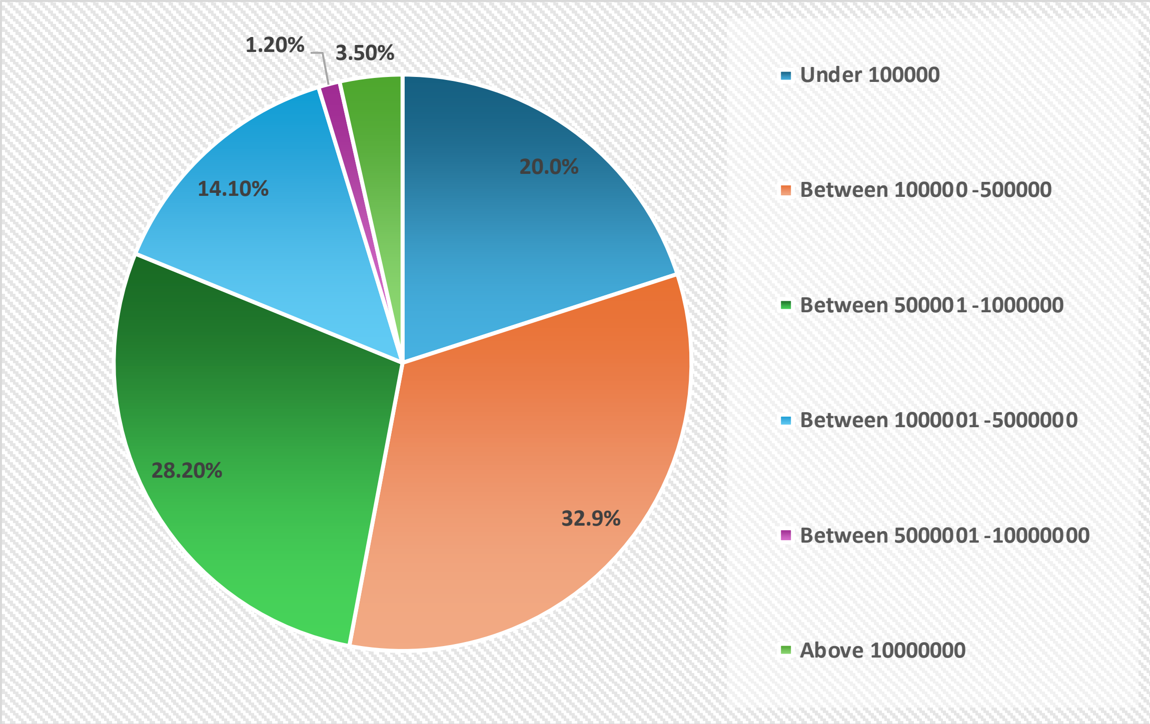

Figure 58. The amount that veterinarians are willing to invest in physiotherapy equipment (n = 85)

Physiotherapy works with a complex set of tools. Manual therapies, such as massage, passive movement of joints, and stretching, play an important role in pain relief and maintaining or improving joint movement paths. At the same time, heat, magnet, and laser therapy are often used, as well as ultrasound treatment, which is most often used in the case of arthrosis, muscle spasms, contractures or tendinitis, or different electrotherapy devices, e.g., TENS device, which can play an important role in pain relief, or selective stimulus current, which can be used to induce muscle contractions [3, 19]. However, physiotherapy can only be completed with active gymnastics, a special version of hydrotherapy, which includes swimming and special movements on an underwater treadmill. This is one of the areas of physiotherapy that is perhaps the most widespread in the public's mind. It is no accident, as it can be used for various problems. Its popularity is shown by the fact that there were already 152 hydrotherapy centers in Great Britain in 2010. Still, it is thought-provoking that only 51% of the employees had formal qualifications in the subject [20].

Almost half of the interviewed veterinarians (47%) would send both a veterinarian and an assistant to physiotherapist training, 19% would train the assistant, and 10% would train the veterinarian (n=100). Most veterinarians, therefore, want to be trained as veterinarians in this field, which suggests that they want professional veterinary supervision in connection with the physiotherapy treatment, regardless of who performs the additional therapy. At the same time, almost a quarter of the responding veterinarians (24%) would not send anyone to such training, which may be because they do not consider physiotherapy important in the surgery. Still, they may already be working with a qualified physiotherapist who is not a veterinarian or an assistant but is satisfied with his work. The importance of training is also shown by the fact that veterinarians typically refer patients to physiotherapists who better understand the methods used in physiotherapy [3, 16]. This resonates with the opinion of veterinarians working in horse practices, who believe that those veterinarians who use alternative and complementary therapies, such as, e.g., physiotherapy, gain proficiency. Their attitude towards these therapeutic methods is also more positive. However, it was proven that equine veterinarians acquired most of their knowledge on this subject after graduating from university, and there was a general demand for further training opportunities on the subject [18].

If the interviewed veterinarians were to send either a veterinarian or an assistant from among their employees to physiotherapist training, 24.7% would finance the entire fee for the training, 35% would finance half of the fee, and 4.3% would finance an even smaller portion of the tuition fee (n=93). In addition, 35.5% of the responding veterinarians did not consider it important to pay their share of the training costs, which may be because many physiotherapists started looking for a job in a veterinary clinic or cooperating with the clinic after completing their self-financed training. The cooperation between the veterinarian and the physiotherapist is beneficial for the physiotherapist because this way, he can gain recognition in the profession and can provide a more comprehensive treatment. It is beneficial for the veterinarian because, in this way, he can gain knowledge in the field of rehabilitation, which is useful even if he were to carry out this activity himself. in addition, he can make a more confident decision during referral [21].

16.6 Conclusions

Most pet owners know the role of physiotherapy in rehabilitating animals' locomotor organs and would typically use it if necessary. Most pet owners would prefer an established handler over a therapist who comes to the house, so establishing and maintaining physiotherapy handlers may be worthwhile, even next to a veterinary clinic. With a suitable physiotherapy caseload, it is probably more worthwhile to maintain a room because the patient traffic per time unit will be higher. Thus, the income and the additional costs incurred (utilities, amortization, etc.) will be distributed among more treated animals. Thus, the average cost per case will also decrease, not to mention the user cost of the travel time during disembarkations. Veterinarians may want to educate themselves in physiotherapy further to confidently decide when to refer a patient to physiotherapy and when not. A significant number of pet owners trust their veterinarian's opinion, which is why it is important to use this capital of trust to refer their patient to a therapist who is also reliable for the veterinarian, or even better if you can provide this service within the practice. It is already possible to use physiotherapy in many veterinary offices, and even more are planning to introduce it. Because of this – despite the increased demands – one must also prepare for the fact that there will be more competition in this area. In the long term, those willing to make even larger investments can remain successful in ensuring that the patients who come for rehabilitation can receive the widest possible range of care.

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Chapter 17: Prevention and control of Eimeria parasite in poultry

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Summary

An important intestinal parasite illness that affects chickens and causes major financial losses for poultry producers across the world is coccidiosis. It has been suggested that different *Eimeria* species, with differing degrees of pathogenicity, impact hens. Coccidiosis incidence is influenced by host, agent, and management variables in addition to environmental risk factors. In addition to histopathology, the presence of lesions and damaged intestinal tissue may aid in a more accurate diagnosis of coccidiosis. Controlling and preventing coccidiosis in domestic hens requires the use of anticcocidials and excellent management practices. So far, chemoprophylaxis has been the primary method used to control coccidiosis. However, the search for different control strategies has been spurred by the emergence of resistance, consumer apprehensions, growing protocols, and probable future bans on the routine of anticoccidial drugs as feed flavors. Vaccines, phytotherapy, aromatherapy, and pre- and probiotics have all been the subject of extensive research. As of now, live vaccinations have shown to be the most reliable and effective alternative therapy for ticoccidiosis. Live coccidiosis vaccines have a variety of disadvantages, but their effectiveness and capacity to make *Eimeria* spp. isolates more susceptible to anticoccidial medications have encouraged their usage. If the immunogenicity of subunit vaccines can be better-quality, they could characterize the next generation of extremely effectual and low-cost anticoccidial approaches.

Key words

Protozoa; Coccidia; Chemotherapy, Drug Resistance

17.1 Introduction

Parasites, especially *Eimeria*, which has a major effect on poultry, impede the development and economic viability of developing agricultural nations(Abbas, Iqbal et al. 2017). *Eimeria*, which affects the digestive system, is frequently contracted by chickens through the use of tainted feed or water. Chickens are affected by 9 species of *Eimeria*, the most virulent of which are E. brunette, E. maxima, E. necatrix, and E. tenella(Abdisa, Hasen et al. 2019). Severe symptoms include bloody diarrhea, enteritis, dysentery, stunted development, and high mortality are transported on by these parasites. The commercial broiler industry agonizes significant financial losses because to the occurrence of *Eimeria* in tropical and subtropical climates. Although coccidiosis is treated with a variety of medications, these therapies are becoming less fruitful due to rising resistance. This calls for other strategies, such immunization. This research aims to investigate strategies for managing and preventing *Eimeria* parasite in chickens(Fatoba and Adeleke 2020).

17.2 Economic Importance

One of the poultry illnesses that obstructs the expansion of this sector is coccidis. It is a complicated illness of chicken brought on by several *Eimeria* parasite types. Both symptomatic and subclinical types are afflicted in birds. The disease's clinical form is considered by conspicuous symptoms of death, morbidity, diarrhea, or bloody stools. Sub-clinical coccidiosis, on the other hand, is mostly categorized by poor weight growth and reduced feed conversion efficiency, which accounts for the main share of overall economic losses(López-Osorio, Chaparro-Gutiérrez et al. 2020).

17.3 General information of avian coccidiosis

Over 1000 species of *Eimeria* have been labelled to communicate a disease to dissimilar host animals such as chicken, turkey, cattle, duck, rabbit, sheep and domestic dog and cat. Four species of chicken—E. tenella, E. maxima, E. mitis, E. acervulina, E. brunetti, E. praecox, and E. necatrix—have been described. The degree of pathogenicity exhibited by these species is correlated with the extent of damage they induce in several parts of the gut(Hou, Wu et al. 2022). The primary cause of coccidiosis in chickens is *Eimeria* oocyst, which is contracted by the birds by consumption of contaminated food, water, or litter. Personnel moving from home to house may also carry the oocyst that sick birds generate on their fecal discharge to the chicken house. The majority of broilers and other commercially raised birds for meat develop in settings that facilitate the disease's fast spread(Haque, Sarker et al. 2020).

17.3.1 Lifecycle

All *Eimeria* species go through 2 or more generations of shizogony, an asexual developmental stage, before entering a sexual phase called gametogony, which produces oocysts. Figure 1 depicts the infective stage, sporulated oocyst, which is bought and emerge sporocysts and sporozoites in the duodenal lumen due to the impact of mechanical and chemical stimuli in the gut (bile salt and trypsin)(Kalita, Sarmah et al. 2018, Burrell, Tomley et al. 2020).

The sporozoites may travel the whole length of the alimentary canal before occupying the mucosa. The next stages of the life cycle contain intracellular development and asexual multiplication, followed by the periodic issue of merozoites that enter the gametogenesis, or sexual, phase of the life cycle. After invading cells, these merozoites mature into macro- or microgametes(López-Osorio, Chaparro-Gutiérrez et al. 2020). Whereas the male gametocyte grows and ruptures to issue seven enormous numbers of minute biflagellate micro-gametes, the former produces a distinct macrogamete. A micro-gamete is created while the micro-gametocyte matures. When the macro gamete is inseminated by a microgamete, a thicker wall around it develops, producing a zygote. The juvenile or immature oocyst is this stage(Burrell, Tomley et al. 2020).

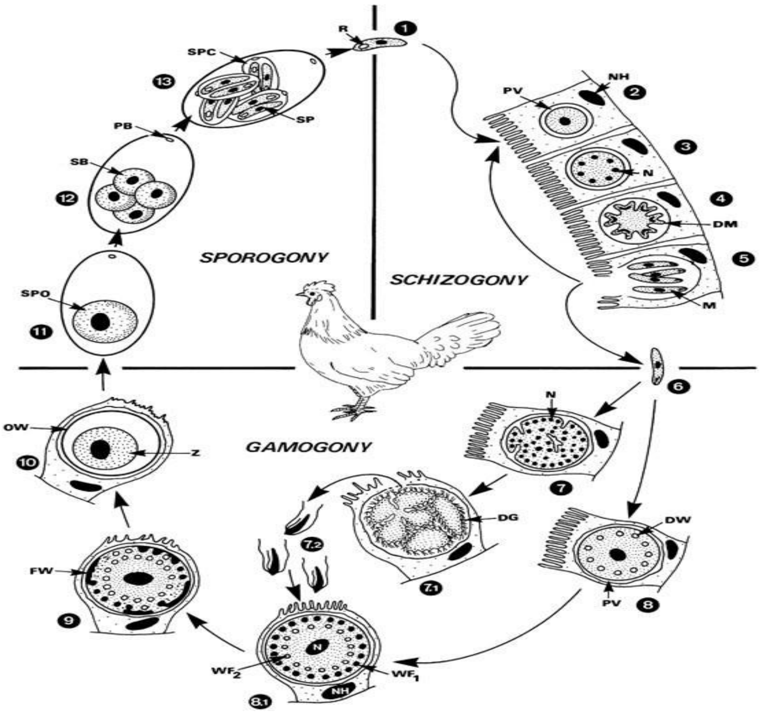

Figure 59 Infective stage proceeding sporulated oocyst, which releases sporocysts and sporozoites due to the impact of mechanical and chemical stimuli in the gut (Heinz 2016)

17.3.2 Epidemiology

Poultry housing and management practices that are subpar encourage the illness coccidiosis. In the intense deep litter system, the illness is more widesprea. Chickens of all ages can contract coccidiosis, although the sickness usually starts in younger birds while their immune systems are still developing(Speer 2019). A coccidia infection epidemic happens when a lot of sporulated oocyst is consumed by chicks. One of the indications of infected birds is reduced food intake; other symptoms include bloody diarrhea and weight loss(Abbas, Iqbal et al. 2017). The no. of *Eimeria* species that co-infect the poultry determines how severe the sickness is. Among the 7 species of *Eimeria* that infect chickens, E. tenella, E. brunetti, E. maxima and E. necatrix have been documented as very pathogenic while E. praecox is the least pathogenic(Matsubayashi, Shibahara et al. 2020).

17.3.3 Prevalence

Parasitic infections are among the many ailments that affect poultry, and they are particularly important in rural hens. Common parasites seen in poultry include helminths, fleas, coccidia, lice, ticks, and mites. The majority of poultry birds have several intestinal parasite infections, which further adds to their widespread dissemination(Diao, Zhao et al. 2022). Numerous studies have documented the occurrence of the *Eimeria* parasite in diverse farming settings throughout different regions of South Africa. Specifically, the frequency rate of 29.46% of *Eimeria* parasites among indigenous chickens in KwaZulu-Natal and Limpopo has been observed. *Eimeria* parasite-related mortality rates of 54.3, 31.7, 70.9, and 52.9% have been documented in Turkey, India, Ethiopia, and Nigeria, in that order(Heinz 2016).

Chicken *Eimeria* infections result from many *Eimeria* species co-infection, which can produce mild to severe lesions. Certain chicken *Eimeria* species inhabit diverse parts of the GI system. For example, E. acervulina grows in the duodenum, E. maxima and E. mitis grows in the interior of the small intestine, and E. tenella, E. brunetti, and E. necatrix raises in the caeca, rectum, and small intestine. It has also been noted that the poultry management system and favorable weather have an impact on the incidence of *Eimeria* infection in hens(Huang, Ruan et al. 2017).

Table 21. Some characteristics of important Eimeria spp. infecting chickens(Bangoura and Daugschies 2018)

Host	Eimeria	Location	Pathogenicity*
Chickens	E. acervulina	Duodenum, Jejunum	*
	E. brunetti	Ileum, Rectum	**
	E. necatrix	Jejunum, Caeca	***
	E. maxima	Duodenum, Jejunum, Ileum	***
	E. tenella	Caeca	***
	E. praecox	Duodenum, Jejunum	*
	E. mitis	Duodenum, Jejunum	*

17.3.4 Identification and Diagnosis

Eimeria species were documented by cross-immunity, lesion investigation, development location, and pathogenicity prior to the outline of genetic tools. Coccidiosis was verified by pathological and morphological investigations, which included oocyst forms and lesion locations. Accurate species identification is vital for successful therapy. Taking curvature, form, and texture into account, COCCIMORPH, a computer software that uses oocyst morphology, provides an alternate identification technique(Kalita, Sarmah et al. 2018). Despite intraspecific and intragenomic differences, PCR-based techniques and qPCR tests using ITS-1, ITS-2, and SCAR markers have also been busy. The investigations of the cytochrome c oxidase subunit 1 (COI) gene are promising, however they do not have a lot of reference sequences. Uniting different loci, such as ITS and COI, is a novel strategy under investigation(Bangoura and Daugschies 2018).

17.4 Treatment, Control and Prevention

Anticoccidial feed additives have greatly extended the chicken sector over the last fifty years, guaranteeing the supply of reasonably priced, high-quality poultry yields. The main methods of preventing coccidiosis, which is taken on by the parasites Isospora and *Eimeria*, are immunization and chemoprophylaxis. The most popular method is chemoprophylaxis, which involves feeding anticoccidial products to animals. Drug resistance, however, has reduced their efficacy, igniting interest in ancillary control tactics such immunization, sanitation, and theoretical genetics. Anticoccidial substances in feed or water, together with appropriate management—with a focus on dry litter, particularly near water and feeding areas—are the keys to boost coccidiosis prevention. Because most harm happens before symptoms manifest and since medications cannot totally stop an outbreak, preventive medications, also known as coccidiosistats, are utilized. Better control measures are obligatory because coccidiosis, which mostly affects the intestines of hens grown in high-density environments, reasons large economic losses—an estimated €2 billion yearly(Bawm and Htun 2021).

17.4.1 Anticoccidial Products

Since the introduction of sulphaquinoxaline and nitrofurazone in 1948, various anticoccidial drugs have been approved by the American Food and Drug Administration to fight coccidiosis in chickens. Table 2 lists the anticoccidial products currently permitted in different regions worldwide. However, national monitoring restrictions may apply to some products. Unfortunately, no new drugs in either group have been approved for use in decades. Resistance to all currently used medicines has been acknowledged, highlighting the critical need for determining new drugs with unique modes of action to guarantee chemotherapy remains a viable primary method for controlling this infection(Abudabos, Alyemni et al. 2017).

Table 22 Common anticoccidial products in specified poultry species(Thabet, Zhang et al. 2017)

Anticoccidial chemicals	**Concentration in feed (ppm)**	**Poultry category**
Amprolium	125–250	Broiler, rearing
Aprinocid	125–250+4	Broiler, rearing
Decoquinate	32	Broiler
Nequinate (methyl benzoquate)	18	Broiler, rearing
Polyether ionophores		

Lasalocid	75–125	Broiler
Monensin	100–120	Broiler, rearing
Narasin	60–80	Broiler
Semduramycin	20	Broiler

17.4.2 Drug Categories

1. Synthetic compounds, commonly known as "chemicals," are made through chemical synthesis and contain several anticoccidial drugs. These drugs act by numerous mechanisms: inhibition of parasite mitochondrial respiration (e.g., decoquinate, clopidol), inhibition of the folic acid pathway (e.g., sulfonamides), competitive inhibition of thiamine acceptance (e.g., amprolium), and some with unknown modes of action (e.g., diclazuril, halofuginone, nicarbazin, robenidine)(Teng, Fuller et al. 2020)
2. Polyether antibiotics, or ionophores, including monensin, narasin, salinomycin, maduramicin, semduramicin, and lasalocid, are made by fermenting Streptomyces spp. or Actinomadura spp. They disrupt ion gradients through the parasite's cell membrane.(Thabet, Zhang et al. 2017)

17.4.3 Mode of action

A general classification of the mechanism of action of anticoccidials on the absorption of the parasite has been attempted, despite the fact that precise knowledge on the discerning action of anticoccidial compounds against certain stages of the organism is sometimes inadequate.

Table 23 Mode of action of Anticoccidial products

	Examples	**Mode of Action**	**Targeted species of *Eimeria***	**Citation**
1. Products affecting cofactor synthesis	Ethopabate	It is a folate antagonist that inhibits a stage in the creation of para-aminobenzoic acid (PABA) and stops the development of vitamin nucleic acids. It is frequently used in conjunction with amprolium to increase the range of activity.	*E. maxima and E. brunetti*	(Noack, Chapman et al. 2019)
	Sulphonamides	By intrusive with the dihydropteroate synthetase process and preventing the conjugation of pteridine and PABA, one can	*E. brunetti, E. maxima, E.*	(Chapman and

		stop the production of dihydrofolate. Only the parasite contains dihydropteroate synthetase.	*acervulina, E. tenella* and *E. necatrix*	Rathinam 2022)
	Pyrimethamine	Pyrimethamine inhibits the reduction of dihydrofolate to tetrahydrofolate by inhibiting dihydrofolate reductase. It has a strong synergistic effect when combined with sulfonamides.	*Eimeria tenella*	(Inoue, Tsujio et al. 2019)
	Amprolium	Completely stop thiamine from being absorbed; this will likely have a negative impact on the availability of vitamin B1, as thiamine is most needed during schizogony.	*E. tenella* and *E. acervulina*	(Aubert and Favennec 2017)
2. Products affecting mitochondrial function	Quinolone drugs	These compounds stop the electron transport in the mitochondria of coccidia, inhibiting their respiration.	*E. tenella*	(El-Shazly, El-Latif et al. 2020)
	Meticlorpindol	It may hinder electron transport in mitochondria on a different level since quinolones do not cause cross-resistance.	*E. acervulina*	(Aubert and Favennec 2017)
	Nicarbazin (4,4′-dinitrocarbanilide)	These compounds inhibit energy-dependent transhydrogenase and calcium ion buildup in rat liver mitochondria, as well as the succinate-linked NAD reduction in mitochondria.	*E. acervulina ,E. tenella, E. gallapavonis , E.* and *meleagrimit*	(Ortega 2018)
	Robenidine (a guanidine derivative)	Robenidine's precise anticoccidial mechanism is still a mystery. Nonetheless, it's thought to prevent mitochondria from undergoing oxidative phosphorylation.	Mixed *Eimeria* Species	(Noack, Chapman et al. 2019)

	Toltrazuril	Toltrazuril lessens the activities of succinate-cytochrome C reductase, NADH oxidase, and succinate oxidase in mouse liver.	*E. falciformis* and *E. tenella*	(Odden, Enemark et al. 2018)
3. Products affecting cell membrane function	Polyether antibiotics	Affect how mono- or divalent cations (Na+, K+, and Ca++) are transported across cell membranes to cause osmotic damage	*E. tenella*	(Thabet, Zhang et al. 2017)
	Ionophore anticoccidials	Permit the development of immunity in contradiction of coccidia	Mixed *Eimeria* Species	(Ferdji, Mimoune et al. 2022)

17.4.4 Products affecting cofactor synthesis

A number of anticoccidial medicines impact a crucial cofactor, which in turn affects vital metabolic processes within the parasite cell. The illness known as coccidiosis, which is brought on by protozoan parasites in the intestines of animals, especially chickens, is prohibited and treated with anticoccidial medications. By focusing on different phases of the parasite's life cycle, these products lessen its capacity to proliferate and spread illness. Their devices of action regularly entail interfering with biochemical actions, such as cofactor synthesis, that are crucial to the parasite's survival and reproduction(Ahmad, El-Sayed et al. 2016).

17.4.5 Products affecting mitochondrial function

Anticoccidial products target vital mechanisms for energy generation and metabolism, impairing mitochondrial activity in coccidian vermin. They prevent electron transfer, which is a necessary step in the creation of ATP, by inhibiting the electron transport chain (ETC) with medications such as atovaquone and decoquinate. ATP group is further hindered by ionophores like monensin and lasalocid, which alter mitochondrial membrane potential and ion gradients. Furthermore, several substances diminish essential protein production by impeding mitochondrial enzymes or interfering with the replication and transcription of mitochondrial DNA. This complex disturbance causes the parasite to lose energy, converts oxidatively stressed, and eventually dies(Awadi 2017).

17.4.6 Products affecting cell membrane function

Anticoccidial products cause cell death by intrusive with the function of the coccidian parasites' cell membranes. They interfere with cellular homeostasis by changing the permeability and integrity of membranes. Antibiotics that are ionophores, like monensin and lasalocid, cartel with cations like Na+, K+, and Ca2+ to create complexes that lead to uncontrollably high ion transport across membranes. This causes osmotic imbalance, bump, and cell rupture by upsetting ion gradients that are essential for cellular activity. Furthermore, several anticoccidial drugs change the fluidity and permeability of membranes by cooperating with the lipids or proteins in the membrane. These perturbations effectively destroy the parasite's capacity to controller waste elimination, intracellular environment, and nutrition intake(Pérez-Fonseca, Gutiérrez et al. 2022).

17.4.7 Products with unknown mode of action

Diclazuril is a nucleoside analogue that may have an impact on the later stages of coccidia differentiation and be tangled in nucleic acid synthesis. In both E. brunetti and E. maxima, it has been demonstrated to impact parasite wall mixture, leading to the development of an strangely thickened, inadequate oocyst wall and zygote necrosis. A derivative of quinazolinone, halofuginone primarily affects the first generation of the schizogony and has an unclear mechanism of action(Balta, Marcu et al. 2021).

17.4.8 Growth-promoting and Antimicrobial properties

It has been discovered that ionophores inhibit mycoplasmas and other Gram-positive bacteria. In hens and turkeys, it has been demonstrated that monensin and narasin suppress Clostridium perfringens (types A and C). Therefore, in certain circumstances, ionophores may have helped suppress necrotizing enteritis. It has been demonstrated that salinomycin decreases the quantity of streptococci (erythromycin and lincomycin) and resistant coliforms. It also appeared to lower the quantity of Salmonella typhimurium germs that were resistant(Wang, Peebles et al. 2019).

17.4.9 European Union regulations

Anticoccidial medications' use as feed additives is disbelieving in the future. The 2008 study from the European Commission suggested keeping them in use since there were no apposite alternatives and because of financial apprehensions. It's uncertain, though, if the European Council would heed this advice. The webpage of the European Commission about feed additives for animal nourishment provides updates on rules(Gilbert, Bellet et al. 2020).

17.4.10 Resistance

Anticoccidial medications are widely used worldwide, although resistance to certain of them—quinolones and pyridinols, for example—develops quickly, while resistance to ionophores takes longer. Because of trickle infections that promote immunity, coccidiosis epidemics have had little effect in Europe despite resistance. Strategies like rotation—using a medicine for no more than two months or two fattening periods—and shuttle programs—using various medicines within a fattening period—are used to reduce resistance. These approaches seek to curtail the period of drug experience and prevent cross-resistance by using medications with different mechanisms of action(Flores, Nguyen et al. 2022).

17.4.11 Vaccines

Passive or active immune responses can produce immunity to coccidiosis, which includes improving bird performance, lowering oocyst output, diminishing lesions, and construction resistance to *Eimeria* spp. infections. Vaccines against coccidiosis have been divided into subunit and live vaccines within the last 20 years. Subunit vaccines, such as CoxAbic®, have not been very positive commercially because they lack powerful antigens, while comprising purified antigens from the parasite's embryonic stages. Live vaccinations can be attenuated or non-attenuated and are made up of sporulated oocysts. Attenuated vaccines, like Livacox®, employ strains with decreased virulence, whereas non-attenuated vaccines use intact wild-type strains. When utilizing drug-sensitive live vaccines, it is authoritative to remove anticoccidial medicines from the feedstuff in order to avoid immunization failures. AdVENT®, Immucox®, Inovocox®, and CocciVac® are a few non-attenuated vaccinations(Venkatas and Adeleke 2019).

17.5 Environmental Management

To reduce the burden of infectious illnesses, good health and an appropriate immune response are necessary. Therefore, general management practices that protect the fundamental needs of poultry should constantly be put into place. These practices include giving birds the right kind of food and water to drink, as well as correct bedding, temperature, humidity, lighting, and ventilation.

In order to prevent the parasite from entering the building and to control its spread in the event that flocks have been sick, management and biosecurity measures for the control of coccidiosis should be prioritized. Strict biosecurity that results in flocks free of coccidiosis is practically unachievable in reality, though. Consequently, the slow development of immunity through recurrent mild (trickle) infections was recommended as a means of controlling this illness already in the early days of coccidiosis research.

Because *Eimeria* parasites are so common and have such a large size for reproduction, contaminated chicken buildings and the areas around them are highly contaminated. The parasite's long-term life in the environment is further safeguarded by the oocyst wall, which shields it from desiccation and chemical disinfectants. From there, it can enter a farm in a number of ways. However, if the farm home was not sufficiently disemboweled and disinfected, oocysts could already be there.

Similar to steps used to avoid other infectious poultry illnesses, biosecurity efforts to stop the entrance of *Eimeria* parasites to the farm should concentrate on the following:

1. Isolation. Fencing should be used to keep birds apart from the surrounding area and to keep other animals, such as rodents and insects, out.
2. Not only should the farm implement traffic management, but there should also be limitations on traffic flow between farms.

3. Cleaning supplies, personnel, and equipment before they reach the farm and poultry house is part of sanitation.

The environment of the chicken house cannot be altered to manage coccidiosis in hens, as the same conditions that encourage the parasite also benefit the birds. The disease is instigated by *Eimeria* spp. To become infectious, *Eimeria* must sporulate after being expelled in feces; crucial variables include temperature, humidity, and aeration. Temperatures between 24 and 28°C are ideal for sporulation, but anything outside 35°C kills oocysts. While dry litter increases the rate of sporulation, moist litter is not as good because of bacteria and ammonia. It is not real-world to increase litter humidity to limit sporulation since this would hurt birds. In addition to being vital for avian health, adequate ventilation encourages sporulation. By lowering the amount of oocysts that accumulate in litter, plummeting bird density can aid in the management of *Eimeria* infections(Aguiar-Martins, Burrell et al. 2023).

17.5.1 Alternative coccidiosis control

Over the centuries, a variety of alternative therapies, including homeopathy, phytotherapy, and aromatherapy, have been employed to treat a wide range of poultry illnesses. Homeopathy is a medical treatment method that involves giving little amounts of a medication that, when taken in large quantities, causes clinical symptoms in healthy people that are identical to the disease's symptoms. The most similar therapy will heal, according to the rule of similars, similia similibus curentur. It is believed to strengthen the body's defensive mechanisms. Herbalism, often referred to as botanical medicine is sometimes mistaken with homeopathy. Its foundation is the application of plants and plant extracts to illness prevention and treatment(Khater, Ziam et al. 2020).

On occasion, its purview is expanded to encompass fungi, items made by bees, minerals, shells, and certain animal components. Aromatherapy, or essential oil therapy, is the use of fragrant molecules derived from plants to treat or prevent disease. It is closely associated with phytotherapy. Utilizing distilled plant volatiles is one of the innovations of the 20th century. Essential oils are not chemically similar to other herbal products since only lighter phytomolecules are recovered during the distillation process(Ademola, Ojo et al. 2019).

The scientific community continues to argue the usefulness of homeopathic treatment for humans, despite the publication of several articles on the subject. A dearth of data supporting the effectiveness of homeopathic therapies is reported by studies assessing the methodological quality of several human homeopathic trials due to publication bias and frequently low methodological worth. Velkers et al. state that "medication efficacy should go beyond the placebo effect and observation and interpretation should be free of subjectivity." Subsequently, a double-blind, randomized clinical study should be used to assess all medications, including homeopathic therapies. Homeopathy in poultry has not been extensively studied in the literature, and like human research, it is occasionally hampered by poor methodology. Scientific studies on the homeopathic remedy are nonexistent(Salman, Abbas et al. 2020).

While certain pharmaceutical medications produced from plants have found their way into mainstream medicine, the majority of herbal remedies were produced without current scientific validation. numerous research has shown unfavorable effects, even if in vitro animal models and/or clinical trials have subsequently revealed numerous herbs to be effective. However, because location bias occurs in the equivalent controlled clinical studies, assessing the data on complementary and alternative therapies is challenging. Published trials are more favorable than negative, with the exception of high-impact conventional medical publications. Additionally, positive research are often of lower quality than equivalent negative studies in publications of complementary and alternative medicine. The latter was not the case with publications of mainstream medicine that published articles on a broader spectrum of treatments.

Alternative approaches to controlling coccidiosis have also included pre- and probiotics, despite the fact that they differ greatly from phytotherapy and aromatherapy. Probiotics are live bacteria or yeasts that are added to food and are thought to improve a person's health. Prebiotics, on the other hand, are indigestible dietary components that also improve the health of the host(Abbas, Iqbal et al. 2017).

17.5.2 Plant (herb) extracts

Various herbal compounds have been discovered as potential dietary supplements to cope coccidiosis, as described below.:

Artemisinin

Artemisinin, an mine from Artemisia annua and Artemisia sieberi, showed encouraging effects against E. tenella, mixed results for E. acervulina, and negative results for E. maximas(Pop, Györke et al. 2015)

Betaine

Sugar beets contain a crystalline alkaloid called betaine, which stabilizes cell membranes to prevent osmotic stress and helps cure metabolic problems. It has the potential to fight coccidiosis by strengthening cell resistance and preventing the growth of parasites. It has a palliative impact on the infection when taken with salinomycin, as seen by the dramatic improvements in feed efficacy and weight gain observed in infected birds. It's possible that betaine's capacity to protect cells from infection is what makes it possible for it to encourage weight increase in chicks. It may also have a beneficial effect on coccidiosis since it strengthens immunological responses in affected chickens(Ademola, Ojo et al. 2019).

Citric extracts

When used in conjunction with organic acids and citric extracts, a product for broiler supplementation demonstrated a reasonable level of efficiency against a variety of *Eimeria* species when coccidiosis lesion scores and oocyst output were taken into account(Ishaq, Sani et al. 2022).

Echinacea purpurea

The immunomodulating characteristics of E. purpurea, which have been extensively studied, have been linked to its anticoccidial efficacy. When broilers were given ground root planning of E. purpurea (0.1–0.5%) for two weeks, it was shown that this decreased weight gain retardation and coccidial lesions following a diverse infection with E. acervulina, E. maxima, E. tenella, and E. necatrix at the age of 28 days.

Gentian violet

Gentian violet, which is generated from coal tar and has antifungal and antibacterial qualities, is also known by the names crystal violet and methyl violet. It has been demonstrated to improve weight growth in birds challenged with *Eimeria* spp. and lower duodenal coccidiosis lesion scores. When used with anticoccidial medications, it increased feed conversion

Mushrooms and their extracts

Because of their ability to boost immunity and have antitumor characteristics, mushrooms and their extracts have received attention in both medicine and as dietary supplements. But, they should be handled with caution since mushrooms may contain radioactive contamination with 137Cs and hazardous quantities of metals (such as arsenic, lead, cadmium, and mercury)(Ademola, Ojo et al. 2019).

Astragalus membranaceus, Tremella fuciformes, and Lentinus edodes polysaccharide extracts all had a beneficial effect on the cellular and humoral immunity of female broilers infected with E. tenella. In a further trial, the birds given the same extracts showed improved growth during vaccination compared to the vaccinated birds who were not treated, and they also had fewer E. tenella oocyst counts following challenge.

When injected into 18-day-old embryos, extracted lectin from the fungus Fomitella fraxinea prevented weight loss in broilers infected with E. acervulina one week post-hatch and was linked to a marked decrease in oocyst shedding in comparison to untreated embryos.

Oregano

It is well known that Origanum vulgare essential oils have antibacterial and anti-parasitic properties. Additionally, albeit less so than lasalocid, several oregano essential oils, particularly thymol and carvacrol, have an anticoccidial action against E. tenella. However, no protective impact of these oils on a coccidiosis vaccine was shown in later tests using diets supplemented with a combination of the essential oils, oregano, thymol, eugenol, curcumin, and piperin(Bozkurt, Ege et al. 2016).

Turmeric (Curcuma longa)

The rhizomes of the C. longa plant provide turmeric, which is a phenolic chemical with antitumor, anti-inflammatory, and antioxidant properties. Diets complemented with 1% curcumin enhanced weight gain, decreased lesion scores, and decreased excretion of oocysts in chicks infected with *Eimeria* maxima. Its effectiveness, however, was restricted to E. maxima rather than E. tenella(Cervantes-Valencia, Alcala-Canto et al. 2015). Curcumin's anticoccidial efficacy may be attributed to its antioxidative properties, which include suppression of NOS induction.

Incidental reports on the anticoccidial activity of other herb(s) extracts

For simplicity, the anticoccidial action of several plants and extracts has been compiled into a single text and is shown in Table 3. To our knowledge, none of the trials that indicated anticoccidial action were replicated(Aguiar-Martins, Burrell et al. 2023), and as a result, none of these drugs have been used widely in run-through.

Pre-and probiotics

Prebiotics, defined as non-digestible food ingredients stimulating beneficial bacteria growth, include Mannan oligosaccharides (MOS), known to enhance gastrointestinal health and immune retorts. MOS reduce severity of *Eimeria* infections in poultry, though efficacy fluctuates with dosage and pathogen strain. Probiotics, live microorganisms conferring health benefits, exhibit promising results against coccidiosis by bolstering protection and reducing pathogen proliferation(Abu-Akkada and Awad 2015). Lactobacillus-based probiotics mitigate coccidia development, while commercial strains like Pediococcus show resistance enhancement and reduced oocyst shedding. Uniting prebiotics and probiotics in synbiotic approaches offers potential synergistic benefits in poultry health management.

17.6 Conclusion and Recommendation

Thus, in light of the foregoing conclusions, the following suggestions are made:

- Appropriate diagnostic techniques and biosecurity precautions ought to be used in order to stop and manage the illness in the hens.
- It is recommended to use appropriate preventive measures and management strategies to mitigate the economic burden of coccidiosis in developing nations.
- More research is required to develop long-lasting and affordable preventive and control measures for this economically significant parasite illness of chickens.

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Chapter 18 : Veterinary Medicine and Microbiome; Uncovering the Hidden World

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Abstract

The prevention, diagnosis, and treatment of diseases, injuries, and disorders in animals are focused on veterinary medicine which encompasses enhancing animal health and productivity. Furthermore, it also plays a crucial role in protecting human health from zoonotic diseases by ensuring food safety through the management of food animal populations and developing new treatments. Moreover, it involves the education and training of veterinarians and animal care professionals and microbiome consists of microorganisms like bacteria, fungi, viruses, and parasites which inhabits different parts of the body including the skin, oral cavity, gut, and reproductive organs. These microorganisms help in digestion, regulating the immune system, and protecting against pathogens due to are essential to maintaining host health. The scope of veterinary medicine is broad in animal welfare and preventive measures such as vaccinations and parasite control, diagnostic procedures, surgical and clinical treatments, emergency care, wildlife conservation, and the development of new therapies. Veterinary medicine is divided into several specialties' large animal medicine, small animal medicine, equine medicine, exotic animal medicine, wildlife medicine, veterinary dentistry and veterinary surgery. The microbiome significantly influences host health by playing roles in nutrient absorption, immune system enhancement, and protection against diseases.

Keywords: Veterinary Medicine, Zoonotic Diseases, Animal Health, Microbiome, Animal Nutrition, Probiotics and Prebiotics, Microbiome-based Therapies

18.1 INTRODUCTION

Veterinary medication is the part of medication that deals with the counteraction, analysis, and treatment of diseases, problems, and wounds in animals (Aderibigbe, 2024). It encompasses advancement of animal wellbeing and productivity. Protection of human wellbeing from zoonotic illnesses (sicknesses contagious among animals and people). Guaranteeing food handling and security through the administration of food animal populations. Innovative work of new medicines, treatments, and items for animal wellbeing. Education and preparation of veterinarians, animal researchers, and animal care experts (Aderibigbe, 2024).

18.1.1 Scope of veterinary medicine

Veterinary medicine covers a broad range of areas focused on animal health and well-being. This includes preventive care like vaccinations and parasite control to keep animals healthy. It also involves diagnosing illnesses through methods like imaging and lab tests and providing treatments including surgery and other medical procedures. Emergency and critical care are crucial for urgent situations. Food animal medicine ensures the health and productivity of animals raised for food. Wildlife medicine helps protect and treat animals in natural habitats. Research is ongoing to develop new treatments and therapies. Education and training are essential for veterinarians and animal care professionals to provide the best care possible (Anderson & Hobson-West, 2024; Mader et al., 2022).

18.1.2 Types of veterinary medicine

There are many specialized fields in veterinary medicine that veterinarian's study. Following types of veterinary medicine are given below:

Large Animal Medicine: In this field veterinarians specialize in the care, health and treatment of larger animals, like cattle, buffalo, horses, pigs, and other livestock. Albendazole, Anthelmintic (to kill helminths in ruminants) and other drugs are used for the treatment of large animals (Khan & Witola, 2023).

Small Animal Medicine: The most common type of veterinary medicine, pay attention on the health and care of companion animals such as cats, dogs and small mammals. Common drugs like penicillin, enrofloxacin that reduce pain, swelling and inflammation in cats and dogs (Khan & Witola, 2023).

Equine Medicine: This branch is related to veterinary medicine of horses and especially focus on the health and well-being of horses, including pleasure horses, performance horses, and racehorses (Keener & Tumlin, 2023).

Alien or non- native Animal Medicine: Some veterinarians particularize in the protection of exotic or non-native animals, involving reptiles, birds, small mammals, and in fact zoo animals (Abbasi et al., 2019).

Wildlife Medicine: This branch of veterinary medicine is linked with uncultured or undomesticated life medicine and veterinarians focus on the health, conservation and management of wild animals, treating damaged or unwell wildlife and driving investigation on wildlife diseases (Aderibigbe, 2024).

Veterinary Dentistry: In veterinary medicine dentistry pay attention on the oral hygiene and teeth care of animals, including extractions, dental cleanings, and oral surgeries (Mader et al., 2022).

Veterinary Surgery: Veterinary surgeons apply surgical procedures on animals, ranging from regular altering and neutering to multiplex orthopedic surgeries (correction of deformities) (Khan & Witola, 2023; Thayer et al., 2022).

18.1.3 Veterinary medicine encompasses various specialized fields based on discipline, approach, and practice type.

Discipline-based specialties include veterinary surgery, cardiology (heart conditions), neurology (nervous system disorders), oncology (cancer treatment), ophthalmology (eye care), dermatology (skin issues), and radiology (imaging techniques). Each of these areas focuses on specific aspects of animal health and requires specialized knowledge and skills.

Approach-based classifications in veterinary medicine include traditional veterinary practices, holistic veterinary medicine which considers the whole animal's health, integrative veterinary medicine combining conventional and alternative therapies like acupuncture and homeopathy, and alternative veterinary medicine that exclusively employs non-traditional treatments.

Practice types within veterinary medicine further delineate its diverse applications. These include private practices where veterinarians treat companion animals or livestock, academic veterinary medicine involving teaching and research at universities, research veterinary medicine focusing on scientific discovery and innovation, government veterinary medicine addressing public health and animal control, and corporate veterinary medicine linked to industry and pharmaceuticals. Each practice type serves distinct purposes in advancing animal health, welfare, and scientific knowledge (Meisner et al., 2022).

Different veterinary medicines along with their effects on different animals are discussed in the Table 1 below.

Table 24:

Veterinary medicines	**Animal**	**Effect**
Tetracycline	Buffalo, Cattle, Sheep, Goat, Poultry	To treat bacterial infections like pneumonia, bronchitis, diarrhea and enteritis. Treat urinary tract infections (Aderibigbe, 2024; Chai et al., 2021).
Fenbendazole	Cattle, Buffalo and other animals	To treat parasitic infections (internal parasites including nematodes, tapeworms and flukes. Treat

		diarrhea, abdominal pain, and weight loss (Chai et al., 2021).
Ketoprofen, Meloxicam	Cattle, Buffalo, Horses, Dogs, Cats	Treat pain and inflammations in animals associated with conditions like arthritis and musculoskeletal injuries. Helps animals move more comfortably (Taylor et al., 2024).
Estradiol	Cattle, Buffalo, Horses, Dogs, Cats	Is a hormone used in reproductive purposes like ovulation, enhance fertility and successful breeding and help induce parturition in pregnancy. Treat heat cycle regulation and uterine infections (GOUGEON, 2004).
Coccidiostats	Cattle, Buffalo, Sheep, Goat, Poultry	Help prevent the development of coccidiosis. They decrease the number of oocysts, breaking the parasite life cycle. Reduce the risk of illness in animals (GOUGEON, 2004).
Xylazine	Horses, Cattle, Buffalo, Sheep and Goat	Reduce pain and anxiety during medical procedures(surgery). Induce soothing effect on animal body during surgeries or vaccination. It comforts the animal.
Kaolin	Cattle, Buffalo, Sheep, Goat, Dogs and Cat	Treat diarrhea in animals but don't treat severe diarrhea in animals. Protect the gut lining, mucous membrane in animals and reduce inflammation in gut (Akdis, 2006).
Monensin	Cattle, Buffalo and Cow	Improve fiber digestion, Feed efficiency, improve the energy status in animals and weight gain.
Ivermectin	Dogs	Used to treat lungworms, roundworms, hookworms, fleas and ticks. Treat heartworm disease that caused by parasitic worm Dirofilarial

		immitis (Langston & Varela-Stokes, 2019).
Betadine	Cattle, Sheep, Dogs, Cats and Horses	Used for wound care, minor cuts, burns, skin irritations. Promote healing, kill bacteria and fungi. Used in surgical preparations (Babalska et al., 2021).
Enrofloxacin	Dogs, Cats	Used to treat bacterial infections. Treat dermal infections, respiratory infections (rhinitis, pneumonia) and urinary tract infections (Ahmad et al., 2021).
Palladia	Dogs, Cats	Used to treat tumor and cancer in dogs and cats. Reducing the growth of tumors. Inhibits protein that promote cancer cell growth and divisions. Treat pancreatic cancer and gastrointestinal tumors. Stimulate the immune response against cancer (MAEDA, 2023).

18.2 Micro-biome

The micro-biome is the assembly of microorganisms including parasites, bacteria, fungi, viruses, and other microbes, that live in and on the bodies of living entities (Zhang et al., 2021).

These microorganisms play crucial roles in sustaining the health and functioning of their host organisms. They were found in different parts of the body like skin, oral cavity, gut and reproductive organs so due to this reason, they involved in prime activities like digestion, immune system regulation, and protect host body against causative agents (Gouba et al., 2019).

18.2.1 Types

There are different types of micro-biome.

Vocal micro-biome

Found in the oral cavity(mouth) including the teeth, gums, and tongue. These are involved in breaking down food and maintaining oral health (Deo et al., 2019).

Nasal micro-biome

Found in the nose serve to filter the air we breathe and save against harmful infectious agents (Shilts et al., 2020).

Cutaneous micro-biome

Present on the cutaneous surface and in skin layers (epidermis, dermis, hypodermis) helps to protect against dangerous pathogens and maintain skin health (Luna et al., 2020).

GIT micro-biome

Found in the gut involved in digestion and absorption of food stuff. They are involved in maintenance of the immune system (Gomaa et al., 2020).

Vaginal miniature biome

Found in the vagina safeguard female conceptive organ and forestall hurtful contaminations (Lewis et al, 2017).

Soil miniature biome

Tracked down in soil that assistance in rotting natural matter, diazotrophy, and upgrade soil fruitfulness (Yadav et al., 2021).

Marine miniature biome

Tracked down in seas, ocean that associated with rotting, upgrading marine supplements and keep up with marine environment (Korajkic et al., 2019).

Air miniature biome

Found in the air that we inhale associated with upgrading indoor and outside air quality (Moelling et al., 2020).

18.3 Job of miniature biome

Microorganisms in the body assume significant parts in keeping us sound. They shield our skin from destructive microorganisms that can cause diseases. Inside our stomach related framework, they assist with separating food and retain supplements that our body needs to develop and remain sound. Microorganisms likewise assist our resistant framework with warding off infections by making it more grounded. They are engaged with making chemicals and different synthetic compounds that our body uses to work appropriately (Qi et al., 2021).

Microorganisms additionally assist with keeping our body's major areas of strength for hindrances, the skin, stomach, and lungs. This forestalls contaminations and aggravation, which can make us debilitated. They even assist with managing significant chemicals like insulin and estrogen, keeping our body adjusted and solid.Experts are also focusing on how microorganisms could help in treating threatening development by supporting our protected structure's ability to fight it.

Microorganisms aren't just important for us — they moreover expect a section in keeping the environment clean by isolating harmful substances like significant metals, toxins, and poisons. As a rule, minute living things have various fundamental places that add to both our prosperity and the sufficiency of our natural elements.

The job of microbiome in drug digestion

The microbiome expects a huge part in staying aware of the body's balance of metabolites, which are central for handling starches, proteins, and fats. These creatures produce metabolites that serve both as fuel for cells and as hailing particles that influence significant metabolic cycles. They also impact energy balance by overseeing hankering and food affirmation, in this way affecting how much energy the body eliminates from food.

Microbially-derived metabolites enter the flow framework and speak with the protected, metabolic, and tangible frameworks of the host, as needs be influencing different physiological abilities. With respect to retention, the microbiome impacts how lipids are absorbed, set aside, and used for energy, which can have ideas for conditions like chubbiness and atherosclerosis. Additionally, in carb handling, the microbiome influences glucose homeostasis, insulin hailing, and glucose processing in the liver.

Moreover, the microbiome expects a section in drug processing by changing the development of synthetic substances and transporters drew in with drug taking care of, hence impacting the feasibility and toxicity of remedies.Moreover, the microbiome uses dietary amino acids like tryptophan, phenylalanine, and tyrosine, which can influence safe reactions, aggravation, and neurological capabilities. Generally speaking, these communications feature the multifaceted manners by which the microbiome impacts different parts of human wellbeing and digestion (Nichols et al., 2019).

18.4 Effect of miniature biome on creature wellbeing

18.4.1 Positive effect:

The microbiome is engaged with food assimilation and supplements retention in the host group of creatures. Manage metabolic exercises. They are engaged with the support of GIT like associated with the forestalling flawed stomach, inside disorder and so on.They play a crucial role in the immune system. Enhance the immune system of the host body and protect it from diseases. They are involved in regulation of mental health like stress, anxiety etc. in the host body. Produces various vitamins and hormones that are required in an animal body. They act as a soldier for skin because they protect host skin from various causative agents. Involved in treatment of cancer by inhibiting the growth of tumor. Micro-biome helpful in reducing the aging processes of the host organisms (Boyajian et al., 2021).

18.4.2 Negative impacts:

Healthy microbes are necessary for digestion and absorption of nutrients, an inequality or disturbance in the micro-biome can causes various health issues. For example, some bacteria present in the gut can produce bad toxins or causes inflammation, leading to digestive issues or diseases. It is crucial for animals to maintain an equilibrium and multiple micro-biomes for optimal health.

18.4.3 A few problems from upset miniature biome

The microbiome, especially in the gastrointestinal plot (GIT) or stomach, assumes a basic part in wellbeing, and unsettling influences can prompt different issues. Awkward nature in the stomach microbiome are related with conditions like the runs, provocative entrail sickness (IBD), and touchy gut disorder (IBS). Past the stomach, the microbiome likewise impacts the insusceptible framework, and interruptions can add to safe related issues like sensitivities and immune system sicknesses. Besides, changes in the microbiome are connected to metabolic issues like hypertension, corpulence, diabetes, and metabolic disorder in creatures. As far as skin wellbeing, disturbances in the skin microbiome can prompt circumstances like sensitivities, contagious contaminations, skin inflammation, and dermatitis. Additionally, changes in the microbiome can expand defenselessness to contaminations, including respiratory plot diseases and urinary parcel diseases (UTIs). Generally speaking, keeping a good arrangement of the microbiome is essential for forestalling and dealing with an extensive variety of medical problems in creatures (Madhogaria et al., 2022).

18.5 Variables influencing the microbiome

A few variables can upset the equilibrium of microorganisms, organisms, and infections in creatures' microbiomes. Anti-toxins, while pivotal for treating bacterial contaminations, can likewise upset gainful microorganisms in the gastrointestinal parcel (GIT), possibly adjusting the generally microbiome structure. Dietary elements assume a critical part too; an eating routine ailing in dietary fiber and fundamental supplements can decrease microbiome variety and wellbeing, while a reasonable eating regimen upholds a flourishing microbiome. Stress and uneasiness additionally influence microbiomes by impacting synapse and chemical creation, which thus can upset the equilibrium of stomach microorganisms. Natural factors like openness to poisons, synthetic compounds, and poisons further add to microbiome changes and effect creature wellbeing. Moreover, the microbiome normally goes through changes with age, which can prompt changes in bacterial arrangement, decreased microbial variety, and an expansion in unsafe microscopic organisms, all influencing GIT wellbeing. Dealing with these elements is essential for keeping a solid microbiome and generally prosperity in creatures. The maturing system causes changes in the stomach climate, for example, diminished stomach motility, changes in pH levels, which causes aggravation in microbiome. Flourishing microbiomes in creatures can be adjusted by keeping away from superfluous utilization of anti-infection agents, keeping a sound way of life, and giving legitimate sustenance (R Gacesa et al., 2022).

18.6 Hardships and restrictions of inspecting and controlling the microbiome

Examining the microbiome represents a few difficulties because of its tremendous variety and intricacy. Containing a huge number of various microorganisms, the microbiome is trying to independently completely distinguish and concentrate on every species. Moreover, microbial networks shift essentially founded on the particular district of the body, with assorted creatures flourishing under various circumstances on the skin and inside different organs. One more hindrance lies in understanding the full utilitarian limit of the microbiome in human wellbeing and illness, as this stays an area of progressing examination and revelation. Both characteristic and outward factors impact the microbiome, including factors like skin area, hereditary qualities, way of life, maturing, and method of conveyance upon entering the world. Moreover, various microorganisms inside the microbiome are problematic or challenging to culture in an examination office setting, which tangles tries to focus on their characteristics and correspondences totally. Overcoming these troubles requires state of the art advancements and interdisciplinary ways of managing unravel the confounded components of the microbiome and its ideas for prosperity and contamination.

Controlling the microbiome presents enormous challenges in view of its marvelous relationship among microorganisms. Attempts to control the microbiome can incite incidental antagonistic outcomes, as the associations between different creatures are awesome and not totally seen. Also, the shortfall of standardized shows for controlling the microbiome makes it hard to see results across changed assessments, obstructing progress in this field. Prosperity concerns in like manner arise, as controlling the microbiome could really introduce perilous microorganisms or upset profitable ones.Moral contemplations add one more layer of intricacy, with discusses encompassing the moral ramifications of modifying the microbiome for purposes past clinical treatment. Administrative obstacles further entangle endeavors to control the microbiome, as clear rules and rules are missing to guarantee the wellbeing and adequacy of such intercessions. Tending to these troubles requires cautious logical examination, moral consultation, and administrative structures to dependably investigate and deal with the microbiome's true capacity for remedial and different applications (TS Rasmussen et al., 2020).

18.7 Connection between veterinary medication and microbiome

The microbiome incorporates trillions of microorganisms (like parasites, microscopic organisms, and so on) that dwell in and on the assemblages of creatures. Veterinary medication recognizes the meaning of the microbiome in supporting the general strength of creatures.

It plays a role in absorption and assimilation of food, natural immunity, and in fact psychological health. Veterinarians are working that how microbiome affects numerous situations in animals and researching different ways to utilize microbiome-based therapies to make better their health (KC Banks et al., 2019).

18.7.1 Microbiome- based therapies

In veterinary medicine an example of microbiome-based therapy is the use of **fecal microbiota transplantation (FMT**). FMT includes passing healthy microbiota from a contributing animal to a beneficial animal to refresh a maintained microbial community in the beneficia's gut. This therapy gives encouraging results in treating some gastrointestinal (GI)disorders in animals, like chronic diarrhea (loose motions) or irritating intestine disease. By refilling the beneficial bacteria in the Gastrointestinal tract, FMT can aid to renovate digestive health (MC Niederwerder, 2018).

18.7.2 Impact of veterinary medicine on microbiome:

Veterinary medicine has both positive and negative effects on the microbiome. On the positive side, it assists in stopping diseases in animals, which can incidentally advantage the microbiome through enhancing overall health. Although, some drugs used in veterinary medicine, like antibiotics can disturb the equilibrium of the microbiome.

Positive impacts

Veterinary medicine plays a crucial role in promoting the well-being of animals, which in turn positively impacts their microbiome.One critical way veterinary medication contributes is through the treatment of diseases brought about by microbes or parasites. By really focusing on these microorganisms, veterinary drugs assist with reestablishing harmony to the microbiome. Moreover, veterinary mediations in parasite control assume an imperative part in disposing of parasites that can upset the microbiome, in this way encouraging a better microbial climate in creatures. Infection counteraction endeavors, including immunizations and viral sickness the executives, likewise add to keeping a different and adjusted microbiome by keeping creatures sound and versatile.

Nourishing direction given by veterinarians is another fundamental viewpoint adding to a sound microbiome. Legitimate nourishment upholds a different microbial local area, upgrading by and large wellbeing results for creatures. Veterinarians likewise assume a significant part in advancing capable anti-infection use, endorsing anti-microbials wisely to limit disturbances to the microbiome. While medical procedures may briefly upset the microbiome because of sedation and post-employable therapies, cautious administration and observing by veterinarians can uphold the microbiome's recuperation over the long haul.

Also, veterinary medication centers around lessening pressure in creatures, as constant pressure can unfavorably influence the microbiome. Stress the executives systems and natural improvement strategies are utilized to assist with keeping a sound microbial equilibrium. In general, veterinary medication's far reaching approach not just treats diseases and advances wellbeing in creatures yet additionally upholds a flourishing microbiome, fundamental for their general prosperity (S Jin Melody et al., 2019).

Adverse consequence

Veterinary meds, while fundamental for working on creature wellbeing, can accidentally upset the fragile equilibrium of the microbiome, pivotal for generally speaking prosperity. Anti-infection use, vital yet problematic, can aimlessly kill both hurtful and gainful microscopic organisms, prompting awkward nature and expected incidental effects. Ill-advised substance addiction, including anti-microbials and antifungals, further risks microbiome variety, possibly affecting long haul wellbeing. Such disturbances can appear in gastrointestinal aggravations like stomach agony and looseness of the bowels, and may stretch out to heart issues, respiratory troubles, blood problems, neurological side effects, unfavorably susceptible responses, and electrolyte irregular characteristics. Natural variables in veterinary settings, like sanitizers, additionally impact microbiome wellbeing.Veterinarians strive to mitigate these risks, often recommending additional measures like probiotics or dietary adjustments to support microbiome restoration and enhance animal well-being (PC Barko et al., 2018).

18.8 Microbiome and animal nutrition

Examine the Impact of diet on the composition and function of microbiome

Here is the effect of diet on the composition and function of microbiome in veterinary animals:

Diet impacts the composition of microbiome that is present in the gut of veterinary animals. The stomach of ruminants has presented various kinds of microorganisms and their important role in stomach of ruminants have disintegration of food mostly cellulose and hemicellulose. Ruminants' gastrointestinal tract is estimated to be occupied by more than 5000 types of microorganisms. Microorganisms are the most abundant in the gastrointestinal tract of ruminants. Their amount and assortment are influenced by many elements like composition of diet, energy requirements and metabolic results, as some of them might be harmful to specific species. The main role of microbiome in rumen is to convert plant-based diet into energy (M Bergamaschi et al., 2020).

18.9 Diet impacts the equilibrium of microbiome

Diet plays a pivotal role in shaping the gastrointestinal microbiome of animals by influencing pH levels and providing essential nutrients. For instance, diets rich in grains can lower rumen pH, promoting the growth of acid-tolerant microorganisms. Nutrient imbalances can disrupt microbial equilibrium. Probiotics, containing beneficial live microorganisms, and prebiotics, such as dietary fibers that stimulate microbial growth, are crucial for maintaining a healthy microbiome. However, the use of antimicrobial agents like antibiotics, antifungals, and antiseptics in feed additives can inadvertently disturb microbiome balance by eliminating beneficial bacteria. A well-established microbiome can help suppress harmful pathogens in the gastrointestinal tract, thereby supporting overall animal health. Veterinary strategies often focus on optimizing diets to enhance microbiome health and minimize disruptions that could impact animal well-being (SA Bahaddad et al., 2023).

18.10 Probiotics and prebiotics

These two concepts work together to promote the gut health and support immune system.

Probiotics are food supplements containing live microorganisms such as bacteria and yeast, akin to beneficial microbes naturally found in animals. These microorganisms confer several health benefits. They promote efficient digestion, aiding in nutrient absorption and utilization, while also bolstering the immune system to help animals combat diseases. Probiotics enhance feed efficiency, leading to improved growth rates and overall productivity, which can reduce the reliance on antibiotics. In dairy animals, probiotics have been shown to enhance milk production and improve milk quality and composition.Moreover, probiotics assume a part in decreasing methane discharges from domesticated animals, adding to natural maintainability. In poultry, they further develop egg quality, increment egg creation, and add to by and large conceptive wellbeing. By keeping a fair microbiome, probiotics support creature wellbeing, diminish illness dangers, and upgrade the personal satisfaction. Normal regular probiotics for veterinary use incorporate Enterococcus faecium, Lactobacillus acidophilus, Saccharomyces cerevisiae, and different types of Streptococcus. Coordinating probiotics into veterinary practices highlights their part in enhancing creature wellbeing and execution across various species (SG Al-Shawi et al., 2020).

Prebiotics are non-edible filaments that act as nourishment for probiotics, the useful microorganisms in the stomach. By advancing the development of these probiotics, prebiotics assist with keeping a solid stomach microbiome in creatures. This makes a few positive impacts, like managing defecations and forestalling stoppage, reinforcing the safe framework, and decreasing irritation because of their calming properties. Prebiotics additionally support in general body capabilities like digestion, assist with keeping a solid body weight, and further develop glucose levels. Furthermore, they can emphatically affect emotional well-being by lessening uneasiness and stress in creatures. Normal wellsprings of prebiotics for veterinary use incorporate high-fiber takes care of, fructans found in plants like chicory and onions, inulin (a kind of fructan), gelatin from natural products like apples and citrus organic products, oligofructose from leafy foods, kelp, and safe starch from vegetables, potatoes, and entire grains, especially valuable for ruminant creatures. Integrating these normal prebiotics into creature counts calories upholds their general wellbeing and prosperity by encouraging a decent and flourishing stomach microbiome (S Jin Tune et al., 2019).

Synbiotics

Synbiotics is a term that alludes to the blend of prebiotics and probiotics in a singular term or condition, expected to participate to propel a decent in general game plan of the microbiome. The possibility of the synbiotics relies upon the likelihood that prebiotics and probiotics have a synergistic effect, overhauling each other's benefits and provoking additionally evolved stomach prosperity and overall success.

Benefits

Synbiotics, which consolidate probiotics and prebiotics, offer various benefits for creature wellbeing. They improve the colonization and endurance of helpful probiotics in the stomach, prompting expanded creation of useful metabolites like short-chain unsaturated fats. This supports stomach wellbeing and lifts resistant capability, working on by and large gastrointestinal flexibility. Synbiotics additionally help in the productive assimilation and retention of supplements, adding to better supplement usage by the creature. They are known to lighten side effects of bad tempered entrail condition and other gastrointestinal issues, advancing stomach related solace.

Furthermore, synbiotics have been linked to improved mental clarity and mood, potentially through their influence on gut-brain interactions. By enhancing the immune system's capabilities, synbiotics help animals maintain robust health and resilience against infections. Overall, synbiotics represent a comprehensive approach to supporting animal health, leveraging the synergistic benefits of probiotics and prebiotics to foster well-being from digestion to immunity. E.g. Prebiotics strands like inulin or fracto oligosaccharides joined with probiotics like Lactobacillus acidophilus (P Markowiak, K Śliżewska, 2018).

Dysbiosis: (Refers to imbalance in microorganisms).

Dysbiosis is an imbalance or irregularity of the microbiome (microorganisms) that lived in a particular environment, like the human body or an animal’s GI tract. This irregularity in microbiome functions can prompt different medical issues and infections. In basic terms, dysbiosis means that the “Great” microscopic organisms in your body are out of equilibrium, prompting issues like stomach related issues, irritation and sicknesses (Slevin et al., 2023).

Factors that effect on dysbiosis

Dysbiosis, an imbalance in the microbiome, can be caused by several factors (Gomaa, 2020a). A diet high in processed foods, sugar, and unhealthy fats can disrupt the microbiome's equilibrium. Antibiotics, corticosteroids, and NSAIDs can also disturb the balance of beneficial bacteria. Persistent stress, poor sleep quality, and exposure to environmental toxins like pesticides and heavy metals contribute to dysbiosis. Infections from viruses, bacteria, or parasites can further disrupt the microbiome, as can hormonal changes such as those during pregnancy or menopause. Aging and genetic predispositions also play roles in microbiome imbalance. Conditions like irritable bowel syndrome and small intestine bacterial overgrowth are examples of gut disorders that can lead to dysbiosis (Gomaa, 2020b).

18.11 Conclusion

Veterinary medicine plays a crucial role in the prevention, diagnosis, and treatment of diseases, disorders, and injuries in animals by enhancing animal health, productivity, and welfare and also safeguards human health from zoonotic diseases and ensures food safety by managing food animal populations. The scope of veterinary medicine is broad due to encompassing surgical and clinical treatments, emergency care, wildlife conservation, and the development of new treatments and educational programs for veterinary professionals. Simultaneously, the microbiome consisting of diverse microorganisms living in and on animals significantly impacts animal health by aiding digestion, regulating the immune system, and protecting against pathogens. These microorganisms are present in and on various body parts and maintain their host's overall well-being The relationship between veterinary medicine and the microbiome is intricate and veterinary practices are medications and antibiotics that can positively or negatively affect the microbiome. Probiotics and prebiotics are utilized to maintain or restore healthy microbial communities by promoting better digestion and immunity with overall health in animals. Understanding and managing the microbiome is essential in veterinary medicine because it influences various physiological processes and disease states. In contrast, the ongoing research aims to leverage microbiome-based therapies like fecal microbiota transplantation, to treat gastrointestinal disorders and enhance animal health. While challenges exist in studying and manipulating the microbiome due to its complexity advances in this field hold promise for improving the health and well-being of animals through veterinary medicine (Jin Song et al., 2019).

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Chapter 19 : Nutritional Profile of Black Soldier Fly (Hermetia illucens) Larvae for Poultry Nutrition

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Abstract

Potential protein-rich food replacements for chicken diets might be insects. The Black Soldier fly (BSF), *Hermetia illucens*, is incredibly ensuring insect species for economic production because of its high-level protein and fat content, which supports its potential use in chicken feed constitution. The protein content and amino acid profile of BSF larvae are similar to those of various feedstuffs that are high in protein, such fish and soybean meal. By transforming organic wastes into beneficial nutrients like proteins, lipids, and chitin, BSF helps lessen the load on the environment and the risk of pollution that results from the expansion of organic waste. This chapter highlights the value of using this insect as a "green" technology to reprocess organic waste in a highly variable way and provide a sustainable supply of protein. It also focusses on the relevance of employing this insect to restore feedstuff that is extreme in protein in chicken feed manufacture.

Keywords: Black Soldier Fly; *Hermetia Illucens*; digestibility; poultry; growth performance; nutritional value

19.1 Introduction:

Since poultry has a lower carbon and water footprint than other animal proteins, it is the most popular category of livestock. This trend is expected to continue as the world's population, which is anticipated to reach 9 billion people till 2050 and demands more animal protein, particularly chicken(Abd El-Hack et al., 2020) (Nijdam, Rood, & Westhoek, 2012) . It is anticipated that dietary preferences would continue to shift away from plant-based proteins and toward animal-based proteins including milk, fish, meat, and eggs. This will put further strain on the chicken industry to fulfill the rising demand for meat and eggs (Hafez & Attia, 2020; Korver, 2023), hence expanding the demand for the need to produce more meat and eggs from poultry. The increasing need for animal products is driving up the demand for components for animal feed, a trend that is expected to persist. To meet this expanding need, non-traditional feed resources are also gradually taking the place of traditional feedstuffs (Belghit et al., 2019).

Furthermore, climate change and global warming , as well as rising feed and energy prices, may have an impact on the amount and quality of animal feedstuff, such as maize and soybean meal, which are important constituents in chicken feed (Nkukwana, 2018). This might have an impact on global food security. For instance, one of the most often used components in chicken feed formulations is soybean meal, which serves as a source of protein (Adli, 2021). But in recent times, the overprice of this component has emerged as a significant ultimatum to the long-term viability of the chicken industry, especially in developing nations.

In multiple nations, insects are being included into human diets as value-added food items (Borrelli et al., 2017), They are also being utilized as a cattle feed substitute in place of traditional protein components like fish or soybean meal (Sajid, Asghar, Tariq, Wilk, & Płatek, 2023; A. A. Shah et al., 2022). Since ancient times, people have utilized insects as food, and many places in the world still consume them now (Liceaga, 2022). However, as there is considerable resentment toward their intake in some countries, utilizing insect meal for poultry feeding is more acceptable to consumers than direct human ingestion (Kröger, Dupont, Büsing, & Fiebelkorn, 2022). Numerous studies have now clearly demonstrated the high protein, necessary amino acid content, mineral, and vitamin content of edible insects (Spranghers et al., 2017).

Numerous studies have shown that using insect meals as food or a feed source can help address the world's food crisis in the long run (De Marco et al., 2015; Kouřimská & Adámková, 2016). Regarding the suitability of certain insect meals as feed requirements for livestock animals (pigs, poultry, and fish), numerous extensive studies have produced some intriguing results (Fitches et al., 2019; Henry, Gasco, Piccolo, & Fountoulaki, 2015; Vernooij, Veldkamp, & Ndambi, 2019). Insects can be directly employed to solve problems related to organic waste, which is produced in enormous quantities and may be environmentally burdensome, in addition to being used as feedstock (Abd El-Hack et al., 2020) . Furthermore, studies have shown that non-pest fly species can reduce manure bulk by 50% and nitrogen and phosphorus wastes by up to 75% in swine and poultry farms. The possibility of using insect meal as feed may potentially have positive environmental impacts because raising insects uses less energy and little land, leaving a smaller environmental impact (Nyakeri, Ogola, Ayieko, & Amimo, 2017). Because of their high edible protein content, insects like black soldier fly larvae (BSFL), mealworm larvae, and crickets are gradually gaining their recognition as an alternative feed resource in swine and poultry diets (Liu et al., 2022; A. A. Shah et al., 2022). Of all these species, BSFL is characterized by high feed conversion ratio, an acceptable survival rate, and a stable provision of phosphorus and nitrogen in the diet. As a result of feeding on a diverse diet of biomolecules and other foods, BSFL has a higher FCR, short generation intervals, and contain plenty of fat, protein, minerals and vitamins than the other two. Thus, BSFL emerges as a more cost-effective method of generating animal feed for consumption that is equally environment and nutritionally friendly(Salam et al., 2021; M. A. H. I. H. Shumo, 2020).

They are also highly sustainable for the same reasons that aquatic feed can be produced on a significantly larger scale in organic streams than can other protein sources, at considerably less environmental cost (Meneguz et al., 2018; Tian, Lu, Paengkoum, & Paengkoum, 2020). From several research it has been postulated that, the black soldier fly larvae may be employed as a food or feed source to feed the increasing world population which in the long run may help in solving the problem of food shortage in the world. Yet, because consumers develop psychological barriers functioning against the consumption of insects, customers prefer to use BSFL in animal feed and not direct use in human diet. (Liu et al., 2022; Raman, Stringer, Bruce, & Chong, 2022). Thus, the inclusion of BSFL in diets appear to have better growth performance and digestibility compared to the normal protein fed diets in pigs and poultry (Zuki et al., 2021). The aim of this chapter is to enlighten the scientists as well as the members of poultry industry about the nutritional quality of BSF, its prospectus as an innovative protein source in poultry feeds, and its usability for the conversion of the undesirable waste into protein rich biomatter by BSF in order to minimize the pollution level.

19.2 BSFL (Hermetia illucens L.)

The primary food source for BSFL (*Hermetia illucens* L. ; Diptera) is indeed this kind of waste matter that constitutes plant residues, animal droppings, debris such as foods, agriculture residues, straw and other forms of organic debris (Nguyen, Tomberlin, & Vanlaerhoven, 2015).To the human and animals, sources of protein from BSFL is an opportunity while recycling organic wastes reduces pollution. Food safety knowledge among cattle farmers was demonstrated with statistics (Purba, Yuangklang, & Paengkoum, 2020; Purba, Yuangklang, Paengkoum, & Paengkoum, 2021). Through its biodegradation process of organic waste, BSFL synthesizes amino acids, peptides, proteins, chitin and vitamins that serves Chemical Industries, Companies dealing in Pharmaceuticals as well as Companies that deal in feeds for pets, pigs and chickens. This process also helps in controlling pests and certain pathogens such as Salmonella and Escherichia coli. Primarily found in South America, BSF may survive in a variety of settings, including temperate, subtropical, and tropical ones. Temperatures between 25 and 30 degrees Celsius are excellent for its survival. (M. Shumo, Khamis, et al., 2019; Wang & Shelomi, 2017). Tentacles and wings are brownish, extending from the head, this huge, thin, black species is divided into three segments: the head, thorax, and belly (Salam et al., 2022). The abdomen has five segments that are speckled with white. Males have smaller wings and end genitals than females, but they are longer overall. Females range in length from 12 to 20 mm, with wingspan of 8 to 14.8 mm (Lu et al., 2022).

Figure 60. Discrimination of female and Male of BSF

There are five phases in their life cycle: egg, larva, pupa, pre-adult, and adult shown in **fig 2**. With around 18–33% fat and 32–53% protein, the larval and pupal stages are the most nutritious and heavily dependent on the quality of diet (Amrul et al., 2022). The lifespan of BSFL is approximately 20 to 22 days, with the first 6 to 8 days spent as a pupa and the latter 14 days spent as an adult metamorphosis (Singh & Kumari, 2019). Adult worms are harmless to other living things, have no taste or digestive system, and have no attraction for fresh food or humans. They thus don't act as disease-transmission vectors.

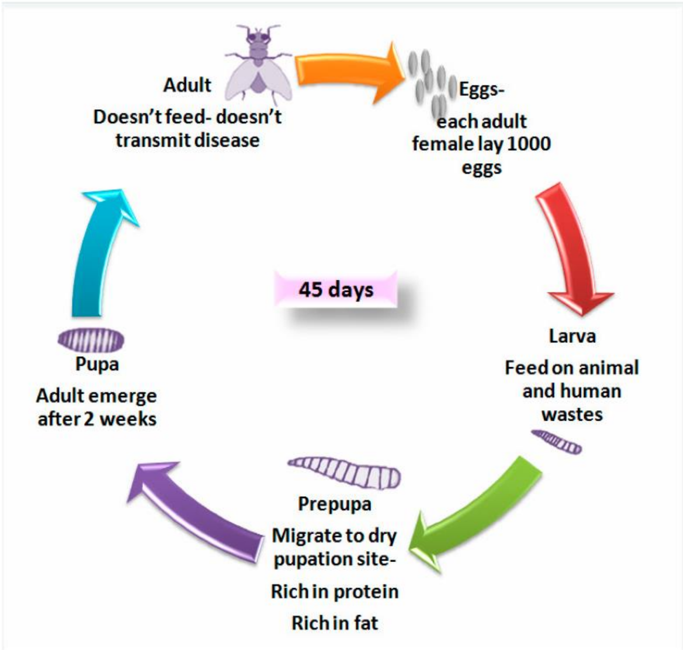

Figure 61. The Black Soldier Fly (BSF) life cycle.

Large volumes of organic wastes, including contaminated animal feed, are transformed into protein mass and soil fertilizer by BSF larvae. Depiction of the process of bioconversion potential of food waste through insect farming for animal feed is shown in **fig. 67**

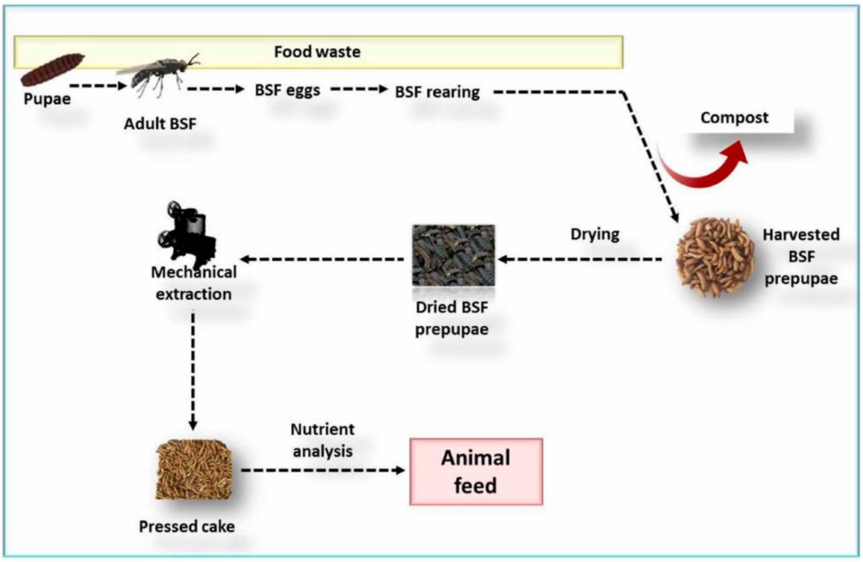

Figure 62: Depiction of the process of bioconversion potential of organic waste through insect farming for animal's feed

19.3 BSFL's (*Hermetia illucens* L.) nutritional value

Black Soldier Fly Larvae (BSFL) are showing promise as a source of protein and fat for animal diets, especially those of chicken. These larvae have a fat content of between 18 and 33 percent and protein level of between 32 and 53 percent, making them incredibly nutritious. A number of variables, including development phase, climatic circumstances, and feed type and quality, affect the nutritional composition of black soybeans. This flexibility enables the nutritional composition to be customized to compensate the unique dietary needs of animals (Yu, Chen, Yu, & Cheng, 2009). When compared to the traditional protein sources such as fish meal and soybean meal, BSFL has higher crude protein level; nonetheless, it is not always constant. Siddquie et.al (Siddiqui et al., 2022) and DE Raedt (De Raedt, 2023) carried out research that show a result that the overall protein percentages range from 21 % to 24 % and 6% to 65. 5%, emphasize this diversity. Thus, while using the percentage compositions of BSFL, it is lower in protein when compared to soybean meal at 49. 4% and fish meal at 67. 5%, they are none the less a source of protein for animal feed. More specifically, broiler chickens' feed compositions are pronounced because fat composition influences the nutritional proportions and energy levels in BSFL (De Marco et al., 2015) . To add to this, these larvae can assimilate a number of micronutrients from their diet which may include minerals and vitamins (Purkayastha & Sarkar, 2021; Rubio, Todolí, Martínez-Sánchez, & Rojo, 2022). For these advantages to be maximized to consumers' benefit, specific suggestions concerning the integration of the BSFL to chickens' consumables must be provided.

The average amount of fiber in BSFL is 9.5%; investigations by Broeckx et al. (Broeckx et al., 2021) showed 4.1%, while Iqbal et al. (Iqbal et al.) found 21.3%. This proportion of fiber is more than fish meal (0.26%) but lower than soybean meal (7.43%). Tyshko et al. (Tyshko et al., 2021),stated that chitin a key ingredient of BSFL is averagely 3. 87% to 7. 21% on average. When BSFL approaches the adult stage of metamorphosis, they tend to have higher fiber contents, which is often correlated with their developmental stage. Thus, literature study on the BSFL has revealed that the chitin content is not constant and it varies with the different research.

19.4 Bio accessibility and stomach digestibility of nutrients from BSFL

Given the fact that its nutritional bioavailability is relatively high as well as its digestibility, this has led to its categorization as a plausible substitute for animal feed primarily for poultry.

19.4.1 Protein Digestibility

Mainly used as the feed ingredient for chickens, Black Soldier Fly Larvae (BSFL) are reported to have high protein content but minimal effects on environmental health. This would indicate the magnitude to which the protein digestibility of the black soldier fly in the chicken larvae changes as the result of the age, processing techniques or the quantity provided as feed to the larvae. Based on the finding of the studies it was observed that, BSFL might be used as a source of protein supplement in animal feed because it could possibly replace fishmeal, insect meal or soybean meal either wholly or partially in feed (Chu et al., 2020; Crosbie, Zhu, Shoveller, & Huber, 2020; Jian et al., 2022; Shelomi, 2020).

In the topic of successors, research established the digestibility of the dried larvae in the form of forage of BSFL is 48 percent. This implies the provision of optimal ways of processing in order to enrich the nutritive value of the protein. BSFL can therefore be identified to be a good replacement of standard protein sources in animal feed, which is about 50% protein. Furthermore, one must mention that the meanings are rising if the inclusion rate is lowered; nonetheless, high protein concentration guarantees the proper absorption of the nutrients (M. Shumo, Osuga, et al., 2019). Conversely, a higher percentage of BSFL at 9% may have negative effect to the digestibility of nutrients and this explains why moderation should always be made at all times when preparing the feed formulation. In other words, this balance is highly important as far as the enhancement of potentials of using the BSFL in animal diets.

19.4.2. Fat Bioavailability

Essential lipid levels are available in Black Soldier Fly Larvae (BSFL), so they are valuable to broilers as an energy source. The ability of hens to utilize these fats is referred to as fats bio availability in poultry and this refers to the health of animals or added diet energy. Chow efficiency of BSFL as an attribute of the feed is positively influenced by their lipid characteristic, and thus any chicken diets need to be formulated with many factors attached to that. The use of BSFL among Laying hen, it is observed that soybean cake and oil can be replaced Fully by BSFL meal and fat which is a very big change from the traditional way of feeding Chickens (Heuel et al., 2021). It was also concluded that diets containing BSFL larval fat reduced the favorability of the chicken meat, and this was particularly true for breast meats (Kierończyk et al., 2023). In essence, it could be concluded that the bioavailability of fat from BSFL diets in chicken avails several prospects. It is in consonance with the need for viable feed production and enhances the nutrient complementary of chicken diets that will positively impact the bird's health and possibly, better quality chicken produce.

19.4.3 Factors Influencing Nutrient Utilization

Among exploitation of larvae, Black Soldier Fly larvae (BSFL) were incorporated into the chicken diets to a greater extent because of numerous nutritional values and the possibility of effective production. Regarding the composition of proteins, BSFL can be classified as those containing greater levels of protein, essential amino acids and fat and that the nutritional structure of BSFL is the primary determinant of the feed supp. These nutritional traits are dependent on the age and the developmental stage of the larvae, what they are fed with which could vary depending on the substrate given to them (Barragan-Fonseca, Dicke, & van Loon, 2017). The methods that are employed in preparing BSFL such as pelleting or drying have great influence on the meals' digestibility and nutrient contents (Lu et al., 2022; Pornsuwan et al., 2023; Weththasinghe et al., 2021). Finally, such parameters as temperature and lodging can effect on feed intake, growth performance and metabolic rate, which in succession affect utilization of nutrients of BSFL (Do et al., 2020; Kierończyk et al., 2022; Maidin et al., 2023). Therefore, knowledge about these relationships is vital for maintaining the chickens and constant production levels and for the efficient incorporation of BSFL into the diet.

19.5 BSFL's Nutritional Value Factors

19.5.1 Growth Performance

In layer diets also when added the defatted BSFL in place of 50% or 100% of soyabean meal there was no impact on feed intake, egg production yolk weight shell weight or mortality (Maurer et al., 2016). But instead of using soybean meal, Heuel et al (Heuel et al., 2022) utilized full-fat BSFL. According to Weththasinghe et al (Weththasinghe et al., 2021) FCR was unaffected on days 1–10 by a 10% BSFL dietary supplement. Chobanova et al. (Chobanova et al., 2023) reported similar outcomes, showing that broiler final weight rose when 10% BSFL was supplemented in the diet. While Kim et al (Kim et al., 2021) found no difference in feed intake or BW when 10% BSFL was supplemented. In concluding the outcome of the above tests, the findings suggest that the positive enrichment of the diet with 10% of BSFL enhances the growth rate of chickens while 100% of SBM retarded it and other ratios have no significant influence.

19.5.2 Immunity and Antioxidants

According to the works of Chen et al (Chen et al., 2022) and Salahuddin et al. (Salahuddin et al., 2024), the impacts of different levels of inclusion of Black Soldier Fly Larvae (BSFL) in the diets of chickens on some of the several metabolic and immune system indicators. What they saw was that some 5%, 10% or 15% of BSFL given to the hens did not influence any parameters of the count of erythrocyte, creatinine level, triglycerides, cholesterol or any other parameter, which defined the TAS (S. R. A. SHAH, 2023). Schiavone et al. point out that these results apply in proving that BSFL possesses antioxidant properties and fails to have unhealthy effects on some biochemical and immunological parameters in hens as a constituent of the diet that is fully BSFL. These outcomes agree with those of Aslam (Aslam, 2023) and KESER (KESER, 2023) who proved that When chicken diets incorporated with BSFL are used, the globulin to albumin ratio increases, followed by a rise in immunoglobulins. This simply means that if no adverse effect or outcome is observed, then the supplementation of BSFL may enhance the immune response and possibly improve the health status of chickens, possibly by increasing their resistance to diseases. The employment of BSFL, as an excellent and efficient basis of chicken feed that supplement the diet, is made possible by its ability to possess antioxidants and boosting the immune system (Abd El-Hack et al., 2020).

19.6 Challenges faced due to Utilization of BSFL *(Hermetia illucens L.)*

19.6.1. Legal and Safety Problems

At this time, food safety laws and regulations are the primary hindrance to the extensive consumption of BSFL in animal feed (Wang & Shelomi, 2017). A substantial obstruction to the growth of insect manufacture units is the stringent European statute regulating the utilization of insects in animal feed, even in the face of significant financing for BSFL research (Rehman et al., 2023). The Global organization Food and Agricultural Organization establishes worldwide food regulations that specify secure conditions for BSFL to meet its needs for producing insects for huff animal feed and farming (Penazzi, 2023). In other words, they shouldn't be protected or categorized as invading persistent species, nor ought they be pathogenic or have any other negative impacts on the health of humans, animals, or plants. They also shouldn't be thought of as carriers of infections that affect any of these.

19.6.2 Consumer Acceptance

It is also important to think about consumer acceptability of animal meat products fed BSFL. The concept of employing insects in animal feed, particularly for poultry and fish feed, is well accepted. In response to a survey, two-thirds of research participants were open to using insects in animal feed (Okello, Otieno, Nzuma, Kidoido, & Tanga, 2023). Similar results were also achieved by Maulu et al. (Maulu et al., 2022), through the utilization of insects in fish feed being the most widely approved. This indicates that there is still room for improvement in consumers' opinions of animal flesh products derived from insects, since there are still certain reservations about these items. Furthermore, consumers may not prefer directly eating BSFL due to the substantial correlation between the probability of CHD and dietary SFA (Kang, 2023).

19.6.3 Production and Price

As of right now, there isn't a single standard for creating and handling BSFL. The small size of the manufacturing equipment, inadequate production, and low efficacy mean that there is just sufficient BSFL for early consumers who will utilize them in minor amounts for scientific research feed (English, Wanger, & Colombo, 2021). The day of bulk manufacturing is still quite a distance away. In this instance, BSFL is not commonly utilized in the feed of chickens or pigs because its accessibility and expense are not comparable with those of soybean meal and fish meal (Lu et al., 2022).

19.7 A Commercial Evaluation of Using BSFL for Poultry Diets

The economic benefits of accumulation of Black Soldier Fly Larvae (BSFL) to chicken diets have been highlighted by recent study, which shows significant feed cost savings without sacrificing production efficiency. The diets containing 20 percent of BSFL had the highest CBR of 2.12 as studied by Waithaka et al . (Waithaka et al., 2022). Lastly, based on this study, it indicates that, indeed, BSFL can reduce the overall feed expenses and also increase the viability of raising meat and eggs production. In a similar way, another study has revealed that establishing higher portion of BSFL in the replacement of the traditional protein sources like fish meal lead the desirability of chicken productions and RoI. These also suggest on the prospectivity of employing BSFL as an economic feed supplement to chicken farming for averting the hitherto associated negative impacts on the environment (Sumbule et al., 2021). Hence, with respect to economic performance of the chicken operations, it is possible to benefit from the application of BSFL as they can maintain or enhance the production returns as compared to more costly proteins demanded (Nampijja et al., 2023).

19.8 Prospects for Research and Development in the Future

The use of BSFL as an organic material as a substitute protein source is receiving more attention due to the increases demand for cheap sources of feeding poultry across the world. However, there are still a lot of study topics that have either not been thoroughly studied or have not been explored yet, which presents an opportunity for more research projects.

19.8.1 Nutritional Optimization

Because of their great fat and protein content, Black Soldier Fly Larvae (BSFL) are becoming more and more acknowledge as a viable source of protein for poultry nutrition. However, depending on the value of the organic waste they eat, BSFL's nutritional makeup might change. When Zulkifli et al (Zulkifli et al., 2022) tried to compare the oven-dried and spray-dried BSFL meals they find out that the former has higher protein content, amino acid and nucleotides. It is for this reason that more focus is placed on understanding the kind of processing techniques that would help retain and perhaps enhance the nutritional value of BSFL in the formulation of chicken feeds.

Therefore, it is essential to look into the various feeding substrates for BSFL and how they affect the nutritional profile of the larvae in order to gain knowledge that might greatly enhance their use in poultry diets.

19.8.2 Effect on the Product Quality and Health of Poultry

In chicken production, feed additives and non-traditional feed components are essential for boosting growth, health, and nutrition while also enhancing product quality (Abbas, 2023; Smolentsev et al., 2023; Vlaicu, Untea, Varzaru, Saracila, & Oancea, 2023). We don't completely understand how BSFL-based diets affect the immune system, resistance to illness, and general physiological health of chickens. To guarantee the safety and caliber of poultry products, further study is thus required to enhance the BSFL's inclusion level, processing technique, and origin in addition to the species, breed, and age of the chickens.

19.8.3 Environmental Techniques for Production and Processing

To improve BSFL's sustainability and feed-sourcing efficiency, production and processing methods must be improved. Numerous factors, like raising circumstances, processing techniques, and the strategy for turning waste into feed, affect the development, survival, and nutritional makeup of BSFL. Improving these variables is essential to raising the yield and quality of BSFL. It was found in studies by Harris & McCabe (Harris & McCabe) that biomass assembly and waste reduction are boosted when a proportion of 100 g of BSFL per Kilogram of substrate is maintained. Further, Tyagi & Aboudi (Tyagi & Aboudi, 2021) discovered that steam exploded and alkali treated maize hay increases bioconversion efficiency and digestibility of feed in the BSFL.

19.8.4 Environmental Impact Assessment

BSFL has been projected to substitute fishmeal and soybean meal conventional feed resources that cause high Green House Gases, deforestation and overfishing. Weththasinghe et al (Weththasinghe et al., 2022) rightly said that the fish oil and fish meal for the aquaculture industry cannot be as efficient as the BSFL oil . As a consequence, it may therefore be proposed that through enhanced usage of BSFL the world may be able to reduce the consumption of traditional feeds, hence the reduction of greenhouse gases output. These might be reduced if renewable energy is used in the generation of electricity for powering the system and nutrient management is enhanced in BSFL systems. Nevertheless, further studies concerning LCA and deeper analysis of broiler production and the valuation of the ecological influences and advantages of BSFL addition to the diet is required to grasp this issue and make it as one of the promising research domains in the future.

19.8.4 Regulatory Aspects and Consumer Acceptance

The two critical factors that affect the suitability of Black Soldier Fly Larvae (BSFL) as a feed supplement within the commercial domains are as follows: The Chicken products consumed by the customers with diets prepared using the chicken soup (BSFL) depend on the information and belief and culture. Another research, carried out by Khaemba et al. (Khaemba, Kidoido, Owuor, & Tanga, 2022), reveals that consumers are more than inclined to buy chicken products that fed with BSFL, and that 71% of them agreed to do so. Analogously, research such as that conducted by Colleta (Colleta, 2023) demonstrates that knowledgeable customers desiring meals based on insects are more likely to purchase meat from hens reared on these kinds of diets. These results highlight the possibility that BSFL will take off in the market, provided that consumers are informed and receptive. Consequently, the improvement and broader acceptance of BSFL as a balanced feed source depend on successfully addressing both consumer and regulatory concerns.

19.8.5 Chronic Health Studies

It is unclear how BSFL-based diets would influence poultry's long-term health. The short-lived consequences of BSFL on the immune system, product quality and growth performance, of poultry have been assessed in a number of studies, with either favorable or neutral findings (Fumo et al., 2023; Lin et al., 2023; Mazzoleni et al., 2023). Thus, to guarantee the security and endurance of this inventive feed cause, it is imperative to carry out longitudinal survey to evaluate the influences of BSFL on the well-being and welfare of weaklings additionally, the possible hazards of disease transmission, allergenicity, and bioaccumulation (Waithaka et al., 2022).

19.8.6 Use of Sub-optimal levels of post-harvest handling particularly in feeding processing and formulation of BSFL in terms of Economics

Thorough investigation should be done to determine whether adding BSFL as an insect or grain to chicken feed is economically feasible in order to maximize the utilization of BSFL. This entails optimizing feed composition to optimize nutritional benefits and feed efficiency, simplifying BSFL manufacturing to cut costs, and enhancing processing methods to guarantee pellet quality and stability(Caminiti, 2023; Kaczor, Bulak, Proc-Pietrycha, Kirichenko-Babko,

& Bieganowski, 2022).According to scientist's research ,the following research questions should be the focus of future research: Relative cost comparisons and cost-benefit analyses of incorporating BSFL into various chicken diets for breeding, race, and layer chickens, suitable market segments and applications for using products produced through the consumption and digesting of BSFL by chickens. Scholarly effort should seek to find ways on better ways of handling BSFLs with a view of achieving the best in feed without compromising on its quality.

19.9 Conclusions

This study shows that insects, when utilized as sources of sustainable protein in poultry diets, can contribute to the realization of the food security challenge and part of the problem-solving process, but with some caveats since this can lead to accumulation of chemicals which are potentially hazardous to food safety. Perhaps the chicken industry will be in a position to generate its source of income independently by utilizing the insects that are fed with organic wastes which in turn are utilized to produce poultry feeds. Based on the literature, the most recent material relating insects as a feed item for chickens can be recommended. It can also be used to replace fish or soybean meal diets of the layer and /or broiler chickens. For this reason, more research has to be carried out to observe whether the high level of BSF larvae meal employed in the manufacture of poultry diets affects the nutritional value, taste, and growth pro les of the meat and eggs. This research must establish the qualitative nature of the fatty acids and chitin substance of the insect before meeting the heath requirements of the poultry or the consumers.

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Chapter 20 : Introduction To Veterinary Parasitology And Life Cycle Of Different Parasites

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20.1 Introduction to Vector Parasitology

Parasitology is the study of eukaryotic parasites such as helminths, protists, and arthropods that consume the host on which they live. Parasites can make an animal sick, so a working knowledge of parasitism is essential for veterinary practice. Parasites can either be endoparasites in the host's body or ectoparasites, organisms that stay outward of their host like on your dog and catch a bedbug. Parasites have long been studied for their relationship with their hosts, from commensalism where a parasite gathers nourishment without harming its host to pathogenicity, which is when parasites cause damage and or disease. Vector parasitology as a field is the study of any known or suspected vectors that transmit parasites to humans and other animals. Understanding and controlling the role of vectors is crucial if endemic parasitic diseases are to be prevented.

20.2 Vectors

Vector is known as a living organism that can transmit a pathogen and disease from one animal to other animal. Arthropods, the largest group of organisms by species up to date and among which insects and arachnids are included, are often deemed as vectors in the field of parasitology. You must know parasites because many important parasitic diseases, particularly agents transmitted to the vertebrate host by arthropod vectors (Table 48-1), are among those entities for which you may be called upon. The result of this with infections in affected areas could be severe health issues to a great deal of financial losses.

Key Vector Groups

1. **Insects**:

20.2.1 Mosquitoes (Culicidae):

Mosquitoes (from the family Culicidae) are small flies with midge-like appearances frequent in many areas of United States which serve as vectors for transmitting a number of diseases, most infamously around here Malaria. Malaria, dengue fever, Zika virus and other diseases are spread by blood-feeding female mosquitoes. They have a life cycle that has eggs, larvae or nymphs and pupae stages from the adult as they pass through various aquatic environments for development till adulthood. Controlling mosquitoes is a major public-health priority, and involves the use of insecticides, environmental management (e.g. clearing potential water-collecting areas such as gutters) and biological control to reduce mosquito populations so that disease virus cannot spread.

20.2.2 Key Facts

- **Size**: The Length of mosquitoes 3 and 6mm.

- **Color**: Brown and gray in general frequently with white green or blue making.
- **Wings**: The size a distinctive vein pattern
- **Antennae**: The male have longer segmented antennae that are thicker than a female.
- **Mouthparts**: Males consume fluid as nourishment, whereas females impale skin with their snouts and suck blood.
- **Life Cycle**

1. A mosquito experiences four unique stages during its metamorphosis.
2. **Egg**

   Laying: The females occasionally store their eggs alone or in groups known as stages in or near standing water.

   Hatching: Depending on the species and the habitat, the time it takes for eggs to hatch into larvae can range from 48 hours to several days.

3. **Larva**

   Appearance: They are aquatic animals with unique heads thoraxes and abdomens. We call them wrigglers.

   Breathing The larvae use siphon tubes to breathe at the water's surface.

   Feeding: They feed on algae, detritus, and marine microorganisms.

   Duration: During their four-day stage, which lasts from four and fourteen days, larvae go through four molts.

   Pupa

   Appearance: Pupae are comma-shaped and more energetic than larvae they are called tumblers.

   Transformation: Pupae do not feed they undergo metamorphosis, transforming into adult mosquitoes.

   Duration: This stage lasts from 1 to 4 days.

4. **Adult**

   Emergence: Adults emerge from the pupal case at the water surface.

   Feeding:

   - **Females**: Seek blood meals for protein to develop eggs. They also devour nectar.

- **Males**: Only nectar and other plant juices should be fed.

Reproduction: After mating, females lay eggs, and the cycle continues.

20.2.3 Habitat and Behavior

- **Breeding Sites**: Mosquitoes breed in stagnant water sources such as ponds, marshes, gutters, containers, and even puddles.
- **Activity**: Most species are crepuscular (active during dawn and dusk), but some are active during the day or night.
- **Lifespan**: Adult females live about 2-4 weeks, while males typically live for a shorter period.

20.2.4 Human Impact and Diseases

Mosquitoes are notorious vectors for several serious diseases:

- **Malaria**: caused by parasites called Plasmodium, which are spread by Anopheles mosquitoes.
- **Dengue Fever**: caused by the dengue virus and spread by Aedes mosquitoes.
- **Zika Virus**: Spread by mosquitoes of the Aedes species, associated with birth defects such as microcephaly.
- **West Nile Virus**: caused by neurological disorders that are spread by Culex mosquitoes.
- **Yellow Fever**: Also spread by *Aedes* mosquitoes, causes jaundice and flu-like symptoms.

20.2.5 Control and Prevention

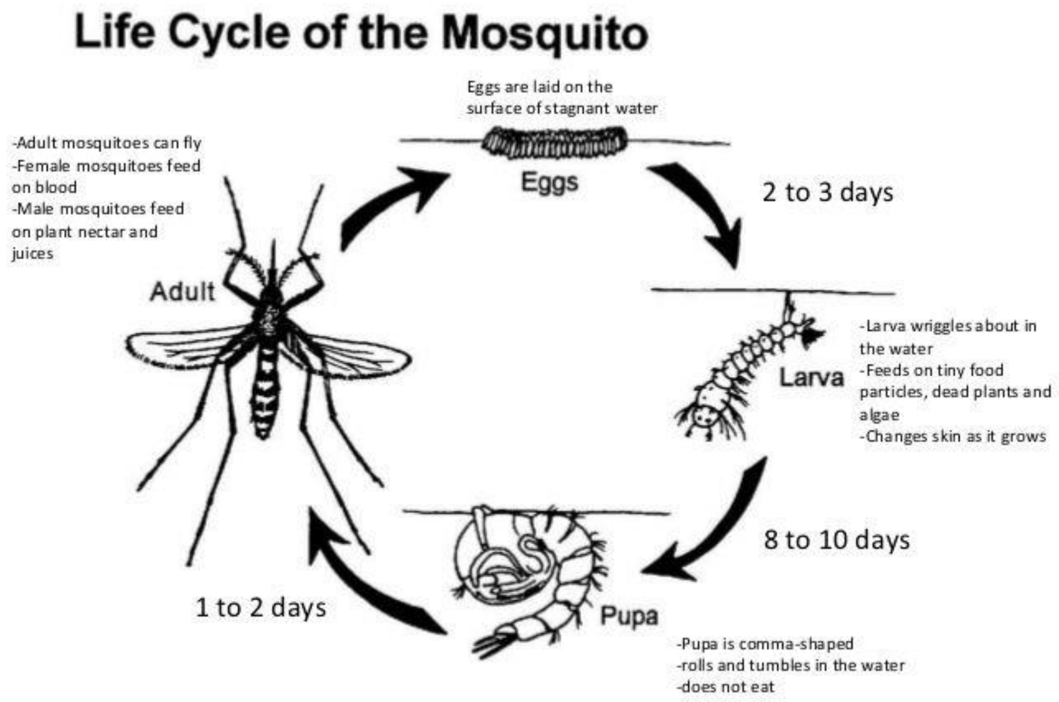
Effective mosquito control involves a combination of methods:

- **Eliminate Breeding Sites**: To stop mosquitoes from breeding near houses and gardens, remove any standing water.
- **Personal Protection**: Apply insect repellents with DEET, picaridin, or lemon eucalyptus oil in them. Use bed nets and dress in long sleeves and pants, especially in places where mosquito activity is intense.
- **Biological Control**: Introduce natural predators like fish (e.g., guppies) and dragonflies to mosquito breeding areas. Use bacteria-based larvicides (e.g., *Bacillus thuringiensis israelensis*) that target mosquito larvae.
- **Chemical Control**: Apply insecticides to reduce adult mosquito populations. Use larvicides in standing water to kill larvae before they mature.

- **Community Efforts**: Participate in community-wide mosquito control programs and public health campaigns to reduce mosquito-borne diseases.

Life Cycle of the Mosquito

The following picture shows the life cycle of a mosquito. There are four stages altogether : (1) eggs, (2) larva (3) pupa and (4) adult.



Fleas (Siphonaptera):

Little, wingless insects called fleas are ectoparasites—that is, they live outside of their hosts. They consume the blood of birds and mammals. and are known for their ability to jump long distances relative to their size.

Classification

Fleas belong to the order Siphonaptera. There are over 2,500 species of fleas, and they are classified based on their host preferences and physical characteristics.

Morphology

Fleas have several distinctive morphological features that make them highly adapted to their parasitic lifestyle:

- **Size**: Fleas are tiny insects that range in length from 1.5 to 3.3 mm.
- **Body Shape**They are laterally compressed, making it simple for them to pass through the fur or feathers of their host.
- **Color**: Usually reddish-brown.

- **Head**: Small with mouthparts adapted for piercing skin and sucking blood.
- **Eyes**: Some species have simple eyes (ocelli), while others are blind.
- **Antennae**: Short and often recessed into grooves on the head.
- **Legs**: The long, strong rear legs are designed for jumping. Fleas have a 150-fold vertical jump ability.
- **Combs**: Many species have combs (ctenidia) on their heads or thoraxes, which help them stay attached to the host.

20.2.6 Life Cycle

Fleas undergo complete metamorphosis, which includes four stages:

1. **Egg**

   Appearance: Small, oval, and white.

   Laying: After the female lays her eggs on the host, the eggs fall off into the surroundings (e.g., bedding, carpets).

   Hatching: Eggs hatch into larvae in about 2-12 days, depending on environmental conditions.

2. **Larva**

   Appearance: Worm-like with a distinct head and chewing mouthparts. They are legless and typically white or translucent.

   Feeding: Larvae consume organic waste, such as the droppings of adult fleas.

   Development: The larval stage lasts about 1-2 weeks, during which they undergo several molts.

3. **Pupa**

   Appearance: After spinning a silk cocoon, the larva becomes a pupa inside.

   Duration: The pupal stage can last from a few days to several months, depending on the environment. Since these indicators indicate the potential presence of a host, fleas can remain in the pupal stage until they sense vibrations, heat, or carbon dioxide.

4. **Adult**

   Emergence: At the point when the climate is correct, grown-up bugs break out of the cover.

   Feeding: Adult fleas begin eating blood as soon as they find a host.

Reproduction: Female fleas, which can lay hundreds of eggs in a lifetime, continue the cycle.

20.2.6 Habitat and Behavior

- **Host Specificity**: Fleas can infest a variety of creatures, including humans, dogs, cats, rodents, and birds, and they are not as host-specific as lice.
- **Feeding**: Adult fleas only consume blood for nourishment. If they do not immediately find a host, they can live for several months without eating.
- **Jumping Ability**: Fleas are well known for their remarkable leaping abilities, which allows them to travel between hosts and throughout their surroundings.

20.2.7 Human Impact and Diseases

Fleas can have a significant impact on both people and animals:

- **Bites**: Flea bites cause itching, irritation, and allergic reactions in some individuals.
- **Disease Transmission**: Numerous diseases are spread by fleas, including:

  Plague: caused by the rat flea Xenopsylla cheopis, which transmits Yersinia pestis.

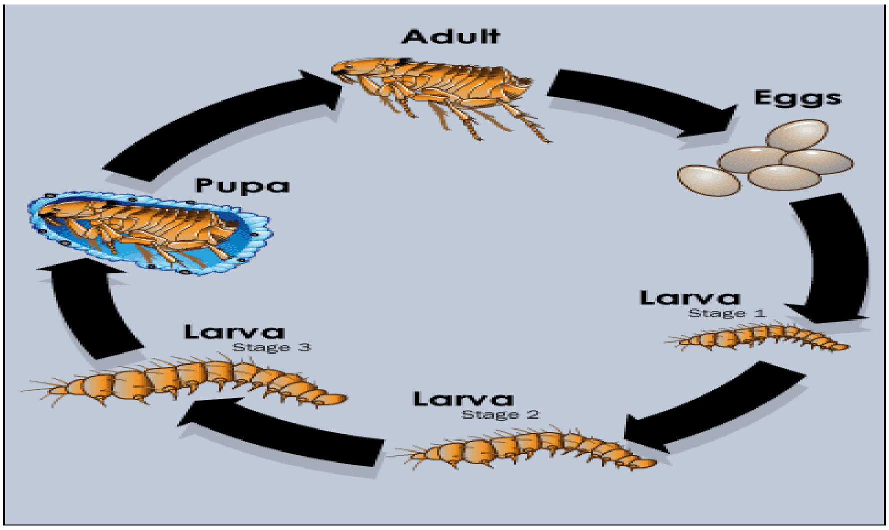
  Murine Typhus: caused by Rickettsia typhi, which fleas can also carry.

  Flea-borne Bartonellosis: causes cat scratch disease and other illnesses; caused by Bartonella henselae and other species.

- **Tapeworms**: Tapeworms (Dipylidium caninum) can be spread by fleas to both humans and pets on occasion.

20.2.8 Control and Treatment

- **Pet Treatment:**The consistent application of flea control solutions, including topical treatments, oral drugs, and pet collars.
- **Control of Environment:** Vacuuming carpets, furniture, and pet bedding frequently to remove eggs, larvae, and adults. Washing pet bedding in hot water.
- **Insecticides**: Use of insect growth regulators (IGRs) and other insecticides in the home to kill fleas at different life stages.
- **Personal Protection**: Avoiding contact with infected animals and using flea repellents when necessary.



20.2.9 Lice (Phthiraptera):

Lice are small, wingless insects that live as ectoparasites on warm-blooded hosts, including humans, birds, and mammals. They belong to the order Phthiraptera and are highly specialized for life on their hosts, where they feed on blood, skin, or debris.

20.2.10 Classification

Phthiraptera is divided into two main suborders:

1. **Anoplura (Sucking Lice)**: These lice feed on blood and are found mainly on mammals.
2. Example: *Pthirus pubis, Pediculus humanus corporis (body louse), and Pediculus humanus capitis (head louse) (pubic louse).*
3. **Mallophaga (Chewing Lice)**: These lice feed on skin, feathers, and debris. They are more commonly found on birds but can also be found on some mammals.

20.2.11 Morphology

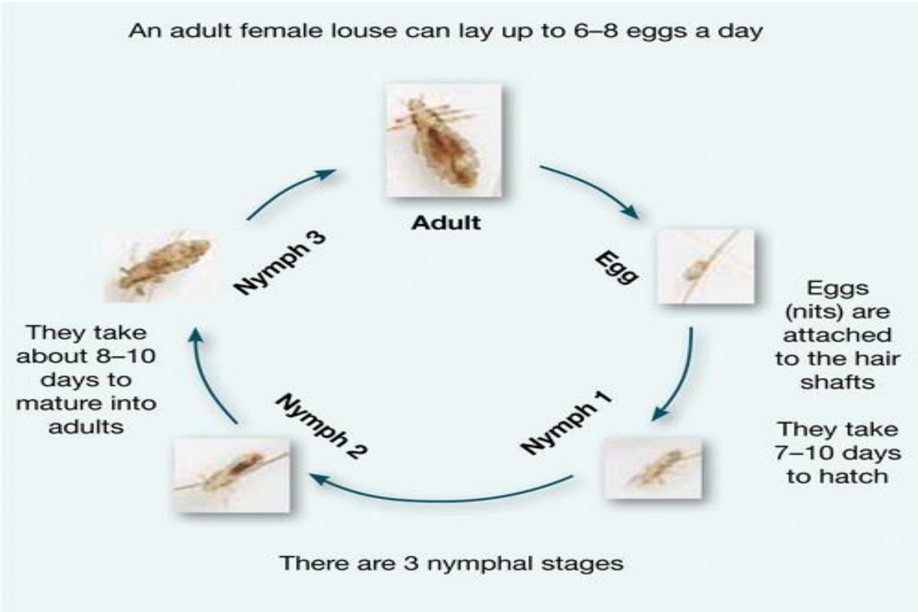
Lice are small insects, usually between 0.5 to 8 mm in length. They have flattened bodies that enable them to cling tightly to their host. Some key morphological features include:

- **Head**: Broad and equipped with mouthparts specialized for piercing skin and sucking blood or for chewing.
- **Thorax**: possesses three pairs of legs designed to cling on feathers or hair.
- **Abdomen**: Generally wider than the thorax, with segments that can expand to hold ingested blood or debris.

20.2.12 Life Cycle

1. There are three primary phases to the partial transformation of lice:

2. **Egg (Nit)**: The eggs that female lice lay, known as nits, are affixed to hair shafts or feathers .
3. **Nymph**: After hatching, the nymph looks like a smaller version of the adult and goes through several molts before maturing.
4. **Adult**: Fully developed lice are capable of reproduction and continue the cycle.



20.2.13 Habitat and Behavior

- **Host Specificity**: Lice are highly host-specific, meaning each species of louse typically parasitizes only one species or a group of closely related species.
- **Feeding**: While chewing lice devour feathers and skin debris, sucking lice feed on blood.
- **Transmission**: Lice are usually transmitted through direct contact between hosts, as they cannot fly or jump.

20.2.14 Human Impact

Certain species of lice are significant to humans due to their role in health and disease:

- **Head Lice (Pediculus humanus capitis)**: Common among school-aged children, causing itching and discomfort.
- **Body Lice (Pediculus humanus corporis)**: recognized for spreading illnesses such as typhus, relapsing fever caused by louses, and trench fever.
- **Pubic Lice (Pthirus pubis)**: Infest the pubic region and sometimes other coarse body hair, causing itching and discomfort.

20.2.15 Control and Treatment

- **Personal Hygiene**: Regular washing and combing can help control lice infestations.
- **Chemical Treatments**: Insecticidal shampoos, lotions, and creams are effective against lice.
- **Environmental Measures**: Washing bedding, clothing, and personal items in hot water to kill lice and nits. The specifics of lice biology, behavior, and control measures is essential for managing infestations and preventing the spread of lice-borne diseases.

20.2.16 Sandflies (Psychodidae):

Sandflies are small, blood-feeding insects belonging to the family Psychodidae, subfamily Phlebotominae. They are known for their role in transmitting various diseases, most notably leishmaniasis. Sandflies thrive in tropical, subtropical, and temperate regions.

Key Facts

- **Size**: Typically, 1.5 to 3.5 mm in length.
- **Color**: Generally light brown or gray.
- **Wings**: Long, hairy wings held at an angle when at rest, giving them a moth-like appearance.
- **Body**: Covered with dense hair, giving them a fuzzy look.

Mouthparts: designed to pierce flesh and draw blood.

20.2.17 Life Cycle

Four separate stages comprise the whole metamorphosis of sandflies:

1. **Egg**

    Laying: In damp, organic-rich settings like soil, leaf litter, and animal burrows, females lay their eggs.

    Hatching: Depending on the surroundings, eggs can hatch into larvae in one to two weeks.

2. **Larva**

    Appearance: Larvae have segmented bodies and a distinct head, like worms.

    Feeding: They eat debris, organic waste, and decomposing vegetation.

    Development: They undergo multiple molts over the course of the several weeks to months-long larval stage.

3. **Pupa**

Appearance: Pupae resemble cocoons and are non-feeding and stationery.

Duration: The larva changes into an adult during this one- to two-week-long stage.

4. **Adult**

Emergence: Pupils exit the pupal case as adults.

Feeding:

- **Females**: The require blood feedings in order to develop eggs. A diverse array of hosts, including humans, animals, and wild animals, are consumed by them.
- **Males**: Feed on plant nectar and do not bite.

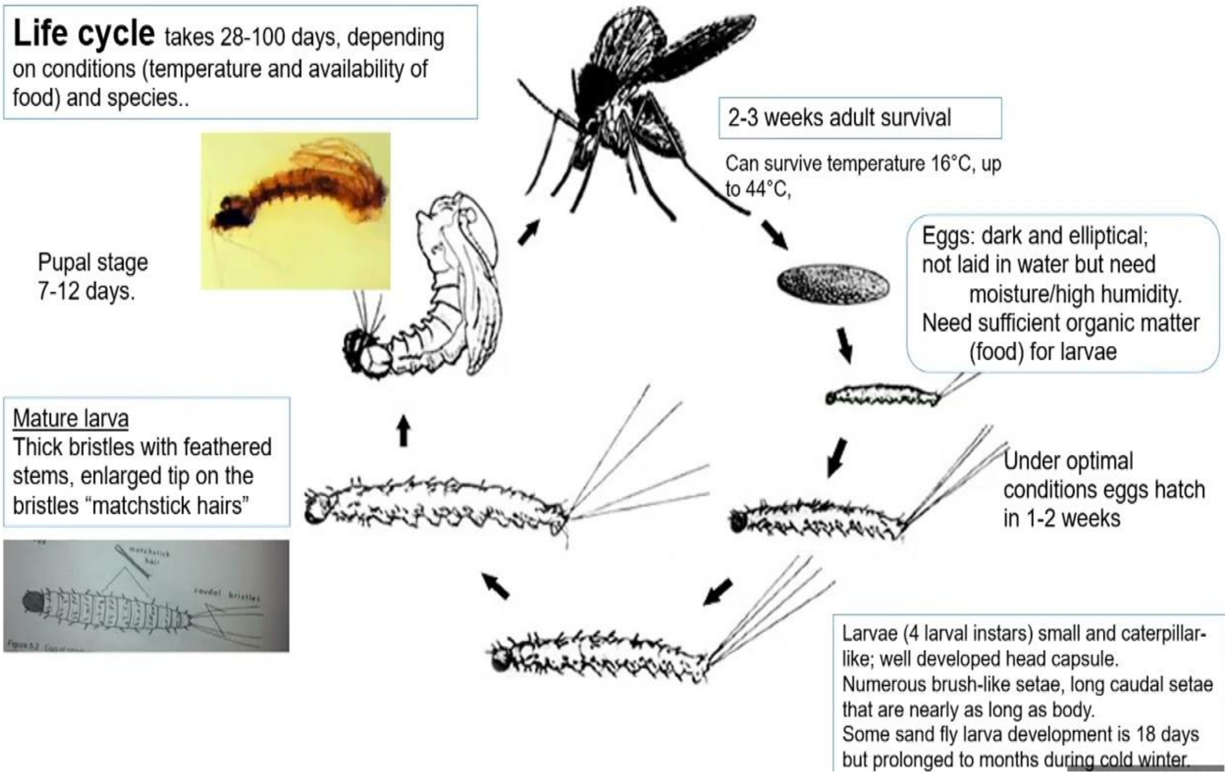
Activity: Sandflies are typically active at dusk and dawn (crepuscular).

20.2.18 Habitat and Behavior

- **Breeding Sites**: Prefer moist, dark environments rich in organic material, such as forest floors, caves, and animal shelters.
- **Resting Sites**: During the day, sandflies rest in cool, humid places such as animal burrows, tree trunks, and house walls.
- **Flight**: Weak fliers; they tend to hop rather than fly long distances and stay close to the ground.

20.2.19 Human Impact and Diseases

Sandflies are vectors for several.



20.2.20 Tsetse flies (Glossinidae):

Tsetse flies, belonging to the family Glossinidae, are hematophagous insects primarily found in sub-Saharan Africa. They are known for their role as vectors of trypanosomiasis, also known as sleeping sickness in humans and nagana in animals. Here are some key points about tsetse flies:

20.2.21 Biology and Behavior

1. **Morphology**: Tsetse flies are about 6 to 15 mm in length and are characterized by their long proboscis, which extends forward when they are at rest. They have a distinctive wing venation pattern with a characteristic hatchet cell.

2. **Lifecycle**: Tsetse flies undergo a unique lifecycle that includes larval development inside the female. Females give birth to fully developed larvae, which immediately pupate in the soil. The pupal stage lasts for several weeks before emerging as an adult fly.

Feeding Habits: Tsetse flies are entirely blood-feeders, both male and female. They pierce the skin of their hosts—which include humans and a variety of animals—with their razor-sharp proboscis in order to obtain blood.

20.2.22 Disease Transmission

1. **Trypanosomiasis** Trypanosomes, a parasitic protozoon that causes sleeping sickness in people (due to Trypanosoma brucei gambiense and Trypanosoma brucei rhodesiense) and nagana in animals, are primarily transmitted by tsetse flies (caused by several species of Trypanosoma).
2. **Transmission Cycle**: When an infected tsetse fly bites a person or animal, the trypanosomes are injected into the host's bloodstream, causing the disease to spread. After that, the parasites proliferate and spread, causing a variety of health problems, including neurological disorders in cases of severe human sleeping sickness.

20.2.23 Control Measures

1. **Insecticide Spraying**: Spraying insecticides in tsetse-infested areas can reduce fly populations. This can be done through ground spraying or aerial spraying.
2. **Traps and Targets**: Specially designed traps and insecticide-treated targets attract and kill tsetse flies. These devices often use colors, shapes, and odors that are attractive to the flies.
3. **Sterile Insect Technique (SIT)**: This procedure involves releasing male tsetse flies that have been sterilized back into the wild. Because these infertile men mate with insane females and have no offspring, the tsetse population steadily declines.
4. **Biological Control**: Scholars are investigating the potential of illnesses, parasites, and native predators to control tsetse fly numbers. However, this method is still being researched and isn't widely used yet.

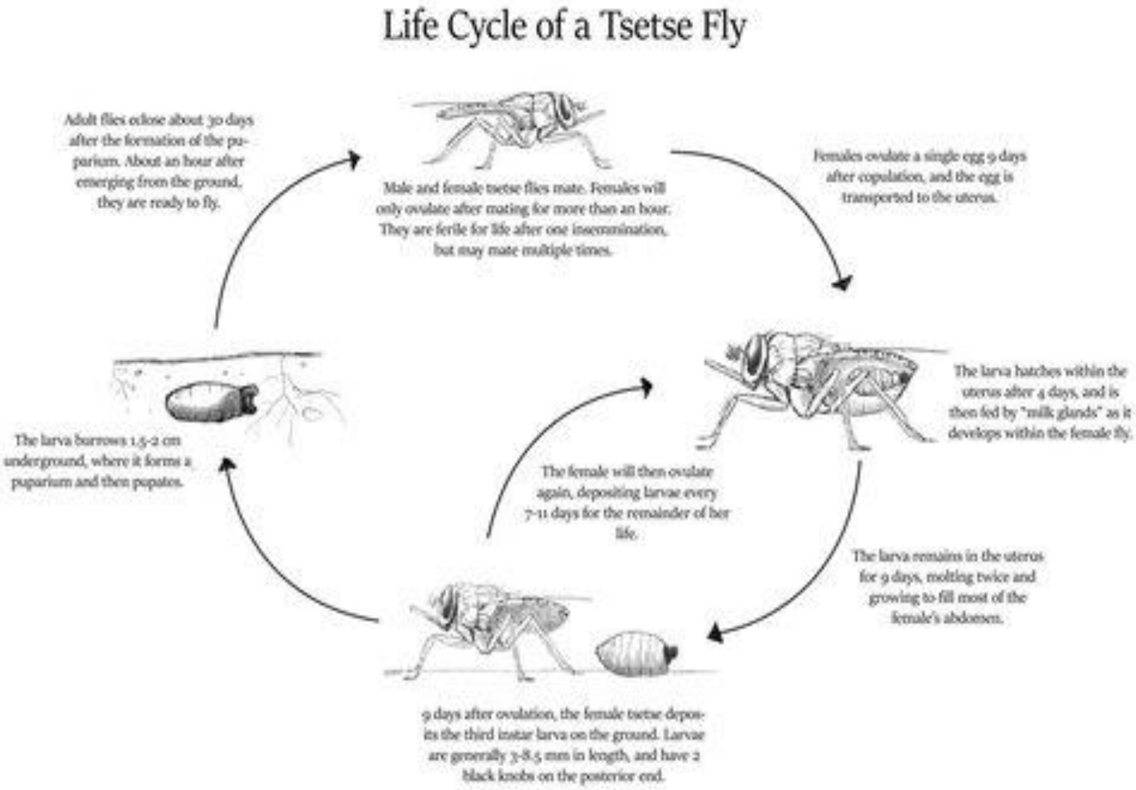
5.Impact and Importance

1. **Economic Impact**: In Sub-Saharan Africa, the tsetse fly has a major effect on livestock productivity and agriculture. The nagana disease affects both food security and economic stability since it lowers cattle productivity and has a high death rate.
2. **Public Health**: In order to reduce the prevalence of resting disorder in humans, which can be fatal if left untreated, tsetse fly populations must be managed. In regions where outbreaks are likely, public health initiatives and campaigns are crucial.

20.2.24 Research and Development

1. **Genetic Studies**: Developments in genetic research are shedding light on the biology and behavior of tsetse flies, which can help create more efficient control strategies.

2. **Vaccines and Treatments**: The goal of advancing research is to provide trypanosomiasis antibodies and improved treatments for those affected.



2- Arachnids:

Ticks (Ixodida): Receiving wires that stick to their hosts to benefit from blood are called ticks.

Numerous illnesses, such as anaplasmosis, tick-borne encephalitis, and Lyme disease (Borrelia burgdorferi), are spread by them. A green, wooded environment is ideal for ticks.

Mites (Acari): Microscopic, mites are carriers of diseases such as scrub typhus (Orientia tsutsugamushi). They can live in soil, on plants, or as parasites on animals.

20.2.25 Life Cycles and Transmission

1. Controlling vector-borne diseases requires an understanding of the life cycles of both the parasites that vectors transmit and the vectors themselves. A vector-borne parasite usually goes through the following stages in its life cycle:

2. **Ingestion**: The vector ingests the parasite while feeding on an infected host. For example, mosquitoes ingest Plasmodium gametocytes when they feed on the blood of a person with malaria.

3. **Development**: The parasite undergoes part of its development within the vector. For instance, in mosquitoes, Plasmodium parasites develop into sporozoites in the gut before migrating to the salivary glands.

4. **Transmission**: The vector transmits the mature, infectious form of the parasite to a new host during subsequent feedings. When an infected mosquito bites another person, it injects sporozoites, which then travel to the liver and begin a new cycle of malaria infection.

20.2.26 Vector Behavior and Ecology

To effectively control vector populations, it is essential to understand their behavior and ecology:

- **Feeding Habits**: Knowing when and where vectors feed helps target interventions. For example, using bed nets to prevent mosquito bites at night.
- **Breeding Sites**: Identifying and managing breeding sites can reduce vector populations. For instance, eliminating standing water can reduce mosquito breeding.
- **Seasonality**: Seasonal population peaks in many vectors are caused by environmental variables including humidity and temperature. Recognizing these trends can aid in the anticipation and containment of epidemics.

20.2.27 Disease Control Strategies

A variety of tactics are needed to control vector-borne diseases:

- **Chemical Control**: Vectors are killed by using insecticides and acaricides. Even though it works well, if it is not handled correctly, resistance may develop.
- **Biological Control**: To lower vector populations, introduce viruses or natural predators. For instance, mosquito larvae can be the target of bacteria such as Bacillus thuringiensis ireless.
- **Environmental Management**: altering environments to minimize breeding grounds. This covers routines like clearing up standing water and keeping spaces tidy.
- **Personal Protection**: protective clothes, bug repellent, and sleeping beneath bed nets sprayed with pesticide are a few examples of preventative measures.
- **Vaccination and Prophylaxis**: Creating vaccinations against diseases carried by vectors and giving at-risk individuals preventive medication.

20.2.28 Emerging Challenges

There are numerous important obstacles for vector control.:

- **Insecticide Resistance**: Chemical control may become less effective as a result of pesticide resistance in vector populations caused by overuse and misuse.

- **Climate Change**: Climate change has the potential to increase the geographic range of vectors and bring vector-borne illnesses to previously unaffected areas.
- **Urbanization**: Fast urbanization may increase the risk of disease transmission by changing vector dynamics and establishing new breeding grounds.

20.3 Classification of Parasites

The three primary groups of parasites are arthropods, helminths, and protozoa. Single-celled organisms known as protozoa are the source of illnesses including giardiasis and coccidiosis. Nematodes, or roundworms like Ascaris, cestodes, or tapeworms like Echinococcus, and trematodes, or flukes like Fasciola, are examples of helminths.

20.3.1 Life Cycles of Parasites

The life cycles of parasites are intricate and may be direct or indirect. The parasite grows only on that one host during a direct life cycle, such as that of Toxocarid Canis. For indirect life cycles, multiple hosts are required. For heartworm, Dirofilarial imcites, for example, the intermediate host is a mosquito, where the parasite partially develops, and the final host is where the parasite matures. To design effective control systems, it is necessary to comprehend these cycles.

20.3.2 Epidemiology

The dissemination and reasons for parasitic sicknesses in creature populaces are researched by epidemiology. Examples of transmission strategies incorporate ingestion (e.g., defiled food or water) and direct contact. (like hookworm infiltration of the skin), vector-borne, (for example, heartworm transmission by mosquitoes), transplacental, (for example, Toxoplasma gondii), and trans mammary (like Strongyloidiasis). Parasitic diseases are significantly influenced by a variety of environmental factors, including climate, location, and host availability.

20.3.3 Pathogenesis and Clinical Signs

The method by which parasites cause illness is referred to as "pathogenesis." Toxin production (such as tick paralysis), mechanical damage (such as hookworms draining intestinal blood), or an immune response (such as flea bite allergies) may all result in this. Numerous clinical signs, such as diarrhea, anemia, weight loss, and respiratory problems, may appear, depending on the parasite and the host species. Early diagnosis and therapy are contingent upon the ability to identify these signs.

20.4 Diagnosis

Clinical examination and scientific tests are combined to detect parasite illnesses. While clinical signs can provide some early guidance, a definite diagnosis often requires test confirmation. A fecal examination is used to check for gastrointestinal parasites, blood tests are used to check for blood-borne parasites like Babesia, serological tests are used to check for antibodies against parasites, polymerase chain reaction (PCR) is used to search for parasite DNA, and so on. An ultrasound is used to check for specific intrarenal parasites.

20.5 Treatment and Control

Antiparasitic drugs are used to treat parasitic infections; the type of drug used varies on the type of parasite. Insecticides fight ectoparasites, anthelmintics target helminths, and antiprotozoals address protozoan illnesses. Preventing parasites from becoming resistant to these treatments requires resistance management, which involves switching up medication classes and implementing integrated control methods. To successfully lower parasite populations, control programs involve chemical treatments, environmental management, and biological control techniques.

20.5.1 Specific Parasitic Diseases

Toxoplasmosis, giardiasis, and cryptosporidiosis are examples of protozoan infections that affect a wide range of animal species and can pose zoonotic risks. Roundworms (like Ascarides), tapeworms (like Taenia), and accidents (like Fasciola, the liver accident) are the guilty parties behind helminth diseases. Diseases like Lyme disease and dermatitis caused by an allergy to fleas can arise from ectoparasitic infestations caused by fleas, ticks, mites, and lice.

20.5.2 Zoonotic Parasites

Zoonotic parasites will be parasites that can spread from creatures to individuals and are very risky for the overall population's wellbeing. Models incorporate Toxoplasma gondii, which can cause toxoplasmosis in people, and Echinococcus granuloses, causing hydatid sickness. Anticipation and control of zoonotic parasites include standard deworming of pets and animals, rehearsing great cleanliness, controlling vector populaces, and instructing general society about the dangers and avoidance procedures related with these parasites. Many viewpoints can be used to analyses zoonotic illnesses. Currently, there are over 150 identified zoonotic illnesses, which encompass all known categories of infectious agents (Acha & Szyfres, 1987). Both the range of animal hosts and the ways in which animals transmit diseases to humans vary widely. Ranking zoonotic illnesses can also be done based on how they affect the economy or public health. This can be investigated further by contrasting the many elements that support the occurrence or consequences of zoonotic illnesses in various socioeconomic contexts. In this sense, the range of zoonotic illnesses and the cost placed on public health in developing nations varies significantly from those in contemporary wealthy nations (McKeown, 1988). Animal-related illnesses are still most prevalent in rural parts of poor nations.

20.5.3 Zoonotic Parasites in fish

Foodborne diseases caused by helminth parasites transmitted by fish and shellfish products pose major public health problems, and worldwide the number of people at risk, including those in developed countries, is more than half a billion (WHO, 1995, WHO, 2004). Some of these parasites are highly pathogenic, and human infection is a result of the consumption of raw or undercooked fish infected by the parasites (WHO, 1995). The reported incidence of these ichthyozoonoses has increased significantly in recent years for several reasons: the development of new and improved diagnosis, by the increase in raw fish consumption in those countries in which such dishes have commonly been eaten, by the increased consumption elsewhere of regional fish dishes such as sushi, sashimi, ceviche, carpaccio based on raw or minimally processed fish, by the growth in the international market in fish and fish products, and by the spectacular development of aquaculture (Keiser and Utzinger, 2005, McCarthy and Moore, 2000, Nawa et al., 2005, Robinson and Dalton, 2009).

20.5.4 Cestodiasis

A parasitic disease carried by fish, human diphyllobothriasis is brought on by cestodes belonging to the Diphyllobothridae family. Diphyllobothrium latum is thought to be the most common of the several species of Diphyllobothrium that are known to be harmful to humans. The principal host of the parasite is piscivorous animals, including humans, and it has a complicated life cycle. A copepod serves as the first of the two intermediate hosts, while a predatory freshwater, anadromous, or marine fish serves as the second.

20.5.5 Anisakiasis

The term "anisakiasis" describes infection with nematodes in the larval stages from the Anisakidae family, namely from the genera Anisakis and Pseudoterranova. The pathogenic species that is most common is Anisakis simplex (Adams et al., 1997, Audicana et al., 2003, Butt et al., 2004, Chai et al., 2005, Lymbery and Cheah, 2007, Sakanari and McKerrow, 1989, Torres et al., 2007). Hardly ever are Contracaecum and Hysterothylacium thought to be able to infect humans (Deardorff and Overstreet, 1980

20.5.6 Prevention and control

Fishbone parasite illnesses are caused by eating raw fish, fish dishes, or fish products that have not been thoroughly boiled or processed to eradicate the parasites. It is obvious that if consumption of such goods stopped, the incidence of disease would decrease or even disappear; but, given the popularity of a wide range of traditional fish meals, many of which have a regional flair, achieving this goal would be quite challenging in practise. Although there is substantial evidence linking wild fish to the spread of parasitic diseases that are contracted through food, there were few reports of aquaculture products directly transferring parasites to humans until recently. Prior to the year 2000, there was essentially no recorded data supporting the notion that fish raised for food could be carriers of illnesses that pose a risk to human health, as stated by several past assessments of fishbone parasites (Howgate, 1998, Paperna, 1991, WHO, 1995). Since then, a significant amount of data about the contribution of aquaculture products to the fishbone zoonoses' epidemiology has been published. The aims of this paper are to review recent knowledge on fishbone parasites in farmed fish and to suggest techniques for prevention and control of parasitic infections in them. The three main categories of fishbone parasites are: cestodes, particularly those belonging to the Diphyllobothriidae family; nematodes, particularly those belonging to the Anisakidae and Gnathostomatidae families; and digenetic trematodes, particularly those belonging to the Opisthorchiidae and Heterophyidae families. Given the morbidities of the diseases they are connected with, the trematodes are the most concerning of them. We will evaluate new data about different parasites and places, but the majority of the new information originates from Asia and South East Asia and relates to fishbone trematode (FBT) infections. The focus is on the epidemiological data and scientific proof linking foodborne zoonoses to aquaculture.

20.5.7 Economic Impact

Animal parasite infections have significant financial ramifications, especially for the production of livestock. Animals with the infection may not grow as much weight, produce less milk, reproduce poorly, and have higher death rates.The expenses linked to managing and averting parasitic illnesses, encompassing veterinarian attention, anthelmintic medications, and putting control mechanisms in place, might be substantial. Furthermore, trade restrictions brought on by parasitic illnesses may have an effect on the cattle sectors' capacity to make a profit.

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Chapter 21 : Phytochemicals activating non-coding RNAs (ncRNAs) to modulate ferroptosis in human cancers and aging

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Abstract

The intricate interplay between phytochemicals, non-coding RNAs (ncRNAs), and ferroptosis represents a promising frontier in cancer biology and aging research. Bioactive compounds (BACs) sourced from medicinal plants, exhibit diverse pharmacological properties and have been shown to modulate cellular processes through their interactions with ncRNAs. Ferroptosis is a kind of controlled cell death characterized by lipid peroxidation dependent on free cellular iron, has become an important route in the development of cancer and diseases associated with aging. This chapter explores the molecular mechanisms by which phytochemicals either activate or inhibit ncRNAs to modulate ferroptosis in human cancers and aging. We discuss the roles of specific phytochemicals and ncRNAs in regulating ferroptosis, highlight the therapeutic implications of targeting ferroptotic pathway, and propose future directions for research in this field. Exploring the interplay between phytochemicals, ncRNAs, and

ferroptosis holds promise for developing novel therapeutic strategies for cancer treatment and aging linked pathologies.

Keywords: Phytochemicals, ncRNA, ferroptosis, cancer, aging, cell death.

21.1. Introduction

An intricate interplay between phytochemicals, ncRNAs, and cellular signaling cascades has attained a significant attention for its potential implications in cancer and aging research [1]. This emerging field highlights underlying molecular mechanisms that how BACs derived from plants modulate ncRNAs to influence cellular functions particularly ferroptosis, an iron flooding death due to excessive oxidation of lipid membranes [2]. Understanding the role of phytochemicals in activating or inhibiting ncRNAs to modulate ferroptosis would possibly help in the development of novel therapeutic strategies targeting cancer and aging associated pathologies [2].

Phytochemicals are BACs sourced from medicinal plants showed numerous pharmacological properties, strong antioxidant potential, suppress cellular inflammatory burst, ameliorate chronic pathologies including cancer and aging. Among these BACs flavonoids, polyphenolic compounds, alkaloids, terpenoids and much more to count [3]. These BACs are not only integral to plant defense mechanisms but also have significant implications for human health. Through dietary intake or supplementation, plant BACs have beneficial impacts on human physiology, prevention and disease cure[4].

RNA molecules are classified into ncRNAs which do not took part in translational activities but regulates transcriptional activity numerous genes and cell signaling cascades [5]. The ncRNAs further classified into microRNAs (miRNAs), long non-coding RNAs (lncRNAs), and circular RNAs (circRNAs) etc. Despite lacking protein-coding capacity, ncRNAs exert regulatory functions through diverse mechanisms including post-transcriptional gene silencing, chromatin remodeling, and protein complex scaffolding [6]. Numerous illnesses such as cancers, neurological conditions, and cardiovascular issues, have been linked to dysregulation of ncRNA expression or activities [1].

Ferroptosis, a kind of regulated cell death caused by iron mediated oxidation of biological membranes and hence accumulates excessive reactive oxygen species (ROS) induced oxidative stress [7]. Unlike apoptosis or necrosis, ferroptosis is distinct in its morphological and biochemical features and is regulated by a unique set of cellular factors. Among ferroptotic regulators cellular iron metabolism, lipid peroxidation (LPO), and redox modulating mechanisms. Recent studies elucidated that dysregulation of ferroptosis may lead to carcinogenesis, neurodegenerative diseases, and aging associated pathologies [8].

Recent studies unveiled the ability of certain BACs to modulate ncRNAs and, consequently, influence ferroptosis in cancer cells and aging-related pathologies [9]. For example, some BACs upregulate specific miRNAs that target key regulators in ferroptosis, thereby cell survival and death depends solely on the cellular redox level. While, many BACs may downregulate protective ncRNAs, leading to enhanced susceptibility to ferroptotic cell death[10].

The intricate crosstalk between BACs, ncRNAs, and ferroptosis holds promising therapeutic implications for cancer treatment and aging linked diseases[11]. By elucidating the underlying molecular mechanisms, it would help identify novel targets for intervention and develop personalized therapeutic strategies. Moreover, insights gained by studying BACs-mediated modulation of ferroptosis could lead to the development of innovative dietary interventions, therapies for cancer prevention, and healthy aging[12](Figure 1).

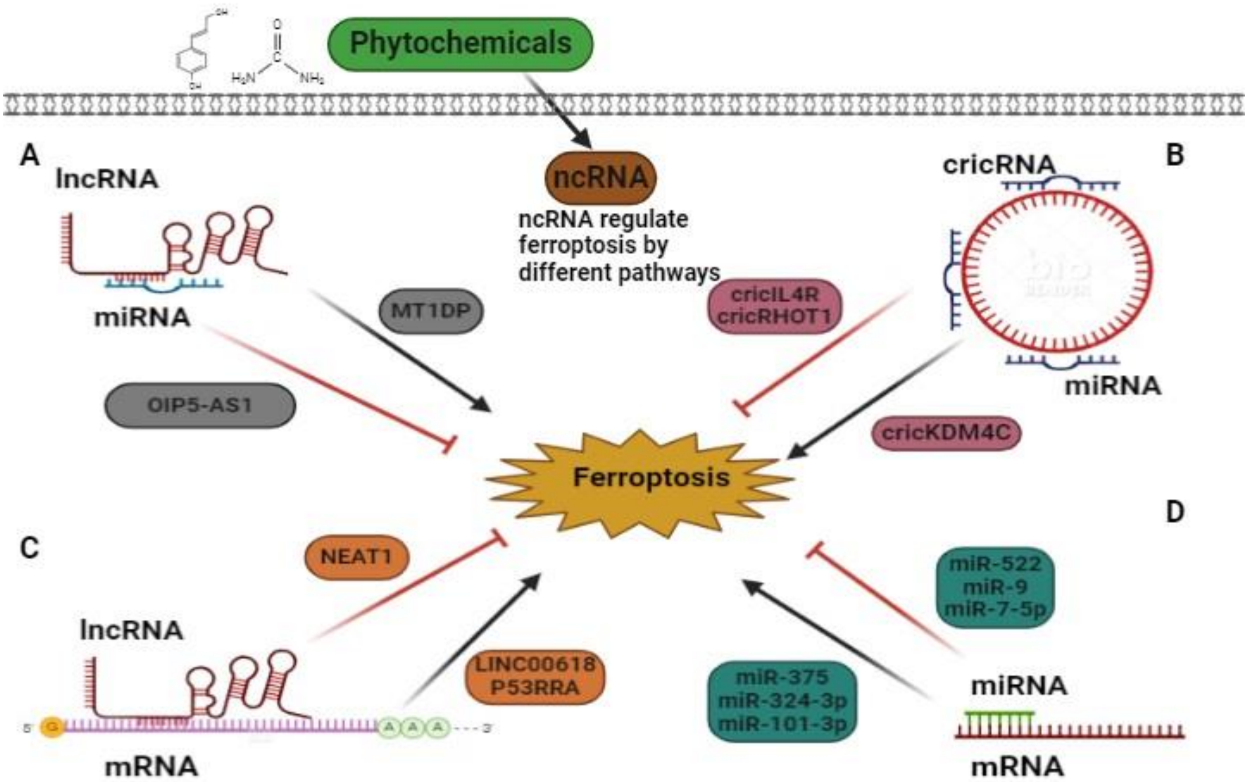

Figure 63: The ncRNAs activated by plant BACs play a crucial role in regulating ferroptosis by modulating the expression levels of its important factors at transcriptional and translational levels. These key players include ferroptosis agonists such as ASCL4, LPCAT3, ALOX, and CYP450, as well as ferroptosis antagonists (GPX4, SLC7A11, SLC3A2, and FSP1). Overexpressed lncRNAs act through pathways A and C, circRNAs through pathways B and miRNAs through pathway D, and inhibit these key players thereby regulating the activity of ferroptosis.

Current manuscript aimed to comprehensively review current understanding of BACs mediated regulation of ferroptosis through ncRNAs in human cancers and aging. Furthermore, underlying molecular mechanisms involved, key BACs and ncRNAs implicated in ferroptosis regulation shall also be highlighted. In addition, therapeutic potential of targeting ferroptotic pathway in the management of aging associated diseases shall also be discussed.

21.2 Ferroptosis

Ferroptosis is an intracellular type of cell death mediated by excessive iron loads, and dissimilar to necrosis and apoptosis is called ferroptosis [13]. Due to excessive LPO, biological membranes got damaged and cellular free iron multiplies the production of ROS many-fold to activate ferroptotic signaling [14]. The concept of ferroptosis likely stemmed from observations of nutrient deprivation-induced cancer cell death [14], as well as from "oxytosis," which denotes neuronal death resulting from glutamate excitotoxicity coupled with blockage of the amino acid antiporter for osmotic carriers family 7 member 11 (SLC7A11/xCT/system) [15]. Beyond cancer and aging, ferroptosis has been studies extensively to confirm its involvement in multiple chronic pathologies (Figure 2).

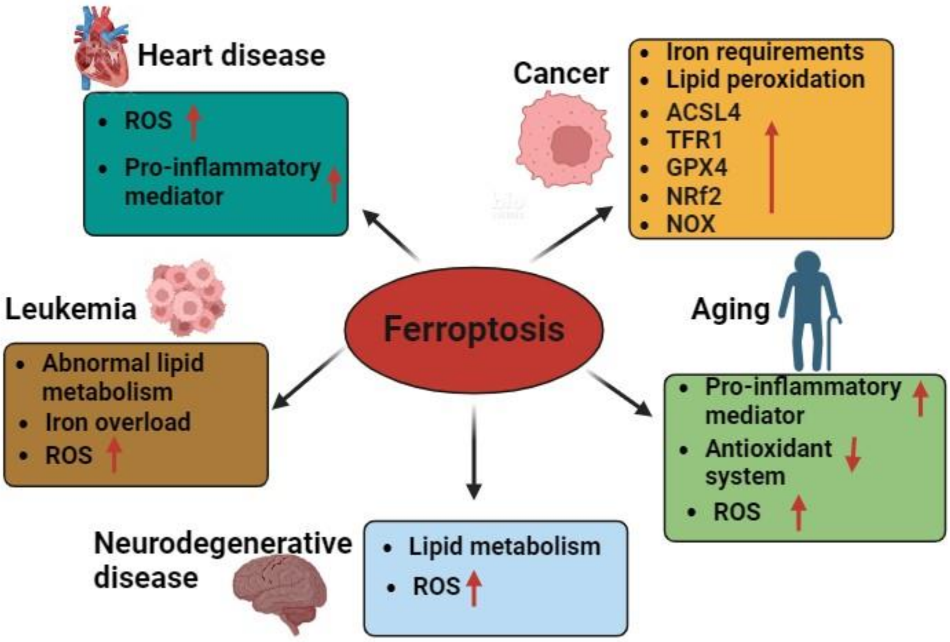

Figure 64: Targeting ferroptosis in different chronic diseases.

21.2.1 Mechanisms of ferroptosis

A. The metabolism of cellular Iron

Iron is an essential nutrient took part in various physiological processes including cellular metabolism, promotes generation of ATP, DNA synthesis, and oxygen, [13] [16]. Since iron is a component of all living organisms, preserving cellular iron homeostasis via iron procurement, utilization, storage, and recycling is a challenging and tightly regulated procedure. The iron level in cells is maintained by redox signaling and iron consumption primarily by Fenton reaction.

Fenton reaction in cells involves the oxidation of Fe^{2+} (ferrous ions) to Fe^{3+} (ferric ions) when H_2O_2 is transformed into reactive hydroxyl radicals ($^{\bullet}OH$). Conversely, superoxide radicals ($O_2^{\bullet-}$) subsequently convert Fe^{3+} back to Fe^{2+}. Extensively expressed transferrin receptor 1 (TFR1)promotes the uptake of free cellular Fe^{3+} ions by binding to transferrin (TF). Fe^{3+} is released from TF upon endocytosis of the TFR1/TF-(Fe^{3+})$_2$ complex upon ingestion. Ferric reductase reduces Fe^{3+} ions into Fe^{2+} ions which translocate across the endosomal membrane facilitated by divalent metal transporter 1 (DMT1) [17] (Figure 3).

Ferroptosis differs from other forms of regulated cell death due to its essential role in iron metabolism. Because iron can receive and lose electrons, it can play a role in the creation of free radicals[18]. When the iron in cells is too much, free radicals build up abnormally and produce more ROS. Oxidative stress brought on by this action causes ferroptotic cell death. On the other hand, aberrant iron metabolism also actively contributes to the development of cancer and encourages the growth of tumors[17].

TFR1 is the main regulator of intracellular iron uptake. It has been observed by researchers that aberrant TFR1 accumulation on the cell surface acts as a particular hallmark for ferroptosis [19]. TFR1 and FTH1 are both upregulated in hepatocellular carcinoma during ferroptotic cell death brought on by erastin and sorafenib, and TFR1 is also more expressed in myeloid leukemia cell lines during erastin-induced cell death. Furthermore, through controlling TFRC, FTH1, and FTL, Calu-1 lungs cancer cells and HT-1080 fibrosarcoma cells, erastin-induced ferroptosis is dependent on iron responsive element- binding protein 2 (IREB2) [20].

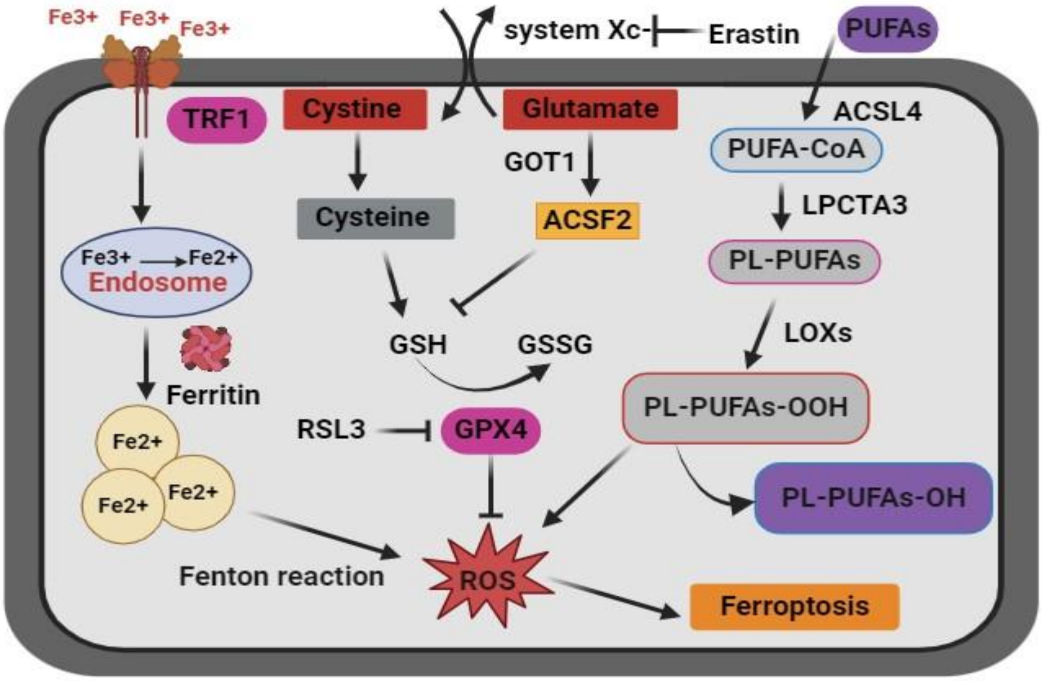

Figure 65: Mechanism of ferroptosis pathway. Three broad categories can be used to classify the primary metabolic processes of ferroptosis viz. LPO, redox homeostasis and metabolism, and iron metabolism.

B. Lipid metabolism

Fatty acid (FA) metabolism contributes essential lipid precursors for various cellular functions, including energy storage, membrane synthesis, signaling molecule generation, and lipid oxidation. This process leads to the accumulation of lipid ROS, playing a significant role in cellular physiology. The most important mechanism leading to ferroptotic cell death is the formation of excessive lipid ROS in cells, even if ferroptosis is induced by various triggers. It appears that ROS from the oxidation of lipids play a part in ferroptosis in addition to ROS produced by the Fenton reaction and iron. LPO is therefore essential for the induction of ferroptosis[17, 21].

The fatty acid substrate arachidonic acid (AA) is used by Acyl-CoA synthase long-chain family member 4 (the ACSL4) to activate AA-CoA [17]. The esterification of AA-CoA into phosphatidyl-(PE)-AA is then facilitated by lysophosphatidylcholine acyltransferase 3 (LPCAT3). Lipoxygenases (LOXs) are activated by Fe2+ and transform PE-AA into the cytotoxic PE-AA-OOH. To prevent oxidative damage to cells, glutathione peroxidase 4 (GPX4) generally transforms harmful PE-AA-OOH into non-cytotoxic PE-AA-OH. However, PE-AA-OOH builds up in the cell and causes ferroptosis when GPX4 is inactive or depleted, which results in its decreased activity. Thus, LPO is primarily responsible for the initiation of ferroptosis [22].

ACSL4 plays a pivotal role when long-chain polyunsaturated fatty acids are being produced, contributing to alterations in cellular lipid composition that heighten cell susceptibility to RSL3-induced ferroptosis[23]. In hepatocellular carcinoma patients undergoing sorafenib-induced ferroptosis, heightened expression of ACSL4 in pretreated specimens of tumors correlates with increased sensitivity to sorafenib. Furthermore, activation of ACSL4 triggers ferroptosis, inhibiting tumor cell proliferation in glioma. Through ACSL4 and LPCAT3, a CRISPR-based genetic display, were identified as facilitators of ferroptosis produced through RSL3 and DPI7, while having no impact on erastin-induced ferroptosis[24]. Studies suggest that oxidation of polyunsaturated fatty acids (PUFAs) generates lipid peroxides, thereby promoting ferroptosis induction. Focusing on the mechanism of lipid metabolism emerges as a promising strategy in tumor therapy[25].

C. Antioxidant metabolism

GSH, a tripeptide abundant in thiol groups, functions as a potent antioxidant, heavily reliant on a steady provision of cysteine and the presence of cystine/cysteine. The system Xc− antiporter facilitates the exchange of cystine for glutamate, importing extracellular cystine while exporting intracellular glutamate[26]. SLC7A11, a regulatory component of the system Xc− transporter, controls cystine uptake at the cell membrane, crucial for intracellular GSH synthesis. Then, intracellular cystine is transformed into cysteine, an essential building block for the creation of GSH. As a crucial regulator of ferroptosis, GPX4 regulates phospholipid peroxidase activity by preventing excessive LPO and enhanced production of lipid alcohols with GSH acting co-factor. Inhibition of SLC7A11 or GPX4 triggers ferroptosis, disrupting cellular antioxidant defenses and fostering ROS accumulation, underscoring the significance of antioxidant pathways in modulating ferroptosis[27].

Strong anti-RAS drug Erastin directly inhibits system Xc activity, which lowers GSH levels in cancer cells and starts ferroptotic cell death. This process initiates ferroptosis. Ferroptosis is also brought on by sulfasalazine, a drug that treats chronic inflammation, which inhibits SLC7A11 directly. Furthermore, the tumor suppressor p53 can enhance cell susceptibility to ferroptosis by suppressing SLC7A11[28]. Additionally, BRCA1-associated protein 1, a tumor suppressor, reduces SLC7A11 transcription by decreasing H2Aub levels, thereby promoting ferroptosis and LPO. Furthermore, by modulating nuclear factor erythroid 2-related factor 2 (Nrf2) expression may modulates antioxidant responses that along with its inhibitor (Kelch-like ECH-associated protein 1 Keap1) controls expression of SLC7A11. Ferroptosis also has a molecular mechanism involving the direct regulation of GPX4 through its degradation or inhibiting its activity [29].

21.2.2. Ferroptosis in Human Cancers and Aging

Several controlled cell death mechanisms, such as autophagy, apoptosis, and necrosis, play vital roles in tumor growth and management[30]. Ferroptosis has acquired significant focus to be involved in multiple tipes of cancers including DLBCL, lung and pancreatic cancers, breast cancer, RCC, HNSCC, and hepatocellular carcinoma (HCC). The induction of cell death, including ferroptosis, has emerged as a promising approach in cancer therapy[31].

Research suggests that the dysregulation of pathways involved in ferroptosis can contribute to cancer development and progression by altering cellular redox balance, promoting tumor cell survival, and facilitating resistance to therapy[32]. Therefore, understanding the mechanisms underlying ferroptosis in human cancer may offer new insights into cancer biology and therapeutic strategies[33]. The build-up of oxidative damage, particularly LPO-a defining characteristic of ferroptosis—is one of the main aspects of aging. As cells age, their antioxidant defense mechanisms may become less efficient, leading to increased vulnerability to oxidative stress and LPO-induced cell death. This process may contribute to tissue dysfunction and organ deterioration observed with aging[34].

Several studies have implicated ferroptosis in age-related diseases such as neurodegenerative disorders, cardiovascular diseases, and metabolic syndromes. For example, in neurodegenerative diseases like Alzheimer's disease (AD) and Parkinson disease (PD), dysregulated iron metabolism and increased LPO have been observed, suggesting a potential role for ferroptosis in neuronal cell death and neurodegeneration[35].

Furthermore, aging is associated with alterations in iron homeostasis and redox regulation, which may predispose cells to ferroptotic cell death. Age-related changes in the expression and activity of proteins involved in iron metabolism, such as TFR1 and ferritin, may contribute to the dysregulation of iron homeostasis and exacerbate oxidative stress-induced damage.

21.3 Noncoding RNAs (ncRNAs)

The ncRNAs encompass a diverse range of RNA molecules that do not encode proteins but significantly impact gene expression in various cellular contexts[6]. ncRNAs can be categorized into two main subtypes based on their function. The first subtype comprises basic structural ncRNAs, also referred to as "consecutive" ncRNAs, such as tRNAs, rRNAs, and snRNAs. These ncRNAs are consistently expressed and play roles in processes like translation and splicing. The second subtype consists of regulatory ncRNAs, which include miRNAs, siRNAs, circRNAs, piRNAs, and lncRNAs shown in (Figure 4) [36]. Despite their non-coding nature, ncRNAs play pivotal roles in fundamental biological processes such as development, differentiation, and cellular homeostasis[37]. Dysregulation of ncRNA expression has been linked to various diseases, including cancer, neurodegenerative disorders, and cardiovascular diseases, underscoring their significance as potential diagnostic markers and therapeutic targets[38].

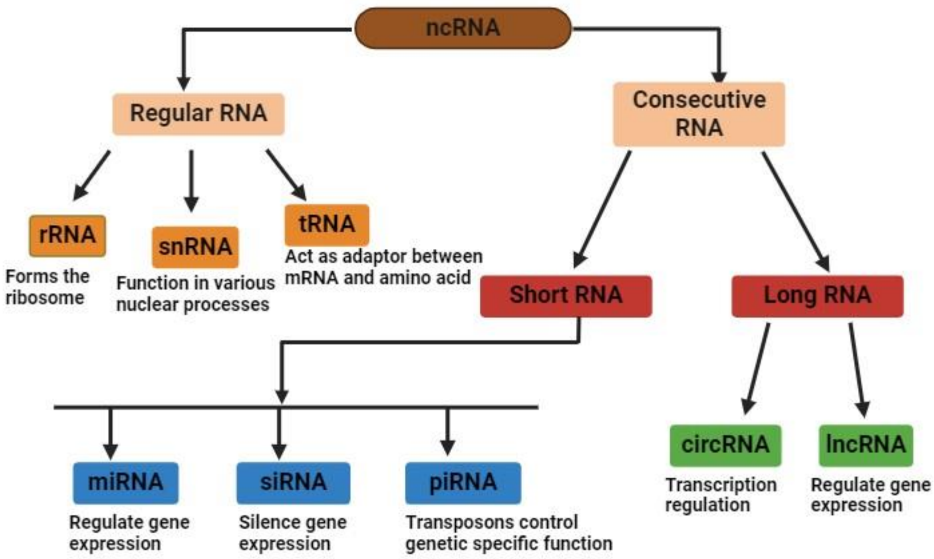

Figure 66**: Classification of ncRNA and their functions in cellular pathways.**

21.3.1 MicroRNAs

The miRNAs are small RNA molecules, typically comprising 18-25 nucleotides, function as post-transcriptional regulators by binding to complementary sequences in target messenger RNAs (mRNAs), thereby modulating their stability and translational efficiency[39]. This interaction can result in mRNA degradation or translational repression, thereby regulating the abundance of specific proteins within the cell. miRNAs play essential roles in various physiological processes, including cell proliferation, differentiation, apoptosis, and immune response regulation. The dysregulation of miRNAs has been linked to a variety of diseases, encompassing cancer, cardiovascular issues, and neurological disorders**[40].**

21.3.2 lncRNAs

The lncRNAs are a diverse class of RNA molecules that are longer than 200 nucleotides yet do not have the ability to code for proteins [41]. Despite their non-coding nature, lncRNAs exhibit diverse functions and play crucial roles in transcriptional regulation, chromatin remodeling, and post-transcriptional processing of RNA. They can interact with DNA, RNA, and proteins to modulate gene expression and cellular processes. lncRNAs are involved in various biological processes, including embryonic development, immune response, and cancer progression[42]. Dysregulated expression of lncRNAs has been associated with the pathogenesis of several diseases, making them potential diagnostic and therapeutic targets[43].

21.3.3 Circular RNAs (circRNAs)

Circular RNAs, formed through covalent back-splicing of pre-mRNA transcripts, possess unique stability and abundance and have been implicated in various cellular functions, including miRNA sponging and regulation of transcription and translation. CircRNAs may be essential for a number of physiological and pathological processes, such as cell division, proliferation, and the emergence of cancer, according to new research [44].

21.3.4 Regulation of ncRNAs by Phytochemicals

Phytochemicals, bioactive compounds derived from plants, have been shown to modulate the expression and activity of ncRNAs, thereby influencing cellular processes and disease outcomes. Several mechanisms by which phytochemicals activate ncRNAs have been proposed [45-46].

- **Direct Binding and Stabilization**

  Some phytochemicals can directly bind to ncRNAs, stabilizing their structure and enhancing their stability. By binding to specific regions of ncRNAs, phytochemicals can modulate their activity and function. For example, resveratrol, a polyphenolic compound found in grapes and red wine, has been shown to interact with specific miRNAs and lncRNAs, leading to their increased stability and activity[47].

- **Indirect Regulation via Transcription Factors**

  Phytochemicals can modulate the activity of transcription factors that regulate ncRNA expression. By activating or inhibiting transcription factors, phytochemicals can indirectly influence the expression of ncRNAs. For instance, sulforaphane, a compound abundant in cruciferous vegetables, activates the transcription factor NRF2, which, in turn, induces the expression of miRNAs and lncRNAs involved in antioxidant defense and cellular stress responses[48].

- **Epigenetic Modifications**

  Phytochemicals can influence epigenetic modifications, such as DNA methylation and histone modifications, which regulate ncRNA expression. By altering the epigenetic landscape of cells, phytochemicals can modulate the expression of ncRNAs. For example, curcumin, a bioactive compound found in turmeric, has been shown to alter histone acetylation patterns, leading to changes in the expression of miRNAs and lncRNAs involved in cancer progression and metastasis[49].

- **Modulation of RNA Processing and Stability**

  Phytochemicals have the potential to influence RNA processing and stability, thereby impacting the levels of ncRNAs. By modulating the activity of RNA-binding proteins and enzymes involved in RNA processing, phytochemicals can regulate the expression of ncRNAs[50]. For example, green tea polyphenols have been reported to modulate the expression of specific miRNAs by altering the activity of RNA-binding proteins involved in miRNA biogenesis and stability[51] [52-53].

The ncRNAs constitute a class of RNA molecules transcribed different segments of the genome devoid of protein-coding sequences. Enabled by advancements in sequencing technologies and computational methodologies, it has been uncovered that a substantial proportion of eukaryotic genes, possibly reaching 90%, possess the capability to yield various types of ncRNAs. These include, but are not restricted to lncRNAs, rRNAs, tRNAs, miRNAs, circRNAs, small interfering RNAs (siRNAs), and PIWI-interacting RNAs.

In contrast to messenger RNAs (mRNAs), which are responsible for encoding proteins, ncRNAs exert substantial influence over transcriptional and posttranscriptional processes, as well as epigenetic modulation of gene expression. Their involvement spans a wide array of biological functions, encompassing both normal physiological activities and pathological conditions. Recent investigations have unveiled a complex regulatory network governing ferroptosis, a form of programmed cell death, across various physiological and pathological scenarios, with an expanding roster of ncRNAs implicated in this process [54]. lncRNAs have emerged as particularly promising candidates for anti-cancer research, owing to their purported tumor-suppressive characteristics. A multitude of studies propose that manipulation of lncRNAs through diverse phytochemicals could offer a novel avenue in cancer therapeutics. Accumulating evidence strongly indicates that specific phytochemical agents exert regulatory control over these lncRNAs [55-56].

Phytochemicals as modulator of ncRNA

I. Camptothecin (CPT)

The Chinese tree *Camptotheca acuminata* (happy tree) is the source of the alkaloid camptothecin (CPT, $C_{20}H_{16}N_2O_4$). CPT has anticancer action and plays an inhibitory function in topoisomerase I [57-58]. It has been shown that the synthesis of HIF-1α-antisense RNA 1 is suppressed by CPT in a variety of human cancer types [59]. Additionally, CPT causes apoptosis in kidney and cardiovascular carcinomas, which boosts the production of antisense lncRNA [60]. It was demonstrated that CPT therapy exerts a synergistic regulatory effect on lncRNA HIF-1α expression in conjunction with miR-17-5-p and miR-155[61]. Through the inhibition of angiogenesis and hypoxia, CPT mitigates the expression of carbonic anhydrase IX (CA IX) within the tumor microenvironment. To synthesize CRLX101, a linear cyclodextrin-polyethylene glycol (CD-PEG) copolymer has been coupled with CPT, a nanoparticle-drug conjugate. The conjugation process has demonstrated enhanced efficacy of CRLX101 compared to CPT alone in terms of inducing apoptosis and suppressing angiogenesis [62].

II. Curcumin

Curcumin, is a polyphenol that is obtained from *Curcuma longa*, evergreen herbaceous plant[63]. In traditional Asian medicine, this fiery yellow powder has anti-inflammatory, antibacterial, and antioxidant properties[64]. Curcumin is a key lncRNA regulator in malignancies and works as a chemopreventive and chemotherapeutic medication against a variety of tumor types [65], Curcumin induce apoptosis in breast cancer and exert inhibitory effects on oncogenic signaling pathways[66]. Moreover, curcumin impede the proliferation of tumor cells by interfering with signaling channels such PI3K/Akt, STAT3, NF-κB, consequently inhibiting the overexpression of GAS5. Curcumin has also been observed to modulate the tumor suppressor HOTAIR in various cancer types including pancreatic cancer [67], prostate cancer [68], hepatocellular carcinoma (HCC), nasopharyngeal carcinoma(NPC) [69], breast cancer. [70], lung cancer, and renal cancer. [71].

Upregulate HOTAIR in breast cancer increases radioresistance, however its impact on the incidence of various cancer type and their response to therapy appears to be debatable [72]. Furthermore, compared to normal kidney cells, renal cell carcinomas express more of the HOTAIR gene, and studies have linked the overexpression of HOTAIR to distant metastases in cases of malignant renal cell carcinomas [73]. Consequently, curcumin affects the miR.-19/PTEN'/AKT/p53 axis in malignancies by acting as a HOTAIR modulator [74].

Curcumin also induces H19, a tumor suppressor that directly prevents p53 activation[66]. Colorectal cancer (CRC) and pancreatic cancers can result from overexpression of p53 [68]. High expression levels of six lncRNAs, namely AF086415, AK095147' RP1-179N16.3, MUDENG' AK056098, and AK294004, have been found in nasopharyngeal cancer [75]. These lncRNAs' expression is suppressed by curcumin, which also has the ability to make cancer cells more radiosensitive [69]. Conversely, curcumin and its phenolic constituents inhibit numerous forms of Cas, such as isozymes. I, II, XI, and the XII which are excessively stated in a number of malignancies [68]. When combined with other elements, curcumin can be a highly effective therapy for tumor cells. Furthermore, curcumin-containing sulfonamides reduces dopamine-related substances such as carbonic anhydrase (CA) I and CA II inhibitors, phenolic sulfonamides, and the combination of curcumin and glucose restriction [76](Table 2).

III. 3,3′-diindolylmethane (DIM)

The phytochemical molecule 3,3′-Diindolylmethane (DIM, $C_{17}H_{14}N_2$) is generated from indole-3-carbinol (I3C) and abundantly found in cruciferous vegetables like broccoli, cabbage, and kale. DIM can control angiogenesis, apoptosis, and cell division in cancer cells in addition to having an effect on signaling pathways [77]. DIM has been shown to cause apoptosis and suppress PCGEM1 expression in prostate cancer. Furthermore, it has been noted that via controlling the expression of the FOXM1 gene, DIM indirectly inhibits the Akt/FOXM1 signaling cascade[78]. In some carcinomas, FOXM1 modulates a variety of lncRNAs. In prostate cancer, androgens receptor variations and AR3 expression are reduced by bioresponse-formulated DIM (BR-DIM) [79](Table 1).

Table 25: Modulation of ncRNA by various phytochemicals (BACs).

Phytochemicals	**Modulated ncRNA**	**Sources**	**References**
Camptothecin (CPT)	HIF-1α	Bark and stem wood of the *Camptotheca acuminata* tree	[54]
Curcumin	RP1-179N16.3, MUDENG, AK056098, AK294004, H19, AF086415, AK095147, GAS5, HOTAIR, and all of them	Turmeric (*Curcuma longa*)	[46-47]
3,3′-diindolylmethane (DIM)	PCGEM1, FOXM1	Crucifers (broccoli, Brussels sprouts, cabbage, kale etc.)	[48]
Epigallocatechin-3-galate (ECGC)	AT102202	Green tea (*Camellia sinensis*)	[80]
Genistein	HOTAIR	Soybean and derived products (tofu, tempeh, soy milk)	[81]
Quercetin	DBH-AS1	Apples, onions, berries, citrus fruits, green leafy vegetables.	[45]
Resveratrol	PCGEM1, PRNCR1, LINC00978	Grapes skin, Red wine, peanuts, certain berries.	[47]
Sulforaphane	miR-155, miR-21, miR-34a, lncRNA HOTAIR, MALAT1	Cruciferous vegetables	[82]
Berberine	miR-223, lncRNA HOTAIR, MALAT1	*Hydrastis canadensis*(Goldenseal), *Berberis vulgaris*, Oregon grape	[83]
Luteolin	miR-34a, miR-21, lncRNA HOTAIR	Parsley, celery, thyme, peppers, certain fruits	[84]
Kaempferol	miR-21, miR-155, miR-34a	Fruits, vegetables	[85]

Apigenin	lncRNA HOTAIR, MALAT1	Parsley, celery, chamomile tea	[86]
Allicin	miR-21, miR-146a	Garlic (*Allium sativum*)	[87]
Ellagic acid	lncRNA HOTAIR	Berries and walnuts.	[87]
Gingerol	lncRNA HOTAIR, MALAT1	Ginger(*Zingiber officinale*)	[49]
Capsaicin	miR-21, miR-155, lncRNA HOTAIR, MALAT1	Chili peppers	[88]
Emodin	miR-21, miR-155	Rhubarb (*Rheum rhabarbarum*), species of buckthorn (*Rhamnus* spp.)	[89]
Naringenin	miR-21, miR-155, lncRNA HOTAIR, MALAT1	Oranges, grapefruits, lemons	[90]
Catechins	RP1-179N16.3, MUDENG	Tea leaves	[91]
Ellagic acid	miR-21, miR-155, lncRNA HOTAIR, MALAT1	Berries (Blackberry, Raspberry, Strawberry, Cranberry, Wolfberry), walnuts, pecans, pomegranate	[87, 92]
Tangeretin	miR-21, miR-34a	Tangerines and oranges.	[93]
Caffeic acid	miR-155, miR-21, miR-34a, lncRNA HOTAIR, MALAT1	Coffee beans, apples, artichokes, and certain grains.	[94]
Piperine	PCGEM1, PRNCR1,	Black pepper (Piper nigrum)	[95]
Polyphenols	GAS5, HOTAIR, H19, miR-21, miR-34a	Fruits, vegetables, tea, coffee, wine, chocolate, and various plant-derived foods	[96]

IV. Epigallocatechin-3-galate (EGCG)

Green tea and almonds are the sources of the well-known polyphenol flavonoid epigallocatechin (EGCG, C15H14O7) [97]. It can efficiently block ncRNA creation in cancers and has strong anti-inflammatory, antioxidant, and anticancer effects. The estrogen receptor (ER) and other signaling pathways likemTORC1, NF-κB, Akt, PI3K, PTEN, and, MAPK, are modulated by EGCG [98]. According to research, EGCG inhibits the lncRNA AT102202, which lowers the amount of 3.-hydroxy-3-methylglutaryl coenzyme A reductase (HMGCR) produced by human hepatocytes. This, in turn, enables the liver to absorb cholesterol. Studies have revealed that in breast cancer cells, the polyphenol epigallocatechin upregulate CA IX, which may have potent antioxidative and antiapoptotic effects. Additionally, it has been shown that the flavonoid component of green tea, EGCG, suppresses CA II.

V. Genistein

Moreover, dietary soy isoflavone genistein (C15H10O5) has anticancer effects both in vivo and in vitro [99]. Numerous human cancer types, including those of the breast, kidney, and prostate, have demonstrated some anti-proliferation effects from it [100-101]. In breast cancer, genistein alters the expression level of HOTAIR, which in turn alters the PI3K/Akt signaling pathway. By blocking HOTAIR, genistein slows the spread of kidney cancer. Genistein was discovered to upregulate the expression of miR-141 in cancer cells while downregulating the expression of HOTAIR. Genistein decreased the expression of miR-34a and HOTAIR in prostate cancer in a synergistic manner. Additionally, a different research revealed that genistein influences HOTAIR, which may have antitumor effects in CRC. Furthermore, genistein inhibits many signaling pathways specifically Wnt and Akt pathways to induce apoptosis in various cancerous cells such as breast cancer, prostate, abdomen, lung, pancreas, skin cancer, and renal cells. In several cancer types, genistein has been shown to promote apoptosis through the reduction of HOTAIR expression. In this instance, the majority of research examined the relationship between phytoisoflavones and estrogens in liver, uterine, cervical, and ovariectomy malignancies via genistein modulatory influence on CA II expression [102-103]

VI. Quercetin

Polyphenolic flavonoid quercetin ($C_{15}H_{10}O_7$) is well known for its chemopreventive qualities [104], present in various fruits and vegetables and hence is an abundant source of antioxidants in foods. Quercetin for its exceptional antiproliferative and antioxidant properties, help downregulate the DBH-AS1 expression in hepatocellular cancer cells. Quercetin has been shown to block a variety of signaling pathways, including PI3K/AkT and Akt/mTOR/P70S6K. The majority of research has verified quercetin's inhibitory effect on CA isoforms. Recently, a new form of Zr-MOF nanoparticles called quercetin-modified metal-organic frameworks (Zr-MOF-QU) has demonstrated outstanding efficacy for CA IX inhibition in tumor cells. Radiotherapy appears to be an effective use of Zr-MOF-QU[105].

VII. Resveratrol

Blueberries, raspberries, mulberries, and grape skin are just a few of the plants and herbs that naturally contain resveratrol (3,4′,5 tri-hydroxystilbene) (C14H12O3) [106].

The anticancer effects of resveratrol have been shown in a number of human malignancies, including those of the prostate, thyroid, CRC, breast, lung, and bladder. By influencing PCGEM1 and PRNCR1, resveratrol suppresses the AR signaling pathway in cancer of prostate [107].Resveratrol has been shown in another investigation on prostate cancer to be a powerful stimulant in lowering PCAT29 expression by cancer cell lines. Resveratrol and AK001796 have been shown to have synergistic growth suppression action in lung cancer. According to a different research, resveratrol suppresses MALAT1 in CRC, which modifies the Wnt/β-Catenin signaling pathway. Resveratrol anticancer activity on estrogen receptor-α in breast cancer is because of u-Eleanor, a lncRNA, being suppressed.

In the realm of breast cancer treatment, resveratrol emerges as a promising anticancer candidate due to its capacity to inhibit aromatase, consequently suppressing the oncogene LINC00978 [108]. Moreover, collaborative efforts between resveratrol and LINC00978 were observed to impede the growth of breast cancer cells in the same investigation [109]. Furthermore, evidence suggests that resveratrol treatment for lung cancer leads to the downregulation of AK001796 expression.

21.4 Function of ncRNAs in Ferroptosis

Two major functional categories may be distinguished among ncRNAs. The first subgroup consists of necessary structural ncRNAs, also known as "consecutive" ncRNAs, and comprises transcribed RNA the rRNAs, snRNAs, and short ncRNAs [110]. These ncRNAs exhibit consistent expression patterns and participate in essential processes such as translation and splicing. The second category includes regulatory ncRNAs, which are involved in the epigenetic regulation of gene expression. These ncRNAs include miRNAs, siRNAs, circRNAs, piRNAs, and lncRNAs. Currently, regulatory ncRNAs, specifically miRNAs, circRNAs, and lncRNAs, are the primary entities engaged in ferroptosis control [111].

21.4.1 MiRNAs in Ferroptosis

Endogenous miRNAs transcripts characterized by their short hairpin structure, comprising tiny, ncRNAs consist of 21-23 nucleotides. These molecules commonly bind to sequences located in the 3′-UTRsof target mRNAs, resulting in various regulatory effects such as translational repression, mRNA cleavage, degradation, or suppression [112].

Recent research conducted over the past few years has unveiled the ability of miRNAs to modulate ferroptosis by targeting genes associated with this process. For example, miRNA-214 functions to mitigate ferroptosis by targeting and inhibiting TFRC; miR-424-5P and miR-4291 similarly target and stop ACSL4 in order to suppress ferroptosis. Conversely, miR-30b-5p and miR-124 act to promote ferroptosis by targeting and inhibiting FPN1, while miR-541-3p and miR-324-3p enhance ferroptosis by targeting and inhibiting GPX4. Additionally, miR-128-3p, and miR-375 induce ferroptosis by targeting and inhibiting SLC7A11[113].

Certain additional ncRNAs also include miRNA-binding regions, in addition to mRNA. These ncRNAs can compete with mRNAs for the same miRNA pool. This results in the control of miRNA activity, which in turn adds another layer of regulation to the miRNA network [114].

21.4.2 LncRNAs in Ferroptosis

The lncRNAs constitute a diverse group of transcripts exceeding 200 nucleotides in length that do not undergo protein translation. These molecules are essential for the regulation of gene expression through a variety of epigenetic processes, including post-translational, transcriptional, translational, and chromatin remodeling [115].

Certain lncRNAs engage in competitive binding with miRNAs, acting as miRNA sponges to inhibit their activity. By downregulating ferroptosis-associated miRNAs, specific lncRNAs exert control over ferroptosis. In brain ischemia/reperfusion (I/R) injury, lncRNAPVT (lncPVT1) was present to upregulate TFRC and p53 by suppressing miR-214, thus promoting ferroptosis. Similarly, by suppressing miR-128-3p, lncOIP5-AS1 enhances SLC7A11 expression, thereby mitigating ferroptosis in prostate cancer [116].

Furthermore, lncRNAs modulate ferroptosis by influencing protein ubiquitination and mRNA stability through interactions with proteins. For instance, lncPMAN may bind to ELAVL1, stabilizing SLC7A11 mRNA to prevent ferroptosis. Conversely, lncHEPFAL may facilitate SLC7A11 protein ubiquitination and degradation [117]. Additionally, lncP53RRA might sequester p53 in the nucleus through interaction with Ras GTPase-activating protein-binding protein 1 (G3BP1), thereby promoting ferroptosis [116].

21.4.3 CircRNAs in Ferroptosis

circRNAs constitute a family of single-stranded RNA molecules abundant within the eukaryotic transcriptome, characterized by their formation of complete continuous loops extending from the 3′ to 5′ end [118]. These molecules arise from exonic regions through alternative mRNA splicing, distinguishing them from linear RNAs due to their possession of covalently bonded circular structures devoid of a 5′ cap and a 3′ poly A tail. In comparison to linear RNAs, which exhibit considerable stability and regulatory potential, circRNAs are notably more resistant to degradation, thereby conferring enhanced stability [118].

The binding of circRNAs with miRNAs as competitive RNAs or RNA sponges, controlling the miRNAs target proteins in the process. Numerous circRNAs modulate ferroptosis-related genes by inhibiting the relevant miRNAs, hence regulating ferroptosis in this manner. For instance, circRNA IL4R (circIL4R) enhances GPX4 by binding to and inhibiting miR-541-3p, which prevents ferroptosis in hepatocellular cancer. CircLMO1 causes ferroptosis in cervical carcinoma by inhibiting miR-4291 and increasing ACSL4 levels [119].

Furthermore, it has been discovered that circRNAs control ferroptosis by binding to proteins directly. It was discovered that the ALKBH5, an RNA-binding protein, is a negative regulator of ferroptosis and ferritinophagy is bound to and blocked by circ-cIARS (has circ 0008367) [120].

In sepsis-induced acute lung injury, CircEXOC5 may directly interact with the Polypyrimidine tract binding protein 1 (PTBP1), an RNA-binding protein, increases ACSL4 stability. This interaction has the potential to induce ferroptosis [116]. As reported,circRAPGEF5 affects endometrial cancer cells' alternative TFRC splicing, consequently binding to and inhibiting splicing modulator fox-1 homolog 2 (RBFOX2), an RNA-binding protein, to provide resistance against ferroptosis.

The phosphorylation of small heat shock protein 1 (HSPB1) at the Ser-15 position, which is involved in the protective reaction against ferroptosis stress, is inhibited by circST6GALNAC6, according to a recent research [121]. Additionally, it was discovered that circLRFN5 interacts to the PRRX2 protein and encourages its breakdown, which causes GCH1 to be downregulated and ferroptosis to occur. Additionally, it has been revealed that circRNA circ101093 (cir93) interacts with and increases fatty-acid-binding protein 3 (FABP3), which improves AA uptake and utilization to prevent ferroptosis and LPO [116]. According to current reports, circRNAs control ferroptosis in the aforementioned ways in addition to their capacity to code for proteins and control transcription[122].

21.4.4 piRNAs in Ferroptosis

Another category of regulatory ncRNAs is piRNAs. These tiny ncRNAs vary from miRNAs in that they are more complicated, large in size, and lack sequence conservation. They can form a piRNA/piwi complex with piwi proteins, which interacts with a target transcript to silence a gene [123].

There aren't many published research on piRNAs and ferroptosis. PiR-36712 was discovered to downregulate SEPW1 expression in breast cancer cells, piR-36712 binds to SEPW1P, a retro processed pseudogene of SEPW1 RNA. This interaction impedes the competitive binding of SWPW1 mRNA with miR-7 and miR-324, while also suppressing p53 ubiquitination, thereby elevating p53 levels and its downstream target p21. Given the regulatory roles of p53 and p21 in ferroptosis, it is not surprising that piR-36712 is implicated in the modulation of this process. Additionally, piR-31470 collaborates with PIWIL4, a piwi-like RNA-mediated gene silencing 4 complex which subsequently recruits methyl-CpG binding domain protein 2, DNMT1, and DNA methyltransferase 3α to induce and induce glutathione S-transferase P1's (GSTP1) hypermethylation and subsequent inactivation according to another study on prostate cancer [54]. According to related research, Inactivating GSTP1 stops tumor cells from avoiding ferroptosis, which promotes tumor development [124]. This suggests that piR-31470 may reduce ferroptosis by inactivating GSTP1.

21.4.5 Structural ncRNAs in Ferroptosis

Ferroptosis regulation involves structural ncRNAs like as tRNAs and rRNAs in addition to regulatory ncRNAs. tRNAs help linking sequence amino acid and mRNAs, facilitating protein synthesis. In eukaryotes, tRNAs typically range in length from 76 to 90 nucleotides [125]. Since they are necessary for the synthesis of proteins linked to ferroptosis, changes in tRNAs may have an effect on how these proteins are expressed, which may then have an effect on ferroptosis. Interestingly, studies have shown that tRNA mutations cause minor changes in ferroptosis by reducing selenoprotein production, with the exception of GPX4 and GPX1 [126]. Furthermore, it was shown that the inhibition of trans-sulfuration and reduction in cysteine levels caused by a loss of cysteinyl-tRNA synthetase lowers GSH production and suppresses ferroptosis caused by erastin. tRNAs appear to promote ferroptosis and decrease GSH synthesis rather than changing GPX4. On the other hand, studies have shown that quinine-modified tRNAs activate GSH reductase, SOD, GPX, and catalase, improving antioxidant defense. Additionally, studies have shown that ROS increase up when the selenocysteine tRNA gene is removed, suggesting that tRNAs may inhibit ferroptosis by enhancing the effectiveness of the antioxidant defense mechanism[127].

Ribosomes are mostly made up of RRNAs. They are also a kind of ribozyme that functions in ribosomes to synthesize proteins. It's interesting to note that the 5′-UTR of human NRF2 mRNA has a highly conserved 18S rRNA binding site that is necessary for internal translation initiation, indicating that the 18S rRNA controls NRF2 expression [128]. Another investigation, which observed that the 16S/18S rRNA ratio in mouse hepatoma cells decreased by 70% following prolonged ethidium bromide treatment, also noted elevated NRF2 expression [129]. Nrf2 is crucially involved in ferroptosis, it is plausible that 18S rRNA may regulate the expression of Nrf2 to modulate ferroptosis. However, for the initiation of early pre-rRNA synthesis, the protein known as nucleus mitotic apparatus protein (NuMA) may bind to 18S and 28S rRNAs and localize to rDNA-promoter regions. NuMA is downregulated in response to ROS. Additionally, the production of rRNA was observed to be suppressed in leukemia HL-60 cells treated with the iron chelator deferoxamine. These findings suggest a potential regulation of rRNA synthesis during ferroptosis. While tRNAs, snRNAs, and snoRNAs all contribute to post-transcriptional splicing in the expression of genes. Both snoRNAs and snRNAs are structural ncRNAs. Nevertheless, no information regarding the association between snRNAs and ferroptosis is currently available. Inspite of all considerations, current day research is mainly concerned with how regulatory ncRNAs, particularly miRNAs, lncRNAs, and circRNAs controls ferroptosis [130].

21.5 Regulation of ferroptosis-linked ncRNAs and their role in cancers

21.5.1 Lung cancer

The primary cause of death from cancer is lung cancer, about 18.0% of fatalities and 11.4% of diagnosed cases. Reduced miR-27a-3p expression reduces SLC7A11 expression in non-small cell lung cancer cells via binding to its 3'-UTR. MiR-4443 acts as a significant inhibitor of the m6A methyltransferase METTL3. METTL3, in turn, regulates the ferroptosis-associated gene FSP1 via m6A modification, promoting resistance to cisplatin. Additionally, miR-302a-3p targets the 3’UTR of ferroportin, suppressing its expression and reducing intracellular iron levels, ultimately leading to LPO and ferroptosis induction. The miR-6077-Keap1-Nrf2-SLC7A11/NQO1 axis is explained by research; it prevents ferroptosis and provides resistance to cisplatin (CDDP)/pemetrexed (PEM)[110].

LncRNAs and circRNAs play regulatory roles in ferroptosis, a pivotal process in lung cancer development. For instance, lncRNA H19 competitively binds to miR-19b-3p, activating ferritin heavy chain 1, a target of miR-19b-3p, thereby reducing intracellular iron levels and suppressing ferroptosis [131]. Overexpression of lncRNA Uc.339 in lung adenocarcinomas inhibits the maturation of miR-339 by binding to pre-miR-339, leading to upregulation of SLC7A11, a downstream target of miR-339 [132].

Numerous bioinformatics analyses have predicted several ferroptosis-related lncRNAs associated with survival in lung adenocarcinoma (LUAD). The lncRNA, miRNA-101-3p control ferroptosis, an oncogene that promotes LUAD progression and metastasis [133]. W. et al. identified CircDTL as a suppressor of ferroptosis through targeting the miR-1287-5p/GPX4 axis. CircDTL is overexpressed in tissues and cell lines of non-small cell lung cancer (NSCLC) patients. In summary, ncRNAs primarily exert their effects through RNA-RNA interactions to modulate the ferroptosis-related genes expression, thereby influencing lung cancer progression. Particularly, several ncRNAs target SLC7A11, suggesting its significant role [134].

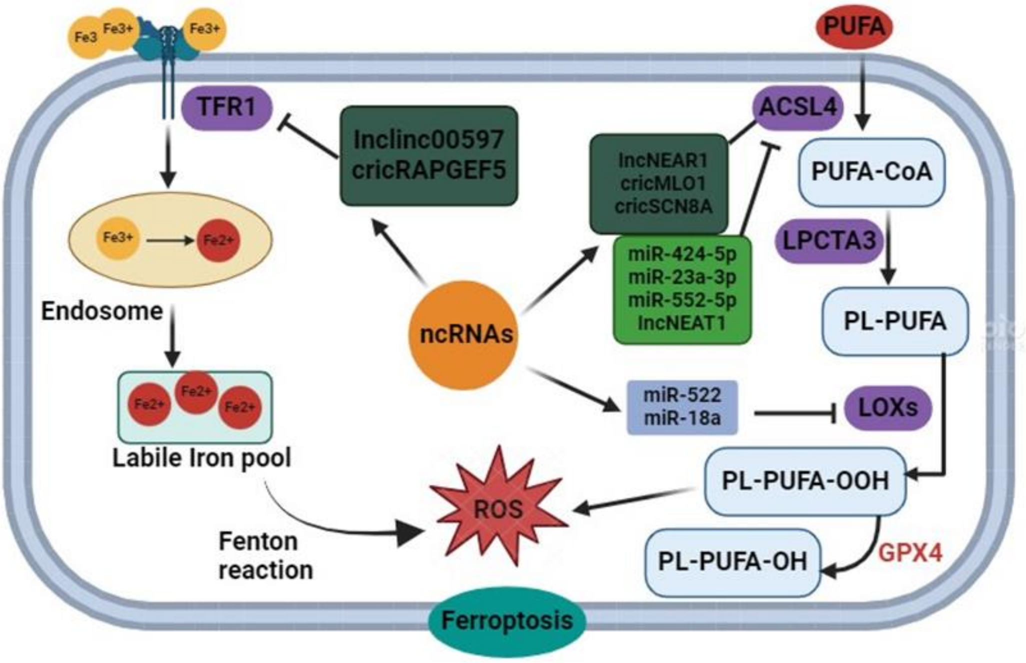

Figure 67: Modulation of ferroptotic pathway by ncRNAs in cancers.

21.5.2 Esophageal cancer

Downregulation of the SLC7A11 levels by upregulating miR-513a-3p, enhances ferroptosis while lowering cell proliferation and metastasis in esophageal squamous cell carcinoma (ESCC) [135]. The pivotal function of circPVT1 in ESCC 5-FU is also explained that ferroptosis can also be managed by miR-30a-5p [136]. Knockdown of circPVT1 led to downregulation of SLC7A11, p-β-catenin, and GPX4, which was reversed by upregulating FZD3 and inhibiting miR-30a-5p [137]. Overexpression of E2F7 caused by hypoxia in esophageal cancer cells activates QKI at transcriptional level, leading to the production of cricBCAR3 [138]. By binding to miR-27a-3p, cricBCAR3 increases the transportin-1 (TNPO1) expressions and stimulates the migration, invasion, and multiplication of cancer cells.

Due to the limited identification of ncRNAs associated with ferroptosis in esophageal cancer, their significance in this cancer type may be comparatively lower than in others[139](Table 2).

Table 26: ncRNAs role in regulating ferroptosis in cancers.

ncRNA species	**Induction of ferroptosis(↑) or suppressed(↓) by ncRNAs in various cancers**	**Mode of action**	**Ref.**
miR-375	↑Gastric cancer	↓SLC7A11	[140]
miR-4715-3p	↑Esophageal carcinoma, ↑Gastric cancer	↓AURKA, ↓GPX4	[141]
miR-214-3p	↑HCC	↓ATF4, ↓SLC7A11	[142]
miR-101-3p	↑Lung cancer	↓GPX4, ↓PTGS2, ↓TBLR1, ↓NF-κB	[143]
miR-324-3p	↑LUAD, ↑Breast cancer	↓GPX4	[144-145]
miR-5096	↑Breast cancer	↓SLC7A11	[146]
miR-1287-5p	↑Osteosarcoma	↓GPX4	[147]
miR-137	↓Melanoma	↓SLC1A5	[148]
miR-9	↓Melanoma	↓GOT1, and inhibit glutaminolysis	[112]
miR-130b-3p	↓Melanoma	↓DKK1, ↑Nrf2/HO-1	[112]
miR-103a-3p	↓Melanoma	↓GLS2, ↓Hydrolysis of glutamine to glutamate	[149]
miR-522	↓Gastric cancer	↓ALOX15	[150]
P53RRA	↑Lung cancer	↓SCL7A11	[151]

(LINC00472)			
MT1DP	↑NSCLC	↑Stability of miR-365a-3p, ↓Nrf2	[152]
GABPB1-ASI	↑HCC	↓GABPB1, ↓PRDX5	[153]
LINC00618	↑Leukemia	↓SLC7A11	[154]
LINC00336	↓Lung cancer	Stabilized by ELAVLI & LSH MiRNA6852 ↑CBS.	[155]
NEAT1	↓NSCLC	↓ACSL4	[156]
H19	↓Breast cancer	↓ROS, ↑glutathione	[157]
lncPVT1	↓HCC	↑GPX4	[158]
OIP5-ASI	↓Prostate cancer	Sequesters miR-128-3p, ↑SLC7A11	[159]
RP11-89	↓Bladder cancer	miR-129-5p, ↑PROM2, facilitates iron export	[160]
MEG8	↓Benign hemangioma	Sequestered by miR-497-5p, ↑SLC7A11, ↑GPX4	[161]
cIARS	↑ Ferritinophagy in sorafenib-treated HCC	↑ ALKBH5, ↑Bcl-2 Transcriptional stability, ↑Interaction of Bcl-2 with BECN1	[161]
cric0000190	↑Gastric cancer	miR-382-5p sponge, ↑ZNRF3, ↓Wnt/β-catenin pathway	[162]

cricKDM4C	↑AML	miRNA let-7b-5p sponge, ↑ACSL4, ↑PTGS2, ↑p53, ↓GPX4, ↓FTH1	[163]
circ-TTBK2	↓Glioma	Sequesters miR-761, ↑ITGB8	[164]
circKIF4A	↓Papillary thyroid cancer	Sequesters miR-1231, ↑GPX4	[120]
circDTL	↓NSCLC	miR-1287-5p sponges, ↑GPX4	[165]
circRHOT1	↓Breast cancer	miR-106a-5p sponges, ↑STAT3	[166]
circIL4R	↓HCC	miR-541-3p sponge, ↑GPX4	[167]
circABCB10	↓CRC	miR-326 sponges, ↑CCL5	[168]
circEPSTI1	↓cervical cancer	miR-375, miR-409-3p & miR-515-5p sponges, ↑SLC7A11	[169]

21.5.3 Gastric cancer

In 2020, gastric cancer resulted in the loss of 769,000 lives worldwide, ranking it as the fifth most prevalent cancer. Another study confirmed that miR-375 can diminish the stemness of GC cells by prompting ferroptosis via SLC7A11 targeting [140]. Paclitaxel and cisplatin facilitate miR-522 secretion from CAFs through the activation of the USP7/hnRNPA1 axis. Moreover, miR-522 inhibits ALOX15, thereby reducing ferroptosis in cancer cells and subsequently decreasing chemosensitivity [150]. A distinct epigenetic mechanism governing the induction of Aurora kinase A (AURKA) was elucidated in a recent study, where suppression of miR-4715-3p in UGCs has also been observed. MiR-4715-3p specifically targets the 3'UTR of AURKA, leading to its downregulation. Goma and colleagues (2019) showed that GPX4 is inhibited and cell ferroptosis is induced by reinstalling miR-4715-3p or suppressing AURKA. lncRNAs also play a significant role in ferroptosis in gastric cancer. HIF-1α stimulates the production of the lncRNA PMAN by binding to the internal HRE site in the promoter.

Enhancement of SLC7A11 mRNA stability by LncRNA PMAN, which is prone to ELAVL1 plasmic distribution [170]. HIF-1α stimulates the synthesis of the lncRNA CBSLR, which forms the CBSLR/YTHDF2/CBS signaling axis with YTHDF2 to decrease the stability of CBS mRNA by improving its attachment to the m6A-modified coded sequence (CDS). Reduced levels of CBS cause the ACSL4 protein to become less methylated, which in turn causes protein polyubiquitination and ACSL4 breakdown. This lowers the pro-ferroptosis of phosphatidylethanolamine (PE) [171]. The lncRNA BDNF-AS has been reported to be significantly expressed in tissues affected by both peritoneal metastasis (PM) and gastric cancer (GC). Subsequent studies revealed that FBXW7 controls VDAC3 protein production by ubiquitination to shield GC cells from ferroptosis. Furthermore, BDNF-AS can affect FBXW7 transcription by attracting WDR5, which in turn affects FBXW7 expression. Additionally, ferroptosis-related lncRNAs that might serve as prognostic markers or references for clinical outcomes were predicted by bioinformatics analysis. In general, environmental stressors that stomach cancer encounters can cause ferroptosis, and this impact might be more reliant on ncRNAs. This property makes it easier to identify ncRNAs linked to ferroptosis in gastric cancer[172].

21.5.4 Colorectal cancer (CRC)

In 2020, CRC ranked third in terms of global incidence, with more than 1.9 million new cases reported. Elevated levels of transferrin inhibited the ability of miR-545 to induce rises in ROS, malondialdehyde (MDA), and iron (Fe^{2+}) levels in HT-29 and HCT-116 cells, consequently promoting ferroptosis in CRC [173]. Zheng, S. et al. discovered that miR-545 increased CRC cell survival by lowering transferrin [174]. An additional investigation revealed that miR-15a-3p stimulates ferroptosis by directly suppressing GPX4 expression by attaching to the 3'-untranslated region of GPX4. This leads to elevated levels of ROS, intracellular Fe^{2+}, and accumulation of MDA both *in vitro* and *in vivo* (Liu et al., 2022a). Furthermore, miR-19a directly targets IREB2, which induces ferroptosis[175].

Ferroptosis, the mechanism by which CRC progresses, has also been demonstrated to be impacted by a multitude of lncRNAs and circRNAs. Another study investigate transcriptome profiles of lncRNAs in primary CRC tissues, LINC00239 is substantially overexpressed in these tissues and its overexpression is associated with a worse prognosis and survival. Functionally, LINC00239 suppresses ferroptosis and enhances resistance to chemotherapy by engaging in interactions between nucleotides 1-315 of LINC00239 and Keap1 Kelch domain, which hinders the ubiquitination of Nrf2 and elevates Nrf2 protein stability [176]. According to research by Yajun Luo et al., LINC01606 thwarts ferroptosis in colon cancer cells by boosting mitochondrial membrane potential while reducing iron levels, lipid derived ROS, and mitochondrial superoxide. By serving as a competitive endogenous RNA that regulates the expression of miR-423-5p and activates the canonical Wnt/β-catenin signaling pathway, LINC01606 mechanistically enhances the expression of stearoyl-CoA desaturase 1 (SCD1). When the transcription factor TFE3 attaches to a specific part called IGHM enhancer 3, it makes LINC01606 gene produce more RNA after joining with the starting regions of LINC01606 [177]. Wang, Y. et al. found that the circ0007142 is a miR-874-3p sponge [54]. In conclusion, a large number of ncRNAs have been linked to CRC, and a variety of downstream pathways imply that CRC may be more susceptible to treatments associated to ferroptosis.

21.5.5 Pancreatic cancer

Extremely malignant is pancreatic cancer. Due of its extremely dismal prognosis, there were 466,000 fatalities and 496,000 new cases [178]. According to another study, poly (rC)-binding protein 3 (PCBP3), which is intimately related to iron metabolism, may directly interact with lncRNA A2M-AS1. Furthermore, it has been found that the AKT-mTOR signaling pathway may be inhibited and p38 activation made easier by the A2M-AS1/PCBP3 axis, and that ferroptosis is regulated by these two routes [179]. Another axis that can prevent ferroptosis and forecast tumor mutation load, immune infiltration, and treatment sensitivity in pancreatic adenocarcinoma is the linc02432/Hsa-miR-98-5p/HK2 axis. Certain lncRNAs associated with ferroptosis, including lncZNF236-DT, lncCASC8, and lncPAN3-AS1 (Ping), were discovered using bioinformatics research in pancreatic cancer. Treatments for ferroptosis may not be inadequate for pancreatic cancer since no numerous ncRNA have yet been identified to regulate ferroptosis in this kind of cancer [180]. Current research has not examined the link between ncRNAs and standard ferroptosis genes.

21.5.6 Hepatocellular carcinoma

In 2020, liver cancer will account for around 906,000 new diagnoses and 830,000 deaths worldwide, making it the sixth most common illness overall [181]. By controlling ferroptosis, the ETS1/miR-23a-3p/ACSL4 axis is essential in hepatic cellular carcinoma (HCC) resistance to sorafenib [134]. By identifying the DNA-binding GGAA/T order, ETS1 transcriptionally activates the promoter of miR-23a-3p. MiR-214-3p promotes intracellular iron buildup and decreases glutathione (GSH) by blocking ATF4, which facilitates ferroptosis. LncRNA HEPFAL is less stable than SLC7A11 and has been shown to have decreased expression in HCC. It can also cause ferroptosis by increasing inside cells lipid ROS and iron levels. Moreover, ferroptosis induced by erastin becomes more pronounced in the presence of lncRNA HEPFAL, possibly through its connection to mTORC1 [182]. Erastin functions with eIF4A and prevents GABPB1 mRNA from adhering to polysomes. This lowers the quantity of GABPB1 protein produced by increasing the lncRNA GABPB1-AS1. This eventually leads to a decrease in cellular antioxidant capability and the silencing of the gene that encodes peroxiredoxin-5 (PRDX5) peroxidase [116]. By promoting p53's binding to the NEAT1 promoter, erematin and RSL3 enhance the expression of the lncRNA NEAT1.To prevent binding to MIOX's 3'UTR, NEAT1 competes with miR-362-3p for interaction. Ferroptosis is exacerbated by this competition, which increases MIOX-mediated ROS generation and decreases intrinsic levels of both NADPH and GSH. For miR-3200-5p, lncRNA HULC functions as a competitive endogenous RNA (ceRNA). By the targeting of ATF4, miR-3200-5p regulates ferroptosis, which inhibits the growth and spread of HCC cells [179]. A positive correlation exists between LINC01134 and GPX4, which is associated with unfavorable clinical outcomes. Silencing LINC01134 enhances oxaliplatin sensitivity by reducing total ROS, lipid ROS, and MDA levels, as well as lowering the GSH/GSSG ratio. Moreover, it promotes Nrf2 recruitment to the GPX4 promoter region through RNA-DNA sequence interaction. Certain ferroptosis related lncRNAs associated with immunosuppression drug and phenotype susceptibility were predicted by Guanghao Li et al. [183]. This could enhance ferroptosis by inducing ferritinophagy and inhibiting ALKBH5's anti-autophagy activity. Furthermore, it was shown that CircIL4R inhibits ferroptosis in HCC via regulating the expression of GPX4 and acting as a sponge for miR-541-3p. A different research found that HCC tissues and cell lines have higher than normal levels of Circ0097009. Ferroptosis was made easier by circ0097009 knockdown via the circ.0097.009/miR-1261/SLC7A.11 axis. It appears that ncRNAs participate in various stages of ferroptosis by regulating key genes or pathways. Ferroptosis-targeting drugs may hold promise for treating HCC, a tumor with significant potential[184].

21.5.7 Renal cell carcinoma

Global data for 2020 showed that kidney cancer claimed the lives of about 431,000 people. Recent research has shown that clear-cell carcinoma of renal cells (CCRC) tissues had lower levels of miR-4735-3p. It has been demonstrated that increased miR-4735-3p expression inhibits SLC40A1 and promotes ferroptosis [183]. Furthermore, icariside II, an anticancer flavonoid, was found to upregulate miR-324-3p, which in turn decreases GPX4 expression and induces ferroptosis in CCRC. Overall, insufficient research currently exists to establish the connection between ncRNAs and ferroptosis pathways. Consequently, the role of ferroptosis-related ncRNAs in CCRC development remains uncertain.

21.5.8 Bladder cancer

Bladder cancer was responsible for around 573,000 cases of diagnosis and 213,000 deaths worldwide in 2020. The incidence rate of the disease was four times greater in men than in women [185]. Recent research suggests that miR-129-5p, which targets PROM2, uses lncRNA RP11-89 as a sponge. PROM2 facilitates iron exportation and defense against ferroptosis by encouraging the development of multivesicular bodies [60]. Moreover, comprehensive studies have revealed strong prognostic associations of several lncRNAs in bladder cancer. CircST6GALNAC6 acts as a suppressor of tumor growth in bladder cancer, potentially promoting ferroptosis through the circST6GALNAC6/HSPB1/p38 MAPK pathway. Furthermore, by attaching to the N-terminus of a small heat shock proteins 1 (HSPB1) and preventing phosphorylation at the Ser-15 position, circST6GALNAC6 may reduce the amount of HSPB1 and activate the P38 MAPK pathway [186]. Current research suggests that ncRNAs influence ferroptosis via the PROM2 and MAPK pathways, while the interaction between ncRNAs and other crucial genes or pathways remains unclear. Consequently, further investigation is warranted to elucidate the phenotype of bladder cancer mediated by ncRNAs through ferroptosis.

21.5.9 Prostate cancer

Prostate cancer is the second most common illness among males worldwide in 2020, accounting for over 375,000 fatalities [187]. According to recent study, miR-15a binds to the 3'UTR of GPX4 and inhibits cell growth, promoting ferroptosis in prostate cancer [188]. Furthermore, Jiang, X. et al. revealed that downregulation of lncRNA PCAT1 had the reverse impact of ferroptosis inhibition and increases resistance to docetaxel. On the one hand, lncRNA

PCAT1 increases the stability of C-Myc protein by connecting with its 151-202 amino acids. This, in turn, increases the production of SLC7A11 by binding to its transcriptional promoter. On the other hand, by enclosing miR-25-3p, lncRNA PCAT1 promotes SLC7A11 overexpression. Additionally, the PCAT1 promoter can bind to the transcription factor TFAP2C, increasing PCAT1 expression [189]. In PC3 and DU145 cells exposed to Cd (Cadmium), lncRNA OIP5-AS1 promotes cell proliferation, colony formation, and cell invasion while inhibiting ferroptosis. LncRNA OIP5-AS1 inhibits ferroptosis through the miR-128-3p/SLC7A11 pathway by acting as a competitive endogenous RNA for miR-128-3p. Compared to other urinary tumors, prostate cancer-associated ncRNAs seem to have a greater propensity to influence fundamental ferroptosis genes like GPX4 and SLC7A11, suggesting that experimental designs targeting these downstream classic genes could be a valuable approach for investigating ferroptosis-related ncRNAs [179].

21.5.10 Breast cancer

Breast cancer mediated mortalities (2.3 million) are highest at global level. New breast cancer cases identified in 2020 were 11.7% of all diagnosed cancers [190]. MiR-5096 stimulates ferroptosis in MDA-MB-231 cells by targeting SLC7A11, as revealed by Yadav, P. et al. This results in a reduction in proliferating, colony formation, migration, and invasion [191]. The diabetic medication metformin causes ferroptosis by binding to the 3 UTR of GPX4 and inhibiting it with miR-324-3p. Trapping the signaling transducers and activator of transcription 3 (STAT3), CircRHOT1 acts as a sponge for miR-106a-5p, which in turn reduces ferroptosis and ROS in malignant breast cells. CircRNA GFRA1 mechanically sequesters miR-1228, which targets the ferroptosis inhibitor AIFM2 (Bazhabayi et al., 2021). Together, STAT3 and AIFM2 are identified as 2 novel mediators of ferroptosis in breast cancer, indicating a potential rise in the frequency of novel ferroptosis-associated signal mediated by ncRNAs [192].

21.5.11 Ovarian cancer and cervical cancer

In 2020, huge number (0.314 million) of cases were diagnosed for ovarian cancer, and 0.2 million deaths reported worldwide. According to recent research, raising miR-424-5p levels reduces the production of ACSL4 by binding directly to its 3'-UTR, which lessens the amount of iron metabolism in ovarian cancer that is caused by erastin and RSL3 [193]. The lncRNA ADAMTS9-AS1 actively suppresses ferroptosis by targeting the miR-587/SLC7A11 axis [194]. Limited evidence supports the regulatory role of ferroptosis governed by ncRNAs. Further investigation into the molecular mechanisms of ncRNA-mediated ferroptosis regulation is warranted. Among women, cervical cancer is the fourth most common cause of cancer-related death. Globally, it is predicted that in 2020 there would be 604,000 new instances and 342,000 fatalities [195]. Studies related to the circRNA-mediated ferroptosis showed decreased level of CircLMO1 in cervical carcinoma cells. CircLMO1 reduces cell proliferation and invasion by acting as a sponge for miR-4291, which in turn increases ACSL4 expression, thereby inducing ferroptosis. Remarkably, researchers discovered that circLMO1 is suppressed by the RNA-binding enzyme DExH-Box Helicase 9 (DHX9), but the reverse complement sequence in intron 3 (I3RC) speeds up the circularization of circLMO1. The circEPSTI1-miR-375/409-3P/515-5p-SLC7A11 axis impacts cervical cancer proliferation through the competing ceRNA mechanism, with circEPSTI1 suppression enhancing ferroptosis mediated by SLC7A11. In SiHa and HeLa cells, the reduction of circACAP2 results in ferroptosis and inhibits cell proliferation. CircaACAP2 functions as a ceRNA for miR-193a-5p, controlling its target gene GPX4. Several traditional signals linked to ferroptosis, including as GPX4, SLC7A11, and ACSL4, were found in cervical cancer, indicating that circRNAs are essential for controlling signals related to ferroptosis [179, 196]. Further investigation is warranted to ascertain the potential of circRNAs as therapeutic targets for breast cancer.

21.5.12 Acute myeloid leukemia (AML)

In 2020, over 474,000 new cases of leukemia were recorded worldwide, with 311,000 deaths (accounting for 3.1% of all fatalities across genders). AML, also known as acute myeloid leukemia, is made worse by ferroptosis when LINC00618 suppresses the production of lymphoid-specific the helicase (LSH). By attaching to the promoter regions of SLC7A11, LSH in turn increases SLC7A11's transcription. It's worth noting that vincristine (VCR), a chemotherapy medication, can induce LINC00618 [197]. Additionally, AML patients exhibited reduced levels of circKDM4C, but overexpression of circKDM4C may encourage ferroptosis and prevent the invasion, migration, and multiplication of cells [198]. Mechanistically, circKDM4C elevates p53 expression by sequestering hsa-let-7b-5p. More comprehensive investigations are necessary to fully elucidate the regulation of ncRNAs associated with ferroptosis in AML, as current information is insufficient.

21.5.13 Glioblastoma

Heterogeneity, invasion, and migration of malignant cells are defining features of glioblastoma, rendering the disease incurable [199]. According to another study, miR-147a mimic targets the 3'UTR of SLC40A1 in U87MG and A172 cells, leading to suppressed cell viability and induction of ferroptosis. Notably, this effect does not extend to the iron storage protein FTH1 or the intake protein TFR [200]. miR-670-3p targets ACSL4 to prevent iron metabolism in U87MG and A172 cells [201]. Depletion of lncRNA TMEM161B-AS1 induces ferroptosis in U87 and U251 cells, consequently suppressing cell proliferation, migration, and invasion. Mechanistically, lncRNA TMEM161B-AS1 binds to the seed area of the 3' UTR sequences of FANCD2 and CD44, acting as a sponge for hsa-miR-27a-3p. Increased levels of lipid ROS and intracellular iron buildup are the results of FANCD2 and CD44 knockdown. Additionally, CircLRFN5 interacts with PRRX2 transcription factor to facilitate its destruction through the proteasomal route controlled by ubiquitin. PRRX2 then causes glioma stem cells (GSCs) to express GCH1 more highly, which prevents ferroptosis by producing the antioxidant tetrahydrobiopterin (BH4) [202]. In glioblastoma, ncRNAs work together to control the iron transporters SLC40A1 and PRRX2's degradation. To clarify the regulation processes of ncRNAs in glioblastoma with respect to other ferroptosis-associated molecules, including GPX4, SLC7A11, and Nrf2, more research is necessary.

21.5.14 Melanoma

Globally, there were about 324,000 new instances of melanoma detected in 2020, which led to 57,000 fatalities. MiR-9 stop ferroptosis by downregulating the production of glutamic-oxaloacetic transaminase (GOT1). Furthermore, in A375 and G-361 melanoma tumor cells, miR-137 reduces ferroptosis by hitting SLC1A5 in a way reliant on glutaminolysis, lowering glutamine uptake. However, the inhibitory impact on ferroptosis may be reversed by over expressing SLC1A5 [203]. Intracellular iron buildup and lipid ROS levels are reduced by miR-130b-3p, but this effect is neutralized by DKK1 overexpression. Additionally, ferroptosis increases chemotherapy's effectiveness. MiR-21-3p targets TXNRD1 to induce ferroptosis and LPO. Furthermore, by causing ferroptosis, anti-PD-1 treatment for melanoma enhances sensitivity due to the miR.-21-3p/TXNRD1 axis. [204]. In contrast to other cancers, ncRNAs predominantly regulate the Xc- subunit SLC1A5 in melanoma, providing a novel perspective on the control of ferroptosis, melanoma suppression, and chemotherapy responsiveness.

21.6 Role of ncRNA in aging

Aging is defined as increasing physiological degradation and, as a result, decreased function, as evidenced by many molecular and cellular markers. Aside from genomic instability and telomere attrition, breakthroughs in ageing science have revealed multiple factors of ageing, making this physiological process complex and convoluted (Figure 6). Senescence affects both cells and organs, leading to age-related disorders such cardiovascular disease, AD, cancer, and sarcopenia, sometimes accompanied by other conditions. Meanwhile, fragility and frailty cause a high rate of death. As the global population over the age of 60 is predicted to double to 22% by 2050, there will be a rise in illness and death among the elderly. As a result, the ageing of the world population has emerged as a serious healthcare concern that requires deeper investigation into the explicit processes behind the ageing process [205-206]. Cellular proteome and transcriptome modifications play a crucial role in physiological changes in cells, tissues, and organ systems as we age. Advancements in microarray and sequencing techniques have improved understanding of mammalian genomes along with transcriptomed. Just 2% of transcript encodes protein RNAs, according to several initiatives including the Encyclopedia of DNA Elements and the Function-Based Annotation of the Mammalian Genome. The vast majority comprise ncRNAs, which serve various roles as versatile RNA molecules [207].

Research into the complex mechanisms of aging and aging-associated pathologies has revealed intricate connections between molecular pathways, and cellular processes such as iron-mediated LPO and ferroptosis. Both miRNAs and lncRNAs, have well recognized for their roles in post-transcriptional activities across various cellular processes, particularly ferroptosis [208].The disruption of ferroptosis regulation has been linked to a range of age-related conditions, such as neurodegenerative disorders, cancer, and cardiovascular ailments[209].

Recent research has shed light on the intricate mechanisms underlying ferroptosis and its implications in aging and age-related diseases. One key aspect of ferroptosis is its dependence on iron and lipid metabolism. Iron is essential for various cellular processes, including DNA synthesis, oxygen transport, and mitochondrial respiration. LPO is a hallmark feature of ferroptosis that leads to the accumulation of lipid hydroperoxides, which disrupt membrane integrity and induce cell death. Key regulators of LPO include the enzyme system composed of arachidonate lipoxygenases (ALOXs), which catalyze the production of lipid hydroperoxides, and GPX4 that reduces lipid hydroperoxides to certain alcohols and shield the cells against ferroptosis [210].

Ferroptosis control is mostly dependent on redox homeostasis in addition to LPO. A redox signaling transcriptional factor Nrf2 regulates antioxidant responses of the cells against oxidative stress. It protects against iron metabolism by upregulating the expression of antioxidant genes specifically HO-1 and NQO1[211].

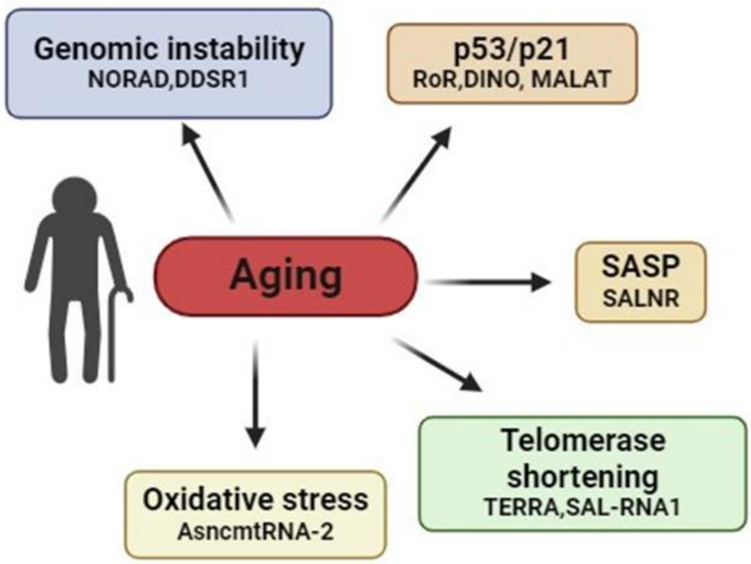

Figure 68: Role of ncRNA in aging and aging associated pathologies.

21.7. Role of lncRNAs in cellular aging

In untransformed cells, telomere attrition, chromosomal instability, DNA damage, mitochondrial dysfunction, oncogene activation, and other cell cycle-related stresses can all result in senescence, a permanent type of growth stop (Figure 7). Senescent cells have distinct molecular, secretory, and morphological features. Flattened, bigger cells, increased SA-*β*-galactosidase activity, and different expressions of senescence-associated pathways (e.g., upregulate p53, p21, p27, and downregulated Sirt1) are characteristics of the senescence-associated secretory phenotype (SASP). Cellular senescence plays a crucial role in the natural aging process. However, the accumulation of senescent cells over time can lead to significant detrimental effects on organisms[211]. These consequences may stem from: (a) diminished regeneration caused by the depletion of stem cells; (b) tissue and organ failure induced by the SASP; and (c) disruptions in energy balance resulting from various stressors[208]. Conversely, cellular senescence provides protection against carcinogenesis, aligning with the intricate interplay between senescence and tumor response pathways. The oncogene-induced senescence (OIS) model is frequently utilized to illustrate this reciprocal interaction concept. Recent research indicates that lncRNAs regulate networks associated with cellular senescence, including those involving p53/p21, pRB/p16, and p14 [212].

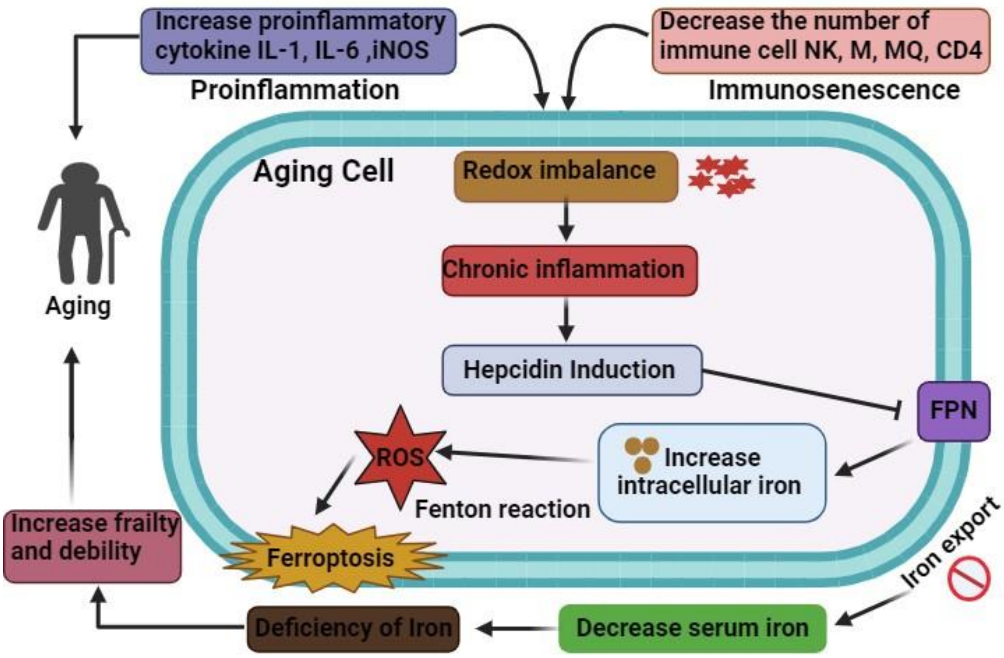

Figure 69: The decline of the immune system and imbalances in cellular redox homeostasis with age can cause ongoing inflammation, triggering the production of hepcidin, a hormone that disrupts the balance of iron in the body. This disruption, along with the occurrence of ferroptosis, significantly contributes to the acceleration of aging, weakening and debility.

21.7.1. Cell cycle–associated lncRNAs

Senescence denotes the permanent halt of typical cell cycle progression triggered by various cellular strains like DNA damage, oxidative stress, telomere shortening, and environmental pressures. Additionally, cell cycle suppressors such as p16, p21, and p53 are recognized for reducing tumors linked to senescence. LncRNAs implicated in cell cycle regulation could influence both senescence and the aging process in organisms [213].

- **MALAT1**

  MALAT1, situated within nuclear speckles, serves as a regulator of the cell cycle. Its heightened expression is a common feature in various solid tumors, contributing to cancer metastasis and recurrence [214]. MALAT1 is involved in facilitating cell cycle progression was elucidated [215]. Numerous studies on cell lines have revealed that depleting MALAT1 leads to G1 or G1/S phase arrest, impeding cell proliferation and growth while promoting a senescent phenotype. Interestingly, MALAT1 knockout mice did not display any apparent abnormalities [216]. Collectively, findings suggest that while MALAT1 may not be essential for organismal development, it could hold significance under specific clinical or environmental circumstances.

- **ANRIL**

ANRIL, an antisense transcript to the p15/CDKN2B/CDKN2B/CDKN2A/ARF gene cluster, has been demonstrated to reduce the expression of CDKN2A and CDKN2B (p15 INK4B) genes locally. This lncRNA is extensively implicated in regulating cell proliferation, senescence, and the aging process. Knocking down ANRIL in WI-38 and IMR-90 cells leads to the upregulation of p15 INK4B, resulting in diminished cell proliferation and the onset of a senescent phenotype. Recent investigations have explored its association with inflammation. The observed positive correlation between ANRIL and TNF-α and NF-κB suggests that ANRIL might play a role in aging and age-related ailments, such as cardiovascular disease and AD [217].

- **7SL**

Many different forms of cancer have been linked to 7SL. It is a highly conserved lncRNA found in the cytoplasm that binds to six signals recognition proteins. It forms a partial duplex with the 3′ untranslated region of p53 mRNA and competes with the HuR protein for binding. Cell cycle arrest and senescence were established in studies incorporating the silencing of 7SL in HeLa and HCT116 cells by increasing p53 translation through an improved relationship between p53 mRNA and HuR [218].

- **MEG3**

A ncRNA called maternally expressed gene 3 (MEG3) shows maternal expression and imprinting. It is involved in several biological processes, including angiogenesis, liver metabolism, and the growth of the nervous system in the body. While MEG3 is abundantly present in certain normal tissues, its expression is suppressed in numerous tumors. MEG3 impacts the activity of several crucial regulators of the cell cycle, including p53, MDM2, GDF15, and RB1[219]. Reinstating MEG3 expression in cell lines like HeLa, C-33A, MCF-7, and H4 resulted in reduced tumor growth by inducing G2/M cell cycle arrest and apoptosis. On the other hand, lowering MEG3 levels promoted autophagy, cell division, and resistance to apoptosis. MEG3 has the potential to be a target for cancer detection, prognosis, and treatment since it is a tumor suppressor. Huntington's disease (HD) and other age-related neurodegenerative disorders have also been linked to decreased MEG3 levels. It may appear incongruous to comprehend the processes of epigenetic gene control in neurons and cancer cells. To gain further insight into its role in brain aging, a deeper understanding of the processes governing senescence and apoptosis is necessary [220].

- **H19**

H19, a conserved lncRNA, is expressed maternally and is located near the IGF2 genes, which are expressed paternally. One kind of RNA that regulates gene expression in our cells is called H19. It has been discovered to promote cell division and proliferation while delaying the aging process of cells. These interrelated consequences may contribute to the onset and progression of cancer. The linkage H19 and IGF2 (H19-IGF2) genes promotes a consistent gene expression pattern, which is essential for cellular activities such as development, proliferation, senescence, and apoptosis. Loss of imprinting at the H19-IGF2 gene has been linked to cellular aging. Notably, hypomethylation of this locus observed during aging results in elevated H19 expression but reduced IGF2 expression, indicating increased longevity and reduced tumor incidence. Conversely, hypermethylation-induced imprinting loss with aging leads to heightened expression of both genes, potentially elevating the risk of cancer later in life. H19 has been implicated in various age-related conditions, including adipose deposition and muscle regeneration [221].

- **UCA1**

Urothelial cancer-associated 1 (UCA1), initially identified in bladder transitional cell carcinoma, promotes cell proliferation and suppresses apoptosis [222]. It also acts as a precursor to other **miRNA**s in malignant tumors. UCA1 overexpression is suggested to induce cellular senescence, which is considered a form of tumor suppression. Research shows that the TBX3/CAPERα repressor complex is essential for delaying senescence in mouse embryos and primary cells. Stress can cause the CAPERα/TBX3 complex to break down, which increases UCA1 RNA synthesis and causes senescence. This complex inhibits the RB pathway and p16 INK4A transcription while controlling chromatin shape. hsRNPA1 interacts with and breaks down p16 INK4A mRNA in actively dividing cells. But UCA1 moves hsRNPA1 out of its binding region on p16 INK4A during senescence, stabilizing p16 INK4A mRNA [223].

- **FAL1**

An extensive examination of the genome identified the initial instance of focal amplification of a lncRNA on chromosome 1, termed FAL1, within somatic alterations of lncRNA copy numbers across 2394 tumor samples from 12 different cancer types. FAL1 exhibits notable oncogenic properties, partly attributed to its ability to reduce the levels of p21 through its interaction with BM1. Conversely, the suppression or reduction of FAL1 expression causes a G0/G1 phase cell cycle halt and induces cellular senescence [206].

- **Gadd7**

Derived from Chinese hamster ovary cells were Gadd7, and its expression levels were assessed following DNA damage. Enhanced expression of gadd7 leads to G1 phase arrest and stimulates apoptosis by interacting with TDP-43 and preventing its association with Cdk6 mRNA. This interference results in the degradation of Cdk6, causing cell cycle arrest and the development of a senescent phenotype. Consequently, the potential impact of gadd7 on aging is anticipated [206].

- **MR31HG**

MR31HG, also known as the MIR31 host genes or the LOC554202, is a region of DNA situated approximately 400 thousand units distant from the p16 INK4A locus on the human genome. This indicates that, in terms of DNA sequence, it comes before p16 INK4A. It contains miR-31, which is downregulated in malignant circumstances but shows upregulation in elderly human umbilical vein endothelial cells (ECs). Literature have indicated that MR31HG might impact cell proliferation and thwart carcinogenesis through miR-31 regulation [206]. A recent investigation discovered that MR31HG expression is heightened during OIS, and suppressing this lncRNA triggers p16 INK4A-dependent senescence. In cells approaching senescence, MR31HG is distributed in both the nucleus and cytoplasm, but upon BRAF activation, it predominantly localizes to the cytoplasm. MR31HG employs polycomb group (PcG) proteins to bind to genomic regions of both p16 INK4A and MR31HG. PcG proteins and elevated amounts of MR31HG are essential for PcG-mediated regulation of the p16 INK4A gene during OIS [206].

PANDAp21-associated ncRNA

PANDA, an acronym for activation of p21-associated ncRNA DNA damage, is a p53-regulated transcript that arises from the p21 promoter. Damage to DNA initiates its activation. It regulates apoptosis, senescence, and cell proliferation in human fetal lung fibroblasts and newborn foreskin cells. PANDA functions by imitating the transcription factor NF-YA, which promotes proliferation. Furthermore, PANDA suppresses the MAPK/ERK pathway in response to p53 activation, which causes G1 cell cycle arrest in lymphomas. Interestingly, PANDA regulates senescence's onset and termination by different ways. Through the formation of the PANDA-SAFA-PRC-BMI complex, PANDA suppresses the expression of pro-senescence genes in proliferating cells. On the other hand, elevated PANDA levels in senescent cells lead it to separate from this complex. By inhibiting genes that encourage proliferation and increasing the expression of pro-senescence genes, this separation causes senescence arrest. Fibroblasts often emerge from senescence with PANDA elimination by siRNA depletion. PANDA targeting has potential for age-related disease therapies due to its dual impact on senescence and proliferation [224-225].

- **lincRNA-p21**

P53-associated lincRNA-p21 recruits hnRNP-K to its promoter region, leading to a decrease in proliferation of cells in mouse embryonic fibroblasts. LincRNA p21 interacts with various factors, such as MDM2 and Rck, to positively modulate p53 transcription. The HuR/Ago2/let-7 complex destabilizeslincRNA-p21, thereby alleviating translational inhibition on target mRNAs. Additional investigations indicate that lincRNA-p21 impedes somatic cell transformation by inducing either senescence or apoptosis. This lncRNA is implicated in both cancer progression and age related coronary artery conditions, including atherosclerosis and myocardial infarction [226-227].

- **PINT**

PINT, a noncoding transcript triggered by p53, is in charge of p53 and modifies TGF-β, MAPK, and p53 signaling pathways via PRC2-mediated alterations in critical gene promoter regions. PINT exhibits an inverse relationship with aging and related conditions.

TUG1

It is commonly known that propagation regulator taurine upregulated genes 1 (TUG1) that is triggered by p53 in response to DNA damage. In addition to inhibiting p53-mediated growth and inducing programmed cell death, TUG1 affects the expression of HOX gene, including HOXB7, which accelerates aging. TUG1 regulates using the miR-455-3p/AMPKβ2 pathway, hexokinase 2 pathway, which in turn affects glycolysis in tumor cell proliferation and metastasis. TUG1, which is strongly exhibited in people human subependymal zone has been linked to age-related neurodegenerative diseases such as HD and ischemic stroke. Through the Wnt/β-catenin and caspase pathways, TUG1 affects tissue-specific aging processes such as intervertebral disc degeneration and age-related cataracts [228]. While TUG1 levels increase in the mouse retina, its exact role in retinal degenerative disorders remains unclear.

21.7.2 Telomere-associated lncRNAs

With every cell division, telomeres—the protective coverings of nucleoproteins located at the ends of chromosomes-shorten. The telomere RNA component (TERC) and telomerase reverse transcriptase are necessary for preserving telomere length. One important indicator of cellular decline and aging in living things is telomere attrition. The regulation of telomere dynamics is influenced by certain lncRNAs on chromosomes 1 and 22, hinting at a possible link with telomere-related disorders [229].

- **TERC**

  TERC acts as a blueprint for synthesizing telomeric DNA through telomerase. The gradual decline in telomerase activity is likely a contributing factor to its involvement in senescence and aging processes. Mice deficient in TERC exhibited manifestations of osteoporosis and accelerated aging in the lungs. The inflammatory state associated with pulmonary senescence might be partly attributed to telomerase-mediated NF-κB transcription [230]. It has been demonstrated that introducing TERC into telomerase-deficient animals can alleviate premature aging features by restoring telomerase function. Additionally, TERC may influence the expression of genes related to angiogenesis and metastasis, independent of its effects on telomere length [231].

TERRA

Ever since telomeric repeat-containing RNA (TERRA) was discovered in yeast, there has been a significant focus on its role as a lncRNA in telomere maintenance during senescence and aging. RNA polymerase II transcribes TERRA in a conserved way. Changes in TERRA expression influence the production of telomeric heterochromatin and the regulation of telomerase activity [232]. However, the correlation between TERRA expression and telomere length varies depending on the species or cell types studied, as well as the methods used [233]. Consequently, data on TERRA expression in cancer have produced conflicting results. While TERRA levels were found to be elevated in various malignancies, they decreased in more advanced stages. Some studies suggest that TERRA overexpression results in telomeric recombination defects and its accumulation, leading to premature senescence[234].

21.7.3 Chromatin-modulating lncRNAs

The aging process and cellular senescence involve alterations in chromatin structure. Changes in histone modifications, DNA methylation, heterochromatin formation, and epigenetic modifications are among the modifications observed in chromatin features. Through the recruitment of several histone and DNA methyltransferases that attach to sites of chromosome inactivity (such as Xist, HOTAIR, and lncRNA-p21), lncRNAs frequently function as regulators, decoys, or guides or by facilitating the interaction of transcription factors with regulatory DNA elements (such as AIR). While ANRIL, TERRA, and H19 are well-known lncRNAs discussed earlier [235].

- **Xist**

  Xist transcription governs X chromosome inactivation and gene imprinting in females by blocking RNA polymerase II access to the inactive X chromosome. While senescent cells exhibit reduced Xist levels, its precise function in senescence remains unclear [236].

- **Kcnq1ot1**

  The paternal allele of the Kcnq1ot1 gene produces the antisense lncRNA known as KCNQ1-overlapping transcript 1 (Kcnq1ot1). The paternal DMR-LIT1 locus attracts chromatin remodeling complexes, which impacts nearby genetically imprinted genes which includes CDKN1C and KCNQ1. Kcnq1ot1 plays big role in the aging process and cellular senescence through its influence on CDKN1C, which is involved in regulating cell cycle progression. Additionally, decreased expression of Kcnq1ot1 is associated with age-related conditions such as type 2 diabetes, heart attacks, atherosclerosis, and a number of cancers [237].

- **ANRASSF1**

  ANRASSF1, an unprocessed, nuclear-localized, intronic antisense lncRNA from an RNA class that is not well understood, targets Ras-associated domain-containing protein 1A (RASSF1A), a tumor suppressor gene that is implicated in DNA damage-induced G1/S cell cycle arrest and apoptosis. Increased DNA methylation of RASSF1A has been linked to age-related chronic gastritis, tumors, and noncancerous liver aging. ANRASSF1 may inhibit RASSF1A transcription via creating an RNA-DNA hybrid and recruiting PRC2 to the RASSF1A promoter region, suggesting its involvement in aging and senescence. While some other lncRNAs have identifiable target genes associated with senescence and age-related processes, it remains unclear how indirectly these lncRNAs contribute to the same area. Similar to Air, an antisense lncRNA produced from the paternal allele and imprinted on the maternally derived Igf2r promoter region is known as antisense Igf2 receptor (Igf2r) RNA [238]. Another illustration is the use of DNMT1 by CEBPA, or extra coding CEBPA, to silence the C/EBP gene. Growth arrest can be induced by encoded C/EBP family proteins through their inhibition of CDK2 and CDK4 [239]. Significantly lower C/EBP levels in aged tissues lead to altered adipose tissue function, decreased adipogenesis, and age-related liver damage. Promoting liver proliferation is achieved by restoring the aged-like isoform of C/EBPα. C/EBPβ and C/EBPγ heterodimerization promotes cell division and inhibits senescence [240]. Similar to this, pRNA forms DNA-RNA triplexes and then induces repression DNA methylation at the rRNA promoter to mute repetitive nucleolar ribosomal RNA (rRNA). The association between pRNA and neurological conditions associated with aging, including AD and Werner syndrome, as well as symptoms like depression, remains unclear, although it is strongly linked to senescence, the aging process, and levels of rRNA1. PTENpg1 is known to negatively regulate PTEN levels, resulting in decreased tumor growth, aging, and senescence [206].

21.8 Conclusion

Ferroptosis, which was recently identified as another kind of controlled cell death, has already been linked to a number of diseases, most notably cancer and aging. This chapter underscores the intricate interplay between phytochemicals, ncRNAs, ferroptosis, and their impact on cancer development and aging processes. This interplay represents a sophisticated network of molecular interactions that regulate cellular functions. The findings suggest that phytochemicals possess significant therapeutic potential in the modulation of ferroptosis. By activating specific ncRNAs, these compounds can regulate the cellular processes involved in cancer progression and aging, offering new avenues for therapeutic intervention. Phytochemicals have shown promise in targeting cancer cells by inducing ferroptosis, LPO that is dependent on iron and is a type of controlled cell death. By modulating ferroptosis-related pathways, phytochemicals can selectively induce cancer cell death while sparing normal cells, offering a potentially effective and less toxic approach to cancer treatment. The study also highlights the role of phytochemicals in modulating aging processes. By targeting ferroptosis and its associated pathways, these compounds may help mitigate age-related cellular damage and promote healthy aging. This suggests that phytochemical-based interventions could have broader implications for promoting longevity and delaying age-related diseases. While the findings are promising, further research is needed to fully elucidate the underlying mechanisms involved in phytochemical-mediated modulation of ferroptosis and ncRNA regulation. Additionally, clinical trials are necessary to evaluate the efficacy and safety of phytochemical-based interventions in human cancers and aging.

Author contribution

AM, MFN and MD wrote the preliminary draft. HF, FS drawn the figures, and revised the draft. MS and MFN revisions and finalize the draft. MFN, FS conceptualize the study.

Conflict of Interest

Authors declare no any conflict of the submission of this book chapter for publication.

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List of Abbreviations

OH	Hydroxyl radicals
ACSL4	Acyl-CoA synthetase long-chain family member 4
AD	Alzheimer's disease
AIFM2	Apoptosis inducing factor mitochondria associated 2
AKT-mTOR	protein kinase B (AKT)/ mammalian target of rapamycin
ALKBH5	AlkB homolog 5, RNA demethylase
ALOX	Arachidonate lypoxygenase
ALOX15	Arachidonate 15-lipoxygenase
AML	Acute myeloid leukemia
ASCL4	Achaete-Scute Family BHLH Transcription Factor 4
ATF4	Activating transcription factor 4
AURKA	Aurora kinase A
BACs	Bioactive compounds
Bcl-2	B-cell lymphoma 2
BECN1	Beclin-1
BMI	Body mass index
BRAF	V-Raf Murine Sarcoma Viral Oncogene Homolog B
CAPERα 1	Coactivator of activating protein-1 (AP-1) and estrogen receptors
CBS	Cystathionine β-synthase
CCL5	Chemokine (C-C motif) ligand 5
CCRC	Clear-cell carcinoma of renal cells
CD44	Cluster of differentiation 44
Cdk6	Cyclin dependent kinase 6
CDKN1C	Cyclin dependent kinase inhibitor 1C
CDKN2A	Cyclin-dependent kinase inhibitor 2A
CDKN2B	Cyclin Dependent Kinase Inhibitor 2B

CDS	Coding Sequence
circRNAs	circular RNAs
C-Myc	Cellular myelocytomatosis oncogene
CRC	Colorectal cancer
CYP450	Cytochrome P450
DHX9	DExH-Box Helicase 9
DKK1	Dickkopf-related protein 1
DMT1	Divalent metal transporter 1
DNMT1	DNA (cytosine-5)-methyltransferase 1
E2F7	E2F transcription factor 7
eIF4A	Eukaryotic initiation factor 4A
ELAVLI	ELAV like RNA binding protein 1
ESCC	Esophageal squamous cell carcinoma
FABP3	Fatty-acid-binding protein 3
FANCD2	Fanconi anemia group D2 protein
FBXW7	F-box and WD repeat domain containing 7
FPN1	Ferroportin 1
FSP1	Ferroptosis suppressor protein 1
FT	Ferritin protein
FTH1	Ferritin heavy chain 1
FTL	Ferritin light chain
FZD3	frizzled class receptor 3
G3BP1	Ras GTPase-activating protein-binding protein 1
GABPB1	GA-binding protein subunit β -1
GCH1	GTP cyclohydrolase 1
GDF15	Growth differentiation factor 15
GLS2	Glutaminase 2

GOT1	Glutamate oxaloacetate transaminase 1
GPX	Glutathione peroxidase
GPX4	Glutathione peroxidase 4
GSCs	Glioma stem cells
GSH	Glutathione
GSH,	Reduced glutathione
GSSG	Glutathione disulfide
GSTP1	Glutathione S-transferase Pi
H_2O_2	Hydrogen peroxide
HCC	Hepatocellular carcinoma
HD	Huntington's Disease
HO-1	Heme oxygenase 1
HOX gene	Homeobox gene
HSPB1	Heat shock protein family B (small) member 1
HuR protein	Human antigen R
IGF2 genes	Insulin-like growth factor 2
Igf2r	Insulin-like growth factor 2 receptor
IL1β	Interleukin-1β
IL6	Interleukin-6
iNOS	Inducible nitric oxide synthase
IREB2	Iron responsive element binding protein 2
KCNQ1	Potassium Voltage-Gated Channel Subfamily Q Member 1
Keap1	Kelch-like ECH-associated protein 1
lncRNA	Long non-coding RNAs
lncRNAPVT	Long non-coding RNA plasmacytoma variant translocation 1
LPCAT3	Lysophosphatidylcholine Acyltransferase 3
LPO	Lipid peroxidation

LSH	Lymphoid-specific the helicase
LUAD	Lung adenocarcinoma
MALAT1	Metastasis associated lung adenocarcinoma transcript 1
MAPK	Mitogen activated protein kinase
MDM2,	Mouse double minute 2 homolog
MECP2	Methyl CpG binding protein 2
MEG3	Maternally expressed gene 3
MIOX	Myo-inositol oxygenase
miRNAs	MicroRNAs
MT1DP	Metallothionein 1D pseudogene
mTORC1	Mammalian target of rapamycin complex 1
NADPH	Nicotinamide adenine dinucleotide phosphate hydrogen
ncRNA	Non-coding RNA
NEAT1	Nuclear Enriched Abundant Transcript 1
NF-κB	Nuclear factor kappa B
NOX	Nitrogen oxides
NQO1	NAD(P) quinone oxidoreductase 1
Nrf2	Nuclear factor erythroid 2–related factor 2
NSCLC	Non-small cell lung cancer
OIP5-AS1	Opa interacting protein 5-antisense RNA 1
p-38	mitogen-activated protein kinases
PANDA	P21-associated ncRNA DNA damage-activated
PCAT1	Prostate cancer associated transcript 1
PCAT29	Prostate cancer associated transcript 29
PCBP3	poly(rC) binding protein 3
piRNA	Piwi-interacting RNA
PIWIL4	Piwi like RNA-mediated gene silencing 4

PRDX5	Peroxiredoxin-5 peroxidase
PRNCR1	**Prostate cancer-associated non-coding RNA1**
PROM2	Prominin 2
PRRX2	Paired mesoderm homeobox protein 2
PTGS2	Prostaglandin-endoperoxide synthase 2
RASSF1A	RAS Association Domain Family Protein 1
RB1	Retinoblastoma protein
RBFOX2	RNA Binding Fox-1 Homolog 2
ROS	Reactive oxygen species
rRNA	Ribosomal ribonucleic acid
RSL3	Ras Selective Lethal 3
SASP	Senescence-associated secretory phenotype
SCD1	stearoyl-CoA desaturase 1
SEPW1	Selenoprotein W1
siRNAs	Short interfering RNAs
Sirt1	Sirtuin-1
SLC1A5	Solute carrier family 1 member 5
SLC3A2	Solute carrier family 3 member 2
SLC7A11	Solute carrier family 7 member 11
snRNA	Small nuclear RNA
SOD	Superoxide dismutase
STAT3	Signal transducer and activator of transcription 3
TBLR1	Transducin β-like 1X-linked receptor 1
TBX3	T-box transcription factor 3
TDP-43	TAR DNA-binding protein 43
TERRA	Telomeric repeat-containing RNA
TFAP2C	Transcription factor AP-2γ

TFE3	transcription factor E3
TFR1	Transferrin receptor 1
TFRC,	Transferrin receptor protein
TGF-β	Transforming growth factor-β
TNF-α	Tumor necrosis factor alpha
TNPO1	Transportin-1
tRNA	Transfer ribonucleic acid
TUG1	Taurine upregulated gene 1
TXNRD1	Thioredoxin reductase 1
UCA1	Urothelial cancer-associated
VDAC3	Voltage dependent anion channel 3
WDR5	WD repeat-containing protein 5
ZNRF3	Zinc and ring finger 3

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Chapter 22 : Pharmacokinetics And Pharmacodynamics In Nanoparticle Drug Delivery In Animals

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Abstract

Drug delivery systems based on nanoparticles (NPs) have drawn the interest of scientists because of nanotechnology, a prominent technology of the 21st century. NPs based drug delivery systems havesignificantly improve current disease therapies by overcoming biological barriers and delivering therapeutic drug at optimal dosage range. For designing these NPs-based formulations alongevaluating their therapeutic potential, understanding their pharmacokinetics (PK) and pharmacodynamics (PD) is crucial. Pharmacokinetics evaluation involves the absorption, distribution, metabolism, and excretion of these NPs-based drugs throughout the body, considering factors including NPs size, route of administration, and surface properties. Pharmacodynamics studies emphasis the interaction between biological system and NPs carriers, including target as well as therapeutic efficiency in animal models along drug release kinetics. Furthermore, integrating pharmacokinetics and pharmacodynamics investigationsfor designing NPs-based drug delivery led to suitabledrug design, significant

optimization, along implementation of these novel therapeutic strategies for better health management and disease control in animals.

Keywords: Nanoparticles, Pharmacokinetics, Pharmacodynamics, Absorption, Distribution, Drug Delivery.

22.1 Introduction

For the last several decades, nanotechnology has been one of the most remarkable fields of study in the development of drugs for disease control. Nano-systems are designed with enhanced functionalitymust having particulate matter that issystematically engineered and manipulated into a specific physical state (1 to 100 nm)[1, 2]. Nanoparticles (NPs) are three dimensional tiny entities, less than 100 nm in size (Figure 1). Nano-based materials offer several advantages due to their smaller size and high surface to volume ratio,which improvestheir reactivity,solubility, and diffusivity while reducing toxicity and enhancing thermal and electrical properties. Many practical domains like electronics industry, textiles, renewable energy, agriculture, synthetic food manufacturing, and medical era are benefited via nanotechnology because of its distinctive features [3, 4].

Nanomaterials advance the possibilities for disease diagnosis, treatment, and prevention to promote health. Low bioavailability and drug accumulation at dangerous level are most common problems in pharmacology.The primary use of nanotechnology is in designing drug delivery systems that specifically transport drugsat precise target to perform specific action. Considering the growing research on nano-drug delivery methods in human medicine, veterinary medicine has also significantly utilized nanomaterials to develop suitable drugs[5]. Micelles, dendrimers, carbon nanotubes, fullerenes, solid lipid NPs, polymeric NPs, liposomes, and metallic NPs are a few examples of typical NPs that frequently used in nano-drug delivery systems in veterinary medicine [6].

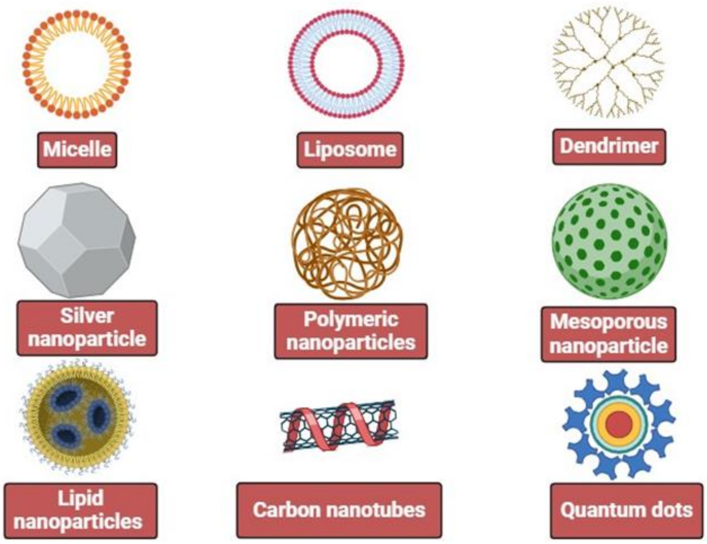

Figure 70: Diversity of NPs being used in drug delivery system.

Pharmacokinetics is a discipline that examines the interaction between a drug molecule and a living system, addressing the absorption, distribution, metabolism, and excretion of drugs in both humans and animal models [7]. It also involves precise dose estimation, bioavailability determination, bioequivalence, toxicity investigations, and drug interaction assessments, including in vitro/in vivo drug correlation.

Drugs alter physiological functions by interacting with their biological targets at the molecular level through receptor binding, chemical interactions, and post-receptor effects to induce therapeutic responses. For example, a medication may bind to an enzyme's active site, interact with cell membranes to affect downstream signaling, or bind to tumor necrosis factor (TNF) [8, 9]. Therefore, careful medication administration is essential to minimize adverse effects[10].

22.2 Pharmacokinetics of NPs Drug Delivery in Animals

Animal model research on NPs pharmacokinetics cover all the aspects of interaction between body system and NPs.

22.2.1 Absorption of NPs

Absorption, also known as integration, is the entry of drug molecule into the blood after administration. The following section will discuss the various delivery routes that have been utilized to investigate NP absorption [11-13]. The overall pharmacokinetics phenomenonof NPs drug delivery system in animal is illustrated in Figure 2.

- **Oral adsorption**

  Oral administration of NPs causes in their excretion through the feces or absorption into the bloodstream[14]. The mucus and epithelium of gastrointestinal tract are the two main barriers in absorption of NPs enclosed drugs. NPs absorption properties of size ranges asn50 nm-200 mm, have been extensively studied at small intestinal lymphoid nodules[15, 16]. In addition, research has also shown that enterocytes in the intestines can absorb NPs. It is more likely that NPs will be absorbed if they are attached to the gastrointestinal tract membrane[17-19].

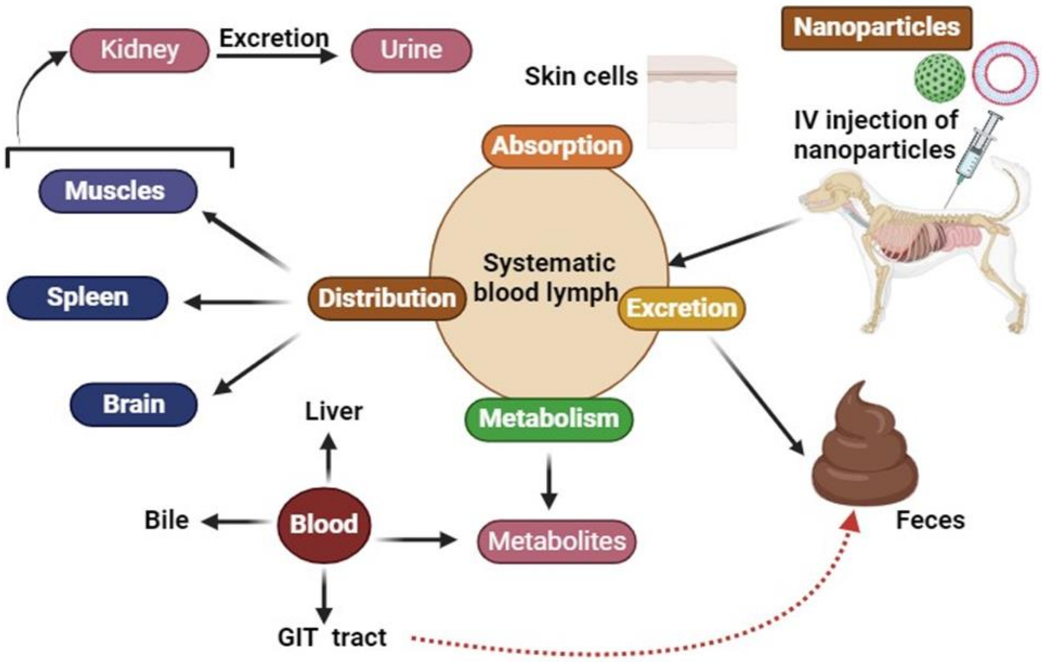

Figure 71: Mechanism of Absorption, Distribution, Metabolism, and Excretion of NPs drug delivery system

- **Nasal absorption**

  Multiple studies have shown that NPs are directly absorbed in the nasal olfactory area, providing easy access to the brain[20, 21]. The nasal route is a typical method for crossing the blood-brain barrier (BBB), as evidenced by data acquired from animal models.

- **Pulmonary absorption**

Both absorptive and non-absorptive excretion processes are observed to compete usually when NPs are absorbed via pulmonary route [22, 23]. Because of their immense surface area, alveoli facilitate the absorption of NPs and subsequent endocytosis. As a result, NPs diffuse smoothly to the lymphatic and circulatory systems after being absorbed via alveoli [24, 25].

- **Percutaneous absorption**

Percutaneous absorption is a widely recognized and extensively investigated method for understanding the interaction between NPs and the epidermis of the skin [26]. The lymphatic system and lymph nodes facilitate the absorption of NPs through the dermis. Adding biocompatible polymers, such as polylactic-co-glycolic acid (PLGA) and phospholipids, to NP compositions enhances their incorporation and interaction with the lipophilic skin segment.

Liposomes (<600 nm) are the only NPs that can penetrate the protective barrier skin [27]. Lipid-based NPs less than 200 nm in size improve skin moisture by loosening corneocyte packing and enhancing medication penetration [28]. Larger NPs (>200 nm) demonstrate time-dependent follicular penetration, while smaller (20 nm) polystyrene-based NPs accumulate in distant follicular segments [29]. Studies indicate that NPs based on polylactic acid (PLA) cannot pass through the stratum corneum, whereas NPs based on PLGA can rely on sebaceous glands and hair follicles for percutaneous absorption. Magnetic NPs smaller than 10 nm can penetrate the stratum granulosum through the skin layers [30, 31].

- **Parenteral absorption**

Parenteral NP absorption has been shown using subcutaneous, intramuscular, intradermal, and intraperitoneal injections[32]. A pharmaceutical molecule must be well-absorbed through many injectable routes to start a therapeutic response. NPs are absorbed intravenously by endocytosis, carrier-mediated transport, active diffusion, and passive diffusion. NPs are more absorbed by dendritic cells, macrophages, and territorial lymph nodes intravenously[33, 34].

22.2.2 Routes of administration

- **Transdermal drug delivery (TDD)**

The body's largest organ, the skin, is composed of three main layers: the epidermis, dermis, and hypodermis.The epidermis consists mainly of keratinocytes (95%), with the remaining 5% made up of Merkel, melanocytes, and Langerhans cells. The stratum corneum, the outermost layer of the epidermis (ranges from 10 to 20 μm), is composed of corneocytes, which are anucleated and physically dead keratinocytes[35].

NPs such as tiny emulsion vascular (liposomes that, ethosomes, noisome, etc.) are particularly formed in order to overcome this barrier [36]. NPs can enter the skin through three different channels: the transcellular, intercellular, and transappendageal pathways [37, 38]. While skin appendages were previously considered minor channel for drug penetration, recent research suggests that hair follicles may be a viable route for drug delivery [39].

Lademann et al. studied the absorption of a dye containing both NPs (320 nm) and non-NPs on human skin. They discovered that non-NPs could only be detected for 4 days, while NPs formulations could still be detected in pilo sebaceous units for up to 10 days [40]. CLSM (confocal laser scanning microscopy) images showed that, after 30 minutes, the accumulation of 20 nm fluorescent polystyrene NPs in skin appendages and hair follicles was similar. However, after 2 hours, hair follicles exhibited greater accumulation, indicating dynamic NP retention over time[41].

- **Oral-route of administration**

Oral drug administration consideredis most frequent method due to its highest patient acceptability rate as it is more convenient, less painful, more effective, has a high rate of patient adherence, and reduces the risks of infections and injuries [42, 43]. The goal of this strategy is to reduce injection-related adverse effects like pain, tissue damage, and noncompliance but solubility and permeability of a medication determine its oral availability[44].Furthermore, acidic environment of gastrointestinal tract (GIT) and enzymatic system commonly cause failed peptide or protein oral delivery via degrading the protein and lowering its therapeutic efficacy[45]. Various key strategies have been explored to enhance the stability and absorption of protein and peptide medications. These include chemical modifications of peptides (such as peptidomimetics or lipophilic derivatives), bioadhesive systems, and the combined use of protease inhibitors or penetration enhancers [46, 47].

- **Inhalation route**

Inhalation had numerous advantages over other delivery methods. It reduces dosage and side effects by eliminating first-pass hepatic metabolism and allows for the local distribution of drugs for respiratory disorders such as chronic obstructive pulmonary disease (COPD), cystic fibrosis, and asthma.Other benefits include its large surface area, which allows for quick absorption because of high vascularization, and its ability to avoid the first-pass effect [48]. The pulmonary route is mainly utilized for the local delivery of medications such as chemotherapeutics [49, 50]interferons[51], vaccines [50, 52].There are sixteen generations of airways that comprise the conducting zone. All the structures involved in gas exchange make up the respiratory zone, which begins at the respiratory bronchioles[53]. Particles smaller than 5–6 µm are absorbed in the trachea and bronchi but nanoscale particles (less than 1 µm) are not absorbed and store into the lower RS, while ultrafine particles (1-2 µm) settle in the bronchioles. Due to their quick bloodstream penetration, ultra-small NPs like dendrimers (<20 nm) frequently demonstrated poor lung retention despite their effective delivery to the alveoli [54-56]. Diffusion (Brownian motion), gravitational sedimentation, and inertial impaction are the three main processes of NPs deposition in the respiratory tract[57].

- **Intravenous administration**

Through IV administration, the rate of medication contribution into the body can be precisely controlled and have immediate response rate. It also solves the problem of medications that cannot be injected into muscles or other tissues or absorbed by the digestive system, while addressing the issue of first-pass metabolism [58]. Intravenous administration is primarily effective in delivery of costly medications like proteins and peptides. Proteolytic enzymes can also be neutralized by intravenous injection. The immediate response and significant bioavailability of medications, even at modest dosages, are the primary benefits of intravenous drug delivery[59]. However, a number of risks are associated to it as the medicine being exposed directly to the systemic circulation. Besides from being expensive and unpleasant for the patient, it also requires the assistance of skilled medical professionals. The FDA approved,a modified form of paclitaxel (Abraxane) was the first intravenously given NPs medication in 2006 [60, 61].

Table 27 NPs based drug delivery in animal model for treatment.

NPs	**Drug**	**Disease**	**Animal model**	**Administration Route**
Liposome	Doxorubicin	Soft tissue sarcoma	Cat	IV
Spherosome	Ribavirin	Feline infectious peritonitis	Cat	PO, IM, IV
Liposome	Clodronate	Malignant histiocytosis	Dogs	IV
Dendrimers	Foot and mouth disease vaccine	Foot and mouth disease	Pigs	IM
Polymeric	E. coli fimbriae vaccine	E. coli	Pigs	PO
Chromium nanocomposite	Chromium	Chromium supplementation	Pigs	PO
Liposome	Streptomycin	Brucellosis	Cattle	IM
Niosome	Flurbiprofen	Analgesic	Cattle	IV

Ring shaped	Human respiratory syncytial	Bovine respiratory	Cattle	Intranasal, IM
Liposome	Diclofenac	Anti-inflammatory and Analgesic	Cattle	Transdermal
Liposome	Diamidine	Treatment of babesiosis	Horse	IM
Polymer nanospheres	Streptococcus equi	Antigens Strangles vaccine	Horse	Intranasal
Liposome	Bovine leukaemia virus	Bovine leukemia virus vaccine	Sheep	IM
CS–PLGA and PLGA	Daunorubicin	Cancer	Wister rats	Oral
Micelle	Newcastle disease vaccine	Newcastle disease vaccine	Birds	PO
Chitosan NPs	Copper	Copper supplementation	Birds	PO
Liposome	Butorphanol	Arthritis in parrots and conures	Birds	SC
Thiolated chitosan	Docetaxel	cancer	Wister rats	Oral
PLA NPs	Aliskiren	Cardiovascular	Male rats	Oral
Chitosan NPs	HCT and HCT-β-CD	Cardiovascular	Rats	Oral

22.2.3 Factors involved in pharmacokinetics of NPs

A number of factors responsible for the variations in the drug molecules or in pharmacokinetics of NPs [62]. Different factors affecting drug delivery NPs have been well described (Figure 3).

- **Size**

Smaller NPs(5.6 nm in size) are quickly filtered via extravasations or renal filtration, whereas bigger NPs are more likely to be removed through cell from the mononuclear system of phagocytes (MPS)[63]. NPs with a diameter of around 100 nm show low MPS uptake rates and prolonged blood circulation residency. The biodistribution of radioisotope-tagged liposomes with sizes from 30 to 400 nm in mouse tumors, livers, spleens, and blood is being studied. The liposome composition was determined by examining tissues four hours after intravenous injection. Twenty percent of plasma NPs were smaller than 50 or 250 nm, while sixty percent of given liposomes were between 100 and 200 nm. In addition, these 100-nm particles made up 20% of the liver's liposome distribution. Reducing particle size below 50 nm increased hepatic NP accumulation. After 4 hours, NPs larger than 400 nm increased spleen absorption by 40-50%[64].

- **Shape**

Specific shape of NPs is another important aspect that might affect binding of NPs, intravascular travel, retention in the body, and disposition at the desired target [65]. The study examined the interactions between macrophages and polystyrene NPs of varying sizes and shapes. The researchers were able to further understand the variations by utilizing Ω, a shape-related element that is dimensionless and connected curvature. The authors found that Ω had a negative correlation with phagocytosis velocity (up to 45 degrees), and that NPs with Ω 45 degrees were efficiently distributed by actin-cup and ring formation. On the other hand, encapsulation did not occur when Ω was more than 45 degrees (ellipsoid) [66, 67].

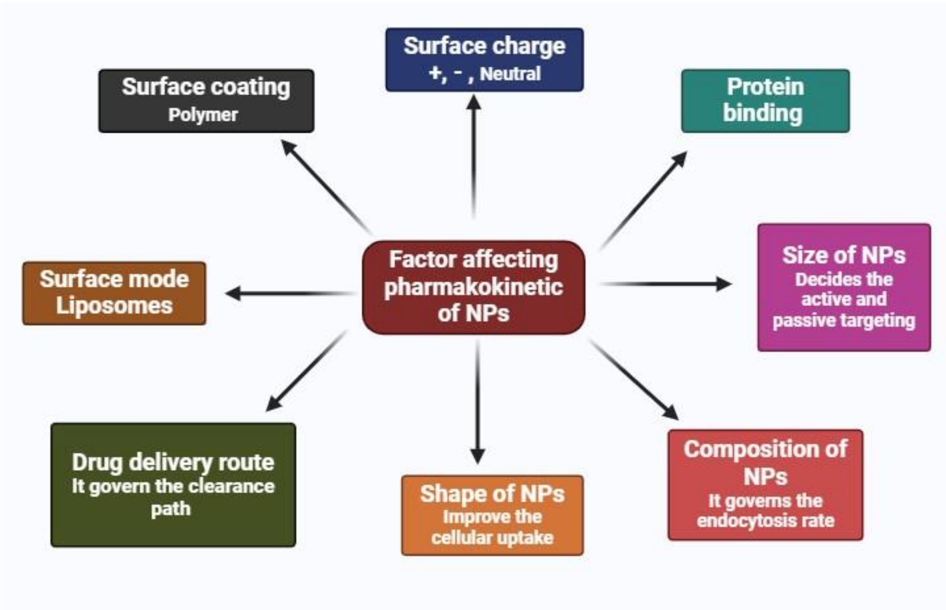

Figure 72: Various factors affecting drug delivery system based on NPs.

- **Surface charge**

Research conducted by Alexis and al. (2008) demonstrated that pharmacokinetics of NPs and MPS absorption are influenced by their surface charge, which is commonly represented by the zeta potential [68, 69]. NPs with +ve charge (> 10 mV) provoke a stronger immune response and do not enhance MPS uptake, in contrast to negatively charged NPs (<−10 mV). Neutrally charged NPs (within ± 10 mV) have the lowest MPS absorption and hence attain the longest circulation duration. Neutral NPs (10 mV) exhibited a clearance of less than 10% after 10 minutes, while 40 mV NPs had a clearance of more than 90% [70, 71].

- **Surface and coating engineering**

  The pharmacokinetics of NPs can potentially be affected by surface modifications and coatings. Therefore, research on hydrophilic coatings has been done to enhance the residence period and to reduce opsonization [68, 72]. For this purpose, polyethylene glycol (PEG) polymers are widely used since they are FDA-approved and have minimal toxicity [73]. Additionally, NPs have their surface modified with poloxamines 906, poloxamer, and a tetrafunctional ethylene dioxide block copolymer to increase their transit time and decrease MPS absorption[74]. Following this up, researchers found that coating NPs with poloxamine 908 and poloxamer 407 decreased the Kupffer [75].

22.2.4 Distribution of NPs in systemic circulation

Tissue distribution is the process by which injected NPs accumulate in different bodily tissues and organs. Comprehending the tissue distribution of NPs is vital for optimizing the delivery of medications, focusing on select tissues, and evaluating potential negative effects. The distribution of NPs throughout tissues is influenced by a multitude of parameters. These include the properties of the NPs, the method of administration, physiological barriers, and interactions with biological components [76]. The movement of drugs between blood and tissues once they reach the systemic circulation is crucial in determining its therapeutic efficacy and negative effects. Medication classes also exhibit significant variations in absorption, distribution kinetics, and distribution mechanism. Specifically, the distribution of small-molecule medications can vary from being restricted to the plasma space to being dispersed throughout the bodydepending upon molecular descriptors and interaction to plasma proteins [77, 78]. In certain tissues distribution of small-molecule medicines might affected by interactions with uptake and/or efflux transporters [79]. Therapeutic protein is largely influenced by their molecular weight to penetrate tissues effectively. Since DDS (Drug Delivery Systems) are usually much larger than the holes that separate endothelial cells, their distribution is usually limited to the arterial space in the absence of specific disease related receptor affinity. However, organs like the liver and spleen, which have larger endothelial pores, may benefit from tissue absorption by bulk fluid flow (convection). Similar to biologics, DDS that bind to transcytosis-prone receptors may enhance tissue absorption at sites of target expression [80, 81].

22.2.5 Factors affecting NPs distribution

The distribution of NPs in body is influenced by a number of variables that are essential for maximizing the therapeutic effectiveness of NP-based drug delivery systems. Their distribution in environmental matrices and biological systems like the human body depends on several factors. These components must be understood to maximize their applications and minimize risks. NPs dispersion is mainly affected by size, shape, surface charge, surface chemistry, and interactions with biological components like proteins.

- **Surface Charge**

  The surface charge of NPs determined by their functional groups or coatings, significantly affects their distribution. NPs with +ve charge tend to interact more with -ve charged cell membranes due to electrostatic attraction, potentially facilitating cellular uptake. But NPswith a negative charge may have the opposite effect and prevent cells from absorbing them. Through influencing cellular absorption rates and clearance mechanisms, these interactions can impact biodistribution [82].

- **Protein Binding**

  Proteins in body fluids, including blood or interstitial fluid, are an immediate target for NPs when they enter biological environments. As a result of these interactions, a protein corona, or layer of adsorbed proteins onto the surface of the NPs, can be formed. The size, shape, and surface chemistry of NPs are some of the variables that determine the composition of this corona. Stability, aggregation propensity of NPs and their interaction with cells or tissues primarily affected by protein binding along their physical characteristics. Protein corona also affects immune system detection of NPs and clearance routes [83, 84].

- **Size and Shape**

  The size and shape of NPs play essential roles in their distribution within biological systems. Smaller NPs can penetrate biological barriers more easily and exhibit longer retention in the bloodstream. Additionally, the shape of NPs influences their interaction with cells and tissues. For example, rod-shaped NPs may have different cellular uptake mechanisms compared to spherical ones. Furthermore, size and shape can affect the rate of clearance from the body via processes such as renal filtration or phagocytosis by immune cells [85, 86].

- **Surface Chemistry**

  The distribution and behavior of NPs in biological systems can be significantly altered by surface changes, such as polymer functionalization or targeting ligands. These modifications can enhance biocompatibility, reduce toxicity, and enable targeted delivery to specific tissues or cells. Functional groups on the NPs surface influence interactions with biomolecules and cellular uptake pathways, thereby affecting biodistribution patterns in cells [87, 88].

- **Biological Barriers**

  NPs encounter various biological barriers as they navigate through the body, such as the blood-brain barrier or the endothelial barrier in capillary walls. The capacity of NPs to pass through these barriers is determined via their physicochemical characteristics (size, charge, surface chemistry). NPs surface modification or receptor-mediated transport ligand usage are common approaches to improving NPs penetration across biological barriers [89].

22.2.6 Metabolic fate of NPs in animals

The immune system removes chitosan-based NPs from the body when they reach the bloodstream, identifying them as foreign bodies via following the events as: (1) Fast PC formation via NP association with plasmatic proteins; (2) protein rearrangement in "hard" and "soft" corona layers; (3) potential activation or inactivation on metabolic enzyme cascades; (4) immune response induction and induction of PC recognition process by immune cells; (5) NPs coated with PC are first taken up by macrophages and removed from the bloodstream; (6) NPs coated with PC are then accumulated in some specialized cells; (7) eventual initiation of particular routes for signaling and apoptotic processes [90, 91].

Some processes that facilitate the entry of NPs into cells include phagocytosis, caveolae/clathrin-independent endocytosis, and caveolae/clathrin-assisted endocytosis. NPs entry in cells is determined by several of their characteristics, primarily due to their size. According to contemporary considering, phagocytosis is the mechanism by which NPs bigger than 500 nm enter cells. The NPs can occasionally form complexes and get swallowed in cells. Protein adsorption also alters the "actual" size of the particle [92, 93]. Adsorptive endocytosis is primarily responsible for controlling the entry of foreign substances (like NPs) into cells, while energy-intensive, saturable endocytic pathways (like receptor-mediated endocytosis) typically control the entry of biological entities (such as pieces of bacteria, viruses, or proteins) into cells. NPs that contain chitosan or are coated with chitosan primarily interact with cell membranes via nonspecific attractive electrostatic forces. This is due to the fact that a specific receptor for chitosan polymer has not been discovered at this time. The general mechanism to attract cell-surface receptors is creating plasmalemma membrane invasions. But, in certain cases, for the internalization of specific NP types, clathrin served as an excellent model.Although it was also suggested that alternative clathrin-independent processes were can also be involved [94, 95].

Three factors impact the fate of NPs in the gut: first, diffusion via mucus, which effectively prevents ingested NPs from entering the body; second, interaction between the NPs and the epithelium[96]. These effects are caused by NPs diffusion over the agglomerated medium, which contains gel-forming mucin-like proteins that inhibit NP transport. Due to heavily glycosylated, negatively charged extracellular proteins in chitosan or chitin matrix, positively charged NPs may penetrate faster into the GI tract. Covering NP surfaces with functionalized groups that interact with mucosa proteins via hydrogen bonds, hydrophobic contacts, or electrostatic interactions can change their fate or duration in the mucosa [97].

Although PEG NPs may approach tumor cells more efficiently than the free drug without chitosan positive charges, their hydrophilic neutral nature limits their entry into the tumor cells. According to certain research, NPs can cause cell death by upsetting cellular, subcellular, and genetic activity via disrupting the integrity of the plasma membrane or damaging the nucleus and mitochondria[98]. To be more precise, NPs have the ability to create reactive oxygen species (ROS)stress as all of the components of a cell (proteins, lipids, and nucleic acids) are impacted by peroxides and free radicals due to imbalance in the normal redox state. Potentially the most dangerous are NPs smaller than 100 nm due to their small volume to surface ratio having ease of cellular absorption, and relatively their high number of chemicals capable of oxidation-reduction cycles causing a redox imbalance[99].

22.2.7 Excretion of NPs

NPs excretion processes and pathways must be understood for determining NP biocompatibility, accumulating potential, and overall safety profile as their removal from body is necessary after exposure. NPs are excreted through a variety of pathways and mechanisms. The kidneys are responsible for excreting the majority of extracellular substances. The intricate process of renal clearance, which includes tubular secretion and glomerular filtration, does not involve tubular resorption [100]. Renal clearance is primarily determined by three physiochemical properties: lipophilicity, pKa, and particle size as well as two types of tubular secretion as organic acid & organic base transfer. NPs (<5.5 nm) are readily eliminated due to their rapid glomerulus passage. The renal elimination of NPs or medications occurs rapidly when they are not bound to a plasma protein. The clearance rate of the drug or NPs is positively correlated with the plasma concentration of the medicine [68, 101].

22.2.8 Renal and hepatic clearance of NPs

Renal and Hepatic clearance of NPs is a crucial process essential for maintaining physiological balance in body. Renal excretion is the ideal way for NP excretion since it minimizes breakdown or catabolism and the kidneys can eliminate molecules from the vascular compartment fast, usually through injections. This helps to prevent any potential unwanted effects. The process of renal clearance of intravascular substances is complex, encompassing tubular secretion, glomerular filtration, and urine excretion as the ultimate step in the removal of the molecule [102, 103]. The filtrate from the glomerulus goes through the fenestrate, GBM, and filtering slits. One of the crucial nanostructural parameters is the slit diaphragm, which has a diameter of around 43 nm. The glomerular capillary wall layers have a cumulative impact that reduces the physiologic or functional pore size to just 4.5-5 nm in diameter [104]. Particles can flow through glomerulus capillaries at the "filtration-size threshold" during filtration. This process depends on molecular size. Filtering intermediate molecules relay on their size and charge as granular filtering is possible for molecules with upto6 nm but not over 8 nm. Significant research agrees that high-definition of filtration-size cutoff is less than 5 nm for globular proteins. Inulin (HD: 3 nm) has a 9-minute blood half-life and is entirely filtered by the kidneys. Furthermore, antibody fragments are cleared more quickly than the whole antibody [105].

There are several physical distinctions between NPs and proteins, while the role of kidneys in protein globule fate provides an important basis for understanding the fundamental features of renal clearance. NPs differ from proteins in their shape, surface chemistry, and internal charge as they made with same surface chemistry and a nearly spherical form but proteins are polydisperse and heterogeneous[106]. These variations might result in different renal responses to NPs than to protein molecules. Clearance experiments were carried out on PAMAM dendrimer-based NPs with near-spherical shape and uniform chemical properties to assess the impact of these features on filtration-size threshold. The findings indicated that PAMAM dendrimers with a diameter of less than six generations, or around 5.4 nm, exhibited efficient glomerular filtration [107, 108].Research assessing the impact of charge on the glomerular filtering of molecules of comparable sizes has demonstrated that ionic molecules are least easily filtered through the renal capillary wall, whereas cationic molecules exhibit the highest filtration efficiency. A charge-modified form of Fab (6 nm in diameter) synthesized by inhibiting branching amine residues using glycolate, has been employed to investigate the comparative glomerular filtration of molecules with varying charge results in significantly less filtering was noted when comparing weakly anionic Fab fragments with weakly cationic Fab. Permanent negative charges on the capillary wall may generate charge-selective filtering. Molecular charge determines whether 6-8 nm molecules can pass through charge-independent filtering since they are too tiny [109].

In the proximal tubule, the kidneys can reabsorb filtered molecules from the tubular fluid or actively discharge unfiltered molecules into the lumen. Renal function depends on this step, which determines compound fate. To demonstrate, glucose resorption can completely reverse glomerular filtration. Understanding particle behavior at the proximal tubule is essential for renal health assessment and exposure control. However, more research is needed to understand NPs renal toxicity.Evaluating how agents interact with the renal processing pathway can enhance their biocompatibility and clearance properties. The process ofrenal and hepatic clearance of NPs through filtration has been well understood (Figure 4).

Within hepatocytes, NPs may undergo metabolic transformations as series of enzymatic reactions for breaking down the NPs components into smaller and more easily excretable molecules. Changes to NPs characteristics brought about by metabolism can influence how these particles travel through the body. After being broken down, NPs are eventually eliminated from the body by the liver via bile excretion and blood clearance. NPs may interact with various liver cells beyond hepatocytes, including Kupffer cells, which are specialized macrophages located in the liver sinusoids. These cells can play a role in the clearance of NPs from the bloodstream through phagocytosis and subsequent processing.The hepatic clearance of NPs can also be influenced by the factors [110, 111].

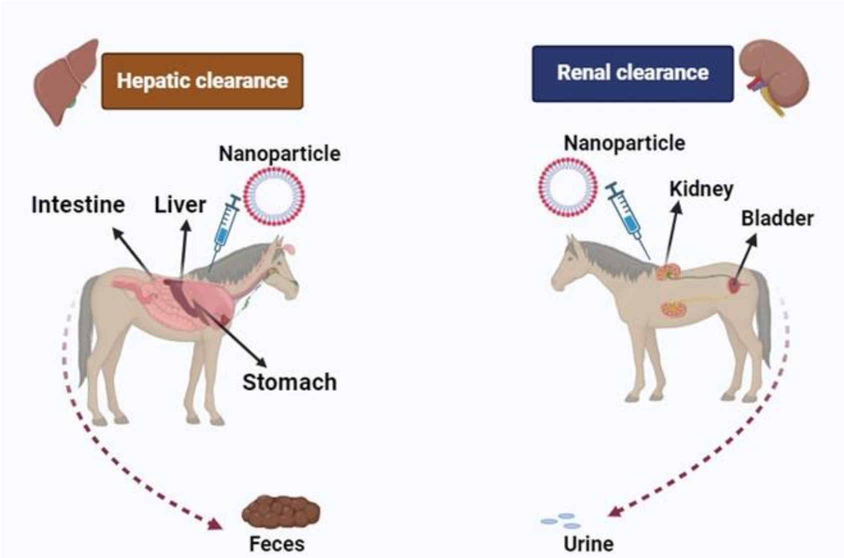

Figure 73: Renal and hepatic clearance of NPs in animals during metgabolism.

22.3 Pharmacodynamics of NPs Drug Delivery in Animals

22.3.1. Mechanisms of NPs-cell interaction

NPs with numerous biomedical applications, offer safer and more efficient solutions for various medical issues [112]. Many NPs must first achieve safe cell entrances before they may achieve high output in terms of therapeutic and prognostic efficacy. The effectiveness of NPs (NPs) depends on their intracellular destination.So, in order to develop safe and effective nanomedicines manipulating their physicochemical properties effectively and a better understanding of the cellular absorption and trafficking pathways of NPs as well as their interactions with CMs is crucial as the efficient and controlled entryof NPsalong transportation in cells still remains a major challenge [113].

22.3.2 Cellular uptake pathways

Most NPs enter cells through endocytosis after reaching their outer membrane and connecting with the extracellular matrix or plasma membrane. After being picked up by endocytosis during membrane invasions, the NPs are transported to various intracellular compartments for sorting and trafficking after they bud and pinch off to form endocytic vesicles [114, 115]. Phagocytosis, clathrin-mediated endocytosis, caveolin-mediated endocytosis, clathrin/caveolae-independent endocytosis, and macropinocytosis are the five primary processes of endocytosis in NPs transportation [116, 117].

- **Phagocytosis**

  Macrophages, dendritic cells, monocytes, and neutrophils are the main players in phagocytosis. In addition to absorbing dead cells and cell debris, these cells are responsible for host defense. While some cell types (such as fibroblasts, epithelial, and endothelial cells) called para-phagocytes as they show some phagocytic activity but not like other defense cell types [118, 119].The physiochemical features of NPs (size, shape, and surface characteristics) control its uptake through the phagocytic pathway. Firstly, Opsonins (complement proteins, immunoglobulins, blood proteins) adsorbed onto the surface of the NPs through particular ligand-receptor interactions via identifying and bindingto specific NP. This initiates a series of processes depending specific receptors that can cause actin to be assembled, cell surface extensions to be created, and, ultimately, the formation of a "phagosome" (structure that can internalize and engulf particles) [120, 121]. As a result, linked receptors influence both the toxicity (inflammatory response) and delivery of NPs. Phagocytes often take up bigger particles more efficiently as it was demonstrated that mouse peritoneal macrophages could absorb particles in the 1000-2000 nm range [122]. Further, the shape of the NPs had a substantial impact on their absorption by cells [123]. Arnida et al. examined PEGylated gold nanorod and nanosphere cell-to-cell absorption in murine macrophages after 6 h incubation. Gold content was measured after washing and lysing the cells. Gold nanorods accumulated less than nanospheres. This clarifies the in vivo investigation on the circulation of injected mice with ovarian tumors using gold nanorods instead of nanospheres [123-125].

- **Clathrin-mediated endocytosis (CME)**

Cells primarily use clathrin-mediated endocytosis to absorb nutrients and plasma membrane components, such as iron from transferrin carriers and cholesterol from low-density lipoproteins (LDLs).Instead of direct contact with membrane components, generic hydrophobic or electrostatic bonds finally initiate the absorption in receptor-independent CME [126].CME occurs at a clathrin-rich region of the plasma membrane, which makes about 0.5–2% of the cell surface[127]. The triskelion also, the clathrin assembly unit, is a three-legged structure made up of three heavy and three light chains [128] is in charge of the complex architecture that spontaneously co-assembles to form membrane curvature and stabilizes it mainly leading to the budding vesicle. Examples of auxiliary proteins that help in the creation and maintenance of membrane curvature include SNX9, amphiphysin, and epsin. The membrane scission protein dynamin can be bound by bin-amphiphysin-rvs BAR proteins and recruited to the budding vesicle to release it into the cytoplasm [129]. Vesicles ranging in diameter from 100 to 150 nm are generated during CME, and they absorb extracellular fluid to commensurate their inner volume. Degradative processes are a common destination for particles that enter cells through this route.

- **Caveolae-dependent endocytosis**

Many biological activities rely on caveolae-dependent endocytosis, including as transcytosis, cell signaling, lipid and fatty acid regulation, membrane protein regulation, and membrane tension control. Furthermore, caveolae-mediated endocytosis influences a wide range of disorders, including diabetes, cancer, and viral infections [130]. Caveolae, membrane incursions shaped like flasks, can be seen in both epithelial and non-epithelial cells, distributed as cytoskeleton anchors across densely populated body areas. Adipocytes of smooth muscle cells are examples of non-epithelial cells whose cell membranes include caveolae increasing surface area by up to three quarters [131, 132]. Albumin is able to target cancer cells in tumor interstitial spaces via its binding to the caveolae-resident albumin receptor, gp60 [133].

- **Clathrin/caveolae independent endocytosis**

  Endocytosis can still occur in cells deficient in either clathrin or caveolae. So, as example primarily the absorption of growth hormones, folic acid, interleukin(IL)-2, and cellular fluids is dependent on a specific lipid composition, particularly high in cholesterol.The growing requirement for folic acid during cancer cell proliferation has prompted extensive research into foliate bio-functionalization for targeted delivery. One chemical that can be ingested via this approach is foliate-coated NPs. The cytoplasm is accessible once foliate-functionalized NPs bind to their receptor in a non-destructive manner. These particles are often able to evade lysosome by residing in endocytic compartments or by being released directly into the cytoplasm after internalization [134, 135].

- **Macropinocytosis**

  Macropinocytosis is distinct from other pinocytosis processes asthis referred as the cytoskeleton rearranges to generate wide membrane outgrowths or frills, which ultimately connect with the plasma membrane to generate a sizable vesicle (0.2–5 μm) that has the capacity to absorb a substantial volume of extracellular fluid. This mechanism represents a non-specific bulk fluid absorption because it absorbs all dissolved elements and particles present in the extracellular fluid without regard to specific receptor presence [136]. Large vesicle formation during macropinocytosis is particularly vital for the absorption of larger NPs that lacks compatibility with clathrin- or caveolae-mediated processes [137].

22.4 Techniques for Studying NPs Pharmacokinetics and Pharmacodynamics in Animals

NPs have emerged as versatile tools in various fields, particularly in medicine, where they hold significant promise for revolutionizing drug delivery, diagnosticapproaches, and therapeutic applications. Their unique properties, including small size, large surface area-to-volume ratio, and flexible surface chemistry, make them highly attractive for biomedical applications. In the context of pharmacokinetics (PK) and pharmacodynamics (PD) studies in animals, NPs offer distinct advantages over conventional drug formulations, presenting researchers the new opportunities to understand and optimize therapeutic outcomes [138, 139]. Unlike traditional small-molecule drugs, NPs exhibit complex behavior in vivo due to their size-dependent interactions with biological barriers and components. Biodistribution, tissue residence time, and total systemic exposure all impact therapeutic efficacy and safety of NPs so, it is essential to understand the PK profile. NPs have pharmacological effects which include altered cellular signaling pathways, regulated release kinetics, and targeted medication administration. These pharmacodynamic features are crucial in deciding the therapeutic efficacy and safety profiles of NPs, as they determine the induction of biological reactions [140].

Therefore, investigating the pharmacodynamics of NPs in animal models provides valuable insights into their mode of action, efficacy, and adverse effects, thereby guiding the rational design and optimization of NPs-based therapies. In recent years, a diverse array of techniques has been developed and employed for studying NPs PK and PD in animal modelslike imaging or analytical chemistry to molecular biology and pharmacology. Each technique offers unique advantages and capabilities, allowing researchers to interrogate different aspects of NPs behavior with high precision and sensitivity [141]. One of the most widely utilized techniques for studying NPs PK in animals is non-invasive imaging, such as positron emission tomography (PET), single-photon emission computed tomography (SPECT), magnetic resonance imaging (MRI), and fluorescence imaging [142, 143]. These imaging modalities enable real-time visualization and quantification of NPs biodistribution and pharmacokinetics in living subjects, offering invaluable information on tissue-specific accumulation, clearance kinetics, and target site localization. Additionally, analytical techniques such as mass spectrometry (MS), chromatography, and spectroscopy play a crucial role in quantifying NPs concentrations in biological samples and elucidating their metabolic fate [144]. Researchers can learn about the ADME characteristics of NPs by tracking their levels in various biological fluids, such as tissues, blood, and urine, and drawing conclusions about the paths of absorption, metabolism, and elimination. It is also expected that these methods will produce significant new discoveries. Despite a shortage of published research on the latest methods, the next section gives the overview of the methodologies used to study NP cellular trafficking [145].

22.4.1 Confocal laser scanning microscopy

CLSM can remove out-of-focus items by using point scanning, unlike the more common epifluorescence methods [146, 147]. Because CLSM has a narrow depth of focus, it may produce successive optical sections that allow for high axial (z) resolution imaging of cell trafficking and interactions as well as accurate subcellular localization. Particle-protein interactions can easily be visualized via CLSM because co-localization reveals information about binding partners and intracellular trafficking routes [148].

22.4.2 Transmission electron microscopy

TEM can differentiate objects by examining how they interact with a focused electron beam [149]. TEM offers resolutions that are better than those of photon-based techniques, frequently on the scale of a single atom, because of the tiny de Broglie wavelength of electrons. Some of its drawbacks include long times for preparing samples, limited multiplexing capabilities, and a lack of variation in biological structures, despite its strength. Because the transmission of TEM is essential, biological samples must also be sectioned using an ultramicrotome (50–100 nm thick). Despite these challenges, the method has the potential to produce a great deal regarding atomic periodicity and other factors including size, electron density, and elemental composition [150, 151].

22.4.3 Atomic force microscopy

Using a piezoelectric-driven scanning cantilever and a scanning probe tip with a radius of around 10 nanometers, atomic force microscopy (AFM) measures interactions with a material to produce high-resolution topographical and mechanical data as its resolution is similar to methods based on electrons [152]. AFM may function in an aqueous environment, maintaining biological structures and natural hydration, in contrast to electron microscopy (EM), based upon vacuum settings. Despite its adaptability, AFM can exhibit inadequate discriminating at times, and a shortage of widely-used labeling. Cryosection skin samples allowed researchers to observe, for instance, the distribution and biomechanical properties of tattoo ink NPs in the dermis and the collagen matrix immediately around the samples [44, 153].

22.4.4 Scanning electron microscopy

SEM has a lateral resolution of around 1.0 nm and offers fundamental data that is helpful in NPs related research [154]. SEM can examine tissues thicknesses in millimeters, unlike transmission electron microscopy (TEM), which necessitates transmission. Additionally, high-quality tomographic data can be obtained by serially slicing and scanning living tissues using focused ion beam (FIB) surface milling. According to FIB-SEM imaging, NPs on the inside seemed to be housed in vesicles, whereas those on the surface of the cells were found to be dispersed. Therefore, it appears that there is a co-factor associated with the membrane that facilitates trafficking [155, 156].

22.4.5 Light-scattering microscopy

Chemical environment of a sample may be extensively studied using elastic and inelastic light scattering microscopy [157]. The former and later are particularly unique since they differentiate between spectrum absorption/scattering and give information on molecule and solid-state vibrations. Palonponet al. demonstrated the real-time mapping of molecular vibrations by the use of live-cell Raman imaging [158]. Researchers utilized 50-nanometer gold (Au) NPs as surface-enhanced Raman scattering (SERS) probes to track individual NPs within murine macrophages. By employing confocal microscopy with high lateral and axial resolution, they successfully monitored the trajectory of the NPs and the chemical milieu surrounding them. This approach enabled the detection of exogenously introduced DNA markers labeled with alkynes [159].

22.4.6 Flow cytometry

The technique of flow cytometry also known as flow-automated cell sorting (FACS), uses a single file suspension cell to pass them via a fluidics channel in order to measure the impedance and multicolor optical scattering. This method allowing for the real-time sorting of cells according to their optical or electrical propertieshandling both fixed and living cells. It can measure hundreds of cells per second and typically offers 10-color multiplexing with spectral adjustment.

From diverse cell populations, statistically stable staining intensity values and distributions may be obtained in high throughput [160]. Forster resonance energy transfer (FRET) methods can also be used to evaluate distribution patterns and interaction partners. Researchers utilized flow cytometry to differentiate between ligand-targeted silver NPs located on the cell surface and those that were internalized. They achieved this by employing a chemical etchant that is impermeable to cell membranes to de-stain surface-bound particles [161, 162].

22.4.7 Dark-field microscopy

Instead of using transmitted light for imaging, dispersed light can be used with dark-field illumination [163]. This technique employs a beam stop, sometimes known as a "spider stop," to obstruct the center of a low-angle conical light beam that is impinging on the sample. Because cells and other transparent objects have refractive indices that are very close to those of their natural surroundings, this imaging technique works very well for these types of things. This means that, on a black background, only items with a high dispersion may be seen. The monitoring of plasmin NPs and their probes in live cells is a common use of this technique and has the potential to be more accurate than fluorescent microscopy in many situations[164]. Researchers looked at the velocity of RSV cell ingress to indicate that 13-nanometer gold NPs probes could be useful dark-field imaging agents without interfering or photobleaching [165].

22.4.8 Correlative microscopy

Combining two or more microscopy methods to examine the same material is known as correlational microscopy [166, 167]. Correlative microscopy aims to use the strengths of distinct methods, such as light, electron, atomic force, and super-resolution microscopy, to gain more detailed information about the material. For instance, the traditional shortcoming of light microscopy poor resolution below 300 nm can be addressed by super-resolution microscopy in a correlative microscopy. As a result, correlative microscopy may open up fascinating new directions in the realm of nanomedicine, particularly in NP intracellular transport [166, 168].

22.5 Future directions and opportunities for optimizing NPs drug delivery in animals

Optimizing **NPs** drug delivery in animals holds significant promise for advancing therapeutic interventions across various fields, including medicine, veterinary science, and biotechnology. Several novel possibilities for improving the safety, efficacy, and efficiency of medication delivery in animal models are opening up as scientists explore further into innovative **NPs** formulations and delivery methods. Future efforts may focus on refining targeted delivery systems that enable precise localization and controlled release of therapeutic payloads within specific tissues or cell types. This primarily includes the development of ligand-targeted NPs capable of selectively binding to specific cell surface receptors that are overexpressed in diseased tissues, thereby enhancing therapeutic efficacy while minimizing off-target effects [169, 170]. It is possible to incorporate stimulus-responsive drug release mechanisms into **NPs** formulations, which can be activated by certain external stimuli or biological signals. Alterations to pH, temperature, enzyme activity, or the existence of disease-related biomarkers are all examples of potential triggers. Responsive drug release strategies enable precise coordinate system control over drug delivery, enhancing therapeutic outcomes while minimizing systemic side effects. Advances in **NPs**-based imaging modalities enable real-time visualization and monitoring of drug delivery processes in animal models. Future developments may focus on the integration of imaging agents, quantum dots, or magnetic NPs, into drug-loaded NPs to facilitate non-invasive tracking of biodistribution, pharmacokinetics, and therapeutic response in vivo. NPs-based drug delivery platforms offer unique opportunities for synergistic combination therapies, wherein multiple therapeutic agents with complementary mechanisms of action are co-delivered within a single NPs formulation. By overcoming drug resistance mechanisms and enhancing therapeutic synergy, combination NPs therapies hold promise for treating complex diseases in animals more effectively [171]. A significant obstacle in the field of NPs drug delivery is the lack of a direct correlation between laboratory studies and actual patient care. Facilitating the translation of promising NPs-based therapies from animal models to veterinary and human patients may require future efforts to streamline the preclinical evaluation process, optimize the flexibility and reproducibility of NPs formulations, and address regulatory considerations [172].

22.6 Conclusion

In conclusion, the exploration of pharmacokinetics and pharmacodynamics in NPs drug delivery systems for animals represents a crucial step toward advancing therapeutic interventions in veterinary medicine and translational research. Through a comprehensive understanding of NPs interaction with biological systems, including absorption, distribution, metabolism, and excretion, researchers can design NPs formulations to optimize drug delivery efficiency, efficacy, and safety profiles in animal models. Moreover, the integration of pharmacodynamic principles, such as drug-receptor interactions, cellular signaling pathways, and therapeutic responses, enables the development of targeted NPs-based therapies customized to the unique physiological and pathological characteristics of animal patients. By investment of emerging technologies, including targeted delivery systems, multifunctional NPs, and responsive drug release mechanisms, and personalized medicine approaches, researchers can enhance the precision, effectiveness, and translational potential of NPs drug delivery in animals. Ultimately, bridging the gap between preclinical investigations and clinical applications holds the promise of transforming veterinary medicine and improving the health and well-being of animals worldwide.

Authors Contribution

AJ, SS, MFN, MR, and AIA wrote various selective parts of the primary draft according to their specialization. MA, HF, MA, and MYW designed and draw the figures. SHM, ZM, MFN revised the draft. MR, MA and MFN added critical discussions. MA, AIA and MFN conceptualization of the study.

Conflict of Interest

No conflict of interest is declared by any of the authors.

Abbreviations

ADME	Absorption, Distribution, Metabolism, and Excretion
AFM	Atomic force microscopy
BBB	Blood-brain barrier
CLSM	Confocal laser scanning microscopy
COPD	Chronic obstructive pulmonary disease
FACS	Flow-Automated Cell Sorting
FRET	Förster resonance energy transfer
GIT	Gastrointestinal tract
MPS	Mononuclear system of phagocytes
MRI	Magnetic resonance imaging
MS	Mass spectrometry
NPs	Nanoparticles
PD	Pharmacodynamics
PET	Positron emission tomography
PK	Pharmacokinetics
PLGA	Polylactic-co-glycolic acid),
ROS	Reactive oxygen species
SEM	Scanning electron microscopy
SPECT	Single-photon emission computed tomography
SWNTs	Single-walled nanotubes of carbon
TEM	Transmission electron microscopy
TNF	Tumor necrosis factor

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Chapter 23 : Safety And Toxicological Considerations Of Nanoparticles In Veterinary Practice

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Abstract

Nanoparticles (NPs) are extensively employed in various industrial and medicinal applications, including veterinary medicine. However, their different physicochemical properties have some medical and environmental risks associated with them. These substances are relatively toxic to normal cells and organs thus their clinical applicability is rather restricted. The huge variety of NPs as well as their properties such as size, shape, and surface area under consideration govern their safety and toxicity. Understanding the mechanism of nanotoxicity and developing techniques to mitigate, it is crucial for ensuring the safe use of nanotechnology in therapeutics. This chapter examines various mechanisms of nanotoxicity and their effects on biological systems and organs, such as the liver, brain, kidneys, and lungs. Therefore, it is important to have accurate data on their impact on different organs to estimate the associated risks. This chapter aims to enhance the comprehension of nanotoxicity by clarifying the patterns of NP transport, accumulation, degradation, and removal. Animals are currently exposed to naturally

occurring and human-made NPs through various means, which highlights the significance of evaluating their impacts and potential hazards. By conducting a systematic review of safety assessment and toxicological considerations, this chapter seeks to enhance the understanding of veterinarians and researchers. It aims to facilitate the safe and efficient utilization of NPs in veterinary medicine by providing information on existing knowledge, limitations, and prospects.

Keywords: Nanotechnology, nanoparticles, nanotoxicity, Drug delivery, biological systems, organ-specific effect, veterinary medicine.

23.1 Introduction

Since its inception around 1974, nanotechnology has made significant progress, primarily focusing on developing novel materials within the size range of 1 to 100 nanometers[1-2]. The term "nano" is derived from the Latin word "nanus," meaning extremely small, and in scientific terms, 1nm is equivalent to 10^{-9} meters[3-4]. Nanomaterials have great potential for biomedical applications and research both *in vivo* and *in vitro*[5-6]. NPs possess unique physicochemical characteristics that exceed those of bulk materials due to their high surface-to-volume ratio, enhanced reactivity, stability, bioactivity, and bioavailability. They also offer controlled particle size, regulated drug release, site-specific targeting, and precise drug delivery [7-8]. Besides size, NPs can be classified based on their physical attributes, such as electrical charge, and chemical properties like core or shell composition. They also vary in shape (tubes, films, rods, etc.), and origin, which encompasses both naturally occurring NPs found in volcanic dust, viral particles, as well as man-made NPs [9].

The potential toxicity of NPs to living organisms is a major concern, which limits their use in disease treatment and diagnosis. In diagnostics, fluorescent labels can be utilized to identify biomolecules and infections, while contrast chemicals improve the effectiveness of magnetic resonance imaging and other research methods. Additionally, NPs are useful for targeted drug delivery, including the transport of proteins and polynucleotides, performing photodynamic therapy, applying heat to destroy tumors, and repairing prosthetics[10].

Challenges and possible detrimental impacts also arise for the researchers when handling NPs because of their toxicity. Hence, it cannot be overemphasized that proper experimental models have to be chosen for determining toxicity, whether cellular (in vitro) or animal (in vivo) models. Modes of NP entry into the body include inhalation, dermal contact, and ingestion depending on the NP's properties and the manufacturing processes[11]. Respiratory, digestive, cutaneous and circulatory contact are some of the biochemical interaction routes between NPs and the body, depending on the substances present in the NPs[12]. Some specific NPs, such as ZnO and TiO2, can block UV rays and are commonly found in different health products. However, there is some controversy regarding its health, safety, and environmental impacts once these NPs are released into the environment. Previous studies show that NPs can find their way into the body through various pathways and reach the crucial organs via the circulatory system thus damaging tissues and cells[12-13]. It is still uncertain how NPs issues arise in toxicity, but research carried out has made a correlation with causes such as morphology, dimensions, distribution surface charge, and the protein corona effect. Numerous investigations on the effects of NPs have revealed that Weighted Genetically they can induce Oxytosis and modulate the genes involved in inflammation[14-15]. Once NPs have been inhaled, ingested, or injected into the body, they circulate through the bloodstream and can deposit within various tissues and organs[16-17]. They can cross BBB, break apart the connections neurons make gain access to cells that have the CXCR6 chemokine receptor, and pass through the tight junctions of the BBB to infiltrate the brain [18].

Moreover, there is a tremendous opportunity for nanotechnology in medicine delivery as it can penetrate cells, tissues, and organs far better than the conventional larger particles. This depicts the poor therapeutic ratio and the high mortality related to present-day drugs[7, 19-20]. Nanomedicines employ different techniques of nanotechnology to deliver solutions to different medical issues and the management of diseases at a faster pace. Besides overcoming the drawbacks of the usual treatment approaches, these therapies help understand different physiological and pathological systems. A thorough understanding of these mechanisms reveals novel opportunities and treatments for current challenges [21]. Nano-theranostics is a field that integrates therapeutics and diagnostics, seeks to evaluate treatment responses and improve the efficacy and safety of medications. They provide a significant opportunity to develop combination therapies that provide treatments while employing detection methods before and during treatment regimens [22-23]. Due to the importance of animals in the economies of many countries, there is a continuous requirement for innovative diagnostic and therapeutic methods to detect and treat animal diseases. The goal is to improve the protein production for human nutrition [24].Nanotechnology has generated significant advancements in veterinary medicine across several areas, such as treatments, diagnostics, tissue engineering, vaccine manufacture, and disinfection[25]. Current uses of nanotechnology are already being utilized in various fields, including animal health, breeding, reproduction, and nutrition. The administration of medication directly into targeted cells, nanotechnology allows for small quantities, thus reducing drug residues and withdrawal periods in animals [26]. Nanotechnology in animal farming offers potential applications such as the delivery of pharmaceuticals, vitamins, probiotics, and nutritional supplements. NPs can be used to detect and eliminate pathogens without requiring surgical intervention. However, excessive use of antibiotics in animal farming could result in residues that affect the end consumer. Applying nanotechnology can reduce the dependence on antibiotics due to their nano-scale characteristics [27]. Nanotechnology can enhance animal production by utilizing nano-fertilizers to deliver nutrients to specific areas in forages in precise amounts, potentially with the assistance of magnets. The integration of NPs with hydrogels or zeolites can significantly improves water quality by effectively absorbing harmful chemicals [28].

Nano minerals provide cost-effective solutions due to their need for smaller quantities while simultaneously promoting growth and enhancing the immune system, providing numerous advantages in animal feed production. They help manage harmful microorganisms in animal feed and enhance rumen fermentation processes. Nano zinc oxide, in particular, is a highly promising nano mineral that improves growth rates, stimulates immunological responses, and alleviates reproductive illnesses in cattle. In addition, studies have demonstrated that nano zinc can effectively decrease diarrhea in young piglets and reduce somatic cell counts in dairy cows with subclinical mastitis [29-30]. Nano silica and nano magnesium oxide are potent nanoscale antifungal agents that efficiently attach to and deactivate aflatoxins, thereby combating mycotoxicosis, a condition that impacts around 25% of animal feed [31]. Nanotechnology is used in several applications to diagnose and treat reproductive disorders, detect estrus, freeze sperm, and manage calving[32]. It efficiently addresses reproductive issues like retained placenta and influences hormone release and protection in reproduction, including steroid and gonadotropic hormones [33]. Despite being expensive in nature and laborious breeding and reproduction procedures, nanotubes are valuable instruments for accurately measuring changes in blood estradiol levels over time. They help identify estrus by attaching to estradiol antibodies through infrared fluorescence. Additionally, microfluidics technology enhances in vitro fertilization procedures [34]. Nanoscopic sensors, specifically cell probes, are employed the diagnose infections in the genital tract, as well as metabolic and hormonal abnormalities, and to detect estrus[35].

Animals can also use different types of NPs in veterinary practices (Figure 1), however, this depends on the low-dose toxicity of metallic NPs such as cadmium. Applying metallic NPs to the reproductive tract improves their effectiveness in that area [26]. Veterinary vaccine manufacture is increasingly incorporating NPs to enhance immunological responses and act as adjuvants for slow antigen release, hence enhancing vaccine efficacy [36]. In pet care, nanotechnology has been used to develop innovative products for surface refreshing and disinfection. For example, shampoos formulated with silver NPs are designed for direct application to skin [37].

Although most NPs are generally considered to be benign, certain types may provide significant hazards. Prolonged inhalation of carbon nanotubes may result in fertility issues in pharmaceutical employees [38]. Furthermore, the formation of magnetic NPs consisting of iron oxide (FeO) within the body, or because of an unstable connection between the drug and particles, might lead to the inadvertent release of the medication in solid tissues instead of the intended target areas [39]. This incomplete release can cause toxicity in healthy tissues and sub-therapeutic dosage at the intended site. Their capacity to navigate different biological barriers within the body, like as the blood-brain barrier, can have substantial internal and ecological implications. As an illustration, there is an increasing need for radionuclides, and carbon nanofibers have been associated with the degradation of the ozone layer in the biosphere [40-41]. Different types of NPs used in veterinary practices have been depicted in detail (Figure 1).

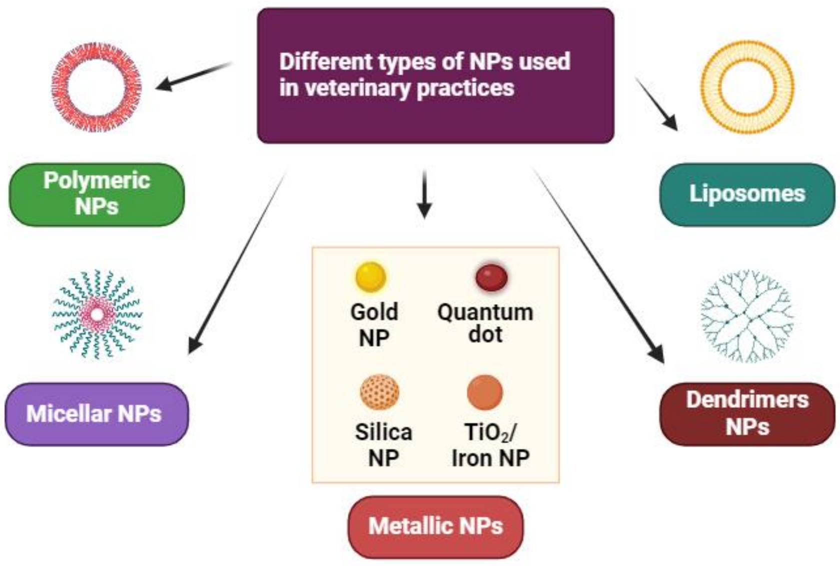

Figure 74: Different types of NPs used in veterinary practices to cure multiple types of diseases.

Aim of this chapter are centered on the movement of NPs across membranes, their behavior, and their effects on cell metabolism[42], examine a specific aspect of NPs performance to determine whether they have harmful and poisonous impacts on organs or they are adequately safe [9]. To develop NPs that are both safe and compatible with animal bodiesto diagnose and treat diseases, it is crucial to have a thorough understanding of the interaction between different elements and the mechanisms that contribute to NP toxicity. Although the safety of numerous chemical components utilized in medicine has been evaluated, the distinct physical and chemical characteristics of NPs might enhance their potential toxicity by determining specific interaction processes with biological organs, tissues, and cells. Therefore, it is imperative to examine the factors and processes that may lead to the toxicity of NPs. The use of NPs in veterinary products and treatments raises concerns about their possible hazards to animal health and the environment. Despite their widespread use, our knowledge of the effects of NPs is still limited, need for a detailed investigation into their safety use [43]. This chapter aims to thoroughly overview the safety and toxicological aspects of NPs in veterinary practice. It will explore the definition and importance of NPs, their uses in veterinary medicine (Table 1), and the possible hazards they may provide to animal health and the environment.

Table 28: Different types of NPs used in veterinary medicine.

Sr. No.	Type of Nano particle	Structural Description	Major use	Reference
1	Polymeric	Composed on polysaccharides (inulin, chitosan), comparable structural foundation to dendrimers, but their branches extend outward from a central core in a reorganized and reconfigured fashion.	Commonly used in the production of antibodies, such as Newcastle.	[21]
2	Liposomes	They are depicted as highly prosperous due to their biodegradable composition. Similarly, particles are dispersed by a bilayer of phospholipid membrane and encapsulate the hydrophobic substances.	Utilizedto immobilize chelated antibodies on their outer surface and for drug delivery due to their exceptional biodegradability and biocompatibility.	[44-45]
3	Carbon NPs and nanotubes	Carbon NPs exhibit a spherical morphology. While nanotubes exhibit tubular or needle-like structures.	Carbon NPs are proficient in successfully communicating with pathogens or cells. While nanotubes can be employed as biosensors for the identification of various components, such as immunoglobulins.	[46-47]
4	Microbivores and respirocytes	Both NPs are designed with exact chemical structures, including tiny pumps, sensors, and moving parts, to accomplish their desired purposes.	Respirocytes mimic the characteristics of both red blood cells and white blood cells autonomously. Microbivores efficiently capture pathogens as they effectively supply	[46-47]

			oxygen to tissues and remove accumulated carbon dioxide through specialized sensors.	
5	Nano shells	Round shaped connected with outer gold layer.	Used for examination of malignant tumors by irradiation with Infrared laser. They diminish the force of X-beam and subsequently can be applied as an adjuvant for radiotherapy.	[47-48]
6	QDs	They consist of a core and an outer layer made of an inorganic substance, along with a fluid coating that can be linked to various biomolecules. A crystal is in the center. The colour of the emitted light is determined by the size of the crystal.	QDshave applications in diagnostics and immunodiagnostics.	[21, 49]
7	Solid lipid NPs	They consist of a lipophilic core. Furthermore, cationic solid lipid NPs can effectively bind nucleic acid components through electrostatic coupling.	They are capable to cross the blood brain barrier thus, effectively deliver the medications inside the central nervous system.	[21, 50]
8	Dendrimers	These are hyperbranched nanomaterials made of polymers that are incredibly small, smaller than human cells, and they are exceptionally dissolved in aqueous solution. When introduced into the flow, their small size and composition arrange them to be strategically removed from the activation of any unwanted resistance	The expansion of solidity and increased restorative productivity can result from the covalent conjugation of dendrimers and stacked drugs. They are essential to the treatment of cancer.	[51-52]

		reactions. They resemble a tree made of enlarged 3D atoms.		
9	Metallic NPs	The metallic center of metallic NPs is covered in a protective coating.	Various metals that are employed in the nanosystem primarily used in cancer treatment one of them is gold. Additionally, metallic NPs are loaded with a variety of antibodies and chelated radionuclide.	[53-54]
10	FeO NPs	Their structure consists of an iron core encircled by an outer fluorescent silica layer, which is where the drug attachment takes place. An outer shell made of natural polymers helps to regulate the particles.	The primary characteristic of this category is their ability to be guided to their target through the bloodstream due to an external magnetic field. They work better for medicine delivery, heat therapy, and imaging.	[47, 50]

23.2 Physicochemical Properties and Toxicity

Some of the important parameters involved in the physicochemical properties of NPs involve Formulation factors that include composition, size, surface chemistry, and form help in deciding the NPs' behaviors in interacting with the biological systems or biomolecules. They influence the biomolecular and cellular communication, kinetics and diffusion of the biological processes, biodistribution of the NPs in the body, and immunogenicity and toxicity of the NPs [55]. Moreover, natural and accidental NPs are not as uniform in terms of their physicochemical properties, that is differences in their material, size, chemical reactivity, and morphology compared to the designed NPs [56]. This is a problem because there are many different forms of NPs and due to this evaluating and understanding the impact and toxicity of NPs within biological environments is difficult. Due to this, there is a new risk area known as nanotoxicology[57].As mentioned earlier, whenever NPs are exposed, they also can affect organs, tissues, cells, and biomolecules, natural, accidental, and engineered are the classifications of NPs [58]. It can cause negative nano-bio interactions hence resulting in negative impacts and nanotoxicity from this interaction. When evaluating toxicity, one also needs to provide detailed information and analyze the structure and the physicochemical characteristics of the NPs. This allows one to administer the observed adverse effects more effectively to particular characteristics of NPs, defining precise relationships between structure and activity/toxicity. Evaluating the safety of NPs with various characteristics of size, shape, and composition, which is typical for natural and toxic NPs, is complicated by the fact that a NPs toxicity depends on its structure [59].

Therefore, it becomes very challenging to generalize information about the toxicity of NPs due to the strong correlation between the characteristics of the examined NPs with the biological environment. In this respect, one of the most promising strategies is to develop universally applicable guidelines that describe the procedures for evaluating the toxicity of NPs only in suitable conditions [60].

23.2.1 Influence of NPs size, shape, and surface chemistry on toxicity

Nanomaterial toxicity depends on factors such as size, surface area, chemical composition, and form that determine how these materials will interact with living organisms or biomolecules. According to several works, size has a critical impact on cytotoxicity. This impact is most connected to the concepts of surface area to volume ratio[61]. Also, NP size influences characteristics like sedimentation velocity, mass diffusivity, attachment efficiency, and deposition velocity[62]. Many scientists consider it as a key parameter of the interaction of these NPs with biological systems, affecting biological functions like endocytosis, cellular uptake of particles, and the effectiveness of the particles' processing in the biological cells[26]. Smaller NPs have a higher ion release rate which means that they are in direct contact with the cell membrane and cellular penetration is higher thus giving more toxicity[63-64].

The toxicity of NPs is typically influenced by their size, as smaller NPs have a greater ability to penetrate biological systems. NPs measuring less than 50 nm, particularly when administered intravenously, exhibit faster tissue penetration compared to those ranging from 100 to 200 nm, leading to more pronounced toxic effects. Decreasing the NP size increases the contact surface area, which elevates levels of oxidation and DNA damage. NPs smaller than 50 nm tend to rapidly spread throughout all tissues, causing harmful outcomes.

Particles over 50 nm in size are removed by the RES, especially the hepatic and splenic organs, where redox stress is highly acknowledged. The size of NP has a direct correlation on the physiological activity of the body. Concentrations less than 1 µm are capable of entering the cells but the implications of such action are still not quite comprehensible. On the other hand, NPs greater than 1µm cannot easily penetrate cell membranes, but they may bind with cell surface receptors, which in turn may affect interferences such as endocytosis[65]. Consequently, the size and number of NPs influence the level of cell survival and the generation of ROS within the cells. LDH release should be used as a reliable marker for necrosis[66].

- **Influence of structure and shape on cytotoxicity:**

NPs come in the form of spherical, rod-shaped filamentous, plate-like, etc., depending on their form, their toxicity varies[67]. The shape of NPs is an important factor in packaging and internalization in the membrane processes such as endocytosis and phagocytosis [68]. Spherical NPs are more readily internalized than tubular ones This is as postulated by [69]. Anything apart from spherical NPs has higher toxicity levels since the shapes have higher contact with the flow of blood. Carbon nanotubes (CNTs) can exist in two forms: single-wall (SWCNTs) or multi-wall (MWCNTs), but the former generates more ROS than the latter. It was also identified that carbon-based NP form and concentration are among the factors affecting NP toxicity [70]. The investigation on TiO_2 NPs revealed that this NPs leads to DNA damage through oxidative stress, lipid peroxidation (LPO), and micronuclei formation under light. These effects produced by the NPs depend on the shape of the NPs [71].

- **Influence of Surface on Cytotoxicity:**

he surface electrical charge of NPs influences absorption, colloidal qualities, plasma protein binding, as well as crossing the BBB [72]. Negatively charged NPs are more readily absorbed by cells compared to positively charged and neutral NPs, as they encounter less resistance from plasma proteins. This occurrence can destroy red blood cells, the clumping together of platelets, and ultimately, the development of harmful effects. The surface characteristics of NPs also impact the absorption of ions and biomolecules, leading to changes in biological responses. In addition, the surface charge of colloids defines the reaction, explaining how organisms interact with differences in sizes and shapes of NPs by storing them at the cellular levels[73]. For instance, analyzing the influence of the silicon surface charge in cell lines, ATP levels and genotoxicity are significantly lower in negatively charged hydrophilic and hydrophobic surfaces as compared to the positively charged hydrophilic amine-modified surfaces. The nature of interaction that takes place between cellular components and NPs strongly depends on the NP surface properties. Cells in contact with NPs can be greatly influenced and some of the properties that may be altered are adhesion and several other aspects which may include morphology, cytoskeleton, proliferation, and survival. This adherence is rather controlled by the surface of NPs and their various surface groups. For example, uncoated FeO NPs with about 50 nm diameter reduce cell adherence by 64% of the NPs coated with PEG. The above effect may be blamed on the fact that surface-coating chemicals may enhance or be used to reduce NP-cell metabolism, specifically about the contacts between NP cells and nano-tubes[74].

- **Influence of concentration on cytotoxicity:**

Different concentrations of silver NPs affect the functioning of mitochondria and the release of LDH, with toxicity levels varying according to changes in concentration [14].

23.2.2 ROS generation and oxidative stress

NPs cause oxidative stress by physicochemical interaction with cell membranes, leading to ion production [22]. Larger NPs exhibit a stronger affinity for cell membrane surfaces, resulting in elevated levels of cellular toxicity. NPs can enter cells and their organelles by mechanisms such as diffusion, endocytosis, and interaction with the cell membrane proteins like the phospholipid layer. Following penetration, the NPs can appear in the endosomes, nucleus or lysosomes depending on mechanisms that have not been well elaborated [23]. The Working group analyzed the cytotoxic effects of NPs of various sizes, which are such as apoptosis, oxidative stress, DNA damage and mutagenesis. Entry of NPs into cells through endocytosis mainly increases the levels of ROS, which can potentially cause inflammatory reactions and mitochondrial damage [23, 75].

Various mechanisms by which NPs make their activity within the cellular system are enlisted below.

1. Interactions between NPs and macromolecules, such as proteins, can alter their structure, which may potentially lead to toxicity. Oxidative stress is a crucial process that disturbs the equilibrium inside cells and compromises the DNA integrity, ultimately resulting in cell death [76]. Typical pathways of NP cytotoxicity encompass: The rise in ROS levels results in the process of oxidation.
2. Perforation of the cell membrane and all cellular membranes.
3. Damage to cytoskeletal components disrupts cellular activities.
4. Disruption in the transcriptional activity and damage to DNA molecule.
5. Energy imbalance resulting from malfunctioning mitochondria.
6. Disruption of lysosome development impedes the process of autophagy.
7. Alterations in the structure of membrane proteins can impact the transportation of substances.
8. Activation of inflammatory mediator production disrupts cellular and tissue metabolism.

NPs can enter cells through diffusion, endocytosis, or interaction with membrane receptors. Once inside, they are found in endosomes, mitochondria, endoplasmic reticulum (ER), or the nucleus. This presence activates signaling pathways that depend on ROS. The accumulation of ROSin the mitochondria degrades oxidative stress, leading to protein misfolding, triggering ER stress, causing DNA damage, and activating pathways that result in cell death (Figure 2) [77].

Although certain NPs, such as silver NPs (Ag NPs), are employed for their antibacterial properties, their incorrect application can damage human cells near the sites of infection. Ag NPs can sterilize wounds by blocking the growth and replication of germs. However, they can unintentionally harm nearby animal cells, causing cell death.

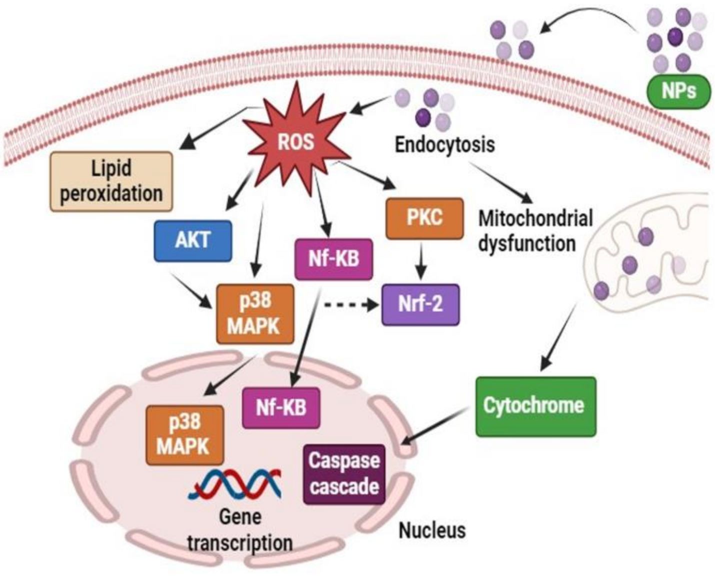

Figure 75: Cellular pathways of NPs to induce oxidative stress and toxicity.

23.2.3 Potential for NPs aggregation and agglomeration in biological systems

The formation of a protein corona around NPs is regulated by several parameters, such as the type of protein, features of the NPs (size, shape, composition), pH, and temperature [78]. For example, the tau protein interacts differently with SWCNTs as compared to MWCNTs, resulting in various alterations in its structure [78]. SWCNTs lead to significant structural changes in tau protein, whereas MWCNTs result in an agglomeration of proteins [78]. The behavior of proteins and NPs is also dependent on the last column which points out that the surface functionalization of the NPs is controlled by the thermodynamic factors [79]. Protein corona formed on NPs influence toxicity by altering the physiological behavior of NPs, namely, cell uptake and aggregation. The characteristics and layout of the corona depend on both, the properties of the NPs and the time the latter spend in contact with the environment, which determines how the NPs behave in the presence of cells. For soft coronas, the molecules are less bound in comparison with the hard ones, which are proteins with higher affinity[80]. In recent years, some types of nanomaterials demonstrated that a protein corona on NPs can decrease their toxicity due to the reduction of cellular uptake[81]. At times corona protein can prevent the membrane dysfunction caused by toxic NPs. The amount of endocytosis depends on the protein corona formed on the surface of NPs which has an influence on the entering of NPs into the cellular realm. Protein corona also impacts the distribution of NPs, some level of pre-coating extends the circulation time of NPs in the bloodstream. Thus, the biological distribution of NPs is a function of the charge and hydrophobicity of surface because they determine the behavior towards plasma proteins, the immune system, and other non-target cells [82].

23.2.4 Impact of NPs composition (metallic, polymeric, lipid-based) on toxicity

Certain subtypes of NPs have been widely employed in drug administrations, diagnostics of diseases and biologically sensing devices. Among such NPs, gold and silver NPs can be discussed in detail because of their optical properties and the availability of modification of their surface[83]. FeO superparamagnetic NPs play a role in drug delivery. as they are amendable to surface functionalization and can interact with magnetic fields, for the site-selective delivery of drugs [84]. Carbon nanotubes and fullerenes are being viewed as favorable for drug delivery because of their size and surface which aid entry into the cellular and surface chemistry [85]. SLNs mean that there is a lipid LLP type of drug delivery system that has a good flow as a drug carrier and that they are good in giving medicines that have poor solubility. Both biopolymers and synthetic polymers have been reported to be used frequently in drug delivery systems based on polymer NPs because they are biocompatible and insoluble in water. Polymer micelles which are formed from block copolymers have other peculiarities such as a rather low value of CMC and enhancement of the solubility of drugs. The advantages of hydrogel NPs are encompassed in the evidence that the hydrogels can swell by response to stimuli and release the drugs [24].

23.3 Routes of Exposure and Distribution

A crucial part of studying the toxicokinetic of NPs is to investigate the various ways they can enter the body. At present, living organisms are mainly exposed to NPs through environmental sources. However, due to the increasing use of NPs in biomedicine, it is also important to consider their therapeutic and diagnostic applications as significant exposure pathways. NPs can enter the body through inhalation, ingestion, skin contact, and injection. Each route of exposure can affect distinct organ systems and result in diverse harmful effects (Figure 3). Pulmonary inhalation is an important unintended exposure route that mainly affects the respiratory system, causing lung lesions, inflammation, fibrosis, and necrosis of lung tissue [86].

23.3.1 Oral exposure to NPs-based products (feed additives, oral medications)

NPs are used in the food sector to improve packaging and preservation, resulting in longer shelf life and better quality [87]. In addition, municipal and industrial waste enters aquatic ecosystems, accumulating along trophic chains [88]. Therefore, dietary supplements, water contamination, and aquatic species such as food fish can be potential sources of oral exposure to NPs [87]. Recent studies have shown that s AgNPs incorporated into food packaging can migrate into the food under different usage conditions [89-90]. Exposure to particles through inhalation during the manufacturing process can also lead to ingestion, as particles cleared by the mucociliary escalator are swallowed and removed through the gastrointestinal tract [91]. When food/feed is consumed, the gastrointestinal system acts as a protective barrier that helps to break down and absorb certain nutrients including carbohydrates, proteins, and fats. NPs can interact with the mucus layer, pass through the epithelium, and reach different organs once they enter the bloodstream. NPs with a diameter smaller than 100 nm are mostly absorbed by epithelial cells through a process called endocytosis [92]. AgNPs have been demonstrated to cause oxidative stress, DNA damage, and inflammation in enterocytes [91].

23.3.2 Inhalation of NPs aerosols in occupational settings or environmental exposure

According to the U.S. Environmental Protection Agency (USEPA), evaluating how nanomaterials interact with animals and the environment is difficult because their properties change at the nanoscale. These altered properties include reactivity, melting point, fluorescence, mechanical strength, and electrical conductivity [86]. The production methods for nanomaterials, including gas phase, vapor deposition, colloidal, attrition, affect their physical structure, which can lead to animal exposure through inhalation, skin absorption, or ingestion. Currently, there is a lack of complete information regarding the several sources, methods, and pathways of NPs exposure. The precise processes via which exposure to NPs induces detrimental interactions with the human body or aquatic and terrestrial creatures are still not fully understood. However, due to the unique features of nanomaterials that are influenced by their size, their impact may be more harmful when compared to exposure to larger materials with the same composition [93-94].

23.3.3 Dermal exposure through topical formulations or wound dressings

Nanomaterials can be released at several stages, including manufacturing, usage, and disposal. Ingestion or skin exposure can occur when drinking water or food additives are contaminated with NPs [95]. In addition, the consumption of fish and shellfish can also lead to direct exposure. The release of industrial effluent containing NPs into water bodies, whether accidental or intentional, increases the risk of exposure through direct skin contact and consumption of polluted water [96]. Although many NPs enter the environment as solid particles, their extensive surface area can cause them to aggregate into larger bulk material through chemical and physical interactions, potentially leading to ingestion or skin absorption upon exposure[97]. Furthermore, formulations used to treat a variety of dermatological disorders or injuries in animals provide numerous benefits, such as enhanced medication delivery, improved wound healing, and targeted treatment NPs in veterinary medicine are frequently encountered by topical preparations for example shampoos are formulated with silver NPs to be applied directly on the skin or wound dressings, leading to dermal exposure [98].

23.3.4 Distribution of NPs in the body and potential for systemic toxicity

After NPs are released into the environment and animals are exposed to them, the response of the body can vary significantly. The biological activity and kinetics of NPs are affected by several aspects, such as their size, shape, chemistry, etc. These factors have the potential to influence biological reactions, such as the movement of substances past protective layers of cells to reach other organs, the induction of oxidative stress, the attachment to proteins and receptors, and localization within specific cellular compartments like mitochondria. Translocation rates are predominantly unclear but are expected to be minimal, however, they may be influenced by impaired or sick conditions [99].

A significant study on the toxicity of NPs in living organisms has mostly concentrated on mammalian systems, specifically exposures affecting the respiratory system. Additionally, alternative exposure pathways, such as the gastrointestinal (GI) tract and the skin, have also been examined. The respiratory system and gastrointestinal tract possess multiple defense mechanisms to maintain clean mucosal surfaces by clearing inhaled cell debris and particles [100]. However, physiological barriers, such as the lung and gastrointestinal mucosa, may not adequately hinder the entry of NPs into organisms.

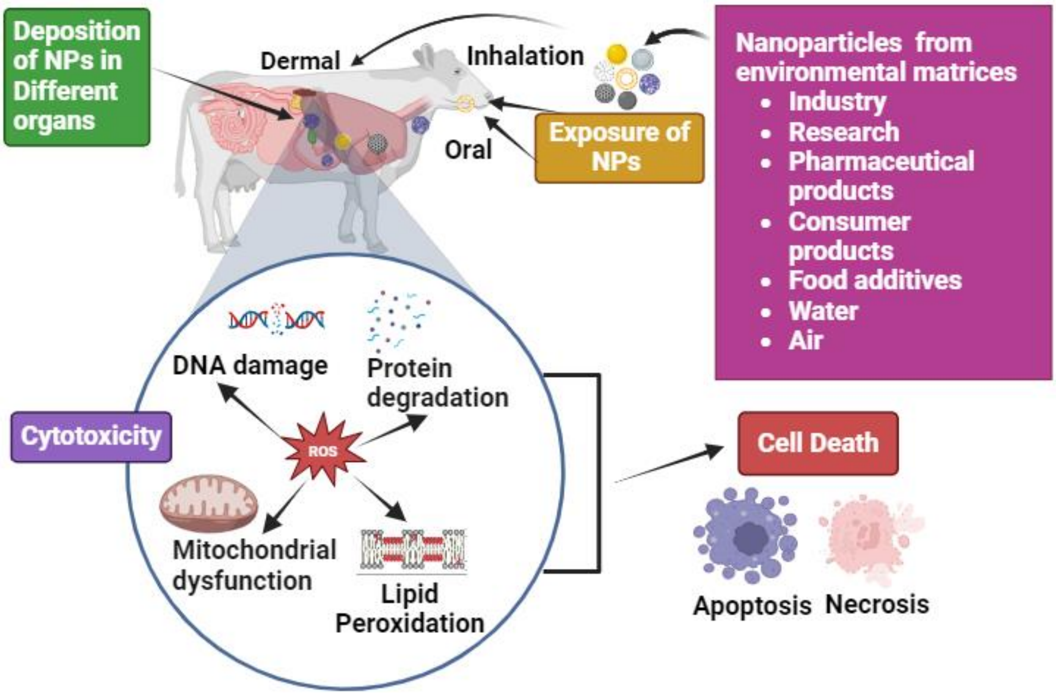

Figure 76: Biodistribution and toxicity of NPs following various routes of exposure.

23.4 Biological Responses to NPs Exposure

NPs can cause harm to specific organs in live beings through multiple processes, such as the production of reactive ROS, DNA harm, protein misfolding, disruption of membranes, and direct physical harm (Figure 4).

23.4.1 Oxidative stress

NPs can induce toxicity through oxidative stress, either directly generating ROS or stimulating the formation of ROS within cells upon interaction. Research suggests that almost all forms of NPs can cause excessive production of ROS in affected organs. Metal or metal oxide NPs are linked to higher levels of lipid peroxidation in the liver, kidneys, and spleen, but not in the heart tissue [101]. Antioxidants have been shown to mitigate impaired mitochondrial function caused by NPs, highlighting the crucial influence of oxidative stress on the toxicity of NPs [102]. The brain, which contains a high amount of unsaturated fatty acids that are easily damaged by peroxidation, is more susceptible to toxicity caused by ROS compared to other organs.

Exposure to NPs frequently leads to oxidative damage to cells, which is mediated by the p38-Nrf-2 signaling system. This exposure leads to the p38, c-Jun N-terminal kinase, nuclear factor κB (NF-κB), Nrf-2, and expression of heme oxygenase-1 (HO-1) pathways [103]. Furthermore, the increase in the level of heme oxygenase-1 (HO-1) after delivery of NPs implies for its role in combating oxidative stress. This process is probably controlled by p38 mitogen-activated protein kinase and the Nrf-2 pathway, which are elements of the cellular response to oxidative stress.

23.4.2 Inflammatory response

An inflammatory response is another widely reported cytotoxic effect of NPs, where the ability of NPs to incite the release of certain pro-inflammatory cytokines and/or increase the expression of related mRNA have been established [Nasrullah, 2023 #244]. For instance, it was ascertained that silica NPs cause a great stimulus of pro-inflammatory cytokines [Li, 2021 #243]. The intentional administration of NPs could lead to the generation of inflammation in organs, well vascularized with macrophages, such as the liver and spleen. That is when macrophages ‘swallow’ these NPs, it garners a cytokines response that in turn causes some impact on tissue morphology and efficacy [104]. Science has shown that suppression of the NF-κB signaling can lessen NP-associated inflammation and fibrosis. This therefore implies that NF-κB pathway is main inflamed crest induced by NPs [105]. This aspect is relevant to the ability of NPs to bind with various proteins creating a protein corona whenever they exposed to the biological fluids; this may in one way or the other affect their ability to interact with the cells and tissues which in turn alter their destiny and outcome.

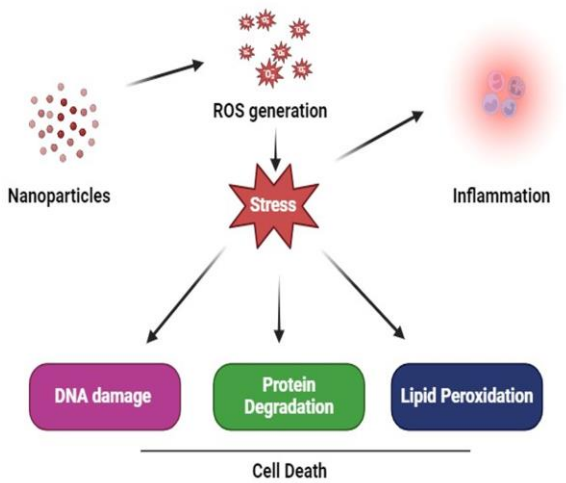

Figure 77: Biological Responses to NPs Exposure.

23.5 Organ-Specific Toxicity

Once NPs overcome biological barriers and enter the bloodstream through several exposure routes, they may accumulate inside important organs like the lungs, liver, and kidneys which can lead to toxic effects (Figure 5, Table 2).

23.5.1 Lung toxicity

Exposure to NPs through inhalation has been associated with various respiratory system injuries, ranging from moderate to severe. Previous studies have consistently observed the presence of inflammatory cells and fibrosis in the lungs of animals exposed to NP[106]. In the case of metal NPs, lung inflammation may persist for several days due to the presence of macrophages and cell infiltrations, yet this inflammation does not result in fibrotic responses

[107]. The presence of sepiolite nano clayparticles can cause temporary inflammation in the lungs, as well as sporadic development of cells with multiple nuclei in the central air spaces and slight thickening of the walls between the alveolar ducts and neighboring alveoli [108]. In addition, CNTs can migrate from the walls of capillary vessels to the cytoplasmic vacuoles in lung tissue. Simultaneous exposure of lung tissue to other harmful substances can worsen the toxicity of NPs, resulting in increased lung inflammation. In their study, Moon et al. (2010) found that when mice treated with lipopolysaccharide (LPS) were exposed to TiO_2 NPs, there was a combined and enhanced effect on the influx of neutrophils, accumulation of proteins in bronchoalveolar lavage fluid (BALF), generation of ROS in BAL cells, and production of pro-inflammatory mediators. Nevertheless, numerous in vivo investigations have indicated that inhaled NPs do not cause severe harm to systems outside of the lungs [108].

23.5.2 Liver toxicity

The liver is an important organ responsible for metabolizing and removing external chemicals when exposed to NPs might cause numerous abnormalities, such as changes in alkaline phosphatase and cholesterol levels, suggesting minor liver injury. Unlike inhalation or oral exposure, systemic methods such as intravenous and intraperitoneal injection lead to significant accumulation of injected material in the liver. They exhibited increased levels of glutamic pyruvic transaminase (GPT) and alkaline phosphatase (ALP), indicating liver damage, along with potential changes in body weight, organ weight reduction, and varying levels of pathological inflammation.

Inflammation is closely linked to the harmful effects of oxidative stress-induced damage, asa well-known consequence of NPs. FeO NPs tend to accumulate mainly in the liver and spleen, which can lead to the development of hepatic lipid peroxidation and oxidative stress [109]. Animals exposed to NPs have shown different forms of liver damage. For example, oral exposure to copper NPs has caused steatosis. Furthermore, the liver exposed to NPs has experienced impairment of respiratory chain complex activity [110], hepatic necrosis, an increase in eosinophilia within the cells, as well as nuclei shrinkage and condensation, bile-duct hyperplasia, with or without fibrosis, and pigmentation.

23.5.3 Kidney toxicity

The kidneys play a crucial role in maintaining homeostasis by eliminating metabolic waste and regulating electrolyte levels. NPs, especially small ones, are prone to elimination via the kidneys, potentially leading to their accumulation and subsequent adverse effects [111]. Exposure to NPs can induce pathological changes in the kidneys, such as enlargement of the glomeruli, epithelial cell degradation, and inflammation of the tubules. The renal response may be attributed to the migration of NPs from the lungs to the systemic circulation and secondary organs because of the release of inflammatory substances from the lungs.

23.5.4 Brain toxicity

The brain presents a significant challenge for drug delivery due to its limited accessibility. Nevertheless, NPs present a hopeful resolution by potentially overcoming the BBB thus transforming the diagnosis and treatment of central nervous system (CNS) disorders. Thus, although NPs have several potential advantages, their higher frequency use may lead to cerebral damage. NPs are frequently conjugated with certain brain sites in mind. For instance, polymeric NPs can be introduced into the wound sites for delivering delivery therapeutic agents resulting in no injury to other organs of the body [112]. Being smaller in size, metal or metal oxide NPs have a higher proclivity of being transported to the brain. For instance, investigations have shown that the TiO2 NPs bring about impressive enlargement of spongiocytes and intracranial hemorrhage besides compromising the trace elements, neurotransmitters, and enzymes for the maintenance of homeostasis [113].In the analysis of histological changes, it is possible to corroborate that FeO NPs can cross the BBB and produce subtle alterations in brain cells and tissues. Some of these changes involve the degradation of fatty substances in neurons and the degradation of membranes [114]. The interaction of the brain with NPs also induces behavioral changes and neurotoxic effects. Inhalation of NPs containing heavy metals has been associated with changes in motor activity, cortical function, and sensory nerve responses. Moreover, hippocampi have been identified as a significant target of NPs, resulting in deficiencies in spatial learning capacity and changes in gene expression associated with memory function [115]. NPs that are inhaled can go to the CNS through olfactory neurons, specifically impacting the olfactory bulb and causing inflammatory reactions. Inhalation of high doses of copper NPs has been linked to severe damage to the olfactory bulb, including reduced numbers of olfactory cells and structural impairment [115].

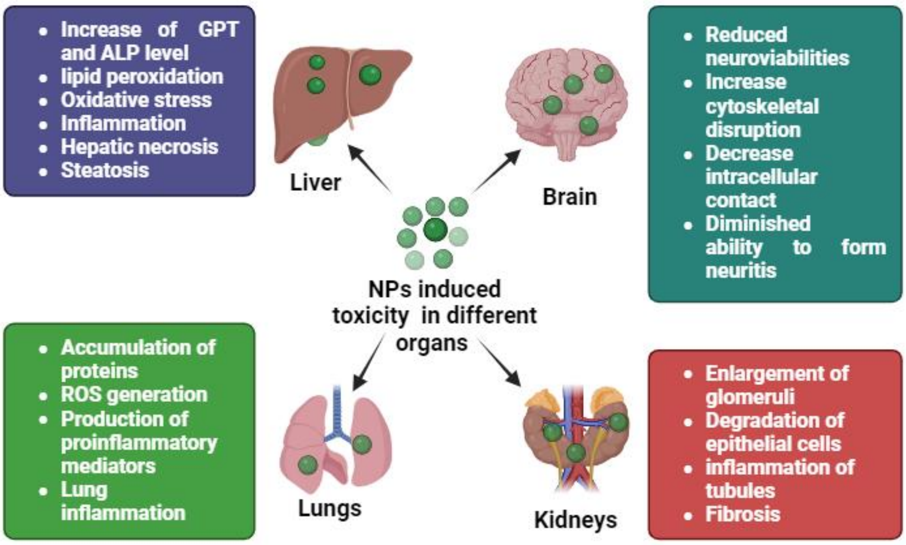

Figure 78: NPs induced toxicity in specific organs.

Table 29: Toxic effect of different types of NPs on specific organs.

Sr. No.	Type of NPs	Target Organ	Toxicity outcomes	Reference
1	Cu, Al	Brain	Increasing the toxicity of neurological and muscular system and improves penetration of NPs in the BBB.	[116]
2	QDs	Brain	Brain tissue had somewhat high concentrations of Cd ions, but no indications of inflammation or parenchymal damage were found.	[116]
3	Ag NPs	Lungs	Blood Ag NP concentration increased in a dose- and time-dependent manner and corresponding increases in alveolar inflammation and tiny granulomatous lesions were noted.	[31]
4	QDs	Lungs	Increase Albumin and LDH level	[117]
5	Al_2O_3	Liver	Accumulation of blood cells and melanoma, together with the necrosis and vacuolation of hepatocytes, and alteration of the portal vein.	[117]
6	QDs	Liver	NPs increased ROS levels in liver	[118]
7	CuO Nps	Kidney	Resulted in the manifestation of DNA fragmentation.	[119]
8	CNT	Kidney	Elevate the concentration of IL-8, LDH, and lipid peroxidation in the serum	[120]

23.6 Methods for Assessing NPs Safety in Veterinary Practice

Evaluating the safety of NPs in veterinary practice involves employing many methodologies, including conducting laboratory research, carrying out trials on suitable animal models, and conducting surveys to explore potential health effects. The objective of these approaches is to assess the toxicity of NPs, biodistribution, bioprocessing, and potential long-term effects in animals exposed to NP-based products such as feed additives or oral therapies. Important considerations include selecting appropriate animal models, analyzing the physicochemical characteristics of NPs, and conducting thorough toxicity studies to identify potential hazards and establish safe levels of exposure. Comprehensive risk assessments, based on these approaches, are crucial for ensuring the safety of veterinary products that utilize NPs [121].

23.7 Regulatory Considerations and Risk Management

Understanding the basic processes of NPs toxicity empowers researchers to devise techniques for reducing harmful effects and nanotoxicity. While certain studies examine the possibility of changing the composition of NP cores, most research focuses on manipulating surface chemistry and characteristics. Few methods such as silica coating and polymer encapsulation can regulate the breakdown of NPs and metal ions release, thus decreasing the harmful effects caused by metal ions and the generation of ROS. Another frequently used method involves applying PEG to the surface to reduce the formation of biomolecular corona and provide camouflage for the NPs. Nevertheless, because of the immunogenic potential of PEG, it is crucial to develop alternative surface-coating technologies that provide similar camouflage capabilities without causing unwanted effects such as hypersensitivity or allergic reactions. This camouflage method shows potential for decreasing the potential nanotoxicity of customized NPs, guaranteeing safer and more efficient nanomedicines, as well as secure uses in consumer items and industrial processes. Nevertheless, the implementation of this technique for natural and incidental NPs present difficulties, as well as issues regarding contamination of unintended agents. Antioxidant therapy or using respiratory protection, such as masks and other protective equipment, can help reduce the harmful effects of these natural and unintentional NPs [60].

23.8 Future Directions and Research Needs

Nanotechnology is increasingly attracting attention, and its application in the animal production sector is expected to further expand. Although the conventional use of nano-enhancements to strengthen animal feed may become possible soon, the complete substitution of antibiotics in feed with NPs will require more time. Before advancing to clinical trials and sanitation assessments, numerous biocidal alternatives must still undergo in vivo testing, as mandated by regulatory standards. it is essential to assess the toxicity of NPs in both cancerous cell lines and normal, healthy cell lines. Dependent exclusively on cancer cells and supposing that the NPs being studied possesses anti-cancer characteristics could be deceptive, as the NPs could be harmful to all types of cells. Validation of NPs capabilities reported in vitro research requires in vivo studies. Nevertheless, creating optimal nanomaterials with the ability to communicate with diseased or injured cells and tissues to initiate the regeneration process remains a difficult endeavor. Furthermore, the use of nanomaterials in regenerative medicine poses significant concern regarding to animal safety, particularly as this field is still in its early stages. The utilization of veterinary animals is progressively acknowledged as an essential translational model for human ailments. To fully understand the fundamental processes of how cells interact with biomaterials at the nanoscale and apply these results in practical medical applications, scientists and veterinary physicians must work closely together [122].

Moreover, advances in veterinary reproductive medicine hold the potential to provide valuable assets to human diseases and facilitate the exchange of knowledge about veterinary treatments to expedite translational medicine. Moreover, studies play a vital role in the animal production industry, specifically in revealing the disparities between knowledge and practical implementation.

Possible future applications of NPs in veterinary medicine encompass their use in nutraceuticals, medication delivery systems, and sperm purification. NP-infused nutraceuticals enhance nutrient absorption, leading to improved growth rates and performance in animals, despite the risk of degradation in the gastrointestinal tract. NPs included into drug delivery systems improve drug targeting and transportation, hence reducing the MIC and enhancing the efficacy against antibiotic-resistant diseases. But still, there are ongoing worries regarding the compatibility of these NPs with live creatures. However, concerns persist regarding the compatibility of these NPs with living organisms [123].

23.9 Conclusion

In veterinary practice, a comprehensive and meticulous assessment of the safety and toxicological implications of NPs is essential. Veterinarians can make informed decisions on the utilization of NP-based products in animal health by possessing a comprehensive knowledge of the physicochemical characteristics, biodistribution, pharmacokinetics, and toxicity profiles of NPs. Conducting comprehensive risk assessments and toxicity studies using both in vitro and in vivo approaches is essential to identify potential hazards and establish safe levels of exposure. Moreover, ongoing scientific investigation is important to address the existing gaps in understanding and concerns surrounding the safety of NPs. This will guarantee the sustainable integration of NPs technology in veterinary medicine. Veterinarians may optimize the benefits of NP-based solutions by prioritizing safety and employing evidence-based approaches to ensure animal health and welfare.

Authors Contribution

AJ, SS, MFN, MR, and AIA wrote various selective parts of the primary draft according to their specialization. MA, HF, MA, and MYW designed and draw the figures. SHM, ZM, MFN revised the draft. MR, MA and MFN added critical discussions. MA, AIA and MFN conceptualization of the study.

Conflict of Interest

No conflict of interest is declared by any of the authors.

Abbreviations

AgNPs	Silver Nanoparticles
ALP	Alkaline phosphatase
ATP	Adenosine triphosphate
AuNPs	Gold Nanoparticles
BALF	Bronchoalveolar lavage fluid
BBB	Blood-Brain Barrier
CNS	Central nervous system
CNTs	Carbon Nanotubes
ER	Endoplasmic reticulum
FeO	Iron Oxide
IL-6	Interleukin-6
LDH	Lactate dehydrogenase
MIC	Minimum inhibitory concentrations
MWCNTs	Multi-walled Carbon Nanotubes
NPs	Nanoparticles
PEG	Polyethylene glycol
QDs	Quantum Dots
RBCs	Red blood cells
RES	Reticuloendothelial system
ROS	Reactive oxygen species
SWCNTs	Single-Walled Carbon Nanotubes
TiO_2	Titanium Oxide
TNF-α	Tumor necrosis factor-α

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Chapter 24 : Antibiotic Resistance and Farm Animals

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Abstract

The development of antibiotic-resistant bacteria in animals is an issue of concern in global health because antibiotic-resistant diseases are an issue for both animals and humans. As the chapter under the extension, it seeks to discuss a literature review on diverse economic, environmental, and nutritional concerns surrounding the use of antibiotics on farm animals. This issue relates to the use of antibiotics in the production of animal growth, the development of antibiotic resistance, and how man is a victim of these antibiotic-resistant organisms from the animals he eats. Moreover, it overviews the risks/sinister characteristics of antibiotic-resistant infections and the outcomes of proper antibiotic use strategies, policy approaches, and international partnerships concentrating on combating antibiotic resistance. The present chapter underlines the need to address sustainable measures in cattle breeding to avoid future issues relating to the inoperativeness of antibiotics for human use.

Keywords

Antibiotic Resistance, Growth Promoters, Public Health, Farm Animals, Antibiotic Use, Policy Interventions, Livestock Production, International Collaboration.

24.1 Introduction

There are many types of farm animals, which include chickens, pigs, cattle, sheep, goats, and geese among others and all these animals are of great benefit to humans. These are crucial in agricultural-dependent economies and most circumstances, the agrarian-based economies contribute significantly to the total GDP. In addition to providing means of financial returns, farm animals also have a use in the environment and food for man through nutrients available from food products that come from farm animals. In addition, farm animals also hold social and economic significance in that they represent a source of income and provision for millions of people globally. Meat, milk, eggs, and wool among others are other important commodities of animal origin that are essential in day-to-day life and livestock source them. Socially, farm animals enhance sustainable farming practices relative to the natural environment such as food production, providing meat, dairy, and eggs (Iwu et al., 2020). They are of importance in conserving the fertility of soils since they are a source of manure, which is an organic form of fertilizer. Grazing animals also help in the regulation of vegetation and the contribution to the maintenance of species diversity The ecosystem is another element that brings an added aspect to the community of inhabitants. This chapter focuses on the significance of farm animals, the consequences of the use of antibiotics, and the emergence of antibiotic-sensitive bacteria and why it is crucial and significant to adopt and apply sustainable practices and policies.

24.2 Farm Animals Overview

24.2.1 Types of Farm Animals

Among the domestic animals, **chickens** are one of the most rare farm birds in the world today. They are mainly reared to produce chicken for stock and consumption known as broilers and for reproduction or laying eggs known as layers. Chickens are among the best feed converters hence the importance of ensuring that these are readily available for producing animal protein at an affordable price. They also do not require a lot of attention or even a professional setting for their keeping: they are bred on industrial, backyard, and small-scale farms. The **pigs** are also prominent farm animals having a high rate of gain and genetic efficiency of reproduction (Kabantiyok et al., 2023). The animals are mainly bred for pork production and since pork is consumed in most societies globally then this simply shows that the production of these animals is very essential. Pigs can also feed on different feed like by-products and leftovers from agriculture, hence they act as versatile animals in the different types of systems (Urban-Chmiel et al., 2022).

Cattle have several uses or products, including beef, milk, and work. Beef cattle on the other hand are reared for meat which is a rich source of protein that accompanies the nutritional needs of human foods diary cattle offer milk which is processed into several dairy products such as cheese butter and yogurt. Besides serving as food, cattle in many cultures are also used for other purposes such as plowing the fields and as carriers of goods and products. **Sheep** farming is the business of rearing sheep for their meat (Lamb or mutton), wool, and milk production. They can look for food in different habitats that include hot and dry areas as well as at high and elevated places (None Muhammad Jamil et al., 2022). Wool from sheep is one natural fiber utilized for making textiles and garments; meanwhile, the milk produced by the sheep is used to prepare specialty cheese and fresh dairy foods.

Goats therefore are different types of farming animals used for the production of beef or mutton, milk, and fiber. Goat meat refers to meat from goats and is commonly used as protein in various societies and is commonly categorized as chevon or mutton. Some specific types of goats are also valued for their fiber and include cashmere goats that are for cashmere produce and angora goats for mohair production. Major products from **geese** include meat, eggs, and feathers; although ducks are mostly used for meat purposes. Meat from goose birds is one of the most preferred ones all over the world, especially during festive moments. Goose eggs are certainly larger and have more nutrients than chicken eggs and therefore they are considered a delicacy.

24.2.2 Role and Services Provided by Farm Animals

Domestic animals play an important role of animals in food production; supplying different products that are useful for human consumption. Poultry, pork, beef, lamb/mutton, chevon, and poultry all play an immense role as a meat source containing proteins and other essential nutrients (Mann et al., 2021). The most vital animal used in providing **wool** is sheep and wool is a natural material that is used in the production of textiles. Wool is one type of fiber that is famous for warmth, durability, and versatile fiber and for this reason, wool has a lot of uses in clothing and other fabrics (Aardema et al., 2024). On most continents, cattle and occasionally goats are used for draught purposes while in tropical areas they are largely used for beef and dairy production. Farm animals also have nutrient cycling functions through manure production which is important in maintaining the fertility of the soil hence reducing dependence on chemical fertilizers.

24.3 Importance of Farm Animals

24.3.1 Economic Contribution

Livestock provides a handy service in the economy of the world. The livestock industry is also a major source of income and foreign exchange as it makes a major contribution to the gross domestic product (GDP) of many nations; more prominently in the developing world where agriculture is the major economic undertaking. Meat products, milk products, eggs, and animal wool contribute a lot of money to the producers and are widely purchased by consumers. Livestock farming across the world gives an approximation of billions of dollars every year. Many of these values involve not only production within the sector of broiler farming but also direct links with other sectors indirectly related to broiler production, including feed production, veterinary services, and meat processing (Bai et al., 2022).

Agriculture and more specifically livestock farming is the source of employment in developing nations. There are millions of people dependent on it in the form of its production in the form of food, its transportation, and its consumption in the form of food processing industries, transport, and retail. In the regions, animal production for food is significant in enhancing employment opportunities, and hence decreasing the rate of poverty. The employment effect created by the industry also touches different related services industries sector to produce a wide range of jobs available. This employment generation is very important for economic stability and growth, especially in employment-starved areas with limited alternative employment opportunities (None Muhammad Jamil et al., 2022).

24.3.2 Environmental Balance

It should be noted that farm animals are very useful in sustainable agriculture production. It serves to conserve the soil fertility by returning nutrients into the ground through manure an organic matter used to fertilize crops (Monger et al., 2021). Thus, this process contributes to the decrease in the use of chemical fertilizers, which makes farming much more environmentally friendly. Also, large animals like cattle and sheep can feed on plants that can hardly be consumed by humans on the field and across the country producing saleable food items like meat. It is an important way to manage the land properly to avoid thicket formation and fire which are dangerous to human lives and to support a healthy ecosystem (Rabello et al., 2020).

Farming has consequences for species richness, as animals are present in the fields. Well-managed livestock farming can prove helpful in preserving habitats for wildlife and, therefore, support numerous and varied ecosystems. For example, nutrient intake, large paws with claws, or grazing animals can help in the elimination of invasive plants, and thus foster the growth of native plants. Still, the question of numbers is a crucial issue because when the amount of cattle increases, they begin to overgraze the land and destroy the soil and the vegetation. Moreover, there must be a reasonable level of the density of the cattle and the frequency of the grazing to avoid negative consequences on the environment (Kabantiyok et al., 2023). Dealing with livestock must admit specific features, primarily directed toward the regulation of population density and ensuring the sustainable functioning of ecosystems.

24.3.3 Nutritional Value

Farm animals play a significant role in supplying the need for essential nutritional values that are core to the human body. Meat, milk, and eggs are a source of protein, essential amino acids, and micronutrients; iron, zinc, and vitamin B12 (Linggawastu Syahrulawal et al., 2023). It is essential to state that these nutrients are important for the healthy growth of the human body as well as for development. For instance, heme iron is present in red meat and dairy products and the consumption of these products provides the body with an excellent absorbable source of iron than the plant-derived products. For instance, vitamin B12 required for the correct functioning of the nervous system and synthesis of erythrocytes is present in animal food (Kim & Ahn, 2022).

The use of animal products in meals can also serve as a balance in the human diet and can make people give balanced meals. Animal proteins are known as complete proteins which are sources of all the necessary amino acids for body requirements. This makes them especially vital in tissue build-up, healing, immunity, and muscle build-up. Dairy products have calcium and vitamin necessary in building strong bones. Meat and fish contain proteins and metals which are important to the body while eggs are foods that are also rich in proteins and other vitamins and minerals. The presence of animal products contributes to nutritional adequacy and improved health and well-being status to complement those given by plant-based foods (Gupta et al., 2022).

24.4 Use of Growth Promoters in Farm Animals

24.4.1 Artificial Growth Promoters

Artificial growth promoters are feed additives that are used to feed the animals to improve their growth rate, feed conversion ratio of the grown feeds, and productivity of the animals. These substances can be hormones or antibiotics that, if administered in designated concentrations, result in the animals experiencing enhanced growth rates, improved feed conversion rate, and market weight achieved more quickly than it might under normal circumstances. The main reason for applying synthetic growth promoters is to produce the largest throughput of animal products including meat, milk, and eggs, given a minimal outgo as feed and sustenance expenses.

Some of the most well-known hormones that are widely used as growth enhancers in farm animals and especially in beef cattle include Bovine Somatotropin (bST) hormone, which is used to stimulate milk production in the cow and other animals that are used for milking. It is pharmacologic because it mimics a substance that is produced by the mammalian nurse in large quantities to allow the production of breast milk (Zalewska et al., 2021). Estradiol is a natural estrogen that is used with other hormones to have effects on the growth of beef cattle. Trenbolone Acetate is an anabolic steroid that has been used in fattening cattle and beef production by stimulating the gaining of muscle mass. Zeranol is an anabolic compound of the estrogen type found in Mexico used to stimulate growth in cattle and sheep. These hormones are administered under many restrictions to curb their effects on animals and people (Laborda et al., 2022).

24.4.2 Antibiotics as Growth Promoters

Antibiotics have been incorporated into meat-producing animals and other animals for growth enhancement for several years. Antibiotics when given in sub-therapeutic levels are not effective in the treatment of diseases, rather, they enhance feed intake and growth rates. They also assist in avoiding the spread of bacteria which are likely to slow down the growth and productivity of crops. Several classes of antibiotics are commonly used as growth promoters in farm animals. Tetracyclines are the classes of antibiotics in this category including oxytetracycline and chlortetracycline used in pigs, poultry, and cattle to prevent respiratory and enteric infections besides stimulating growth. Penicillins are hormones, including growth-promoting substances, and antibiotics such as penicillin (amoxicillin) are administered to control bacterial infections and facilitate growth in different animals on farms. Some of these antibiotics include sulfamethazine which is used for fighting bacterial infection and promoting growth in stock. These antibiotics are usually mixed with the feed or water given to the livestock in a bid to enhance their consumption to optimal levels that will allow the antibiotics to work (Xu et al., 2022).

24.4.3 Benefits of Growth Promoters

Growth promoters are produced and used especially in farm animals to increase productivity since they increase the growth rates and feed conversion efficiency. Animals grow to market size faster and therefore, more often than not, their production cycle is faster hence optimality in the utilization of resources that are incurred. This culminated in an improved efficiency which saw higher production of meat, milk, and eggs to feed the people (Chiara La Torre et al., 2024). The use of growth promoters has impacts on the economic aspects of farmers in a positive manner. It is evident that when production is enriched, the growth rate and other parameters such as feed efficiency can also be enhanced hence bringing down the costs of feed and other materials used in raising the animals (Nalepa & Lidia Hanna Markiewicz, 2022). These-cutting down on the costs –directly increases the profit levels in any business organization. Finally, some types of animals can be brought to market quickly, which will increase the turnover of animals and, thus, the income. Besides growth promotion, many growth-promoting antibiotics also exhibit advantageous features of acting as growth controllers and preventive measures against bacterial infections. This two-in-one role aids in the general management of the health of the animals, with low incidences of illness and death among them. Healthy animals yield better results in terms of reproducing and Arlington's herds rarely need treatment from vets, hence something that will improve the economic profitability of livestock enterprises (Gupta et al., 2022).

24.5 Negative Aspects of Antibiotic Use

24.5.1 Irrational Use of Antibiotics

Antibiotic misuse involves the unnecessary and/or excessive administration of these drugs in animal populations with no supervision from a veterinarian or according to the right check-up practices. Some of the instances include prescribing antibiotics at low concentrations, prescribing them for conditions that are not bacterial in nature, or turning to antibiotics as a substitute for cleanliness as well as other proper management of animals. It also includes excessive use of antibiotics, especially in cases where its use in growth promoters is not warranted. Examples of misuse and overuse of antibiotics in farm animals include (Lim et al., 2020).

Feeding livestock antibiotics at subtherapeutic levels to compound for disease, and malnutrition and promote growth leads to the emergence of antibiotic-resistant bacteria within the community. Using antibiotics to control diseases among animals, especially in regions where animals are penned close together and within stressful conditions. Consumers who administer antibiotics to their animals at their own will without the supervision of a veterinarian, hence, choose the wrong antibiotic type and wrong dosage. Failure to adhere to the recommended days of withdrawal before slaughter leads to antibiotics entering the food chain, especially in meats and other related products (Nalepa & Lidia Hanna Markiewicz, 2022).

24.5.2 Development of Antibiotic Resistance

Resistance to antibiotics is known to occur through many processes but the most common ones entail inheritable genetic mutations and via acquisition of resistance genes from other antibiotic-resistant bacteria. Antibiotics work by antagonizing bacterial microbes and killing them on the spot while those that are resistant to the antibiotic remain intact to multiply. Extensive use of antibiotics over the years allows these resistant bacteria to replace the normal bacteria in that region. Key mechanisms include are, forces acting upon Bacterial DNA Bacteria mutations and evolutions, genetic mutations can switch specific proteins of the bacteria for the antibiotics, making the drugs ineffective (Mann et al., 2021). Gene transfer is when bacteria on their own can obtain resistance genes from other bacteria through a horizontal gene transfer mechanism through conjugation, transformation, and transduction. Based on the classification of resistance genes discovered in the previous step, identify what specific antibiotic resistance genes are associated with the resistant E. coli.

Antibiotic resistance in farms is a complex event dining process influenced by the factors resulting from the high use of antibiotics. The following are some of the most popular antibiotic-resistance genes prevalent in farm animals, thus embracing this rampant world health concern. Some common antibiotic-resistance genes found in farm animals include. *tet* genes are normally detected in poultry and swine and the results presented by Hossain et.al., (2008) indicate that this gene is mostly transmitted through poultry. According to the findings of Chiara, 70% of poultry farms and 60% of swine farms contain *tet* genes that code for proteins that protect the bacterial ribosome from the action of tetracyclines, which indicates resistance (Chiara La Torre et al., 2024).

bla Genes are also found in cattle and pigs and encode beta-lactamase making the beta-lactam antibiotics like the penicillins and the cephalosporin system inactive. Previous studies identified that about 55 % of cattle farms and 50 % of pig farms were positive for *bla* genes (Smith et al., 2018). *sul* genes, which offer resistance to sulfonamides, have been reported in around 65 % of the poultry industry and 45% of the cattle industry. They change the target enzyme of sulfonamides, its functionality being thus eliminated (Hu et al., 2016). *mecA* gene is situated in one of the subspecies of methicillin-resistant Staphylococcus aureus (MRSA) and has been reported in approximately 40% of the cattle farms and 30% of the pig farms. It is named *bl2A* mainly because and encodes a protein that has a low-level binding affinity with penicillin, making it resistant to beta-lactam antibiotics (Bosch et al., 2016).

24.5.3 Prevalence of Antibiotic Resistance in Farm Animals

Antibiotic-resistant genotypes in farm animals are a growing issue in the world today. There is a high variability of the carrier rates of antibiotic-resistant bacteria in the different species of animals and parts of the world. For instance, a study carried out in the United States observed that poultry and swine farms had high MRSA and multidrug-resistant *Escherichia coli* infection rates (Blau et al., 2018). Likewise, a study carried out in China revealed that Asians acquiring over 60% of chicken farms harboring at least one Salmonella strain with multiple antibiotic resistances (Yang & Hassan, 2019). Spain has reported antibiotic resistant bacteria in animals and Food safety Authority in Europe (EFSA) has reported high resistance in *Campylobacter* from poultry and *Salmonella* from animals. Low- and middle-income countries have relatively more relaxed laws in the use of antibiotics, and this has been cited to worsen antibiotic resistance as noted in various studies conducted in South Asian and African countries (Boeckel et al., 2019). With the rising prevalence of resistance particularly in the developing world, these studies underscore the issue’s trans-border character and the importance of driven international cooperation in tracking and regulating antibiotic consumption in animal farming

The growth of antibiotic-resistant bacteria in farm animals that are likely to spread to humans, leads to various health complications. Pathogens that relate to the resistant type pose quite a challenge when it comes to treatment; hence, animals will take longer time sick periods, high morbidity/mortality rates, and much suffering. Paling and Kennedy (2004) reported that these infections are endemic and can be quickly transmittable among livestock, especially in intensive production systems. The problem of antibiotic-resistant bacteria minimizes the efficacy of routine conventional therapies. This can result in the utilization of other and, usually, more costly or less efficient forms of antibiotics when treating animals, and this would mean a rise in the cost of animal medical care. There are occasions when no remedy can be offered and, in such situations, health impact/failure and welfare consequences to the affected animals are inevitable. They also cause reduced efficiency in the management of diseases across the farm, which in turn means that there will be high levels of biosecurity needed for future control plans, and more reliance on vaccination and other measures (Lim et al., 2020). The overall net effect observed is a decline in production efficiency or more appropriately described as a less efficient and costly livestock production system.

24.6 Impact on Human Health

24.6.1 Transmission of Antibiotic-Resistant Genes

The transmission of antibiotic-resistant genes from farm animals to humans can occur through several pathways. Farm animals are potent sources of resistant bacteria and caretakers including farmers, veterinarians, and slaughterhouse workers can directly get infected by contact with these animals or their handlers (Katarzyna Kazimierska et al., 2021). Foodborne transmission of such bacteria can spread through meat, milk, and eggs at a processing stage or during transportation. When people use these products in their homes, they can hence eat food that contains antibiotic-resistant bacteria. One example of waste that undergoes recycling is dung that is produced by farm animals, which is used to fertilize crops. Bacteria can survive in manure and can be transferred to specific ecosystems such as soil, water, and food crops where it can indirectly affect humans through contaminated foods or water sources. The effect of antibiotics on antibiotic resistance can also be highly illustrated by giving out real examples of infections that are associated with humans (Kim & Ahn, 2022).

Several antibiotic-resistant infections in humans have been linked to the use of antibiotics in farm animals. Methicillin-resistant Staphylococcus aureus **(MRSA)** infections can be acquired from infected animals through direct contact or coming across infected animals from other farms without or with limited aqua treatments (Ribeiro et al., 2023). A specific type, known as livestock-associated MRSA (LA-MRSA) poses a great danger to people working with pigs and cattle. Extended-Spectrum Beta-Lactamase **(ESBL)-**Producing Bacteria. These bacteria comprise *E. coli* and *Klebsiella pneumoniae* which pose a threat to humans and may cause fatal infections. ESBLs are enzymes present in bacteria and researchers found out that many antibiotics cannot treat infections that are caused by these bacteria and they also blamed these bacteria on contaminated meat. ***Campylobacter* and *Salmonella*** these common foodborne pathogens possess resistant strains to various antibiotic agents; resistant strains primarily due to antibiotic usage in poultry and other livestock (Schwarz et al., 2018).

The development of antibiotic resistance in agricultural animals has far-reaching implications for human health. Animal-resistant bacteria can enter the human food chain via meat and dairy products, causing more difficult-to-treat foodborne diseases. For example, *Campylobacter* and *Salmonella* resistant to conventional medications have caused widespread outbreaks of foodborne illness (Scallan et al., 2011). Furthermore, farm runoff containing resistant bacteria might contaminate local water supplies, posing further dangers to human health. Antibiotic-resistant bacteria in water sources near farms have been demonstrated in studies to colonize human gut flora, resulting in asymptomatic carriage and possible transmission throughout communities (Tasho and Cho, 2016). Antibiotic-resistant illnesses also have a significant economic impact, resulting in longer hospital admissions, more intensive care, and the use of alternative, often more expensive, medications. Addressing these effects necessitates comprehensive tactics such as monitoring antibiotic usage in agriculture, enforcing restrictions, and encouraging research into alternate therapies and prevention measures.

24.6.2 Public Health Risks

The broader implications of antibiotic resistance for public health are profound. That is, infections by resistant bacteria prolong hospital stays and necessitate the utilization of multiple antibiotics and/or higher costs that may not be easily accessible, thus raising the cost of healthcare. Hence, antibiotics lose efficacy partly due to drug resistance, a factor that leads to increased morbidity and mortality rates (Bai et al., 2022). New strains of some particular infections might emerge and resist being controlled using the conventional and more economical forms of antibiotics. The antibiotic-resistant bacteria have the potential to increase the spread of community epidemics and complications within healthcare facilities. Further, the ability to pass on the genetic mechanisms of resistance also applies to other bacterial species and thus makes the conditions much broader (Linggawastu Syahrulawal et al., 2023).

Antibiotics are essential for the success of many surgical, oncological, and transplanting interventions. The resistant bacteria pose a threat to the safety and effectiveness of these procedures such as surgery. Case Study "An LA-MRSA" in the Netherlands showed that the prevalence of human MRSA is considerably high, and most of the cases are contracted from livestock, especially pigs (Smith et al., 2018). This has however been partially rectified by techniques like better hygiene as well as policies that have banned the overuse of antibiotics in animals. Real-World Lastly, ESBL-producing E. coli in UK supermarkets; a study carried out in the United Kingdom revealed that a high percentage of retail chicken meat was positive for ESBL-producing E. coli hence posing food-borne risks. Measures have been taken by the government through public health and awareness that advocated for proper preparation of meat and its handling (Nalepa & Lidia Hanna Markiewicz, 2022).

The World Health Organization (WHO) promotes the One Health approach, the concept suggesting that global health is not only about human health but also about the health of animals and the environment (Hu & Zhu, 2016). It encourages joint efforts to contain the usage of antibiotics in farming and increasing monitoring of antibiotic resistance as well as boosting new measures for safe agriculture and food supply. Such examples give credence to the necessity of adopting multi-faceted interventions in responding to the AR from farm animals' implications to human health. This should involve an intersectoral approach between the Ministries of Agriculture and Health; the community needs to be made aware of the problem.

24.7 Addressing Antibiotic Resistance

24.7.1 Best Practices for Antibiotic Use

Thus, proper measures should be taken to prevent, control, and regulate using antibiotics in farm animals as a way of preventing antibiotic resistance. Veterinary oversight is important to consult your vet before administering antibiotics to your pets through, they must only be prescribed and given out by a qualified vet. This results in higher percentage chances of using the correct type and dosage of antibiotics when needed. Abstain the use of antibiotics only in cases of confirmed bacterial infections, refrain from using them for reinforcement or acceleration of growth (Draper et al., 2020). This targeted antibiotic approach limits the chances of resistance which may be caused by the constant use of a single antibiotic. Prescribe anti-infectives for the right dose as well as duration to be sure the infection has been cleared and to lessen the possibility of the bacteria evolving resistance (Ribeiro et al., 2023). Record keeping documents every instance of antibiotic administration including the administered antibiotic brand, the dose taken, the days it has taken, and the animals treated. All the following helps in keeping track of the use of antibiotics and possibly containing them where necessary. Hygiene and biosecurity ensure that there is strict observation of hygiene and biosecurity to minimize the incidences of infections hence low antibiotic usage (Katarzyna Kazimierska et al., 2021).

Several alternatives to antibiotics can help maintain animal health and productivity without contributing to antibiotic resistance. Proposes the production of breast-enhancing bacteria that will provide better digestion and a better immune system that will not require a doctor. By enlisting the use of vaccines, species can be shielded against particular bacterial agents, thus lowering instances where antibiotics are required. Phytogenics are plant-based products that offer capabilities such as anti-microbial, anti-inflammatory, and anti-immune boosting. A good feeding regime apart from proper feeding systems that are supposed to offer adequate feeds to the animals to enhance their health and diminish the cases of diseases that may affect the animals, and proper living conditions in a bid to improve the standards of those animals' lives.

24.7.2 Policy and Regulation

Some of the current regulations and the extent to which they have been effective are as follows. All firms agreed that several governments have put measures in place to try and contain the usage of antibiotics in the animals that are reared on farms. Recently many countries ban the growth and promotion of antibiotics such as the European Union have prohibited the application of antibiotics as feed additives. Prescription requirements drugs can only be sold and dispensed with a veterinarian's prescription and can only be used for veterinary purposes (Kim & Ahn, 2022). Supervising programs that will help in recording the use of antibiotics in animals and the outcome on resistant organisms. Despite these regulations having proved effective in terms of reducing losses, the extent of their effectiveness in curbing loss-making is pegged on the enforcement and compliance levels. However, there is a problem; due to weak or inept implementation and oversights in the rules set, they are less effective in some areas (Mann et al., 2021).

Recommendations for Improved Policies

To enhance the effectiveness of policies addressing antibiotic resistance, the following recommendations can be considered. Stricter enforcement makes it mandatory for the existing regulations to be fully implemented by auditing or inspecting businesses and/or organizations and penalizing them when they fail to meet the prescribed standards (Monger et al., 2021).

There should be concerted efforts to establish and particularly use antibiotics worldwide to endorse the standardized global approaches. Launch incentive programs and aid in the promotion and acquisition of practices and goods that can replace the use of antibiotics. Such actions as broadening the scope of surveillance efforts to capture larger quantities of data on antibiotic usage and resistance patterns to support improved policy-making.

24.7.3 Role of Farmers and Veterinarians

Some of the measures that need to be taken include greater education and awareness of the risks of using antibiotics. Both farmers and veterinarians play crucial roles. It recommends that training the farmers should be conducted to educate them on the correct use of antibiotics, standard biosecurity measures, and practices not to use antibiotics. Awareness Campaigns can be conducted through organized public health campaigns that help the public become aware of antibiotic resistance dangers and the right time for usage (Kim & Ahn, 2022). Give constant opportunities for veterinarians to update their knowledge of innovative strategies and information regarding the care of animals. Encourage cooperation between farmers, veterinarians, and researchers to create the desired collaboration in terms of knowledge production and technology transfer.

Create and disseminate actionable recommendations and best practices for portion control healthy eating habits and other safe food-handling practices to reduce or eliminate the need for antibiotics. Provide technical assistance and information services to the farmers to assist them in the good implementation of these practices on their farms. Feedback should be put in place to help check if the practices that have been put in place are effective or not and other useful tips for change management. Educating itself and its teams through these strategies, farmers and veterinaries can play an important role in combating antibiotic resistance and encouraging sustainable animal farming. Thus, it is primarily the task of policymakers, researchers in the field of agriculture, and scientifically oriented people to effectively meet this challenge nowadays available for the mankind (Iwu et al., 2020).

24.8 Future Directions

24.8.1 Research and Innovation

Antibiotic resistance remains an ongoing issue around the world, which means that research efforts and advancements will be vital in this area. Consider and promote other therapies like phage therapy which is the use of viruses that produce antibiotics to act specifically only on pathogenic bacteria without interfering with the useful bacteria (Urban-Chmiel et al., 2022). As part of the research question, they should examine the efficacy of combined doses where there is more than one antibiotic or when antibiotics are administered with other substances. Improve targeted molecular-based drug therapy for bacterial infections and regulate the development of bacterial antibiotic resistance based on genetic and phenotypic frameworks. Increase and support further the antibiotic stewardship initiatives and interventions that focus on the rational utilization of antibiotics and the tracking of resistance trends (Laborda et al., 2022). The researchers have also noted that improving technology is a work in progress in combating antibiotic resistance. Use molecular data from genomic sequencing and biomathematics to monitor the spread of antibiotic resistance genes and new patterns.

24.8.2 Global Collaboration

Combating antibiotic resistance needs to be achieved through strategic and collective regional initiatives. Collaboration knows no boundaries and allows the pooling of various forms of capital as well as strategies for combating antibiotic resistance globally. Towards this end, effective policies and guidelines need to be put in place and implemented for the rational use of antibiotics, their monitoring, and control that can be followed globally (Rabello et al., 2020). Collaborate for synthesis of existing data, awareness, and development of research works by other researchers to form evidence-based approaches for understanding resistance mechanisms. Foster the development of more WASH infrastructure and capacity-building projects in low- and middle-income countries to enhance the early identification of resistance, promote better stewardship, and prevent future emergence. The global action plan of the WHO is aimed to offer guidelines for countries to formulate their national plans of action, to put into practice surveillance systems as well as encourage scientific and innovative activities. **CARB-X** (Combating Antibiotic-Resistant Bacteria Biopharmaceutical Accelerator) is an international public/private partnership aimed at identifying and fostering the development of novel antibacterial agents (Xu et al., 2022). The **JPIAMR** is an organization of the National Research Funding Organizations that collaboratively designs research agendas; provides funding for multi-disciplinary research projects; and focuses on key thematic areas in the field of AMR (Draper et al., 2020).

24.9 Conclusion

Antibiotic resistance in farm animals is a complex process that requires immediate and proper intervention to prevent both animal and human ailments. The authors discuss this problem throughout this chapter, using various approaches and different aspects to explain it. Farm Animals are imperative being chickens, pigs, cattle, sheep, goats, and geese, and are part of food security, business, and conserving the environment. The utilization of growth promoters, especially antibiotics has remained pervasive in driving productivity as well as maintaining animal health. However, problems such as antibiotic resistance have been realized from the careless use of antibiotics in health facilities and farms, and these are dangerous to human and animal health. They are contracted directly from the infected person, contaminated food items, or through contact with objects that bear the genes of resistance. Antibiotic resistance poses a significant threat to public health including more expenses for treatment, the failure rate of treatment, and the risk to successful medical procedures.

This chapter comprehensively advocates for contemporary measures to implement in strategies to address antibiotic resistance, namely, responsible use of antibiotics, embracing of other treatment forms, enhancing of policies and legal frameworks, and international partnerships. These are not only the concerns of humane society towards animals and livestock, or a food production health issue but a universal health problem. This is true because it involves measures such as practicing good antibiotic use, embracing non-antibiotic options, and strengthening international cooperation as a way of slowing down the emergence of resistant bacteria. There is therefore a need to practice sustainable livestock production and have sound public health policies to meet our food needs base our health on a strong footing and support a balanced ecology. Moving ahead, more research, development and cooperative efforts on the global level will remain imperative for finding ways to counter the threats from antibiotic resistance that threaten the health of the world's population. This chapter champions change by appealing to policymakers, HC individuals, farmers, veterinarians, and consumers with a common message to strive for a world in which such antibiotics remain useful in fighting infections, yet without compromising the welfare of farm animals and humans alike.

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Chapter 25 : Bovine mastitis,Risk factors, therapeutic stratigies and alternative treatment

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Abstract

Bovine mastitis one of the major threats to modern dairy farming , is the inflammation of mammary gland of the bovine. It is an inflammation of the tissues of the udder brought on by damage from the elements given as chemical irritability, or infection from certain microorganisms. Although the cause of bovine mastitis has long been comprehended, its multifactorial nature and complicated etiology make management more challenging. *Staphylococcus aureus*, *Streptococcus agalactiae*, other *Streptococcus* species, and *coliforms* are the most prevalent bacterial pathogens causing bovine mastitis.Moreover, mastitis can lead to the development of antibiotic-resistant which is an emerging threat to human and could have a detrimental effect on human health. Dairy sector is facing serious set back due to the high incidenc and prevalence of mastitis. Economically Mastitis affects dairy farmer due to a loss of milk yield, compromises milk status in term of qualtiy, the probabality of permanent loss of one or more quarter, or several mortility issues, premature culling rate and cost of treatment. There are many factors like hygienic nutrition , stage and lactation , body weight of animal , age, milk yield , udder type, teat size, parity, and period of lactation are directly linked with the pathogens involved in developing mastitis. Consequently, alternative therapies in particular, natural compounds derived from plants and animals have been explored for the preventive and therapeutic stratigies in order to control bovine mastitis.

25.1 Introduction

Mastitis, an inflammation of the mammary gland, has significant effects on animal well-being and the economic aspect of dairy farming. Even after recovery, it can lead to prolonged reductions in milk production. Bovine mastitis is considered a principal threat of a notable decrease in milk production and immense financial losses to the dairy industry of any country (Abebe et al., 2016). It not only shows the serious concerns regarding the quantity of milk but also deteriorates the milk quality by posing some physical, chemical, or microbiological changes which contributes undesirable and detrimental cahnges to a variety of dairy products (Heeschen, 2005).

In Asia Cow and Buffalo are the most frequently domesticated animals in order to meet the demands of peoples for Milk , meat and dairy products. In Pakistan, Population of dairy animals accounts for 97.1 million (including cattle 53.4 million and buffalo 43.7 million). Those providing 63,741,000 tons of milk available for human consumption. The total milk consumption by humans consists of 24,238,000 tones from cattle while 39,503,000 tones from buffalo (GOP, 2021-22).

One of the main contaminants in milk is Staphylococcus aureus. The primary cause of Bovine mastitis is the presence of the *Staphylococcus aureus* in surrounding environment of the animal. The primary bacterium that which is common worldwide as a result of inadequate hygiene and Cattle that are given antibiotics for an extended period of time become more resistant to methicillin and other beta-lactam antibiotics that are being used by the owners . The pathogen is not being eliminated from the local environment in spite of the stringent control measures. *Staphylococcus aureus's* susceptibility to antibiotics may be useful to veterinarians for Controlling staphylococcal mastitis can be achieved with antimicrobial therapy. Finding the clinical isolates' antibiotic susceptibilities is essential for both treatment and stopping the spread of resistant isolates (Aarestrup et, al., 2019).

The most transmissible bacteria that cause intramammary inflammation are Streptococcus aureus, Streptomycosis agalactiae, and Streptomyces uberis.Together with the mammary gland, the rectal, rumen, and genital regions are the primary reservoirs for these infectious pathogens. Whenever cows and buffaloes come into direct contact with an infected animal, or when milking occurs and infected milk comes into contact with an uninfected udder, the bacteria from the mastitis organisms enter the teat canal (Sharif et al., 2020).

Bovine mastitis is categorised into Clinical, Subclinical and Chronic Mastitis. Visual clots or discolorations in the milk, frequently accompanied by a tender and swollen udder, are indicative of clinical mastitis. Fever, appetite loss, etc., are also occasionally present. The most often isolated pathogens in clinical cases of mastitis are Streptococcus aureus, E. coli, Streptomyces dysgalactiae, and Streptococcus uberis (Bengtsson *et al*., 2005).

Affected quarter's milk has changed; it may have clots or flakes in it or be very thin. In addition to being grouped according to their place of origin, mastitis-causing agents can also be classified as major or minor pathogens based on the frequency and severity of their symptoms (Heikkilä et al. and Saidani *et al*.2021)

It is arguable that Str. uberis is primarily spread by transmission from cow to cow, though this is not the only way it can cause disease. The majority of writers have classified Str. uberis as an environmental causative agent because it originates in the vicinity of the animal. The increased prevalence rates of Str. agalactiae show that this bacterium is a major contributor to mastitis, particularly in herds with inadequate management and hygiene (Zadoks *et al*.and Davies *et al*., 2016).

Considerable losses in term of milk production occur through both clinical and subclinical mastitis. The milk yield does not reach upto the mark even the animal recovered from subclinical mastitis, so the financial loss is irreversible. Antibiotic treatment for cases of subclinical mastitis stops the condition from developing into a clinical form during lactation, but the subclinical form still exists even after treatment (St. Rose *et al*., 2019).

Mastitis costs dairy farmers finance in a number of ways. Direct expenses include labor, medicine, evaluations, and disposed of milk; indirect costs include lost future milk output and reproductive losses (Rollin *et al.,* 2015).Numerous risk factors, such as Bacteria involved in causing mastitis, host, and environmental conditions, are known reasons linked to the incidence of bovine mastitis. The mastitis control stratigies took into account in each of these parameters that responsible for directly influencing the disease. All the pathogen from environmental class do not contained in cow's udder and they usually occupy the surrounding environment e.g bedding material.In other words they are best referred as opportunistic Microorganisms , looking for the possibility of the desired conditions to cause an infection. For example, they may enter the teat during milking due to liner slippage, or they may cause clinical mastitis when a cow's natural immunity is weakened by stress or other nutritional deficiencies. Environmental pathogens like *Strep. uberis* and *Escherichia coli* (*E. coli*) invade and multiply in the udder's epithelial tissues, trigger the host immune response, and quickly disappear. Numerous bacterial species, including Streptococcus species (e.g., *Strep.uberis*), coliform species (e.g., *E. Coli, Klebsiella spp*., *Enterobacter spp*.), and *Pseudomonas spp*. have been identified as being responsible for environmental mastitis (Chen and Gu Han *et., al* 2020).

This Study is aimed at :

1. To Scrutinize and discuss variuos Risk Factors involved in Bovine Mastits.
2. To develope and design different control measures and stratigies to combat with the Economic losses produced by various forms of Mastitis.
3. Treatment regime alternative to antibiotic therapy used in war against mastitis.

25.2 MASTITIS:

Mastitis is a term that comes from the Greek words mammae,or mastos which means mammay gland, and "itis" which means inflammation. It is an inflammatory, agonizing bacterial infection of the Mammary tissues. Numerous different kinds of bacteria, both Gram-positive and Gram-negative, involved in the process of the disease. The disease might be moderate to severe, transient, or develop into a chronic condition depending upon the exact cause of disease .Research based data shows there have been a total of 137 distinct microbial species, subspecies, and serovars have been isolated and identified from udder of cow and buffalo. Studies using nucleic acid hybridization have changed how many mastitis pathogens are classified. Somatic cell count (SCC) is an effective tool and can be used as an index of udder well-being. Cows having good health status or those already recovered from mastitis should have an SCC lower than 200,000 cells/mL, and cows with counts over 400,000 cells/mL should be marked as having an intramammary infection.

25.3 CLASSIFICATION AND ETIOLOGY

25.3.1 Types of mastitis

According to the agent of infection involved Bovine mastitis can be graded into three different categories: Contagious, Environmental and Summer Mastitis (Heeschen *et.,al* 2012).

- **Contagious Mastitis**

  The mammary gland of an animal suffering from mastitis is the primary target of infection by infectious pathogens.These Microorganisms have adopted the conditions to sustain within the udder of the host, where they use to produce infections and may transmit to other animals.This type of mastitis is further differentiated into Sub-clinical and Clinical mastitis. Mastitis costs dairy farmers finance in a number of ways. Direct expenses include labor, medicine, evaluations, and disposed of milk; indirect costs include lost future milk output, reproductive losses, and the early culling and replacement of mastitic cows.

- **Subclinical mastitis:**

  It is distinguished by alterations in the content of milk (SCC, leukocytes, variations in the pH and ion concentration), although there are no outward manifestations of severe inflammation or anomalies in the milk. Probable indications of a subclinical infection include a decline in the quantity and quality of milk . Subclinical mastitis is attributed to the symptoms mild in nature by means of inflammation that the cow experiences. . Furthermore , the confirmation of sub-clinical mastitis is completed through SCC test on given milk samples.When subclinical mastitis occurs, the milk SCC in a healthy lactating mammary gland is 1,000,000 cells/ml of milk (Guidry, 2007).

  Clinical Mastitis: This stage is characterised by prominant alteration in the balance between host defence and pathogenic bacteria. This form is further classified into Acut and and chronic mastitis . Acute mastitis is attributed by the sudden onset with severe condition while chronic mastitis is characterised by th loger duration without severe conditions.. In addition, if other cows in the same herd show clinical signs of mastitis, farmers may cull the animals to avoid a more pervasive mastitis outbreak within the herd.Clinical mastitis is mainly focused by stratigies of prevention and elimination of the causitive agent (Sears et al., 1993).

  Chronic mastitis: This type of mastitis is an inflammatory response that remains for longer period f time for months, and may keep on with from lactation to next.

- **Environmental Mastitis:**

  The organisms that cause illness, such Escherichia coli, are generally found on the skin or in the udder, but when a cow comes into contact with a contaminated environment, they enter the teat canal. The pathogens often present in feed, bedding, and animal waste (Heeschen, 2012).

- **Summer Mastitis:**

A third type of mastitis, suggested as summer mastitis. This type is characterised by the an sudden onset of sign in dry cows and heifers and lead to severe and extreme damage to the udder. The cow must be culled early because the diseased quarter sustains irreversible harm. Cows are more prone to become infected in environments with high fly populations.

Table 30 Outline the different types of mastitis & their clinical signs

Type of Mastitis	General signs	Changes in udder
1.Peracute	• High fever • Tachycardia • Loss of apitite • Sunken eyes	• Abnormality of milk • Clumped mass • Swelling • Pain
2. Acute	• Low grade fever • Less depressed state	• Pain • Swelling • Abnomal milk (Yellow or brown flakes)
3. Subacute	• Absent	• Watery consistency of milk
4.Chronic	• Absent	• No visible change (only detected through tests e.g CMT)

25.4 Major & minor contagious pathogens

Infectious agents of bovine mastitis are also divided into major and minor pathogens ac-cording to their prevalence and the severity of symptoms. Coagulase-negative staphylococci (CoNS) and Corynebacterium bovis are regarded as minor contagious pathogens, whereas major contagious pathogens are Staphylococcus aureus, Streptococcus agalactiae, and Mycoplasma bovis. Escherichia coli and Streptococcus uberis are the two environmental pathogens that are most commonly isolated from cases of mastitis among many others.

25.5 Infectious or non-infectious Causes

It is observed that bacteria having cell wall in their strucutre are considered to be the major mastitis-causative agents. The main reservoir and mode of transmission have led to the classification of mastitis pathogens as either environmental or contagious. Keeping in view the pathogenesis of disease. These Microorganisms have adopted the conditions to survive in host, where they use to produce infections and may transmit to other animals. Environmental or opportunistic invaders have gained the entry to cause infection from the surroundings of the cow preferably than from other in-fected cows. These microorganisms are non-host-adapted and can be instantly done away with by the animal. In perspective of causing infection Fungi specifically yeasts of candida genus ,mostly associated with mastitis and are categorised asminor environmental pathogens.

Table 31 Different Causes of Masttis in Bovine

Species of animals	Organisms involved (Bacteria, Virus, Fungus)
Cattle	Bacteria: *Staphylococcus aureus; Streptococcus agalactiae; Str.agalactiae,Klebsiella spp. ; E.Coli ; Brucella abortus ; Pasteurella multocida ; Pseudomonas pyocyaneus ; Mycobacterium bovis ; Corynebacterium pyogenes.* Virus: *Vesicular stomatitis ; Infectious rhinotracheitis* has been documented as the cause mastitis n cattle (Blood et. al., 1983). Fungus : *Tichosoron spp ; Candida spp ; Cryptococcus neoformans*
Buffalo	Bacteria: *Staphylococcus aureus ; Streptococcus agalactiae; Str.agalactiae;* *Str. uberis ; Str. bovis; E.Coli; Pseudomonas spp ; Mycoplasma .* Fungus: *Tichosoron spp ; Candida spp ; Cryptococcus neoformans*

25.6 ECONOMICS OF MASTITIS

For most people in the world, dairy products—especially milk—are among the most important foods that people should eat on a daily basis. The demand for milk is rising due to the world's population growth, which is driving up the average amount of milk produced per cow. For meeting the demand of milk the increase in the milk yield resulted through genetic selection, additionally by proper cow nutrition and managemental stratigies. Mastitis is identified as the one of the economically damaging disease of the dairy cow affecting the dairy sector by means of reduction in milk quality and quantitiy. Presence of variety of bacteria in milk makes it unfit for human use . In comparison to the normal cow the mastitis has been documented as major threat leads to overall reduction in milk by 21% and butter fat 25% in affected cows.The milk of infected cow reduces in charactersitics in terms quality and quantity with the loss of butter fat and sugar contents. Mastitis not only leads to reduction of market value of infected cow but also the gross structure of udder is seriously damaged(Dhanda and Sethi, 1946). Mastitis affects the economics of dairy farmer in two ways : Direct and indirect Costs

25.6.1 Direct costs:

1.Use of antibiotics and abnormal characteristics of milk leads to disposal of milk.

2. Treatment costs and veterinary services.

25.6.2 Indirect costs:

1.Mortility in severe cases.

2.Decreased milk production in remaining period of laction

3.Extra labour required for the management of infected animals.

4. Increased cell leads to extra penalities by Milk collecting companies.

5. Increased culling rates.

25.7 RISK FACTORS ASSOCIATED WITH BOVINE MASTITIS

25.7.1 Risk factor:

An attribute (given as age, teat injuiry or season) that directly involved in the processes that increases the chance to predispose an individual to develop disease or negative outcome. In perspective of mastitis various factors are used to judge the unwanted events that exist throughout the period disease . These factors are deteremental in predicting potential threats that could lead to severe conditions. There are many factors like hygienic nutrition , stage and lactation , body weight of animal , age, milk yield , udder type, teat size are directly linked with the Microorganism involved in developing mastitis.

25.7.2 Pathogens and Infectious Agents

As we have discussed earlier there are several Pathogen which are the potential causes of bovine mastitis such as bacteria (e.g., *Staphylococcus aureus, Streptococcus agalactiae*), mycoplasmas, and fungi. The point of entry of these pathogen is teat canal of udder through environmental source such as contaiminated bedding and soil or through contagious transmission resulted from infected animas during milking.

- **Environmental Pathogens:**

  Bacteria such as *Escherichia coli* and environmental streptococci have the best adaptation to survive in wet and dirty environment. Poor condition and average sanitation and hygiene programme lead to the chance of lowering udder health.

- **Contagious Pathogens:**

  During milking, cows can contact with one other and transfer pathogenic microorganisms such as *Streptococcus agalactiae* and *Staphylococcus aureus*. Transmission is aided by improper milking practices, such as using infected equipment or not thoroughly disinfecting the teat.

25.7.3 Host Factors

The susceptibility of cows to mastitis varies based on intrinsic and extrinsic factors:

- **Genetics**: Some breeds and individual cows may have genetic predispositions to mastitis resistance or susceptibility. Selective breeding for resistance traits can reduce susceptibility.

- **Age** : Age influence many factors that directly link to cause mastitis or icreases its chance of ocuurence. The aged cows having more body weight are more prone to mastitis. An aging animal's immune system is vulnerable to stress encountered with it.

- **Species:** Because of firmly closed teat orifice structure of buffaloes they are considerably less subjected to mastitis than cows.

- **Breed:** Compared to Holstein Friesian (16.33%) and local breeds (13.27%), cross breed jersey (70.41%) is more prone to mastitis. (Acharya et., al 2022). In case buffalo prevalence rates have been documented higher in Jaffarabadi breed i.e. 20.00% while in comparison to Murrah buffaloes were 18.18 percent. (Shelke et., al 2022).

- **Herd Size:** Depending on previous reports and data available it can be concluded that there are higher prevalence rates in large size herd than with smaller one. These variation may due to poor maintinance conditions and housing facilities.

- **Season:** The rainy season has the highest frequency of mastitis (62.24%), followed by the summer (26.53%) and the winter (4.08%).It appears that mastitis outbreaks are more common in colder weather. This states that low-temperature environments are better for bacterial development.

- **Udder type:** Higher prevalence rates are recorded in cows having cup shaped udder than incase of bowl shaped udder.This may may be due to the fact that cup shaped udder remaind more closer to ground which is suitable microorganisms to thrive.
- **Milk Yield :** In high producing cows there are more chances of mastitis than those low producing cows. Heavy body weight of cross breed animals is suggestive of more surface area to hosts environmental microorganisms could lead to mastitis. Moreover , In Cows due to high production of milk teat spincter remained opened for longer duration which is definitive to microbes to colonize.
- **Stage of Lactation**: Primiparous cows (first-time mothers) and cows in initial period of lactation are more predispose because of juvenile or compromised mammary gland. In cows with late lactation they are higher risk to decreased immune function which lead to develope mastitis.
- **Udder Conformation**: Susceptibility can be influenced by variables such teat width, length, and orientation. Poorly conforming cows run the risk of not milking out completely, which puts them at risk for mastitis.

25.7.4 Management Practices

Farm management plays a vital role in mastitis prevention :

- **Milking Procedures**: The risk of mastitis is increased by poor milking practices, deficient pre- and post-milking cleanliness, and incorrect machine settings (such as vacuum levels and pulsation rate). It's critical to thoroughly sanitize teats both before and after milking.
- **Cow Comfort and Housing**: Keeping animals in larger number in compromised space , faulty ventilation, and inadequate bedding material increases stress levels in cows, which increases the chances to develope mastitis
- **Nutrition and General Health**: The immune system of cows can be weakened by inadequate nutrition, metabolic abnormalities (such as ketosis), stress from transportation, or environmental changes. As a result, the cows are more vulnerable to diseases.

25.7.5 Environmental Factors

The farm environment can harbor mastitis-causing pathogens:

- **Bedding and Housing**: Moist, dirty bedding invites microbes and stimulate their gowth. Adopting proper cleaning stratigies using good quality bedding decreases the chancs f contamination.

- **Climate and Weather**: The rainy season has the highest frequency of mastitis (62.24%), followed by the summer (26.53%) and the winter (4.08%).It appears that mastitis outbreaks are more common in colder weather. This shows that low-temperature environments are better for bacterial development.

25.7.6 Milking Equipment and Hygiene

Proper maintenance and cleaning of milking equipment are critical:

- **Method of Milking:** Mastitis is greatly influenced by the method of milking. It is recorded that in hand milking which require proper washng of hands there are more chances of carying bacteria from one animal to other which could lead to mastitis than milking with machine.In machine there is always advantage of safe halthy mlk.

- **Equipment Maintenance**: Teat damage or inadequate milking can result from defective equipment, such as broken liners or broken milking machines, which increases the risk of developing mastitis.

25.7.7 Human Factors

It is crucial for staff and works to play a part in preventing mastitis:

- **Proper Training and Education**: Incidence rates can be greatly decreased by providing farm personnel with the necessary training on sanitary milking techniques, managing cows, and early diagnosis of mastitis signs.

25.8 Alternative To Antibiotics For The Treatment Of Bovine Mastitis

Mastitis is one of the major threat and conomical damage for using antibiotics in treating the high producing cows and buffaloes.The conventional dairy sector is using 80% antibiotics for the purpose of treatment and prevention of bovine mastitis. The impulsive use of antibiotics is leading to devastating net results given as Milk tainted with antibiotics residues , developement of resistant bacteria and direct threats to human health. Another disadvantage of using milk containing antibiotics residues is reduction of shelf life , quality of milk and milk products. Although high standard preventive and control stratigies being applied in a war against mastitis but it continues to be the most prevalent herd health issue.Due to the several issues discussed above with use of antibiotics this could referred to the development of approaches and stratigies that may serve as alternative to antibiotics.

25.8.1 Application of Ethno-Veterinary Medicine in the Treatment of Mastitis

Ethno-veterinary medicine is referred to local healthcare approach based on methods , knowledge, tradtional trust, skills and practices.This local system consist of treating diseases of animals and as well as covers spirtual features of treatment. Different types of product extracted from medicinal (Outlined in table 1.6) plants in the form of infusions, pastes, drips, ointments, and minerals accessed by the animal owners. These product can be given to animal through drenches, as vaccine , vapors and massage.

Table 32 Different plant extract and their Uses (Abbasi et., al 2013).

Plant name	Plant Part used	Description and uses

1.*Citrus limon* 2.*Withania somnifera* 3.*Peganum harmala* 4.*Rumex nepalensis* 5.*Calotropis procera* 6.*Malva parviflora* 7.*Brucea antidysenterica*	Fruit juice Fresh crushed roots Leaves and branches Leaves in fresh state Stem Whole plant Leaf	• Fruit juice mixed with sugar and applied topically on udder for 15 days to treat bovine mastitis. • Application of paste of roots on udder to treat mastitis. • Smoke of plant leaves and branches applied on udder to treat mastitis. • Anti-inflammatory properties when applied topically on udder. • Paste applied on udder provide relief in pain swelling. • Provide relief in Mastitis especially in chronic cases. • Bovine mastitis

25.8.2 Nanotherapy

Nanotherapy is the use of nanoparticles in medical industry for the therapeutic purpose.In a broader aspect this technology promises for increasing effectveness of treatment especially in the field of engineering, electronics, environment, medicine.This technology provides the new ways material at nanoscale level and has the prime importance in view of common public health .Nanoparticles such as silver, copper nanoparticles according to previous research data have been reported to provide best results in case of mastitis management. Worldwide these new therapeutic stratigies getting popularity for managing *S. aureus* infections. Various studies states that nanoparticles such as silver shows synergistic effect when used with antibiotics (erythromycin) to treat *S. aureus* infection.

25.8.3 Vaccines

There are two arms to the mammary gland's immune response: the innate and the adaptive. An antigen is specifically responded to by the adaptive immune response arm. Every vaccine contains a killed or weakened version of the particular microorganism (such as Escherichia coli, Staphylococcus aureus, etc.) that causes the illness (like mastitis). The portion of the organism that stimulates the immune system to react to a "antigen" is still present in the vaccine, even though it has been modified so that it will not cause illness. Mastitis vaccines against S. aureus are made up of exopolysaccharides, which are sugar residues secreted by bacteria in the environment, or bacterins, which are killed or avirulent/weakened strains of S. aureus. The last and fourth measure of vaccine effectiveness is the decrease in the infection's progression from subclinical to clinical mastitis.

25.8.4 Bacteriophages

As an alternative to antibiotics, bacteriophage therapy can be used to treat intra-mammary infections. Viruses that can infect and destroy bacteria are known as bacteriophages. It has been shown that phages have promise as novel antimicrobial agents for use in veterinary medicine. As a preventative measure against *S. aureus*-caused intramammary infections, phage K has been utilized. The fact that milk and udder secretions inhibit the latter phage is one of its primary drawbacks.

25.8.5 Cytokines

The study of recombinant cytokines' immunotherapeutic application in the management of bovine mastitis has been spurred by the rise in antibiotic resistance. Small proteins called cytokines play a crucial part in cell signaling. Regretfully, the improved homeopathy The German physician Samuel Hahneman created homeopathy in 1810, and it is frequently described as a "complementary or alternative medicine." Homeopathic remedies are seen as advantageous by producers as a way to avoid antibiotic resistance development. Nevertheless, there is insufficient scientific proof of homeopathy's efficacy. Current veterinary research has concentrated on comparing the cure risk of therapy and control groups. However, some research on homeopathic treatment for mastitis revealed no positive results.

25.8.6 Bacteria and bacteria-derived inhibitory substances

A number of scientists propose that natural compounds with antimicrobial properties, generated by bacteria, could serve as viable substitutes for antibiotics in the management of bovine mastitis. Staphylococcus chromogene specrtum colonization of the teat apex in dairy heifers provides protection against elevated somatic cell count in the udder quarters. It's interesting to note that two of the ten S. chromogenes isolates, which came from two separate teats from the same heifer, consistently prevented all strains of S. aureus, Streptococcus dysgalactiae, and S. uberis from growing in vitro, but none of the strains of E. coli did.

25.8.7 Animal-derived inhibitory substances

There has also been discussion about the potential use of immunomodulators—a naturally occurring substance made by mammals—as non-antibiotic antimicrobial agents for the management and avoidance of bovine mastitis. Most well-known natural immunomodulator is lactoferrin. A glycoprotein called lactoferrin is present in a variety of bodily secretions, including milk, tears, saliva, and bronchial mucus. According to HAFEZ et al. (2013), this molecule has an antibacterial effect on NAS as well as several important mastitis-causing pathogens, including S. Aureus.

25.9 Plant-derived inhibitory substances and medium-chain fatty acids

New biologically active substances with antimicrobial activity may be found in plants, which is a promising source. The fact that plant-derived medications do not cause resistance after repeated exposure is one of their main benefits (DOMADIA et al., 2007). Diterpenes , trans-cinnamaldehyde, eugenol, carvacrol, and thymol are a few examples of naturally occurring compounds derived from plants. All exhibit antibacterial activity against the main pathogens associated with mastitis.

25.9.1 Nutritional Supplementation

An animal's general health is closely linked to its nutritional state, and an ability to fend off illness has long been linked to optimal nutrition. Many of the scientists has clearly discussed the roles of micronutrients in the processes of infection and immunity.

Table 33 Summary of micronutrient effects on mammary gland immunity.

Micronutrient	**Observation**
Selenium	• Decrease in neutrophil activity • Enahance bactericidal competence of neutrophils
Vitamin E	• Play vital role in neutrophil bacte1icidal activity • Se and Vitamin E when used in combination they reduces the chances of IMI at calving
Vitamin A	• Decreases SCC
B- carotene	• Role in bactericidal activities of phagocytes
Copper	• Cu deficiency causes reduction in neutrophil killing capability
Zinc	• Zn deficiency increases susceptibility to bacteial infection

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Chapter 26 : Pathogenesis, Diagnosis, and Vaccination Strategies for Infectious Bronchitis in Poultry

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Abstract

Avian infectious bronchitis infection is characterized by upper respiratory, alimentary, reproductive, and renal diseases in chickens, contributing to substantial biosecurity and financial challenges for the poultry industry. The economic impact of infectious bronchitis includes a decline in the egg production rate of about 20-70%, poor quality of the eggs for trade and hatching, stunted chicken growth, and an increased mortality rate (20-60%) in poultry. In infected flocks, cystic oviduct and edema of the shell gland lead toward false layer syndrome and shell-less egg syndrome, respectively. The causative agent for IBV is an RNA-enveloped virus with great potential for recombination and mutation, leading to new viral serotypes/variants. Furthermore, chickens respond to viral attacks via stimulating cell-mediated, mucosal, and humoral immunity. This chapter covers sufficient knowledge of IBV pathogenesis, specific geographical epidemiology, and advanced diagnostic methods that are significant for developing effective prevention and control strategies for IB. Efficient diagnostic techniques include viral isolation in the embryonated eggs, immunohistochemistry,

in situ hybridization, ELISA, neutralization assay, and RT-PCR. However, there is hope in the form of multivalent vaccines. These vaccines, with their ability to optimize the host's immune response against a broad spectrum of IBV serotypes, show promising outcomes in limiting the disease burden in the poultry industry. An advanced technology: nanoparticles are engineered for the optimized and efficient delivery of viral vaccines.

Keywords: vaccination, infectious bronchitis virus, clinical pathology, control of IBV

26.1 Introduction:

Avian infectious bronchitis: an infectious viral disease of chickens, causes economic losses along with several respiratory and reproductive syndromes in poultry due to its contagious nature and emergence of multiple viral serotypes of infectious bronchitis virus (Ramakrishnan & Kappala., 2019). IBV is an enveloped virus belonging to the genus Gemmacoronavirus (γCoV; group 3 coronavirus) within the family coronaviridae and having (+) ssRNA of genomic size about ~27kb (Jiang et al., 2017; Marandino et al., 2023). Moreover, direct contact with respiratory secretions of infected chickens and poultry products contaminated with the infectious bronchitis virus or inhalation results in acquiring this pathogenic viral disease from one flock to another (Tsukamoto et al., 2018). It can be found on the hatched egg's shell surface via shedding from either the alimentary tract or the oviduct of chickens. In chickens, IBV causes nephrotic syndrome (mortality rate due to IBV neuropathogenic strains is ~5-90%), renal dysfunction, reduced feed efficiency, epithelial hyperplasia, impaired oviduct function, abnormal development of eggs (aberrant oogenesis), limited egg production (up to 12%) and quality decline (Roberts et al., 2011; Ignjatovic et al., 2002). It also predisposes them to secondary infections such as pericarditis and bacterial airsacculitis, which may worsen the infection outcomes. IBV results in drastic economic losses by changing the quality and quantity of eggs or meat obtained via layers, breeders, or meat-type (broiler) chickens. Shells of poultry eggs may be pigmented, giving pale shade and susceptible to breakage. Infected eggs' albumen shows watery viscosity. This results in efforts to limit and control the hazard of infectious bronchitis (IB) in poultry by developing live and inactivated vaccines and other therapeutic practices (Samad et al., 2021; Samad et al., 2022; Khera et al., 2022).

IBV was first observed in the US in North Dakota in 1930, while the first report of infectious bronchitis was reported in 1931 by Schalk and Hawn during laboratory studies of clinically confirmed cases (Jackwood & de Wit., 2013). Beaudette and Hudson first cultivated IBV in embryonating chicken eggs (Ferreira et al., 2003). Primarily, IBV was recognized as an infectious disease only in young chickens, but later studies revealed it is associated with causing infectious illnesses in laying or semi-mature flocks. Some IBV strains are geographically restricted, while others transmit from one geographic area to another, with high rates in countries performing livestock practices (Bande et al., 2017). Moreover, respiratory illness and renal lesions associated with IBV were observed in the 1940s and 1960s, respectively (Cook et al., 2012). Van Roekel sheds light on the vaccination program with his initial steps 1941 (Jackwood & de Wit., 2013).

The causative agent of IB, infectious bronchitis virus, is typically ~120nm in diameter with spikes length of about 20nm. Structural proteins associated with IBV are spikes (made up of glycopolypeptides; S1 and S2 subunits), internal nucleoprotein (surrounds viral RNA), and membrane glycoproteins (~10% exposed to outside) (Mo., 2018). Additionally, small membrane proteins vital for virion formation are also present. IBV can be replicated in the respiratory tract, gastrointestinal tract, renal organs, and oviduct (Ramakrishnan & Kappala, 2019). According to classification, the former two groups (1,2) of the Gemmacoronavirus genus include mammalian coronaviruses that show a distinct nature from infectious bronchitis virus (which is also named avian coronavirus) concerning their genomics, genome organization of IBV: 5' (an untranslated region) UTR-1a/1ab-S-3a-3b-E-M-5a-5b-N-3' UTR (Sun & Liu., 2016). Moreover, new viral progeny starts to appear within three hours post-viral infection. Furthermore, IBV strains become inactivated when heated at 60^C and 45^C for 15 and 90 minutes, respectively (Tan et al., 2020). It shows stability at 6-6.5 pH in the culture medium. To limit its infectivity, treat the equipment with 50% chloroform (at RT) or 0.1% sodium deoxycholate (at 4°C) (Cavanagh & Naqi., 2003).

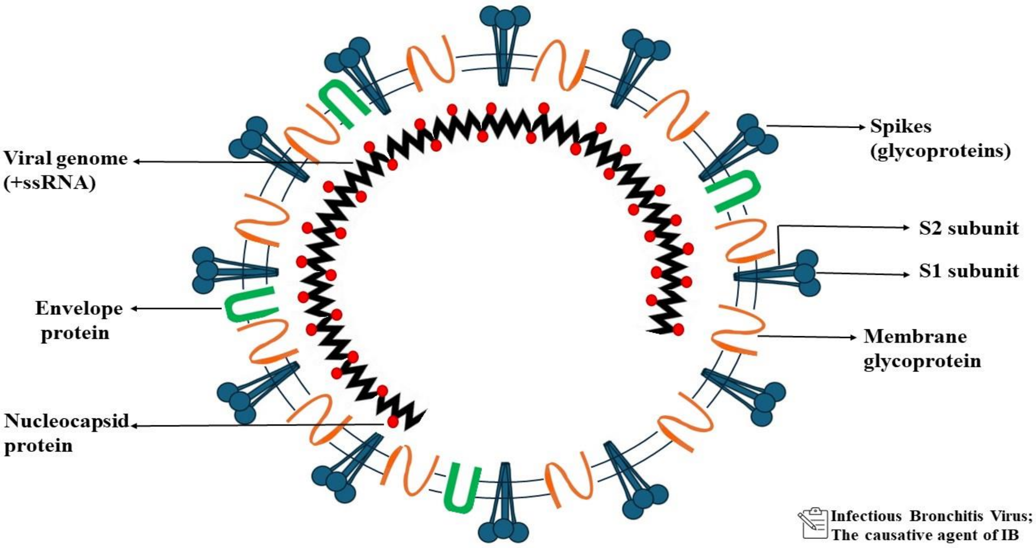

Figure 79: Avian infectious bronchitis virus (IBV); the causative agent of IB in poultry

The IB inoculation period is ~18 and 35 hours for tracheal immunization and ocular inoculation, respectively (Pohuang et al., 2018). It is generally dose-dependent. Live and inactivated virus vaccines are routine practices in handling IB in poultry. Humoral immune responses are triggered via an inactivated virus vaccine with a limited advantage over cytotoxic T-cell proliferation inside the host (Kreijtz et al., 2011). Inactivated vaccines, especially oil-emulsion vaccines, are inoculated before egg production in layers (10-18 weeks) and broilers (de Wit et al., 2022). The administration route of inactivated vaccine can either be subcutaneous or intramuscular injections. These vaccines have an advantage as they assist in minimizing the frequency of viral antigens present in the respiratory tract, thus limiting further viral spread. Broilers (meat-type chickens) are usually vaccinated with live IB vaccines as the initial vaccination; the period for administration varies depending upon the method used for the vaccination process and maternal antibody titer in chickens (Bhuiyan et al., 2021). The booster shot is administered to 10–18 day-old broiler (Yan et al., 2013). IBV strain attenuation is correctly achieved by growing them within embryonated chicken ova to produce live vaccines. Live vaccines illustrate some problematic consequences as they might revert to virulent form via genetic modifications and further accelerate in pathogenic capacities (Shoemaker et al., 2009). The diverse variety of viral serotypes makes it challenging to develop safe, economical, and reliable vaccines that are effective against most serovars. Therefore, employing limited vaccine variety with good management practices and vaccine rotation strategically is an effective control strategy to combat infectious bronchitis disease. One of the effective ways to enhance a host's immunity or to boost its defense mechanism comprises the production and administration of multi-valent vaccines. Prospects for vaccination management aim at genetic manipulation systems for this lethal disease, which can be beneficial (Kim et al., 2021). Vaccination programs for infectious bronchitis constituting live priming are effective and aid in safeguarding flocks from pathogenic viruses (Guo et al., 2010).

Detection of IBV can be achieved via isolation of the virus from embryonated eggs, antibody-based detection/antigen competitive ELISA, nucleic acid-based detection (RT-PCR), neutralization assay, immunohistochemistry and hemagglutination inhibition test (Bronzoni et al., 2001). For convenience and as a standard practice, RT-PCR and ELISA are performed as diagnostic techniques for IBV due to their accurate and rapid outcomes. Multiple tests using appropriate diagnostic techniques are vital to achieve the complete picture of the virulent stage. The finest diagnostic method can be explicitly established to suit disease outbreaks' specific needs, thus increasing diagnostic techniques' efficacy and accuracy (Villarreal., 2010).

26.2 Epidemiology:

The fatality rate in young folks is about 20-30%, although it can cause extreme economic losses when factors such as viral strain-associated and host immune status-associated factors are involved (Bhuiyan et al., 2019). Coinfection or secondary infections in infected folks result in an increase in morbidity and mortality rates in poultry folks. Moreover, neuropathogenic IBV strains are relatively highly pathogenic as compared to remaining IBV strains and cause 45% mortality (Cavanagh & Naqi., 2003). Proventriculus infections result in 70%- 95% mortality in young chickens (Yu et al., 2001).

Avian infectious bronchitis is widely distributed in Europe, Australia, Africa, the United States, and Africa (Khataby et al., 2016). Most infectious bronchitis strains are unique or specific geographically, while few strains of IBV correlate to the vaccine strains. Several Delaware IBV strains (reported in the 1990s, distributed across the US), Connecticut IBV, and Arkansas IBV strains have been reported (Bande et al., 2017). IBV proceeds to alter or modify as indicated by Ark-like strains. Delaware IBV strain is of medical importance as the vaccine against the DE072 strain gives rise to viral variants such as GA08 and GA98 (Cook et al., 2012). California variants causing respiratory distress were also reported in the 1990s. CAL99 (a unique but related variant) was reported in 1999, indicating California variants evolve with time. Other California variants include CA/1737/04 and CA/557/03 (Jackwood et al., 2012).

In South Carolina and Georgina broilers, IBV was reported in 2007, and Ark or Mass vaccines could not decrease broilers' disease burden. Such viruses were reported as GA07 (similar to CA1737/04 IBV serotype) and GA08 strains (similar to CA/557/03) (Ennaji et al., 2020).

In 1986, Ark-like variants affected Brazilian chickens, and Mass IBV serovar indicated maximal tropism for various organs such as the trachea and gonads (Roh., 2013). In Mexico, most of the chickens are shown to be affected by IBV strains in the early 20s. Four distinct IBV serovars were designated as D1466, D3128, D207, and D3896 in 1980. France, Italy, Spain, and Belgium were also affected by IBV strains (Davelaar et al., 1986).

26.3 Pathogenesis:

In poultry, young chickens risk developing IB disease and show more disease severity than other inclusive chicken varieties—chickens' ability to resist disease increases with their life span. IBV attaches to the host cell via S1 spikes (receptor binding domain). IBV illustrates virulence nature along with tissue tropism due to variation in its S1 glycoprotein (Wickramasinghe et al., 2021). Interaction of the host's sialic acid receptor with the receptor binding domain of spikes facilitates the virus to infect the reproductive and renal systems (Wielgat et al., 2020). The interaction between the putative heparan sulfate-binding site and IBV (attenuated Baudette strain) contributes to the broad host range. Subsequently, in the S2 subunit, COOH terminus, membrane fusion occurs following viral entry inside the host cell, releasing its genome, triggering replication, facilitating assembly, and promoting release (Abolnik., 2015).

Contact with bronchial secretions (mucus, sputum), fecal droplets of infected flocks, and aerosol transmission induces the spread of infection (Jackwood & de Wit., 2013). The aerosol transmission was observed within a day when the virus was detected in the Bursa of Fabricius, renal system, and respiratory system (trachea) (Abdel-Moneim., 2017). The incubation period depends on the infection route and the infectious dose. The respiratory route and ocular inoculation incubation period is ~18 h and 35 h, respectively.

26.3.1 Clinical manifestations:

The primary route of infection in chickens is via the respiratory tract, which infects the host's epithelial cells by producing infectious lesions all over the trachea, air sacs, and lungs. Proceeding to the next phase of infection targets the renal system and urogenital tract (oviduct or testes) (Cavanagh., 2007). It also infects the duodenum, cecal tonsils, and jejunum and often persists in the chicken alimentary canal. Clinical signs include nasal discharge, crackles during inhalation, and sneezing. Infected chickens appear to be dull, along with feather fluffing. Moreover, infected chickens exhibit edema, conjunctivitis condition indicating froth or foam appearance in the eye, profuse lacrimation, and a reluctance to move. Reproductive tract infection includes disrupting oviduct functions, leading to diminished egg quality (soft, pigmented shells along with watery albumen) and quantity (egg production) (Hoerr., 2021). Unless appropriate strategies are implemented, diminished production of eggs cannot revert to normal egg-laying in flocks. Thus leading to significant economic losses in the poultry industry.

26.3.2 Histopathological manifestations of IBV infection:

- **Respiratory tract:**

  Several histopathological changes are observed in the IBV-infected chickens, such as respiratory lesions formation, epithelial hyperplasia, epithelial cell sloughing, congestion, lymphoid cell infiltration, cilia loss along with infection in the Harderian gland of the eye (Hassan et al., 2021). Harderian glands become enlarged due to cellular responses against IBV and severe inflammation depending on the bird's age, IBV strain, and immune status of infected flocks (Chousalkar et al., 2007). The mucosal secretions of the host's goblet cell and dyspnea are also observed in infected chickens. Viremia occurs by infecting monocytes and macrophages (Sun et al., 2021).

- **Urinary tract:**

  Renal nephritis is characterized by the infection of a nephropathogenic strain of the infectious bronchitis virus (Munuswamy et al., 2021). Here, the enlarged and swollen kidneys become pale, along with tubular degenerative changes (Chong & Apostolov., 1982). Urate starts to deposit inside them. Moreover, secondary bacterial infections also occur because of IBV infection in chickens. Immunosuppressed infected flocks coinfected with bacterial infections, such as encounters with Escherichia coli, ultimately led to death in most chickens. Thus, significant economic losses occur in poultry (Wickramasuriya et al., 2020). Interactions of combinations of IB vaccine, Escherichia coli, Newcastle illness, and M. gallisepticum result in drastic body conditions with the formation of persistent lesions and infections leading to high morbidity and mortality rates in chickens; inoculated at the age of ~1 week (Bhuiyan et al., 2021). Clinical signs of IBV in the urinary tract include diarrhea, weight loss, and depression in the infected chickens (Najimudeen et al., 2020).

- **Reproductive tract:**

  Besides infecting the respiratory system, IBV also infects the renal, digestive, and reproductive systems of chickens. It affects the oviduct of laying hens and appears to be hypoglandular or nonpatent along with regressed ovaries in severe cases of infection (Cavanagh & Naqi., 2003). In young chicks, cyst formation occurs in the oviduct, and there is inflammation in the mucosa of the oviduct. Furthermore, egg yolk accumulation in the abdomen, epithelial degeneration, micro bleeding, epithelial deciliation, and pale and enlarged liver with proventriculus hemorrhages are also observed in IBV-infected chickens (Najimudeen & Shahnas., 2021). In male IBV-infected chickens, diminished sperm production has been reported besides infecting or targeting their testes (Yan et al., 2023).

- **Gastrointestinal tract:**

  Avian infectious bronchitis causes histological changes in the gastrointestinal tract of chickens, such as mononuclear cell infiltration, shedding of the host's epithelial cells, and alternations in the texture and consistency of feces (Najimudeen et al., 2022). Field and vaccine strains of IBV demonstrate persistence in the intestinal tract, primarily in caecal tonsils > seven months following viral shedding in feces ~20 weeks, even after recovery from the disease (Cook et al., 2012; Najimudeen et al., 2020).

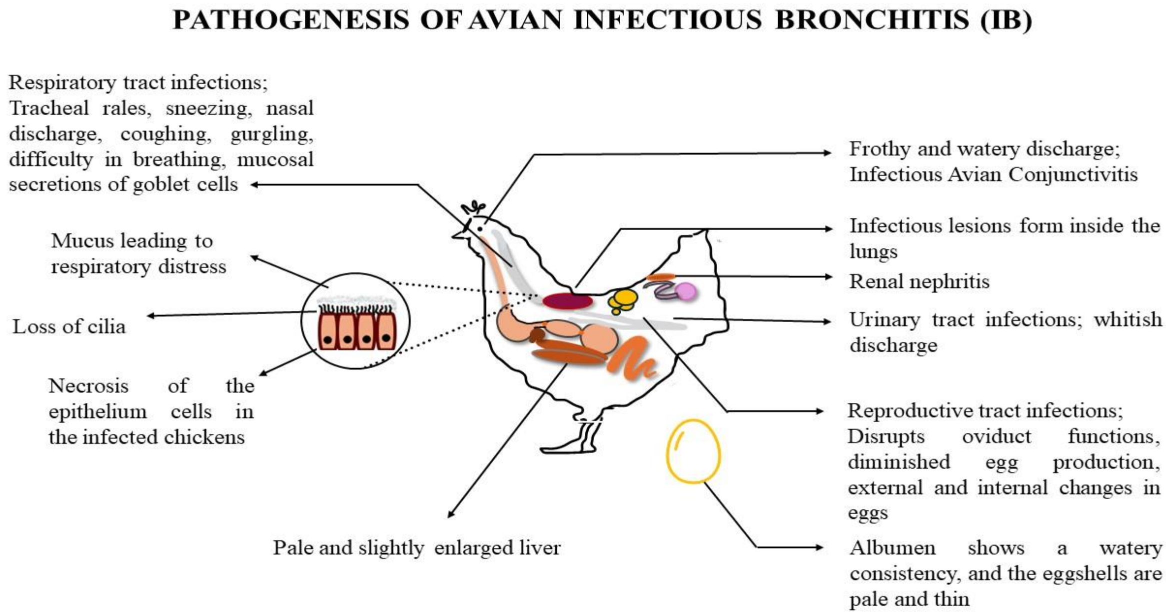

Figure 80: Clinical manifestations and histological changes associated with infectious bronchitis disease in chickens

26.4 Diagnosis:

An enhanced level of antibody titer and the presence of viral genome or antigens present in the suspectable sample; the sample may be a tissue sample or blood sample, indicating the presence of infection (Chhabra et al., 2015). Viral antigens' titer is at the peak level during 4-5 days of infection. Moreover, diagnosis is also based on virus isolation, clinical history, and RNA detection of IBV strains. As the primary target of infectious bronchitis virus is the respiratory tract, it can be isolated from the trachea (within the first week of viral infection), lungs, or renal organs, mainly the kidney. The spleen and trachea are sites suitable for histological diagnosis, while the pancreas and liver are sites for detecting viral antigens (Tang et al., 2022).

Sometimes, infected flocks' suspectable samples are authorized to pass through a medium such as embryonated eggs or various cell cultures (i.e., tracheal organ cultures) from an SPE source. However, viral antigens do not need to be detected in the first trial. Thus, at least 3-5 blind passages may be required before being reported as unfavorable as it does not cause any infection or persistent lesions in chick embryos (Awad et al., 2014). Embryo from infection-free chickens can be used to successfully isolate IBV as there will not be any interference of IBV with any other pathogen. Furthermore, TOCs (tracheal organ cultures) also facilitate the isolation and identification of infectious bronchitis viruses requiring intensive labor (Thilakarathne et al., 2020). Blind methods are time-consuming and tedious, while TOCs and SPE embryos (specific pathogen-free embryos) speed up the detection and viral identification process.

Direct detection of pathogenic strains of IBV in the infected flocks can be achieved via immunohistochemistry, in situ hybridization, nucleic acid detection, and electron microscopy (Cavanagh, 2001).

26.4.1 Viral isolation in the embryonated chicken eggs (ECE):

Various effects are linked to IBV infection in embryonated eggs, such as curling, embryo dwarfism, several hemorrhagic effects, formation of lesions (severity depends upon viral serotype/strain), and ultimately mortality (Tsai et al., 2020). To isolate IBV from chickens' embryonated eggs; 10-11 days chickens' embryonated eggs are grown in the allantoidal cavity. It contains 0.1mL of suspension sample; the sample may be cloacal or tracheal swabs and incubated for 4-5 days. Allantoidal fluid is collected later, and analysis of embryonated eggs is performed after 2-3 days of sample inoculation (Villarreal, 2010).

26.4.2 Antibody-based detection:

Tracheal scrapping taken during postmortem can be examined using immunoperoxidase or immunofluorescence assay and specific monoclonal antibodies (Abdel-Moneim et al., 2006). These scrapping or sections can also be examined by agar gel precipitin tests that have high sensitivity by propagating the virus in the embryonated eggs. Moreover, Allantois-derived cells or CAM sections (chorioallantoic membrane sections) can be employed for immunofluorescence assays (Wang et al., 1999). Viral presence inside the allantoic fluid or in tracheal organ cultures can be detected using specific monoclonal antibodies via ELISA (usually antigen-captured, indirect ELISA) (Basbaum et al., 1986). The majority of ELISA tests are unable to differentiate viral strains. This is due to the plate surface saturation with the viral suspensions for the interaction between antigen and antibody (Coetzee., 2018). Positive results indicated the presence of viral strain. IgG presence indicates humoral immunity. ELISA detects antibodies against spikes protein (S1) of infectious bronchitis virus effectively (Wang et al., 2002). Furthermore, competitive ELISA or monoclonal antibody-based blocking ELISA efficiently detects strains such as Arkansas and Massachusetts (Karaca & Syed, 1993). A sample containing infectious bronchitis virus antibodies is added to a microtiter plate encoded with the virus. Further monoclonal antibodies that are specific for specific strains are also added. Instead of IBV strain binding to specific monoclonal antibodies, it binds to chickens' antibodies. The higher the level of chicken's antibody, the more the blockage of binding between monoclonal antibody and IBV strain (Klestova et al., 2022).

26.4.3 Nucleic acid-based detection:

Samples obtained from chickens suffering from IB disease or amplified viral particles from embryonated eggs are employed for RT-PCR (Bande et al., 2016). The viral passage in the embryonated eggs can enhance the viral titer before analysis. RT-PCR technique is sensitive in detecting viral genome (RNA) extracted from clonal or tracheal swabs (Callison et al., 2006). Positive results are not sufficient for accurate detection, but the sequence obtained via RT-PCR should be compared with the corresponding viral strains sequence. RT-PCR has replaced VN and HI serotyping techniques due to its reliability and sensitivity of results. Furthermore, antigenic variations have been examined, such as nucleotide sequencing of genes coding for

S1 subunits (spikes) of various IBV strains (Legnardi et al., 2020). It is important to note that it is currently impracticable to distinguish viral strains from vaccine strains based on sequencing the S gene. This is due to the genetic diversity of IBV strains, their similarity with vaccine strains, the potential for recombination between viral strains, and the influence of vaccine strains on the genome of existing viral strains (Van Borm et al., 2021).

S1 genotype-specific RT-PCR and restriction fragment length polymorphism RT-PCR techniques are also used to identify IBV strains (Ameen et al., 2022).

26.4.4 Neutralization assay:

This test is specific for specific serotypes and detects antibodies produced by the S1 protein of spikes. This technique utilizes tracheal ring cultures or cell cultures, thus detecting cytopathic effects, any embryonic changes with time, or cryostasis. Here, no cross-reaction with serotypes other than the desired serotype occurs. These neutralizing antibodies target viral antigens and play a significant role in protecting effects towards the serum. These neutralization antibodies inside the serum are successfully detected by microneutralization test (Zhou et al., 2022). The limitation of this technique is the lack of standardization among various neutralization systems of viruses.

26.4.5 Hemagglutination inhibition test:

The infectious bronchitis virus doesn't directly clump erythrocytes (agglutination of 1% RBCs) of chicks (Zhou et al., 2004). Specific HI antibodies are produced post-viral infection, targeting viral spikes protein (especially the S1 subunit). Their role is to prevent the clumping of RBCs. Such antibody's titer detection and their presence are detected, usually 2-3 weeks after the viral infection. Samples collected from infected chickens were diluted and mixed with RBCS and IBV to perform this test. Positive results are indicated by the inhibition of agglutination, which indicates the presence of specific antibodies. Type C phospholipase enzyme exposes hemagglutinin to the virus. Thus, this test facilitates accessing the immune response against the infectious bronchitis virus in chickens (Villarreal., 2010).

26.4.6 Immunohistochemistry:

Immunohistochemistry detects antigens of IBV (viral proteins) in infected tissue samples, especially of the renal system and bronchial epithelium, providing a confirmatory diagnosis of the disease. To perform this technique, a suspected tissue sample is collected, followed by fixation, and immunoglobulins specific to viral antigens are added. Positive results are demonstrated by the detection of bounded antibodies to viral antigens (IBV antigens), and further staining confirms the presence of infection (Abdel-Moneim et al., 2009).

Moreover, in situ hybridization and microscopy aid in diagnosing or detecting positive sense RNA viruses, such as avian infectious bronchitis virus. N proteins of IBV, vital for viral replication inside the host cell, can be detected by using techniques such as electron microscopy (TEM; transmission electron microscope) (Maier et al., 2013).

26.5 Preventive and control measures:

26.5.1 Vaccination:

Vaccination is necessary to boost the host's immune system, thus limiting the disease burden of any infectious agent, as live and inactivated vaccines against IBV enhance chickens' immune systems to combat the infection (Guzmán & Hidalgo., 2020). One of the finest or easiest methods for inoculation is via water. It is important to practice vaccines having low virulence. Young chickens have low immunity, and highly virulent vaccines can be harmful or have negative effects on their respiratory system (Bhuiyan et al., 2021). Live vaccines are commonly used as initial vaccines for commercial layers, breeders, and chickens that are frequently exposed to pathogenic strains of IBV, thus providing long-term local immunity against pathogens or infections.

- **Live attenuated vaccine:**

  The live vaccine provides local respiratory tract protection in young layers and breeders and is formed by the serial, consecutive passages of IBV serotypes in ovo (inside the growing embryo) (Bande et al., 2015). The advantage of live vaccines involves the stimulation of the host's immunoglobulins (primarily IgA) along with T cells, as they are significant for providing long-term immunity (cellular immunity) to the host cell (Caron., 2010). Immunity at the specific infection sites, as well as systemic immunity providing broader defense against infectious agents, is provided by live vaccines. For example, Massachusetts strain H 120 is a mild and live vaccine used in poultry (Bijlenga et al., 2004). The immunogen is primarily administered, ensuring strong immunity, particularly in areas where the disease burden is extreme, to sustain robust respiratory immunity of the host. Vaccination routes include ophthalmic administration (eye drops), intra-tracheal administration, intranasal administration, In Ovo vaccine administration, and mass vaccination (immunization to the whole community, either by drinking water or broad spraying) (Nazmi et al., 2017). Limitations of live vaccines include side reactions post-vaccine inoculation. A booster dose of live vaccine is administered around a week ago. Multivalent combinations, including different antigenic type vaccines such as Ma5 (mild-single component) with inactivated vaccine and IB 4/9 vaccine, safeguard flocks from diverse infectious bronchitis virus strains/serovars. Another combination comprising of IBV 274 (consisting of D274 viral strain) or IBV 4/91 (consisting of 4/9 IBV strain) with IB multi-vaccine and Ma5 facilitates in maximizing the vaccines' potential to achieve widespread immunity to the flocks and shield them from further IBV attacks (Bhuiyan et al., 2021)

  One of the major challenges faced nowadays is the emergence of diverse variants of IBV with time, which further complicates preventive procedures and control measures. Precise selection of vaccines specific for strains in particular geographical areas is necessary, thus improving control and preventive strategies for IBV infections in poultry. When multiple IBV strains circulate in the field, cross-protection in specific pathogen-free chickens is necessary, which provides a degree of immunity as well as lessens the disease severity in infected flocks even if they are exposed to genetically different variants. To achieve this goal, the combination of the Connecticut IBV strain and mass strain with the JMK IBV strain shows promising results (Umar et al., 2016).

- **5.1.2-Inactivated/killed vaccine:**

Inactivated vaccines either contain killed organisms or antigens, usually in purified form, providing immunity to the host cell (chickens), but the immunity is not long-lasting as the killed form of organisms or antigens are unable to sufficiently stimulate the host defense system as live vaccines do (Guo et al., 2010). A positive aspect of inactivated vaccines is that they generally don't result in pathological changes inside the host body due to their inability to undergo replication inside the tissues or cells as live vaccines do. Advanced and highly complex techniques are involved in the inactivation and later purification of antigens, making this an expensive process. Moreover, inactivated oil emulsion vaccines are administered to poultry breeders before the egg-laying period (Lopes et al., 2021).

An inactivated vaccine plays a significant role in providing maternal immunoglobulin protection to the offspring (chicks). As inactivated vaccines stimulate immunoglobulin synthesis, the level of immunoglobulin starts increasing inside the host body, circulates in the blood, and later passes to offspring, facilitating early protection against certain diseases. Furthermore, streamlined vaccination programs reveal positive prospects in protecting chickens against pathogenic serotypes of IBV. This technique involves the combination of distinct antigens, particularly inactivated ones, which enables broader immunity only with a few doses (Samad et al., 2021)

The overall objective of killed vaccines is to provide maternal-derived immunity to the young ones later, with the booster dose providing sufficient immunity generally at 6-7 weeks of age (Chhabra et al., 2015). Inactivated immunogens are efficacious in eliciting broad-spectrum protection against a decline in the rate of poultry eggs since the virulence and potency of live vaccines might not be sustained until the onset of the oviposition phase (egg laying phase). However, limitations of manual delivery of killed vaccines followed by accidental puncturing of the thoracic area might result in extreme stress with increased morbidity and mortality rates of flocks (Birhane & Fesseha., 2020).

Table 34: Live attenuated and inactivated/killed vaccines to combat IB in poultry

Vaccine type	**Administration route**	**Characteristics**	**Efficacy**	**Side effects and limitations**	**References**
Live attenuation vaccine	-Oral administration -Nasal inhalants (intratracheal route) -Subdermal injections -Ophthalmic administration (eye drops) -*In ovo* route (embryonic vaccination)	-Serial attenuation of infectious strains in embryonated eggs; diminishing virulency -Initial IBV immunization -Recommended for endemic areas -Immune responses: 1)Systemic immunity 2)Local immunity	-Multivalent vaccines (IBV H120 & H50) elicit cross-protection combating a broad spectrum of IBV serotypes -Live vaccine (IB 88) combined with a mass-type booster dose with H120; provides immunity -Booster dose: Recommended at day:8-10	-Tissue damage Recombinant & mutation effects -Risk of secondary bacterial infections (airsacculitis) -Adverse side effects in day-old chick (DOC) -Time consuming process	(Bhuiyan et al., 2021) (Cavanagh & Naqi., 2003) (Sultan et al., 2019)
Inactivated/killed vaccine	-Subcutaneous administration -Intramuscular injections	-Infectious strains are killed or inactivated chemically or thermally -Infected allantoic fluid of embryonated eggs generally	-High potential for multivalent formulations / combinations targeting a broad spectrum of IBV serotypes	-Cost-effective -Side reactions: fever, breast tenderness, injection site irritation -Diminished	(Javadov et al., 2019)

		serves as the basis of such vaccine production (oil emulsion-based vaccines) -Immune responses: Humoral immunity, along with narrow-range mucosal protection	-Booster dose: Recommended to be administered at 6-7 weeks of chickens' age	mucosal IgA secretion -Minor local immunity & Ab-mediated immunity	

- **Recombinant vaccines:**

  Recombinant vaccines include nucleic acid-based vaccines, viral vector-based vaccines, and peptide-based vaccines. Viral vector-based vaccines elicit enhanced immune responses in chickens even when the maternal immunoglobulins are involved in neutralizing conventional vaccines (Ravikumar et al., 2022). Furthermore, they also help deal with issues related to viral genomic mutations, such as RNA mutations in the case of IBV, and provide robust protection against infectious strains of IBV. To produce the perfect vaccine, accurate protein folding is vital. These vaccines safeguard ~95% of chickens from N1/62 IBV serotypes (Zegpi., 2019). Genetics and immune responses of the host, along with appropriate vaccine application and combination, are essential while formulating a vaccine.

  Viral peptides are generally used to construct subunit or peptide-based vaccines. Viral spike proteins, together with nucleocapsid protein epitopes, are used to construct novel IBV vaccine. These types of vaccines, generated from multiple viral epitopes, elicit cell-mediated as well as humoral immunity in the host cell, thus providing >80% of immunity from IBV in chickens (Yang et al., 2009).

  Furthermore, the backbone of the plasmid DNA vaccine is DNA that contains the antigenic principle and later undergoes transcription as well as translation inside the host cell. There are two ways to deliver plasmid DNA vaccines. It can either be monovalent, requiring a single promotor for a single antigen, or multivalent, requiring one or >1 promotor for single or multiple antigens. DNA plasmid encoding IBV nucleocapsid protein, membrane protein, and spikes protein (S1 protein) shows effective and significant results in chickens. To enhance the efficacy of vaccination, administration of plasmid DNA vaccine (multivalent; double dose) with a booster dose of killer vaccine (1x) shows protective efficiency against IBV (Yan et al., 2013).

Table 35: Types of recombinant vaccines to combat IB in poultry

Recombinant vaccines	Administration route	Characteristics	Efficacy	Side effects limitations	References
Nucleic acid-based vaccine (DNA based vaccines)	-Administration directly in embryonated egg (*In Ovo*) -Nasal sprays -Oral administration	-Contains nucleocapsid as well as spikes protein generally, S1 protein -Triggers immune responses in chickens by producing viral proteins inside the host cell	-Robust immune responses with the potential to provide cross-protection against IBV strains (i.e.M41)	-Antibodies against viral genome may be produced Post-translational modifications may alter the sequence of proteins	(Bande et al., 2020)
Viral vector-based vaccine	-*In Ovo* route of administration	-Modified vector (non-infectious virus) stimulates immune responses by expressing viral genes - Elicits long-term enhanced protection against IBV -Demonstrates positive outcomes in mitigating complications from viral RNA mutations	-Safeguard ~95% flocks from N1/62 IBV serotypes -illustrates effectiveness even with a single dose -cost-effective -Cross-protection efficacy	-Protein (epitopes) arrangement may alter, affecting vaccine efficacy	(Bhuiyan et al., 2021)

Subunit protein vaccine	-Intra-ocular administration -nasal sprays -intramuscular administration	-Elicits immune responses in host: 1)Humoral immunity 2)Mucosal immunity 3)Cross-protection immunity 4)Cellular immunity	-Shows maximal multi-cross-protection (broad spectrum of immunity)	-Administration of repeated vaccine doses is required -Systemic side reactions include fatigue and fever with local reactions at the injection site -Restriction to protein-based antigens	(Bande et al., 2015)

26.5.2 Nanotechnology as a therapeutic strategy:

Nanotechnology is one of the advanced and intensive techniques, possessing a broad application spectrum due to its extraordinary utilization potential, used to correctly diagnose and control the disease burden of infectious outbreaks in many fields, including the global poultry industry. The word ‘nano’ is derived from ‘nanus’ (a Latin word), meaning a minute/tiny unit ~10-9m (one billionth) or 1nm (Yadav et al., 2021). Nowadays, a diverse variety of nanoparticles are being used. Some of them include Quil A and chitosan-based nanoscale adjuvant complexes (less than o.1μm), G-Ag nanocomposites, polymeric carbonized nano-gels, and self-assembled protein nanoparticles (Chandrasekar et al., 2020). All of them show virostatic natures and improve host immune responses.

During anaerobic conditions following high temperatures, lysine hydrochloride pyrolysis occurs, and carbonized nano-gels are manufactured. Even its small concentration of about 30 μg/ml elicits favorable outcomes in providing immunity and lessening viral spread by hindering the interaction between host cells and viral spikes glycoprotein (S1 and S2 proteins). Furthermore, self-assembled protein nanoparticles facilitate the immunogen (viral antigen) presentation of spikes protein to the host’s defense system in the best possible way, employing flagellin’s adjuvant properties to amplify antibody responses or enhance immune responses (Abbas et al., 2022).

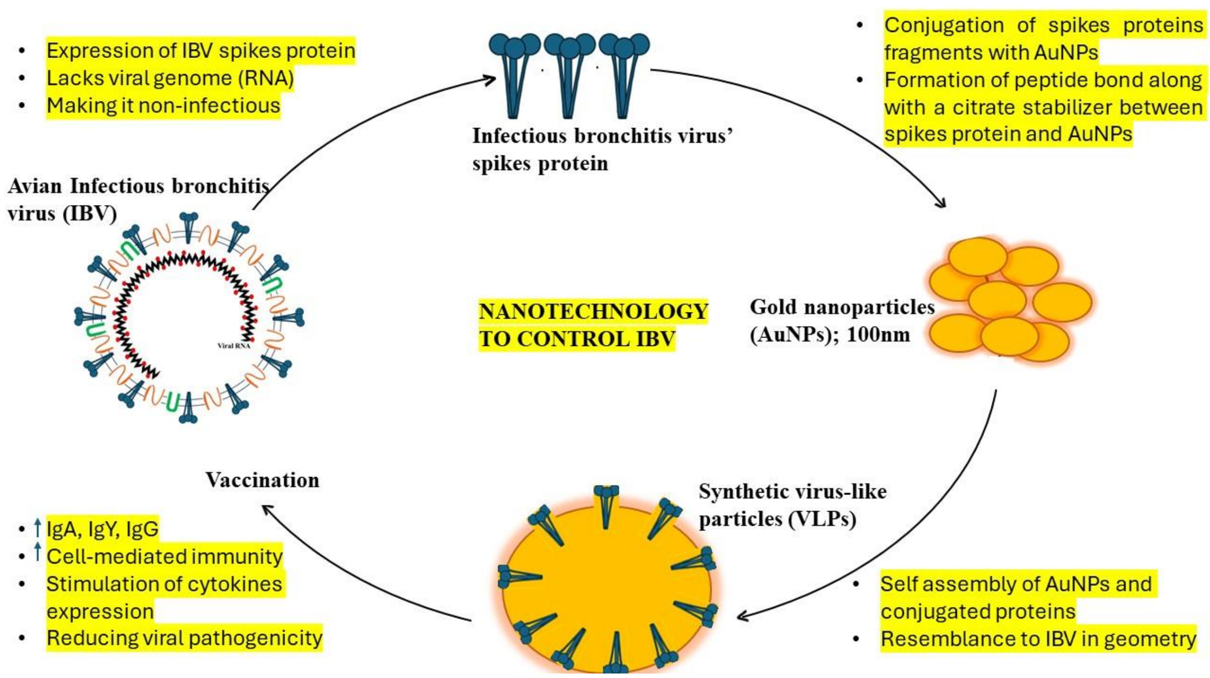

Figure 81: Nanotechnology as a control measure for infectious bronchitis disease in poultry.

For example, a DNA vaccine (monovalent and bivalent) that encodes viral spikes protein (S1) encapsulated within a nanoparticle (chitosan-saponin nanoparticle) helps to provide enhanced immunity against homologous viral challenge (Bande et al., 2020).

26.6 Treatment and environmental monitoring:

No specific treatment is available for infectious bronchitis virus, but some precautionary measures help to reduce the disease burden of IBV in poultry. These measures include preventing overcrowding of flocks, maintaining sufficient feed consumption, and supplying heat to avoid cold stress (Jackwood & de Wit., 2013). Antibiotics may be employed to lessen the mortality rate in infected chickens resulting from secondary bacterial infections such as airsacculitis. Moreover, electrolyte replacement and maintaining sufficient hydration status of the body also serve as effective therapeutical measures. 72 mEq of potassium and sodium with 1-3rd bicarbonate salt form is recommended for treatment (Rana et al., 2021).

Several environmental factors influence the spread of IBV in chickens. By maintenance of proper environmental monitoring strategies, disease burden can be controlled. The following are the major environmental monitoring strategies for the limitation of IBV spread:

- Appropriate bird density
- Biosecurity protocols
- Hygiene measures (disinfection techniques)
- Good air quality
- Vaccination programs (such as H120-vaccinated birds exhibits lessen viral transmission)
- Appropriate management of physical and chemical environmental conditions (temperature, pH, humidity) (Islam et al., 2013)

26.7 Future Prospectives:

Research on DNA, subunit, and vector vaccines for infectious bronchitis virus integrating the spikes protein (S1 glycoprotein gene) as well as reverse genetics processes to strengthen immunogenicity have been performed. Recombinant or vector-based vaccines are significant in representing >1 antigen type to the host, thus providing broad-spectrum protection from different strains/serotypes of infectious bronchitis virus. These combinations aid in accomplishing the multivalent immunization procedures. Another vaccine type contributing towards lessening IB pathogenicity and providing host immunity is the DNA vaccine that is administered in the growing embryonated egg (*In Ovo;* route of delivery) or to the live chickens. New and advanced diagnosis and management techniques, along with therapeutic measures, help us combat these pathogenic microbial diseases.

26.8 Conclusion:

After decades of comprehensive and detailed research on avian coronavirus (IBV), along with the multiple investigations related to IBV vaccine efficacy, it remains a center of attention for poultry researchers due to its virulent nature. The infectious bronchitis virus is associated with the occurrence of multi-systemic infections in chickens, primarily affecting their respiratory and reproductive systems. Multivalent vaccines/multivalent combinations enable broader protection in poultry chicken even with few doses of vaccines, thus aiding in attaining cross-protection against heterologous IBV challenges. Live and inactivated vaccines play a pivotal role in poultry immunization. Live vaccines are paramount for inducing mucosal and cellular immunity and safeguarding commercial broilers and layers. On the other hand, inactivated immunization plays a significant role in the induction of robust antibody-medicated immunity (humoral responses) that is integral for the establishment of maternal immunization in poultry (aid to permit protective maternal immunoglobulins to progeny).

The emergence of new variants of IBV pressures healthcare societies and poultry producers to evaluate their vaccination plans and the generation of modern vaccines. Furthermore, a prevalent tool aiding modern drug delivery techniques, such as the administration of AuNPs (gold nanoparticles) for effective IBV vaccine delivery, enhances the host's immune responses effectively. Accurately diagnosing or detecting illnesses in infected birds is necessary for the timely prevention of the disease through the application of appropriate control and management protocols. Ultimately, these findings illustrate the economic importance, detection procedures, and modern knowledge of IB with the utilization of advanced multivalent combination programs and recombinant vaccines to deal with IBV encounters.

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Chapter 27 : Non Conventional feeding regimens: The future of Livestock

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Abstract

Knowing the challenges faced by traditional livestock feeding needed a multifaceted method. Land management, efficient Water usage, and adoption of alternative feeding crop resources are sustainable and can also help environmental impact mitigate. Effective measures and modern practices are essential to mitigate the risks of disease transmission in traditional livestock feeding. Implementing proper sanitation and handling procedures can significantly improve livestock health and productivity. Finally, for the guaranteed safe and consistent supply of nutrients through better feed management which prevents diseases and promotes the health and production of livestock feeding. The sectors of livestock can move towards a more and more sustainable and economically viable future through the addressed challenges.

To cut costs associated with standard feed, cost-efficient non-conventional feeding methods investigate alternate sources like insects, algae, or by-products. These techniques provide sustainable solutions to the cattle and aquaculture sectors by optimizing nutritional value while reducing production costs and environmental effects. Using unconventional sources, such as insects or byproducts, to increase nutritional intake while cutting expenses is known as non-conventional feeding. Non-conventional refers to traditionally valuable livestock feeding practices that are economically valuable. Farmers who embrace innovation and strategies see improvements in the productivity and health of their animals. For environmental protection, animals of various sizes, from huge ones like cows and buffaloes to little ones like goats, etc.

Beyond Traditions: Non-conventional Feeding Regimens, the Future of Livestock Feeding

27.1 Introduction

27.1.1 Overview of traditional livestock feeding practices

The growing amount of livestock waste has created significant issues for the management of the growing demand for animal protein products. Selecting the right treatment technology is still a challenging undertaking, and there is still much dispute on this topic. The livestock industry is expanding very quickly, but overall, the trend indicates a major uptick in reaction to the quickly rising demand for animal products, particularly in developing nations. Huge amounts of manure have been produced concurrently with the increase in husbandry production. Even while manure is a useful soil supplement and provides nutrients for crop growth, misuse and excess of it could hurt the environment (Khoshnevisan, Duan, Tsapekos, Awasthi, & Liu, 2021).

27.1.2 Explanation of the need for innovation in feeding regimens

Concerns over the contribution of livestock agriculture to greenhouse gas emissions have arisen as a result of the expanding global human population and the rising demand for meat and dairy products. For many people, especially in developing nations, ruminant cattle nevertheless provide nutrient-rich food despite these worries. Additionally, they produce human-edible protein from plants that people cannot digest, giving food to places that are unsuited for farming because of poor soil or unfavorable weather. The task at hand is to maximize ruminant feed efficiency while reducing its negative environmental impact. Since the beginning of time, various feeds and animal methods for feeding have been developed. With the right advances in technology, ruminant livestock may still be a major component of environmentally viable agricultural systems (J.M. Moorbya, 2021).

27.2 Current Challenges in Traditional Livestock Feeding

With the practice of traditional livestock feeding global warming, the environment and the industry of livestock face many more challenges and also face the impact on the environment and livestock industries. Through these a vital range of animals large and small, every type of animal is challenged by the effects (Patel, Kendra, & Prajapati, 2020).

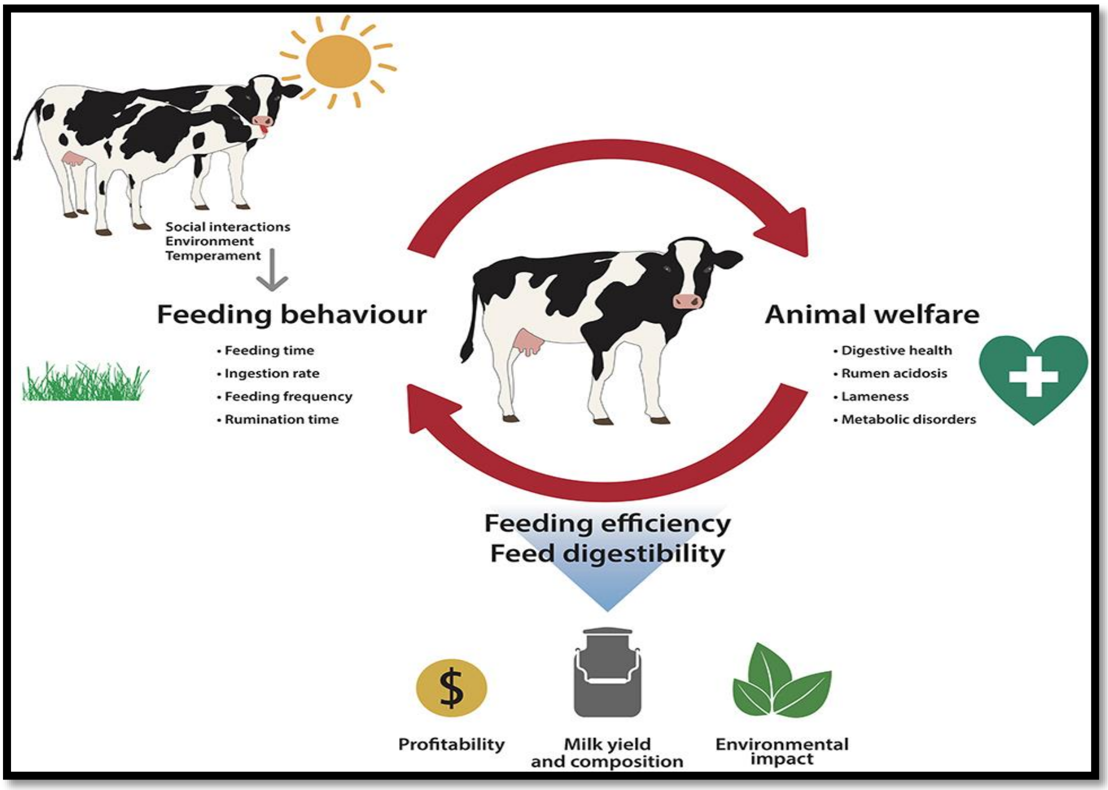

Figure 82 Feeding Circle

27.2.1 Environmental impact

- **Land use and deforestation**

  It is one of the primary environmental problems that is related to livestock feeding traditionally in the usage of land. Land use and deforestation are significant environmental concerns associated with traditional livestock feeding practices. Additionally, the cultivation of feed crops like soy and maize further exacerbates deforestation, impacting soil health and contributing to environmental degradation. Addressing these issues requires sustainable land management practices to minimize the ecological footprint of livestock farming. Due to deforestation not only loss of biodiversity is faced by the people but also interrupted in the soil erosion contribution and the ecosystems. However, A large land is needed for the cultivation of feeding crops like wheat, maize, and soybean. Which can further the problems of land usage and deforestation (Müller, 2022).

- **Water consumption**

  In the demands of livestock feeding another demand is significant Water resources. Highly water intensive is used for the production of feeding crops. Such as 15000-litre Water is required for the production of 1 kg of beef. If we focus on the consumption of water used for the growth of feeds and the consumption of water by the animals this problem immensely consumes water placing a large stain on many areas, especially on local areas and in particularly in arid regions where the scarcity of water is common. In addition to the overall Water footprint of livestock feeding significance of water resources is required (Frédéric, et al., 2022).

- **Greenhouse gas emissions**

  Large animals like buffaloes and cow livestock significantly contribute to the emissions of greenhouse gases like methane, and during the digestive process of larger animals, a potent greenhouse gas is released. Additionally, the highly global warming potential gasses like methane and nitrous oxide both are related to the manure production of livestock feeding.

27.2.2 Economic challenges

- **Rising feed costs**

  The production of livestock economy faces a major challenge of the cost of feeding. Due to several factors, the cost of feeds is rising day by day. Which main factors are increasing demands for grain, adverse weather conditions so crops are affected, and biofuel production competition for land. All these problems like rising cost prices it makes difficult for small-scale farmers to sustain their operations and remain profitable. Poultry animals are mainly based on grain-feeding (Gerald, 2020).

- **Market volatility**

  In the market, livestock is inherently volatile, due to the factors of disease outbreaks, changes in consumer preferences, and international trade policies. The challenges faced by poultry farms in production are significant, including disease management and fluctuating feed costs. Disease outbreaks can devastate flocks, while high feed prices strain budgets highly (Peter & Blaustein-Rejto, 2021).

27.2.3 Nutritional issues

- **Inconsistent nutrient supply**

  For the productivity of livestock and the health continuous supply of nutrients is essential. However, traditional feeding practices often lead to Inconsistent Nutrients Supply. For example, nutrient intake by animals like buffaloes and goats is affected and the quality of pastures is very seasonal. The nutrient content of feed crops can fluctuate which is based on the crop's growth.

- **Disease transmission**

The diseases spread through traditional livestock feeding practices. Introducing the pathogens into the livestock population is due to water sources and tainted feeding. Disease transmission in traditional livestock feeding can be significant due to close animal proximity and shared feed sources, which facilitate the spread of pathogens. Moreover, in the farmer's practices, the animals are mainly kept close, and the repaired spread of diseases is facilitated. The diseases among cows and buffaloes are the diseases of mouth and foot in large animals, while in poultry farms avian influenza is spreading (Laura, et al., 2020).

27.3 Overview of Non-conventional Feeding Regimens

27.3.1 Definition and types of non-conventional feeding

Non -non-conventional feeding regimens refer to the substitute to the strategies of feeding that divergence traditional pasture-based and grain-based methods. By utilizing the diverse unconventional resources for feeding these routines include insect-based proteins, novel feeding components, industrial wasting, and agriculture by-products.

- **Agriculture By-products:** Efficient management of agricultural by-products maximum is crucial for the waste and maximizing the resources utilized in sustainable agriculture practices.
- **Industrial waste**: Industrial waste is the byproduct of manufacturing processes that can include chemicals, solvents, and other harmful materials. The organic waste of industrial products can easily and safely be utilized the livestock feeding.
- **Insect-based proteins:** Larvae and mealworm Insect-based which are rich in protein. These Insects are black like soldier flies and they are also can cultivate organic waste. These Insects are necessary for the feeding of fish, and poultry and also for large animals like goats etc.
- **Hydroponics and Aquaponic system**: In these systems all the plants that are growing without soil, often nutrient-rich water is used those plants are involved. Fresh fodder livestock can be used for the production of these plants.
- **Algae and seaweed:** All those plants that can grow in the water means aquatic plants are highly Rich with nutrients and can be responsible for cultivation. The continued increase of these is being used in the farming of fish and other animals like cows and poultry (Hartog & Sijtsma, 2013).

27.3.2 Advantages over traditional methods

For the non-conventional many advantages are used over traditional methods for the offer of routines (Hrishitva, Samad, Hamza, Muazzam, & Khoiruddin, 2022).

- **Environmental Benefits:** industrial wasting and agriculture by-products are utilized for the water resources and land; Famer has minimal environmental footprints which can help in the organic waste recycling in which Insect-based proteins and algae are included (Susan, 2017).
- **Cost Efficient:** By utilizing the locally available wasted material and the by-products, the farmers can reduce the cost of feeding and can make it economically available and

a small scale of these can particularly produce. These methods aim to optimize nutritional value while minimizing environmental impact and production expenses, offering sustainable solutions for livestock and aquaculture industries.

- **Nutrition value:** In the essential nutrients rich ingredients are fed by the many non-confrontational. So, the algae used is provided with important fatty acids and vitamins.
- **Disease management:** Some feeds used in non-conventional feeding have antimicrobial properties that can help in the reduction of disease in livestock.

27.4 Non-conventional Feeding Techniques and Approaches

27.4.1 Insect-based Feeds

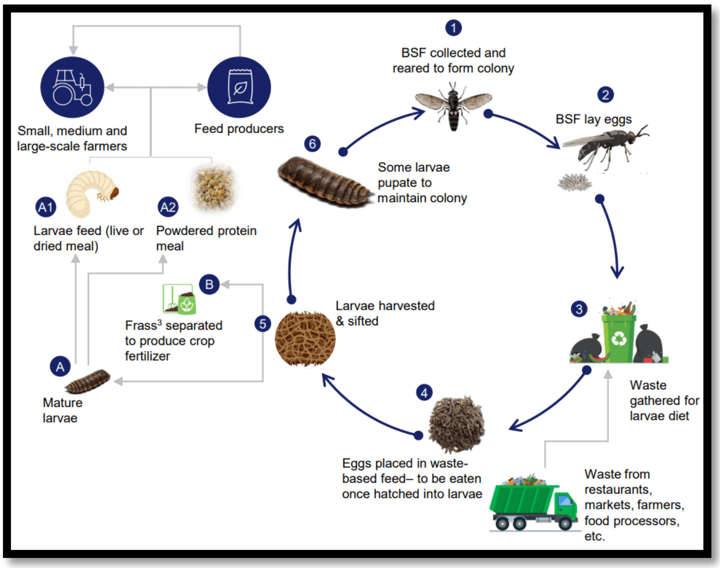

Figure 83 Insect-based Feeds

27.4.2 Overview of Insect Farming for Animal Feed:

Insect farming is very useful for the growth of the animals which includes raising significant insects like black soldier flies, mealworms, and crickets. Because of its efficiency and sustainability, it become a very popular technique. It is possible to cultivate the Insects with organic waste, converting it into high-protein biomass, that becomes beneficial as well as suitable for animal consumption.

27.4.3 Nutritional Benefits of Insect-based Feeds:

Insect-based feeds are rich in amino acids, and high levels of fats which are the main components of protein diets. Along with protein, there are also calcium and phosphorus, found in this type of feed and thus is very healthy for animal growth as compared to traditional feed ingredients like soy or fishmeal feed.

27.4.4 Examples of Successful Implementation in Livestock Production:

Insect-based feeds have been successfully implemented into the food of poultry and fish for their better growth and development. Most importantly black soldier fly larvae are used in poultry farms in Europe and the United States at large scale. This way of feeding is leading to increased growth rates and feed conversion ratios, where it serves as a sustainable alternative to fishmeal.

27.5 Plant-based Feeds

27.5.1 Discussion of Novel Plant Sources for Animal Feed

Novel plant sources highly including algae and hemp are becoming viable and perfect options for livestock feed. Microalgae like spirulina and chlorella are the main components of the Algae and are too much rich in high protein and lipids. Hemp contains protein and omega-3 fatty acids.

27.5.2 Nutritional Benefits and Advantages Over Traditional Plant-based Feeds:

Algae-based feeds offer high levels of protein, essential fatty acids, vitamins, and antioxidants. They enhance the immune system of livestock. Hemp seeds and meal provide a complete amino acid profile to animals and they get much-needed energy from this feeding.

27.5.3 Case Studies of Successful Implementation:

In Denmark, algae-based feeds are highly used in dairy farms to increase the productivity of milk and reproduction of the livestock. Hemp meal is used in poultry diets in Canada, resulting in healthier birds with better weight gain. Moreover, it has a very low cost (Comerford, Miller, Kapsak, & Brown, 2021).

27.6 Single-cell Protein Feeds

27.6.1 Explanation of Single-cell Protein Production

Single-cell proteins (SCP) is also a great level of feed that is derived from microorganisms. Microorganisms like bacteria, yeast, and fungi are the source of the SCP. These microorganisms are cultivated in controlled environments, often using waste materials. Furthermore, it can be produced by the method of by-products as substrates, which results in biomass being harvested at a great level for feeding the livestock.

27.6.2 Nutritional Benefits and Advantages of Traditional Protein Sources:

Single-cell proteins provide high protein content, amino acids, and vitamins and gaining popularity among farms and diaries. This is because these methods of feeding are highly digestible. Unlike traditional protein sources like soy or fishmeal, SCP production is more productive and useful.

27.6.3 Examples of Successful Implementation in Livestock Production:

Yeast-based feeds are widely used in poultry production in Europe, enhancing growth performance. Bacterial protein meals prove to very effective way in aquaculture, by offering a sustainable alternative to fishmeal or soy.

27.7 Food Waste Recycling

27.7.1 Types of Food Waste Suitable for Livestock:

Food waste includes fruit and vegetable scraps, bakery waste, brewer's grains, and dairy by-products. Food wastes are rich in nutrients and can be repurposed to feed livestock that can provide energy to them.

27.7.2 Processing Methods:

Food waste must be processed to ensure safety and nutritional adequacy mainly by popular methods of anaerobic digestion, ensiling, and dehydration. These processes reduce the risk of pathogen contamination in the waste food. Processing methods increase the life span and digestibility of the waste.

27.7.3 Environmental and Economic Impacts:

There are many benefits of recycling food waste into animal feed reducing landfill waste and eventually diminishing the carbon footprint. Farmers enhance livestock production more sustainable and economically viable. The Netherlands and Japan have demonstrated significant environmental and economic benefits from food waste recycling.

27.8 Lab-grown and Synthetic Feeds

27.8.1 Development and Production:

Lab-grown and synthetic feed are biomass that is produced in controlled environments. Animal cells, synthetic amino acids, and vitamins all combinedly make lab-grown and synthetic feeds. The production process meets the nutritional needs of livestock.

27.8.2 Nutritional Considerations:

Lab-grown feeds are designed to maintain nutrient profiles, including proteins, fats, carbohydrates, and vitamins to be tailored to livestock requirements, improving health.

27.8.3 Prospects and Scalability:

Lab-gr own and synthetic feeds have the ability for scalability. Moreover, innovation in biotechnology and bioprocessing can reduce costs that are reported much less than the traditional ways of feeding. Pilot projects in the U.S. and European countries are exploring the feasibility of expanding the production revealing a transformative future for livestock feeding.

27.9 Alternative Feed Sources

A. Discussion of Unconventional Feed Sources

Unconventional feeding by food waste and biofuel co-products is earning much fame and is praised by farmers as it provides sustainable and cost-effective nutrition for livestock. Food waste involves the material fruit and vegetable crumbs, bakery, and dairy items waste. On the other hand, biofuel co-products include distillers' grains and glycerol components. These resources provide an alternative to traditional feed ingredients (Sekhar, Neeradi, & Mahender, 2017).

B. Nutritional Benefits and Challenges of Implementation

Nutritional Benefits:

Unconventional feed sources are very rich in vitamins, minerals, and energy, while biofuel co-products like distillers' grains con high levels of protein and fibre. Both resources can enhance the nutrient profile of livestock diets, supporting health, and productivity.

Challenges of Implementation:

However, identified issues and challenges should be taken into account for the proper dealing that will result in successful implementation. Food waste must be processed to ensure safety and consistency, eliminating risks of contamination and pollution by dumping waste material. Challenges like in collection; and transportation are also reduced by these processes and environmental issues are also decreased. Therefore, careful formulation and monitoring to meet the nutritional needs of growing livestock.

C. Case Studies of Successful Use in Livestock Production

Case Study 1:

Food Waste in Swine Diets:

In South Korea, swine producers have successfully integrated food waste into pig diets at very low costs. The government has supported this initiative with strict regulations to ensure food safety and nutritional adequacy for the animals on their farms.

Case Study 2:

Distillers' Grains in Dairy Production:

In the United States, distillers' grains, a by-product of ethanol production are very popular and are used in dairy animal food. The farmers have gained very good quality milk production and composition with very little expense. Dairy farms have developed partnerships with biofuel producers to provide high-protein feed ingredients.

These case studies justify that unconventional feed sources improve livestock production while addressing severe sustainability and economic issues. With proper management and regulation, these alternative feeds will lead to the future of livestock production (Tasneem & Abbasi, 2016).

27.10 Technological Innovations in Livestock and Poultry Feeding

Precision Feeding and Smart Farming

Precision feeding and smart farming are very essential techniques. These methods can be understood by the following points such as:

1. IoT and Sensor Technologies:

Precision feeding the use of the Internet of Things (IoT) and sensor technologies to optimize feed delivery to livestock and poultry farms to broaden the production level. Sensors monitor significant elements in the production of the animals like animal weight, feed consumption, and the environment of the areas where these are kept (den & Sijtsma, 2011).

2. Data Analytics and AI in Feed Management

Data analytics and artificial intelligence (AI) evaluate and interpret gathered data and information by IoT devices. AI algorithms analyze this data to forecast the nutritional requirements of livestock and detect early signs of diseases. Machine learning reveals growth rates and adjusts feed compositions to ensure animals' demands for nutrients for their growth and health (Neila, Averós, & Estevez, 2016).

B. Fermentation Technology

1. Fermented Feeds:

Fermentation technology various types of is utilized for producing fermented feeds. The fermentation food is produced by ingredients like silage, fermented soybeans, and probiotics to make high-quality feeds. Similarly, silage is also another technique of fermentation feed by using corn and grass and further storing in anaerobic conditions.

2. Benefits for Livestock Health:

There are many benefits of fermentation feed for the health of livestock. Fermentation feed breaks down the structure of carbohydrates into very simple elements. Fermentation nutrients help in digestion through better nutrient absorption that boosts the immune system (Yonggan, Zhang, Law, & Yang, 2024).

C. Genetic Engineering and Feed Additives

1. Genetically Modified Organisms (GMOs) in Feed:

GMOs are used to enhance the nutritional value of the feed process. GMGs also efficiency of animals' health. Bt corn and Roundup Ready soybeans are the main components of this method. Moreover, it is resistant to pests and illness (Goetz, 2009).

2. Probiotics and Enzymes:

Probiotics and enzymes absorb the feed nutrients. Probiotics are live beneficial bacteria that promote a healthy gut. Moreover, the microbiome strengthens the immune system of the livestock. Both phytase and protease are the kinds of enzymes that break down feed components such as proteins, to bioavailable them. These improve health and feed efficiency (Dajić, Bošnjak-Neumüller, Pajić-Lijaković, Raj, & Vasiljević, 2018).

3. Case Studies of Technological Innovations

Case Study 1:

Precision Feeding in Poultry Farms:

In the Netherlands, poultry farms have adopted precision feeding systems using IoT and AI. Sensors placed in feeding stations collect data on individual birds' weight and feed intake. AI algorithms analyze this data to adjust feed delivery in real-time, ensuring each bird receives the optimal amount of nutrients. This approach has led to significant improvements in feed conversion ratios and overall flock health. In this way, the farmers have to spend money on the production of their livestock.

Case Study 2:

Fermentation Technology in Dairy Farms:

Another case study is taken in Germany, where dairy farms have integrated fermentation technology to yield the best quality silage from corn and grass production. The use of fermented feeds has improved milk quality and production. It has also increased the health of dairy cows and buffaloes. The fermentation process has eventually lowered the demand for synthetic additives and increased the natural methods.

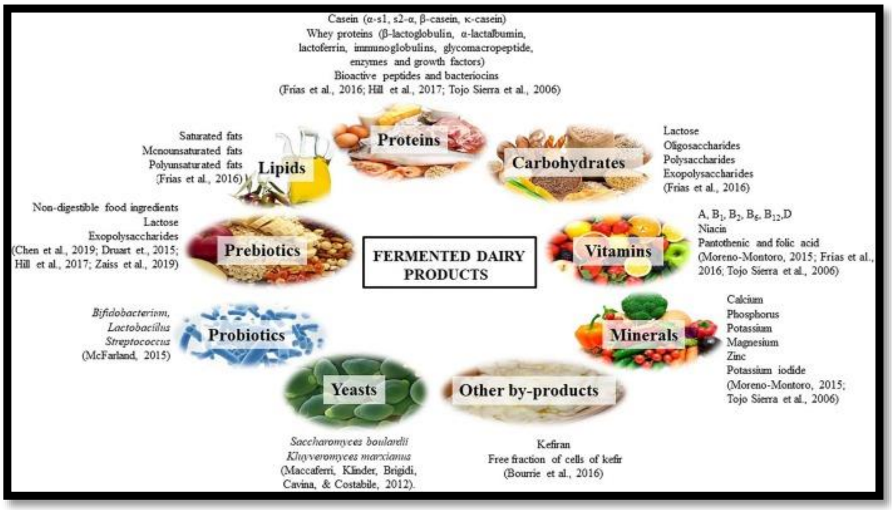

Figure 84 Fermentation Technology

Case Study 3:

GMOs and Feed Additives in Swine Production:

In the United States, in the farms of swine, producers are using GMO corn and soybeans combined with probiotics and enzymes. As a result, the producers gain effective and productive. GM crops are rich in nutrients that make the feed a qualitative resource. On the other hand, the additives improve digestion by strengthening the immune system of the animals and nutrient absorption. This combination has resulted in better growth rates, and improved feed efficiency which helps in expanding the pig's production. These techniques also play a crucial role in lowering environmental impact due to lower nutrient waste (Dariusz, et al., 2013).

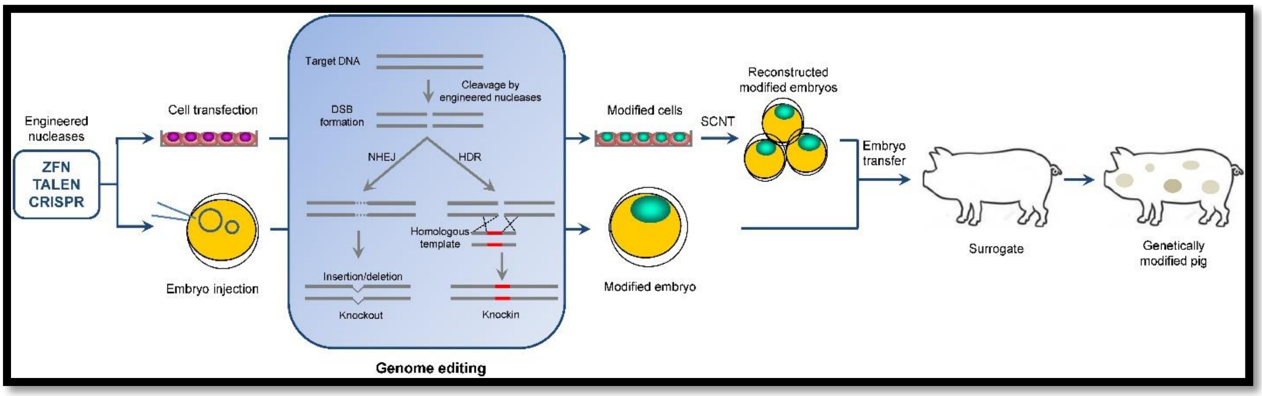

Figure 85 GMOs

So, the technological innovations in the procedures and methods of livestock and poultry farm feeding, like precision feeding, fermentation technology, and genetic engineering, are transforming the livestock business. The fermentation feeding also provided benefits such as improved feed efficiency, and better animal health. Further, diminishing the carbon footprint is a very important advantage for societies. Farmers can achieve more sustainable and profitable livestock production with the utilization of innovative techniques.

27.10 Regulatory and Policy Considerations

A. Current Regulations on Non-conventional Feeds

Current regulations on non-conventional feeds depend on various stages like there are different regions and types of feed. For example, in the European Union, insect-based feeds for aquaculture are being used on a large scale. However, this method in other livestock like goats, cows, and buffaloes is still under review and needs further research and development to provide clear-cut results. Similarly, in the United States, the Food and Drug Administration is more popular and is immensely used for food waste and by-products. In both methods, they are processed to be preserved for animal feed and production. Regulations and policies greatly emphasize saving feed, preventing contamination, and maintaining nutrient quality in the feeding of livestock.

B. Challenges in Regulatory Approval

The regulatory approval process for non-conventional feeds has multiple issues and challenges that hinder their implementation as well. At the top of the discussion, the main threat is reported that the security of the data is not ensured and that is why it is difficult for regulatory bodies to assess identified risks. This is because the previous regulations are mostly suited to traditional feed ingredients, which are hard to classify and approve innovative animal feeding ways and techniques. This can lead to lengthy approval processes. Moreover, the more the approval will take time the more costs for producers will be increased. Furthermore, it is also noticed that along with the approval of regulation, resistance from stakeholders is also being observed, who are wary of changing behavior, practices, and standards.

C. Policy Recommendations for Fostering Innovation

To promote innovation in non-conventional animal feed ways, several policies are suggested:

1. **Streamline Regulatory Processes**: Streamline regulatory, processes are very important that simplify and expedite the approval process for new feed ingredients. The governmental department takes too much time to finalize the process. These instructions and guidelines will create common understandings for non-conventional feeds.

2. **Encourage Research and Development**: Moreover, the researchers and the stockholders must invest their time to provide funding and incentives for research into safety and security. These will also encourage to development and efficacy of non-conventional feeds and enhance the productivity of the farms.

3. **Promote Collaboration**: similarly promoting collaboration and effective communication between regulatory bodies, researchers, and dairy and farm stakeholders to ensure an approach to feed with innovative and new techniques.

4. **Develop Standards**: Establishing clear standards and guidelines for the production is very crucial and therefore, non-conventional feeds are surely safer and provide quality of the feeding process. These standards will also make more effective outcomes.

D. Specific Examples and Applications

1. Insect-based Feeds

- **Example**: Black soldier fly larvae meal
- **Potential Benefits**: High protein content, amino acids, sustainable production.
- **Limitations**: Regulatory hurdles, and scaling production of the animals. These limitations lower the trust of the farmers and researchers to bring immediate results.
- **Real-world Example**: The insect-based feed is embarrassed in the Netherlands. Furthermore, black soldier fly larvae are used in poultry diets, which improve growth rates and feed efficiency of the livestock including animals.

2. Plant-based Feeds

- **Example**: Algae-based feeds.
- **Potential Benefits**: Rich in protein, essential fatty acids, and micronutrients; sustainable and can be grown on non-arable land.
- **Limitations**: High production costs and limited large-scale availability.

- **Real-world Example**: Danish dairy farms use algae-based feeds to improve milk quality and yield.

Figure 86 Plant Based Food

3. Single-cell Protein Feeds

- **Example**: Yeast-based feeds.
- **Potential Benefits**: High protein content, consistent quality, and sustainable production.
- **Limitations**: Cost of production and consumer acceptance.
- **Real-world Example**: European poultry farms use yeast-based feeds to enhance growth performance and feed efficiency.

4. Food Waste Recycling

- **Example**: Brewer's grains.
- **Potential Benefits**: Reduces waste, is cost-effective, and is rich in nutrients.
- **Limitations**: Variability in nutrient content and potential contamination risks.
- **Real-world Example**: South Korean swine farms incorporate food waste, leading to reduced feed costs and improved weight gain.

5. Lab-grown and Synthetic Feeds

- **Example**: Cultured meat cells used as feed.
- **Potential Benefits**: Controlled nutrient profile, sustainable, and reduces reliance on traditional agriculture.
- **Limitations**: High production costs and technological barriers.
- **Real-world Example**: Pilot projects in the U.S. exploring the use of cultured cells in livestock diets.

It can be difficult to navigate the regulatory environment for non-conventional feeds, but existing policy suggests collaboration to promote advanced technological approaches. Real-world examples of non-conventional feeds in action demonstrate their potential benefits, these benefits can be concluded sustainability of the environment by reducing the carbon quantity, cost-effectiveness, and enhanced animal nutrition like improving digestion system. Moreover, the most important point that is addressing regulatory and practical challenges should be taken into consideration which will be key to integrating these innovative feed sources into mainstream livestock production.

27.11 Nutritional Considerations and Challenges

Importance of Proper Diet Formulation

Proper diet formulation is critical in livestock production to ensure that animals receive the necessary nutrients for growth, health, and productivity. Diets must be tailored to specific needs, considering factors such as age, breed, production stage, and physiological status. For instance, young animals require higher protein levels for growth, while lactating females need more energy and calcium to support milk production (Devendra & Leng, 2011).

A. Potential Challenges of Non-conventional Feed Sources

1. Palatability and Acceptance by Animals: Palatability and the acceptance by animals in dairy and poultry farms is a very serious issue. Animals may be reluctant to eat feeds. Insects or algae might not be as readily accepted by all societies.

2. Nutrient Deficiencies and the Need for Supplementation: Insects are high in protein; they might be deficient in vitamins. Therefore, supplementation keeps a balanced diet containing main dietary elements including vitamins, proteins, and amino acids.

3. Anti-nutritional Factors: It is observed that non-conventional feeds several times interfere with nutrient absorption or digestion of dairy animals and poultry farms.

4. Regulatory Hurdles and Potential Consumer Concerns Regulatory approval for non-conventional feeds can be complex. The late approval lacks safety and ethics of feeding livestock with non-traditional feeds (McAllister, et al., 2020).

B. Strategies for Overcoming These Challenges

1. Feed Processing Techniques to Improve Palatability and Digestibility: Insects can be processed into meals or oil can help in the process of acceptable to animals. Fermentation and extrusion also improve the taste and nutrient availability of dairy and poultry feeds such as algae and food waste (Neethirajan, 2023).

2. Development of Balanced Rations Combining Non-conventional and Conventional Feedstuffs: Non-conventional feeds should be integrated with conventional feedstuffs as their combined result will develop a balanced rations and diet. Like, incorporating insect meals with traditional grains enhances the nutrient profile of poultry.

3. Research on Mitigating Anti-nutritional Factors: Techniques such as fermentation, heat treatment, and the use of enzymes demand a great level of research to reduce the effect of anti-nutritional factors.

4. Addressing Public Concerns Through Education and Transparency: Addressing public issues about the safety, benefits, and environmental impacts will mitigate feed concerns. Certification and labeling are the best ways to ensure feed quality and safety standards are met.

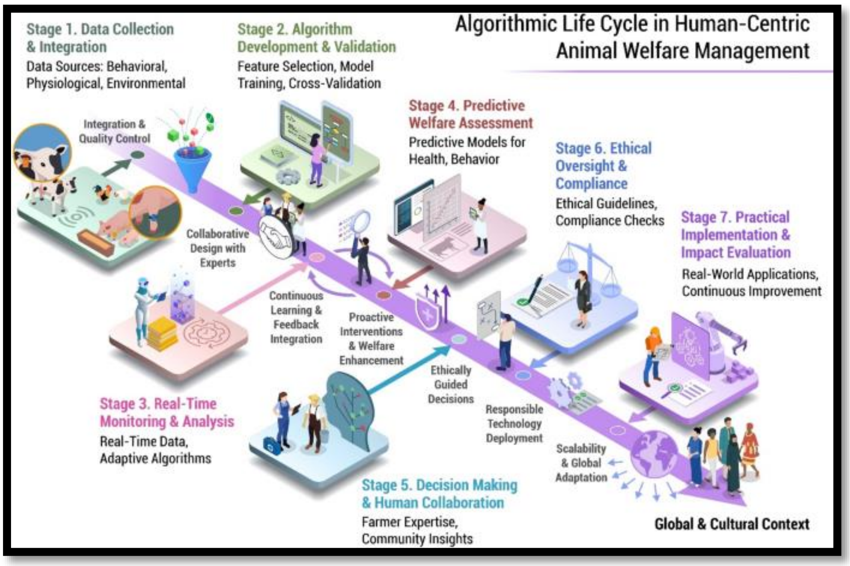

Figure 87 Animal Welfare Management

C. Case Studies of Successful Implementation

1. Insect-based Feeds in Poultry: In the Netherlands, poultry farms entail black soldier fly larvae meal in their feed formulations. Furthermore, it is combined with traditional grains to provide a balanced diet. This integration has led to improved growth rates and feed efficiency to increase the number of poultry animals (Hovi, Sundrum, & Thamsborg, 2003).

2. Algae-based Feeds in Dairy Cattle like Cows and Buffaloes: In Denmark, algae-based feeds are highly used in dairy farms to supplement conventional silages. The algae are processed to enhance the digestibility and sustainability of milk production. This approach has resulted in better milk quality and yield that is much more in demand in the current market.

3. Fermented Food Waste in Swine Production: Similarly, in South Korea, swine producers have incorporated fermented food waste into pig diets to strengthen their immune system and to make digestion within a short time. The fermentation process improves palatability and reduces ANF which enables the dairy and farms' stack holders nutritious and acceptable to pigs. This practice has significantly reduced feed costs and improved weight gain in pigs also increasing their health chances.

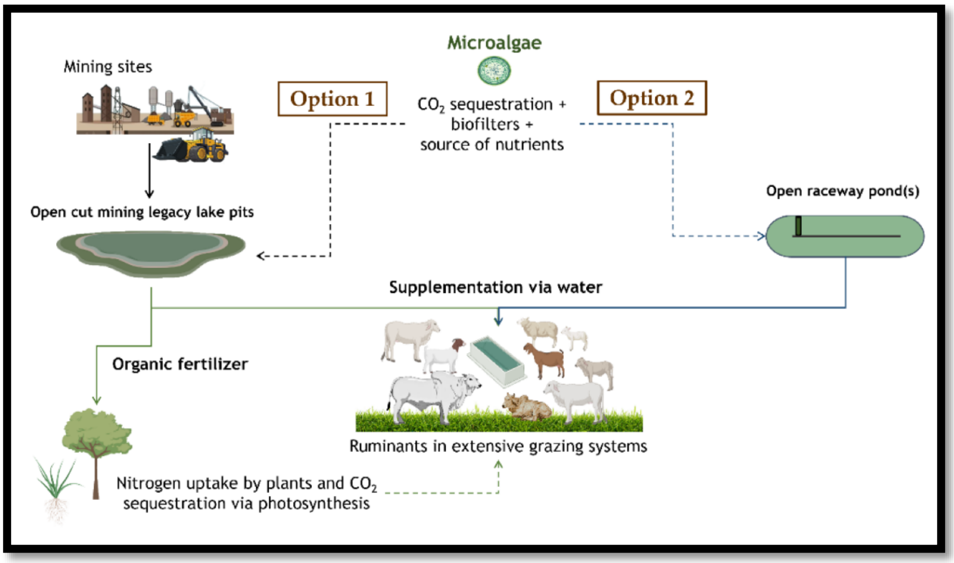

Figure 88 Microalgae

Addressing the nutritional challenges of non-conventional feeds requires an effective and valuable approach like proper diet formulation, processing techniques, and balanced rations. Moreover, there is also a need to account for the consideration of nutritional concerns. By addressing these identified issues and challenges, non-conventional feeds can become a viable and sustainable option in livestock production, for the farmers in the whole world. The adoption of this approach will provide multiple benefits such as nutrition, cost-effectiveness, and environmental impact.

27.12 The Future of Livestock Feeding

Potential long-term benefits of adopting non-conventional feeding regimens:

Water availability and fodder productivity will be significantly impacted by changes in ruminant systems for raising livestock brought about by climate change. While warm-season and tropical grasses are frequently utilized, pasture-based agriculture in temperate climates is dominated by cool-season grasses. In these areas, pasture maize and Miscanthus varieties can be employed as a means of adapting to climate change.

Reduced reliance on traditional feed grains and improved food security: To create novel, performance-tested varieties for fodder breeding programs, research endeavours are required. For many, grassland-based livestock farming shall grow unfeasible in the absence of reliable, affordable forages, and consumers or governments may find it inappropriate to move to concentrate-based diets (Ignacio, et al., 2019).

Decreased environmental impact of livestock production (e.g., land use, greenhouse gas emissions): Employing unconventional feed can reduce livestock production's environmental impact. The total use of land and emissions of greenhouse gases related to animal production can be reduced by recycling food scraps and employing flies or algae, which need less land and water.

Enhanced animal health and well-being through more diverse and nutritious diets: Global differences in livestock production can be attributed to factors such as species raised, goals, size, magnitude, agroecology, and resources used. It has changed quickly as a result of growing needs, population expansion, rising affluence, urbanization, and evolving technology. Livestock is retained for transportation, draught power, social and cultural assets, and a variety of products.

Emerging technologies and innovations in livestock feeding:

Precision feeding and personalized nutrition for animals: Numerous technical solutions have been introduced in response to the increased demand for animal feed, to produce more high-quality feed with fewer resources. Feed productivity increase, feed quality enhancement, feed quality maintenance or preservation, and feed nutritional status enhancement are the five groups into which these solutions can be divided. These are the technologies that several LMICs are creating and putting into practice to increase feed supply. Improved forage varieties, improved crop wastes and roughages, and promoting private sector involvement are examples of commonly utilized technology (Balehegn, et al., 2020).

Utilization of artificial intelligence (AI) and machine learning to optimize feed formulation: Several industries, notably farm planning, livestock management, environmentally friendly resource utilization, and disease management, have been transformed by AI technologies. Artificial Intelligence (AI) is revolutionizing farming operations by boosting productivity, cutting waste, and lessening environmental impact through the use of drones, robots, and sophisticated monitoring systems. It helps farmers anticipate customer behaviour, improve animal health, and solve challenges without lowering productivity. AI systems can save expenses, automate processes, and enhance the quality of cattle. Farmers can automate the monitoring of animal welfare and behaviour, anticipate disease outbreaks, and plan feeding regimens (Melak, Aseged, & Shitaw, 2024).

Development of novel feed supplements and additives: When switching from pasture to mixed rainfall and flooded systems, the density of animals increases. Livestock systems are interdependent with agricultural operations and land use; they support crop production and regulate ecosystem services. Livestock has an impact on economic security and biodiversity, either positively or badly (Alders, et al., 2021).

The Role of research and Collaboration in advancing non-conventional Feeding Practices:

Collaboration government organizations, business partners, and academic institutions must work together to carry out research, create novel technologies, and set up legal frameworks. The adoption of innovative feeding regimens can be sped up by working together that enable the exchange of information, resources, and standards of excellence.

27.13 Conclusion

Raising important insects such as mealworms, crickets, and black soldier flies is known as insect farming, and it is a widely used method of animal growth. Compared to conventional feed ingredients like soy or fishmeal feed, insect-based feeds are healthier for animal growth since they are high in lipids, rich in amino acids, and low in calcium and phosphorus. Algae and hemp are emerging as promising feed alternatives for animals because they are rich in protein, important fatty acids, vitamins, and antioxidants.

Because of their high protein, vitamin, and amino acid content, single-cell proteins (SCP) are likewise becoming more and more popular. Their source is microorganisms such as yeast, fungi, and bacteria, and they can be made using byproducts as a substrate. Recycling food waste, such as leftover fruit and vegetable scraps, bread scraps, brewer's grains, and dairy byproducts, is another effective strategy that can be used to feed animals.

Currently, different locations and feed types are subject to different restrictions regarding non-conventional feeds. For example, food waste and by-products are employed in the US while insect-based feeds are commonly used in aquaculture in the European Union. In animal feeding, these standards place a strong emphasis on preserving feed, avoiding contamination, and upholding nutrient quality. Regulators must overcome obstacles such as data security, protracted approval procedures, higher producer costs, and stakeholder opposition to approve these feeds.

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Chapter 28 :Navigating Hematological Disorders from Diagnosis to Treatment

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28.1 Introduction

The blood consists of a fluid part called plasma and the formed elements called cells. The blood cells are of three types: red blood cells (RBC), white blood cells (WBC) and platelets (PLT). White blood cells are further divided into three main groups, granulocytes (neutrophils, eosinophils, and basophils), monocytes and lymphocytes (Patel & Radia, 2017). Blood cells are continuously destroyed, either by aging or because of their functional activities and are replaced by new cells. There is a fine balance between the rates of formation and destruction of these cells in healthy people. The production of blood cells is termed haemopoiesis (De Pater & Dzierzak, 2015).

Hematological disorders include a wide range of conditions that affect the blood and its components. These disorders can have severe consequences on the body's capacity to transport oxygen, fight infections, and form clots (Arboix et al., 2016). This chapter delves into the several types of hematological abnormalities, their etiology, signs, symptoms, diagnostic approaches, and treatment methods.

28.2 Classification of Hematological Disorders

There are three major groups of blood disorders:

i. Blood disorders affecting red blood cells
ii. Blood disorders affecting white blood cells
iii. Blood disorders affecting platelets

28.1 Blood disorders affecting red blood cells

Red blood cell-related blood diseases can have a significant impact on the body's capacity to carry oxygen and remove carbon dioxide. The following are some typical red blood cell disorders:

- **Iron Deficiency Anemia**

  The most frequent reason is iron deficiency in the body, which prevents the body from producing sufficient hemoglobin, a protein which helps in carrying out oxygen and found in red blood cells. In this disease a patient may feel fatigue, dizziness, dyspnea, pale skin, alopecia and restlessness (Jimenez et al., 2015). The condition may be diagnosed through signs, symptoms, physical examination, on complete blood count (CBC) a patient may have decreased hemoglobin and hematocrit levels, on microscopic examination red blood cells may show microcytic and hypochromic characteristics. Moreover, serum ferritin and iron level also suggest the iron deficiency in body (Camaschella, 2017). In addition, elevated level of total iron-binding capacity (TIBC) can indicate iron deficiency. Iron deficiency in body can be recovered by oral iron supplements including ferrous sulfate, intravenous iron, intake of iron rich foods (meat, beans, cereals and lentils) and use of vitamin C rich citrus fruits can improve the absorption of iron (Naigamwaila et al., 2012).

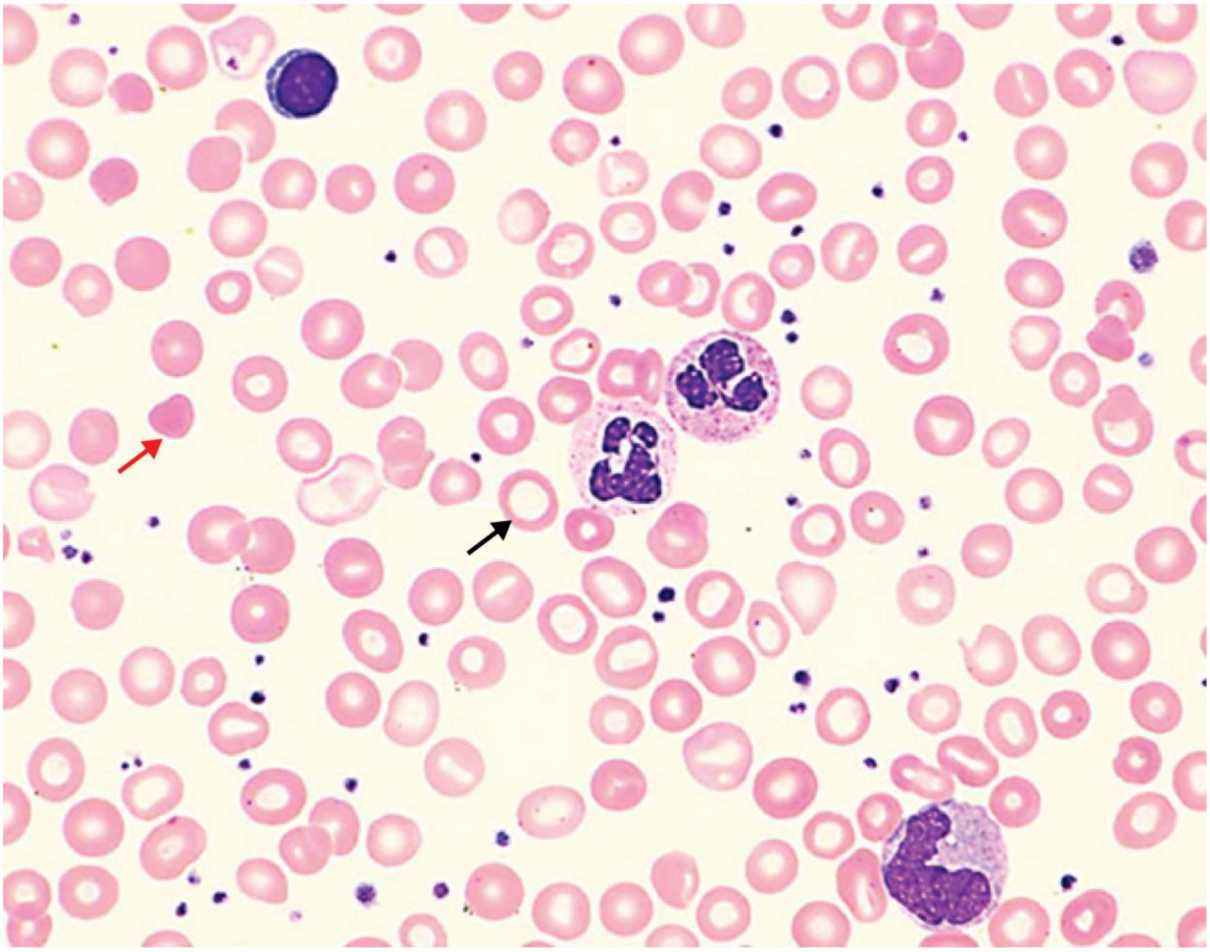

Figure 89: Microscopic examinations of peripheral blood smear showing iron deficiency anemia with microcytic (red arrow) and hypochromic (black arrow) red blood cells (40X).

- **Pernicious Anemia**

Pernicious anemia may develop by the body failure to absorb vitamin B12 which is necessary for red blood cell formation and normal brain function. The condition is induced in response to an autoimmune disease which damages the internal lining of stomach particularly those that make intrinsic factor, a protein required for vitamin B12 absorption (Toh, 2017). Also, gastric and intestinal surgeries can lead to decrease production of intrinsic factor. Though less common, but a diet deficient in vitamin B12 can also cause pernicious anemia. A patient with pernicious anemia may feel fatigue, pale skin, dyspnea, dizziness, muscular weakness, memory loss, glossitis and mouth ulcers (Annibale et al., 2011). Pernicious anemia can be diagnosed by CBC showing macrocytosis, decreased red blood cells, low levels of vitamin B12, and the presence of antibodies against intrinsic factors or stomach cells. Schilling tests are also used to measure the absorption of vitamin B12 by the digestive system. Regular injections of vitamin B12, oral high dose of vitamin B12 supplements and healthy diet rich in vitamin B12 such as eggs, meat, cereals and dairy products (Bizzaro & Antico, 2014).

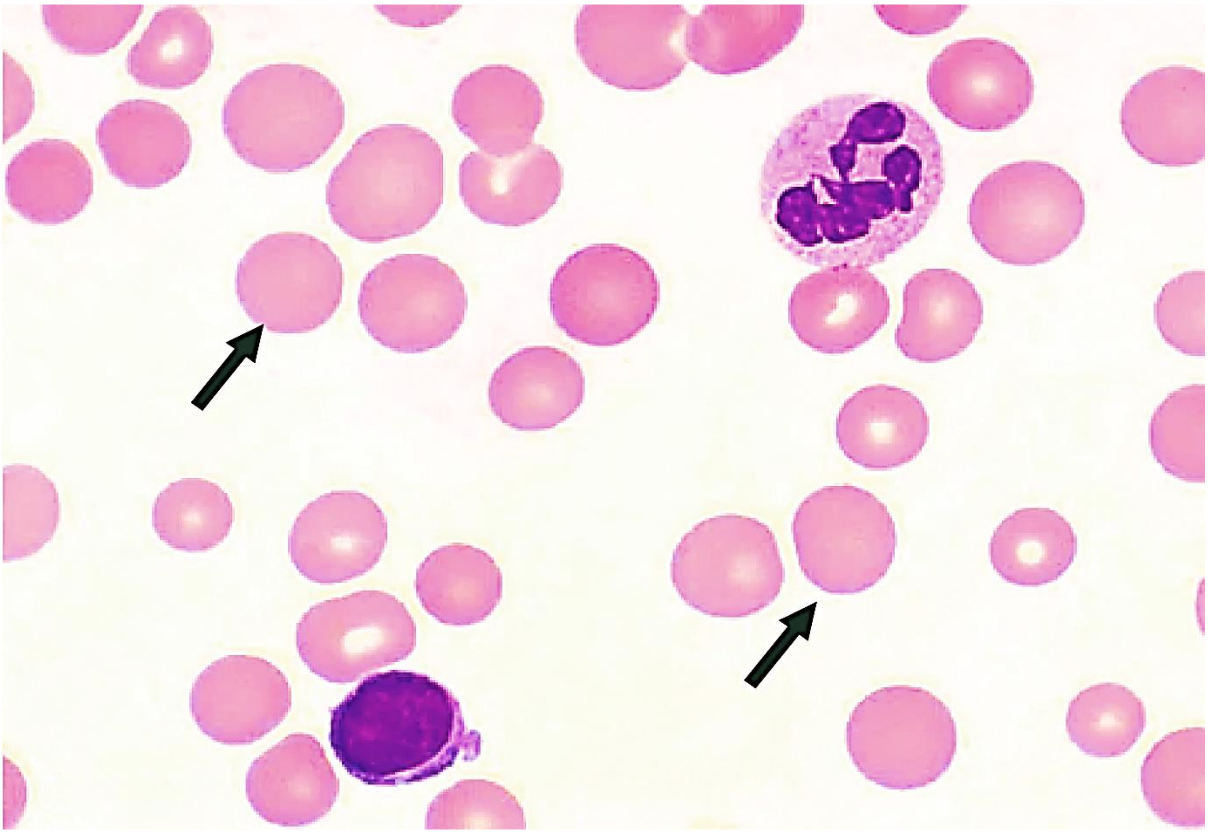

Figure 90: Microscopic examinations of peripheral blood smear showing pernicious anemia with macrocytic red blood cells (black arrow) (40X).

- **Aplastic Anemia**

Aplastic anemia is a rare blood disorder which is caused by the inability of bone marrow to produce new blood cells. In this conditions all three types of blood cells i.e., RBCs, WBCs and platelets are deficient, which can be developed at any stage of life. Aplastic anemia can be inherited or acquired with unknown etiological reasons, however some factors including autoimmune diseases, radiation, chemotherapy, medication, certain toxins and viral infections may contribute to developing the aplastic anemia (Young et al., 2010). Due to decreased number of RBCs, a person with aplastic anemia may develop frequent infections due to leukopenia, weakness, tachycardia, easy bleeding due to thrombocytopenia and dyspnea. Aplastic anemia can be diagnosed through multiple tests such as CBC, bone marrow biopsy and reticulocyte count (Peslak et al., 2017). The severity of disease can determine the possible choices of treatment i.e., blood transfusion, bone marrow transplant, stem cells transplant, use of certain antibiotic and antiviral drugs, and use of beneficial drugs such as erythropoietin to produce the sufficient number of RBCs (Miano & Dufour, 2015).

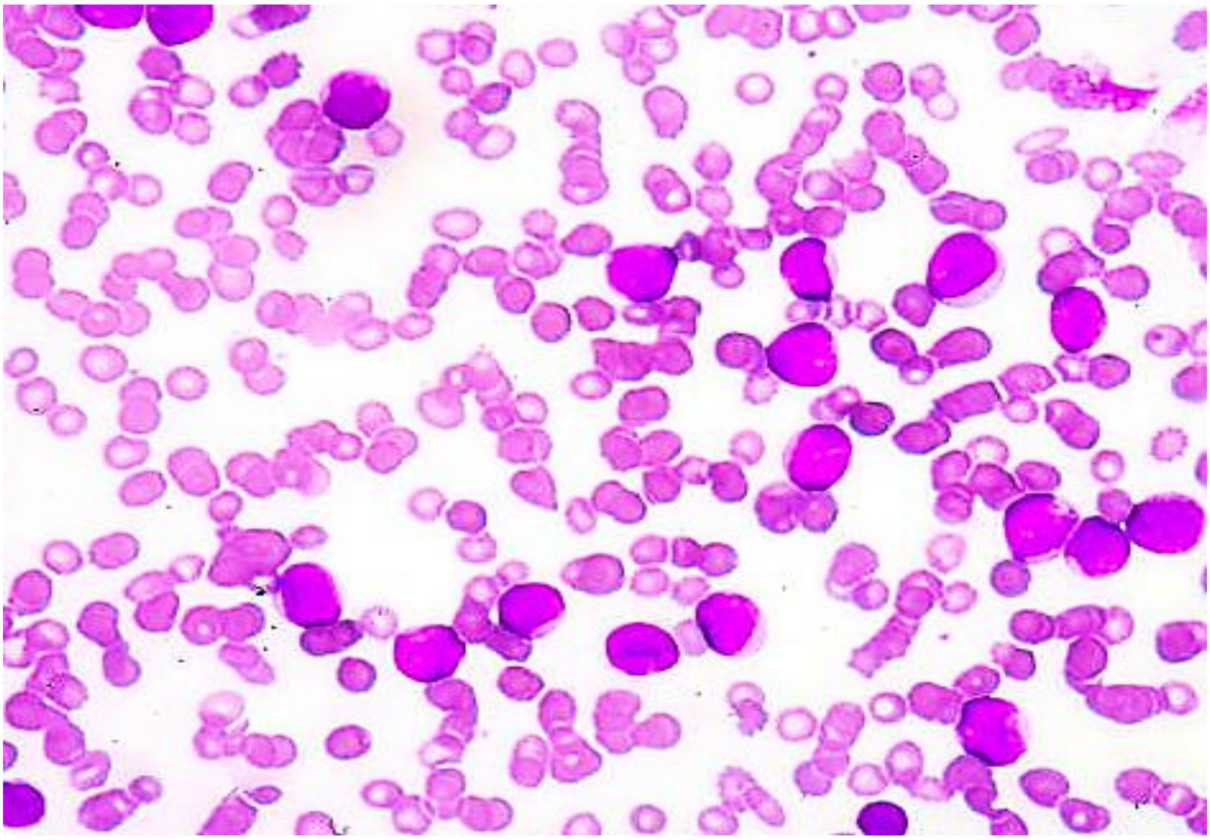

Figure 91: Microscopic examinations of peripheral blood smear showing aplastic anemia (40X).

- **Hemolytic Anemia**

Hemolytic anemia is a group of blood disorders (inherited or acquired) in which RBCs are destroyed more quicky compared to their production rate. The hemolysis can be intravascular or extravascular. This is caused by mutation in hemoglobin gene leading to abnormal production of hemoglobin which may cause sickle shaped RBCs (Barcellini & Fattizzo, 2015). Thalassemia, a genetic disorder in which abnormal hemoglobin is produced leading to destruction of RBCs, is another cause of hemolytic anemia. Hemolytic anemia can also result from spherocytosis, a condition in which sphere-shaped RBCs are produced. The other factors which can cause hemolytic anemia are autoimmune diseases, paroxysmal nocturnal hemoglobinuria, infections like malaria and some drugs (Hill & Hill, 2018). The prominent signs and symptoms of hemolytic anemia are dyspnea, dark colored urine, tachycardia, splenomegaly, hepatomegaly and gallstones (Barcellini & Fattizzo, 2015). Hemolytic anemia can be diagnosed through CBC, reticulocyte count, microscopic examination of peripheral blood smear, direct Coombs test, haptoglobin test and lactate dehydrogenase (LDH) test. This condition can be treated using corticosteroids and immunosuppressive drugs, blood transfusion, splenectomy, use of folic acid supplements, and bone marrow transplant (Phillips & Henderson, 2018).

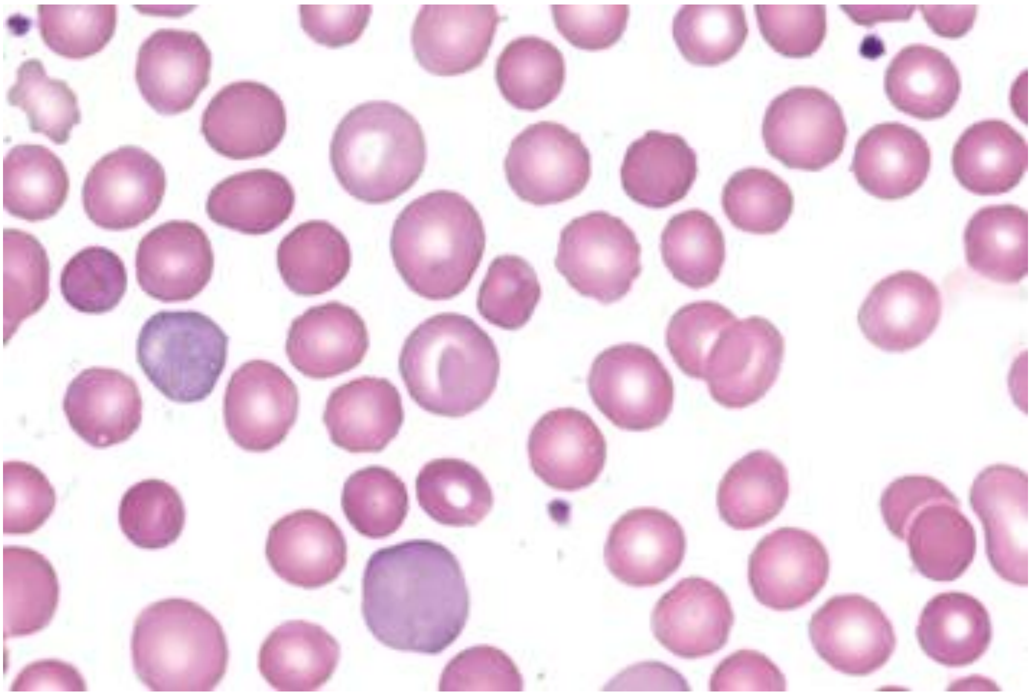

Figure 92: Microscopic examinations of peripheral blood smear showing hemolytic anemia (40X).

- **Sickle Cell Anemia (SCA)**

  Sickle cell anemia may refer to production of RBCs with abnormal morphological features. In this condition, abnormal hemoglobin, known as hemoglobin S is produced due to mutation in HBB gene. Such misshapen RBCs can lodge into blood vessels which may cause narrowing of the Blood vessels and other health complications (Pinto et al., 2019). Sickle cell anemia is an inherited disease which is shifted to the next generation in autosomal recessive pattern. Clinical signs of sickle cell anemia may include fatigue, weakness, retarded growth, swelling of hands and feet, frequent infections, and vision problems (Rees et al., 2010). The other complications induced by SCA are acute chest syndrome, stroke, liver, kidney and spleen damage, pulmonary hypertension and gallstones. Diagnosis may include newborn screening, blood tests and hemoglobin electrophoresis. Various approaches can reduce the complications of SCA such as use of Hydroxyurea, pain relievers and antibiotics specially penicillin, blood transfusion, gene therapy and bone marrow transplant (Williams & Thein, 2018).

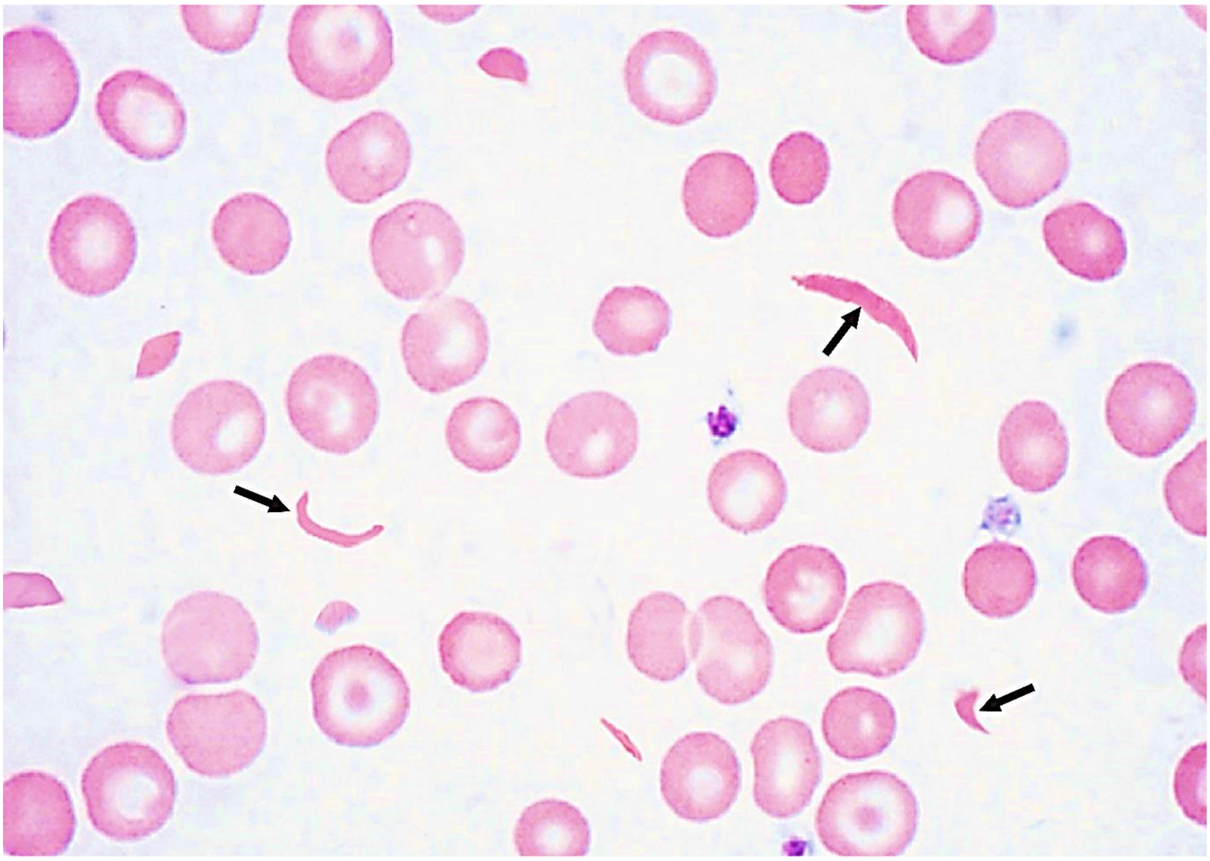

Figure 93: Microscopic examinations of peripheral blood smear showing sickle cell anemia with misshapen RBCs (black arrow) (40X).

28.3 Blood disorders affecting white blood cells

Blood disorders affecting white blood cells (WBCs) have a significant impact on the body's ability to fight against infections. White blood cell disorders can result in either an overproduction or decreased production of WBCs, or in dysfunctional WBCs. Here are some common disorders affecting white blood cells.

- **Leukopenia**

  Leukopenia refers to the decreased number of white blood cells resulting in weak immune system and increased risk for infections. Leukopenia can be caused by bone marrow disorders (myelodysplastic syndromes), autoimmune diseases (lupus, rheumatoid arthritis), certain infections (HIV/AIDS, viral hepatitis, typhoid fever), various medicines (used for chemotherapy, certain antibiotics, immunosuppressants), nutritional deficiencies (specifically lack of vitamin B12, folate and radiation therapy) (Carli et al., 2015). The clinical signs and symptoms may include frequent infections, fever, mouth ulcers, sore throat and fatigue. Leukopenia can be diagnosed by CBC, bone marrow biopsy and other screening tests. This condition can be treated by discontinuing causative medications, use of granulocyte colony-stimulating factor, antibiotics or antivirals for infections and balanced diet (Cargnin et al., 2018).

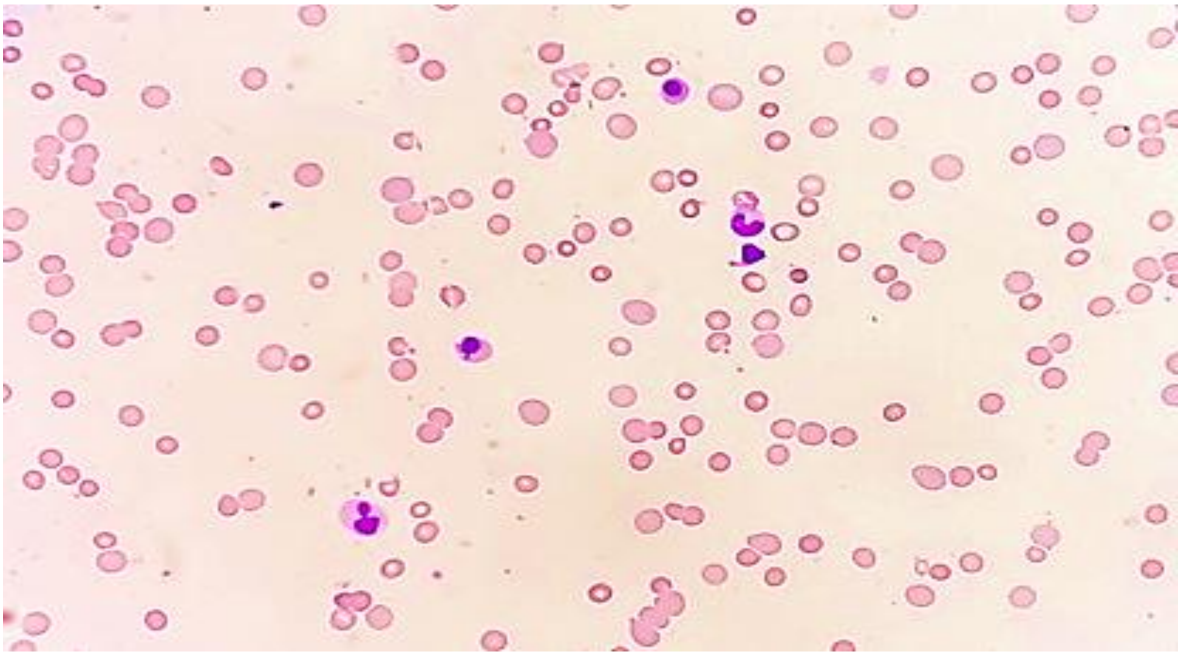

Figure 94: Microscopic examinations of peripheral blood smear showing leukopenia with decreased number of WBCs (10X).

- **Leukocytosis**

Leukocytosis refers to an increased number of white blood cells, usually in response to infection or inflammation, but can also indicate bone marrow diseases. The factors associated with leukocytosis are bacterial, viral and fungal infections, inflammatory conditions i.e., rheumatoid arthritis, inflammatory bowel disease, acute or chronic leukemia, myeloproliferative disorders such as polycythemia vera, essential thrombocythemia, physical or emotional stress, trauma and certain medications such as corticosteroids (Chabot-Richards & George, 2014). The clinical signs and symptoms are fever, fatigue, pain or discomfort in the upper left abdomen (due to splenectomy) and weight loss (in cases of leukemia). The diagnostic approaches of leukocytosis are CBC, differential WBC count, bone marrow biopsy and additional tests to diagnose infections or inflammatory conditions. Leukocytosis can be treated by handling leukapheresis (in severe leukocytosis) to reduce WBC count and use of antibiotics for infections, chemotherapy for leukemia (Connolly et al., 2010).

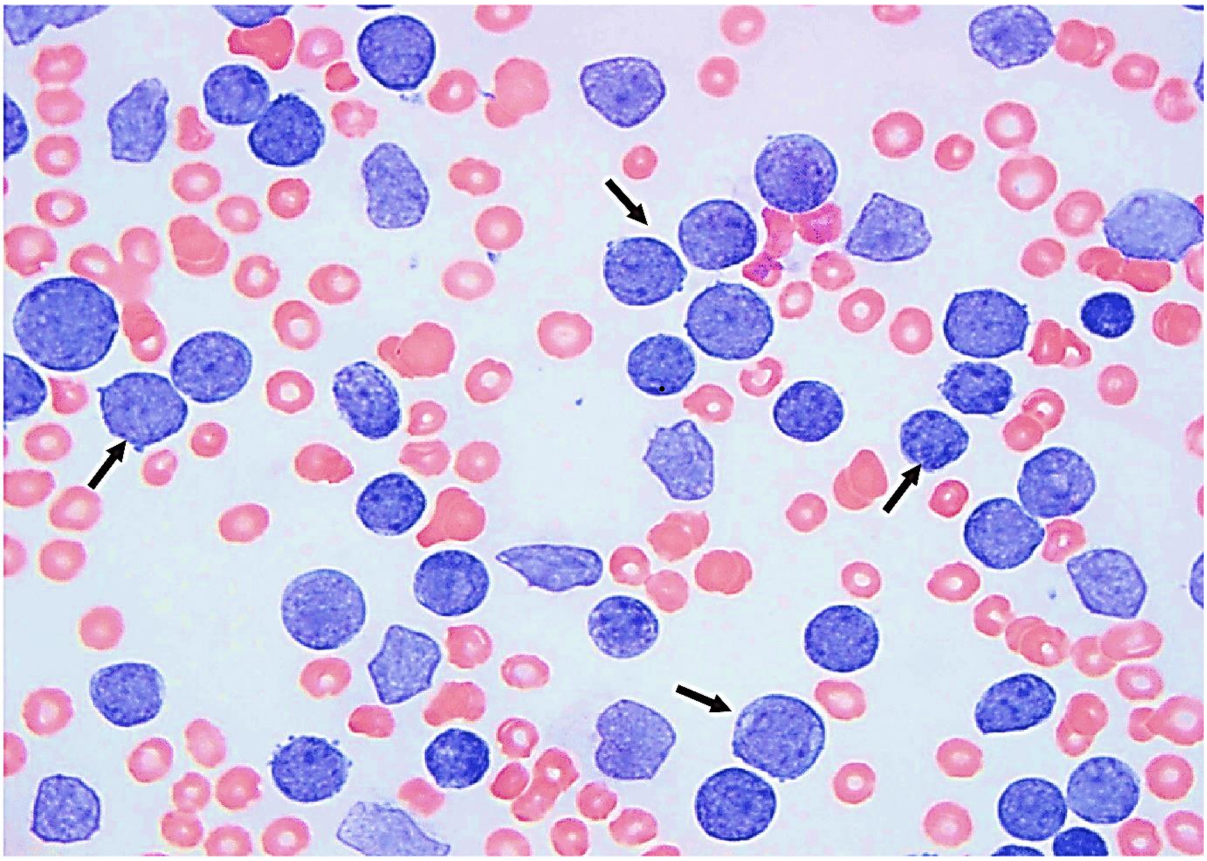

Figure 95: Microscopic examinations of peripheral blood smear showing leukocytosis with increased number of WBCs (black arrow) (40X).

- **Leukemia**

Leukemia is a type of cancer that affects the blood and bone marrow. It can be identified by a high level of abnormal white blood cells. There are following four most common types of leukemia:

i. Acute Lymphocytic Leukemia (ALL)
ii. Acute Myeloid Leukemia (AML)
iii. Chronic Lymphocytic Leukemia (CLL)
iv. Chronic Myeloid Leukemia (CML)

The clinical signs and symptoms of leukemia may include inflamed lymph nodes, frequent infections, fever, easy bruising and osteoarthritis. Leukemia can be diagnosed by CBC, bone marrow biopsy and genetic testing to identify specific mutations associated with leukemia. Leukemia can be treated with immunotherapy, bone marrow transplant, radiation therapy and chemotherapy (Saultz & Garzon, 2016; Terwilliger & Abdul-Hay, 2017).

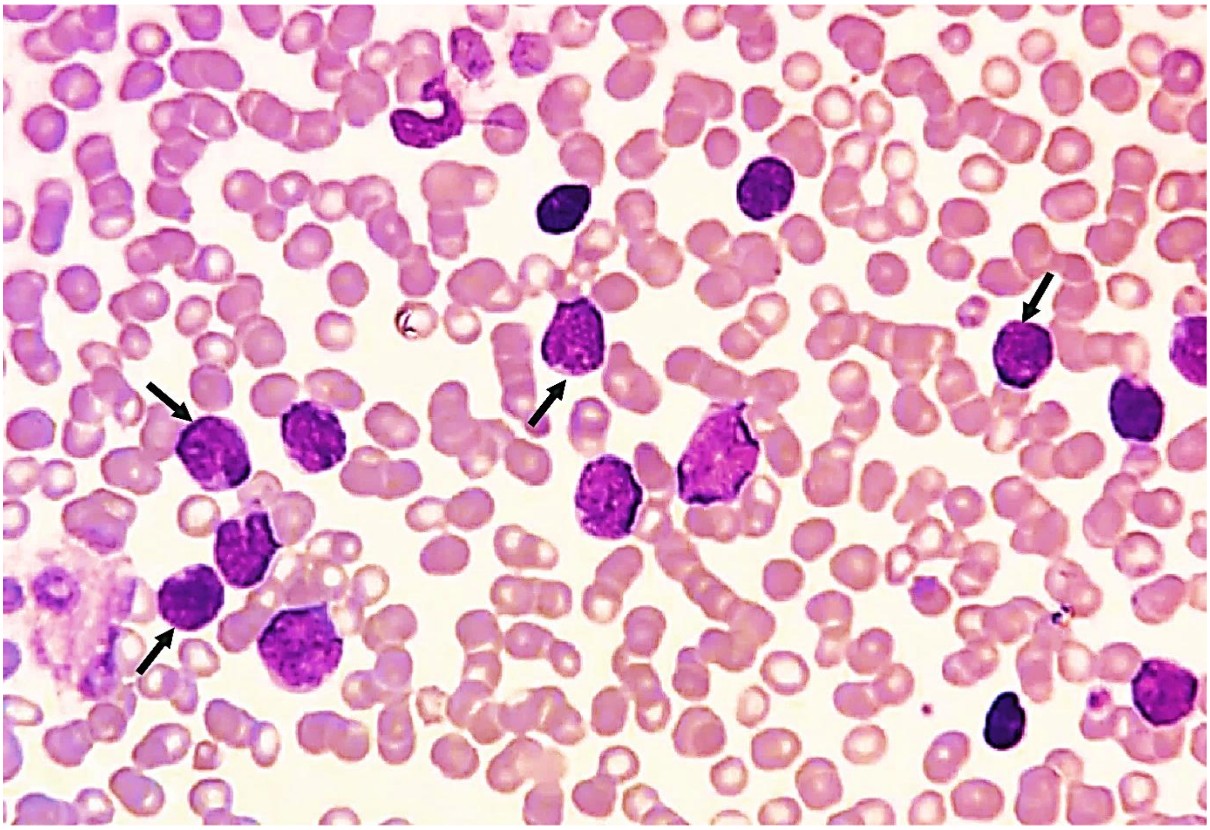

Figure 96: Microscopic examinations of peripheral blood smear showing leukemia with increased number of abnormal WBCs (black arrow) (40X).

- **Lymphoma**

  Lymphoma is a form of cancer that develops in the lymphatic system and mostly affects lymphocytes. There are following two most common types of lymphoma, Hodgkin lymphoma: which is characterized by the presence of Reed-Sternberg cells and Non-Hodgkin Lymphoma which affects B-cells, T-cells, or natural killer (NK) cells (Armitage et al., 2017). The clinical signs and symptoms include inflamed lymph node, excessive sweating at night, abrupt weigh loss, weakness and fatigue. Lymphoma can be detected through lymph node biopsy and imaging tests such as a CT scan or a PET scan to determine the extent of the disease and other blood tests. Chemotherapy, radiation therapy, immunotherapy and stem cell transplantation are used for treatment purposes (Molyneux et al., 2012; Singh et al., 2020)

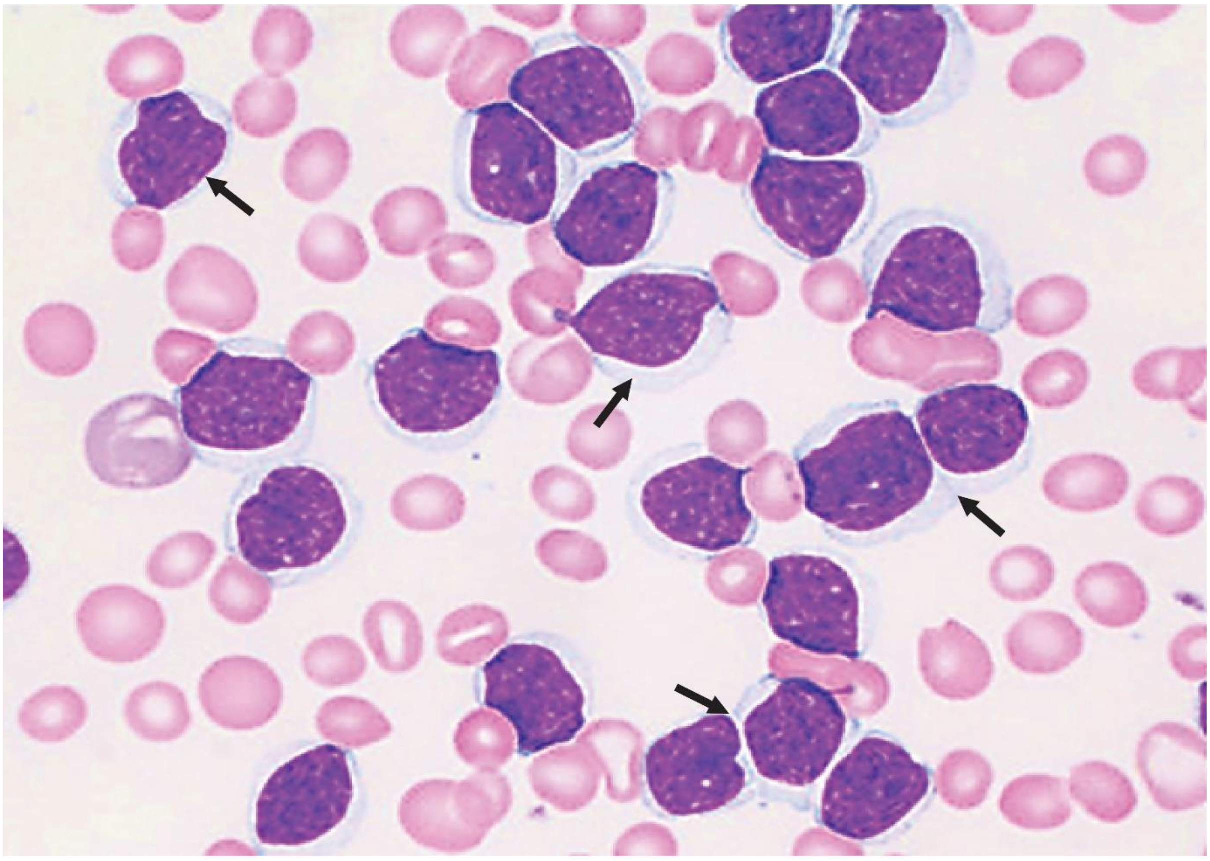

Figure 97: Microscopic examinations of peripheral blood smear showing lymphoma with increased number of abnormal lymphocytes (black arrow) (40X).

28.4 Blood Disorders Affecting Platelets

Platelets or thrombocytes are anucleated cells that originate from stem cells through the process of megakaryopoiesis and mature within 4-5 days. They have a size range of 2 to 4 µm with discoid shape and varies in number ranging from 150,000 to 400,000 cells/mL of blood and may remain for 10 days or less in the peripheral circulation (Koupenova et al., 2017). Following are some important platelets oriented blood disorders.

- **Thrombocytopenia**

  Thrombocytopenia refers to abnormal low number of platelets in the blood leading to increased bleeding and bruising. The condition can be mild to severe with various etiological factors. Decreased platelet production due to bone marrow disorders, nutritional deficiencies, viral infections like hepatitis, HIV, Epstein Barr virus chemotherapy drugs, certain antibiotics, and other medications that affect bone marrow, can induce thrombocytopenia (Stasi, 2012). Similarly, increased platelet destruction due to Autoimmune Disorders i.e., immune thrombocytopenia (ITP), lupus, certain medicines i.e., heparin-induced thrombocytopenia (HIT), certain antibiotics, anticonvulsants, various infections such as sepsis, mononucleosis and pregnancy can also induce thrombocytopenia. Moreover, increased platelet consumption due to disseminated intravascular coagulation (DIC), thrombotic thrombocytopenic purpura (TTP), hemolytic uremic syndrome (HUS) and splenomegaly are the other factors of thrombocytopenia (Barcellini & Fattizzo, 2015). The clinical signs and symptoms of thrombocytopenia include easy or excessive bruising, prolonged bleeding time, epistaxis, hematuria, melena, petechiae on skin and weakness. Thrombocytopenia can be diagnosed by CBC, microscopic examination of peripheral blood smear, assessment of bone marrow biopsy as well as antibody tests (Izak & Bussel, 2014). Thrombocytopenia can be treated with the use of corticosteroids, intravenous immunoglobulin (IVIG), immunosuppressive drugs, and thrombopoietin receptor agonists. Furthermore, blood or platelet transfusions, splenectomy and avoiding certain medications such as aspirin and nonsteroidal anti-inflammatory drugs (NSAIDs) can also decrease the chances of thrombocytopenia (Koupenova et al., 2017).

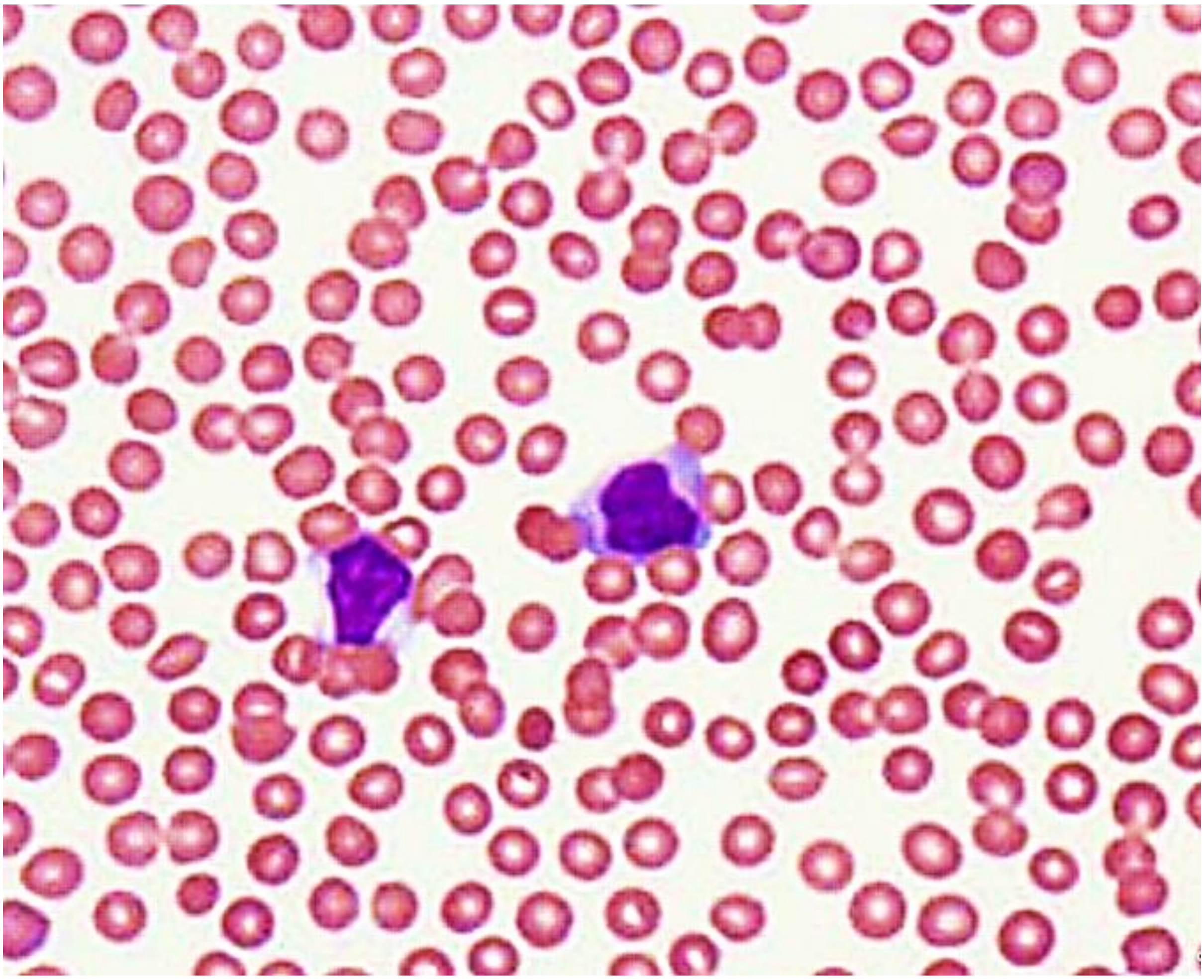

Figure 98: Microscopic examinations of peripheral blood smear showing thrombocytopenia with decreased number of platelets (10X).

- **Thrombocytosis**

Thrombocytosis is characterized by an excess high platelet count, which increases the risk of blood clots. This is caused by bone marrow disorders like essential thrombocythemia or polycythemia vera and may be induced in response to other diseases including inflammation, cancer and iron deficiency (Sulai & Tefferi, 2012). Clinical signs and symptoms may include thrombosis leading to strokes and heart attack, dizziness and chest pain the condition can be diagnosed by CBC, genetic testing and bone marrow biopsy. Thrombocytosis can be treated with use of low dose of aspirin and other drugs which can decrease platelet count (Chiarello et al., 2011).

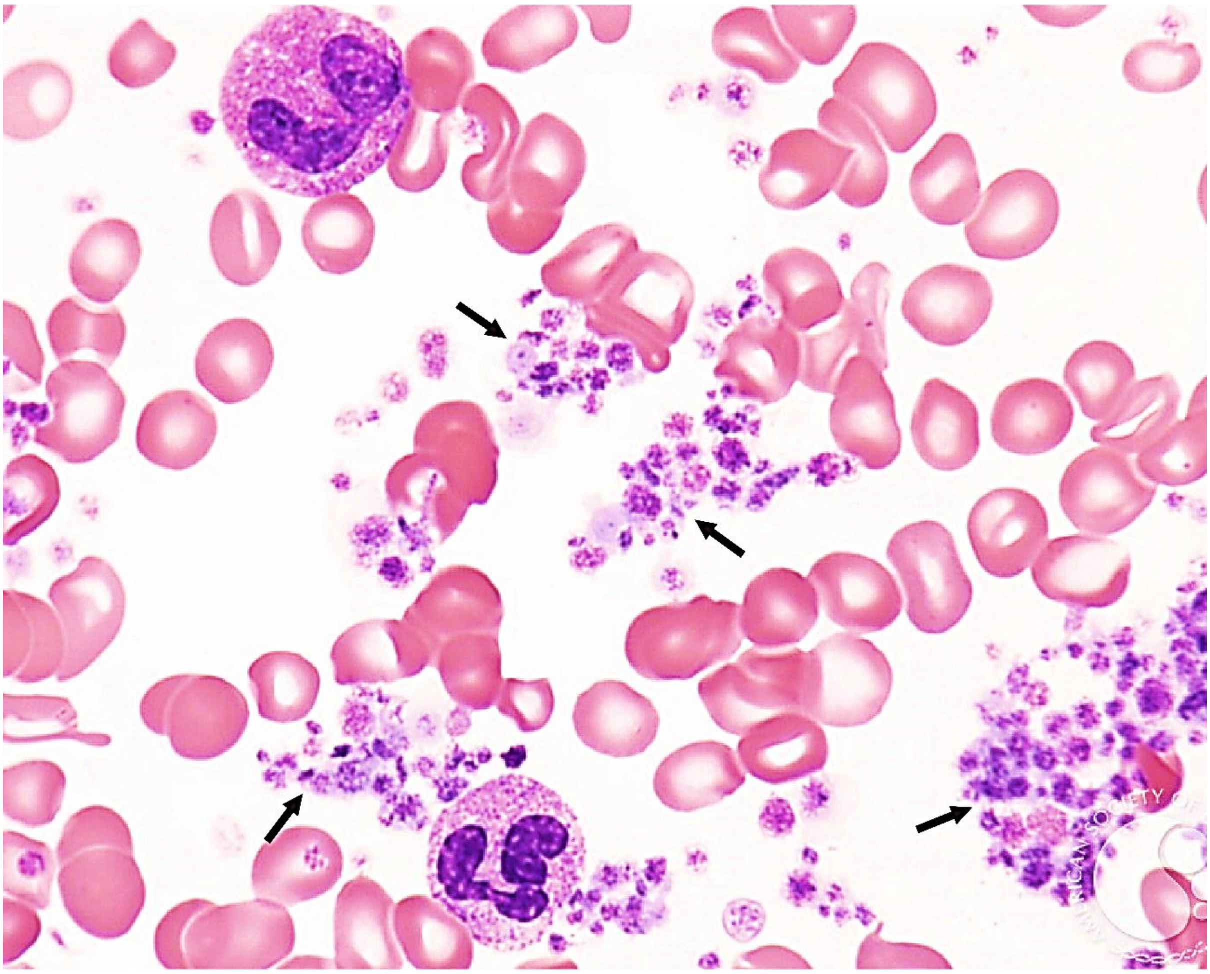

Figure 99: Microscopic examinations of peripheral blood smear showing thrombocytopenia with increased number of platelets (40X).

- **Platelet Function Disorders**

These disorders involve abnormalities in platelets function, leading to bleeding problems despite a normal platelet count. These disorders are categorized into two categories i.e., inherited disorders, such as Glanzmann thrombasthenia and Bernard-Soulier syndrome and acquired disorders, caused by medications (like aspirin or clopidogrel), medical conditions (such as uremia), or surgery (Gresele et al., 2015). The symptoms include easy bruising, nosebleeds, prolonged bleeding from cuts or surgery and bleeding gums. The diagnostic approaches may include platelet function tests (to assess how well platelets are working), genetic testing (for inherited platelet function disorders) and bleeding time test (to measure how quickly blood clots form) (Paniccia et al., 2015). This disease can be treated by avoiding medications that impair platelet function such as desmopressin. In severe cases platelet transfusions are recommended.

- **Heparin-Induced Thrombocytopenia (HIT)**

Heparin-induced thrombocytopenia is an immune-mediated adverse reaction to heparin, leading to a drop in platelet count and an increased risk of thrombosis. This disease is caused by the formation of antibodies against heparin-platelet factor 4 complexes, leading to platelet activation and clot formation (Greinacher, 2015). The symptoms of this condition include low platelet count, new blood clots, red or purple spots on the skin (petechiae), and pain, redness, and swelling in the extremities. Heparin-induced thrombocytopenia can be diagnosed by blood tests (to detect antibodies against heparin-platelet factor 4 complexes) and by clinical assessment, based on the timing of heparin exposure and the presence of thrombosis. Heparin-induced thrombocytopenia can be avoided by discontinuation of heparin, alternative anticoagulation therapy (e.g., direct thrombin inhibitors) and monitoring and managing complications of thrombosis (Cuker & Cines, 2012).

Managing platelet-based blood clotting disorders often involves regular monitoring, lifestyle adjustments, and avoiding activities that could increase the risk of bleeding or clotting. Patients should work closely with their healthcare providers to manage their condition and prevent complications.

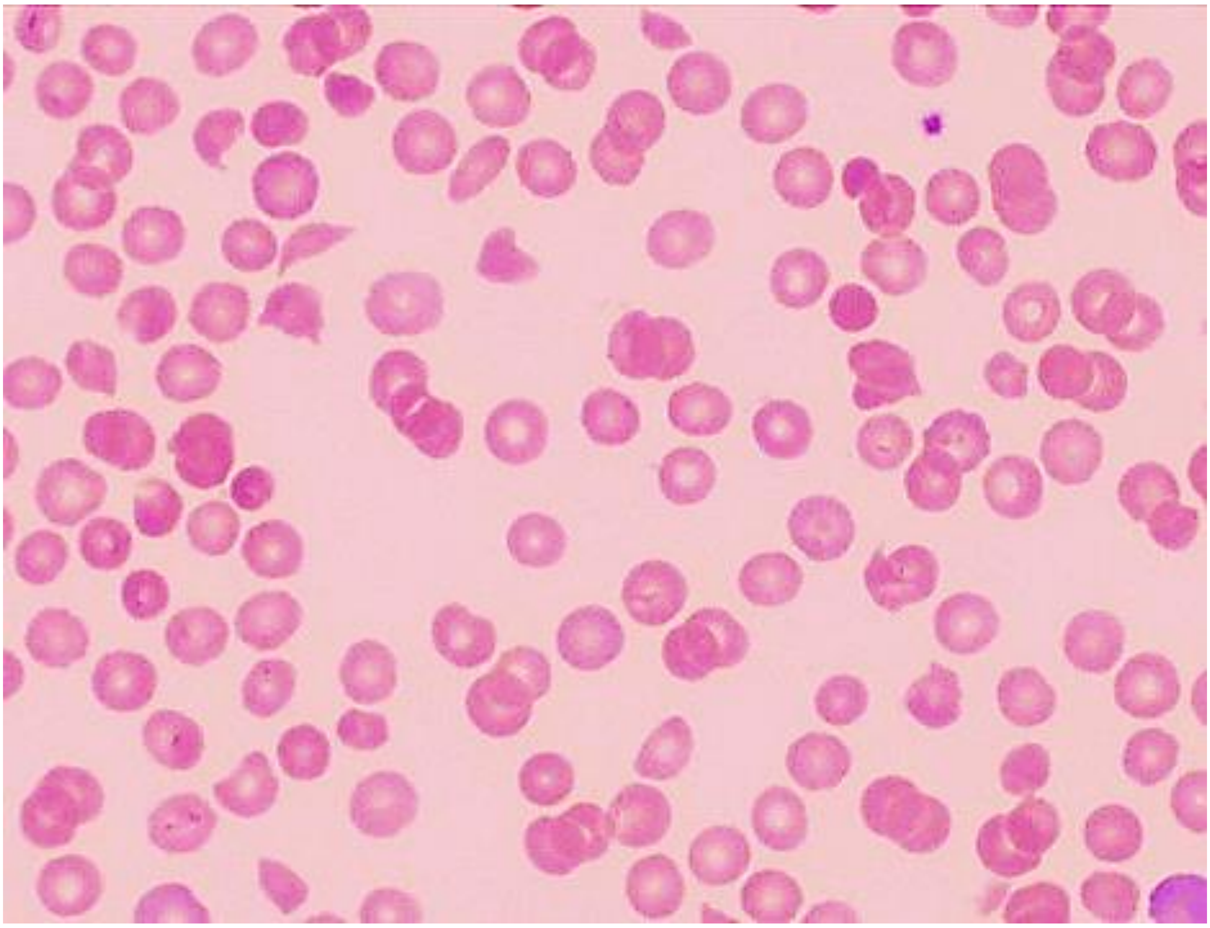

Figure 100: Microscopic examinations of peripheral blood smear showing heparin-induced thrombocytopenia (40X).

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Chapter 29: Antimicrobial Peptides Are A Substitute For Antibiotics In The Farming Industry, Particularly For Livestock And Poultry

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Abstract

The food security and agricultural productivity has been historically boosted by the overuse of antibiotics in livestock and poultry farming but has led to various problems. Some of the notable problems that arose are antimicrobial resistance (AMR), human health risks, and environmental degradation. As bacteria continue to develop resistance to antibiotics, the conventional treatments become ineffective, making it a serious health threat globally. On the basis of this development, this chapter discusses the potential of using antimicrobial peptides as substitutes for traditional antibiotics in farming. AMPs are naturally occurring molecules with broad-spectrum antimicrobial properties derived from various organisms; animals, plants, and microorganisms. More interestingly, they have unique mechanisms of action through which they mostly act by disruption of microbial membranes, therefore leading to a rapid destruction of the pathogen with a lower probability of resistance development. In vitro and in vivo studies have already proven that AMPs display fair effectiveness against common pathogens of livestock and poultry. Examples of these peptides, including cecropin and nisin. They possess high activities against *Salmonella* and *Staphylococcus aureus* pathogenic bacteria. Application of these peptides within farming practice involve considerations for their cost-effectiveness, delivery system, and regulatory compliance. Scientific progress in the field of synthetic biology and engineering of peptides reduces the price of manufacturing, enhance AMP's stability, universality, and selectivity agents adapted for agricultural purposes. The effective design of delivery methods, like encapsulation, guarantee protection from degradation of the AMPs and ensure their bioavailability in target animals. Despite all the progress and advances, many challenges remain an obstacle. These include high production costs, developing potential resistance, and strict regulatory requirements. Strategic investment, public-private partnership, and well-structured training programs for farmers and veterinary professionals could clear the path of such barriers. By harnessing AMPs, agriculture will significantly reduce its reliance on conventional antibiotics to avert the impending AMR crisis and thereby ensure safe and sustainable farming practices for public health.

Keywords: antimicrobial peptides, antimicrobial resistance, livestock farming, pathogen control, sustainable agriculture

29.1 Introduction

Antimicrobials are chemical substances that are either derived naturally from living organisms or manufactured synthetically in the laboratories. They can be administered through oral, parenteral, or topical pathway to kill or inhibit the growth of pathogens. The use of antibiotics in farming has been a common practice for decades. This is primarily done to promote growth and prevent disease in livestock and poultry (Hosain et al., 2021). This intensive use of antibiotics has enormously played a role in enhancing the production of food and improved food security. However, massive use and abuse of antibiotics in animal farming have occasioned the unavoidable repercussions (Van Boeckel et al., 2015). These effects are unfavourable to humans, animals and environment. The use of antibiotics also leaves its residues in meat and animal products making it dangerous to human health. These could result im allergic reactions or even interference with the digestive system and the microbiets. Furthermore, the contamination by running off into the soil and water systems has an enormous effect on the environment and presents the major ecological issues (Kyuchukova, 2020). The mounting of increased usage of antibiotics in food animals is one of the foundational challenges associated with the occurrence of antimicrobial resistance. In this process, it modifies its structures to prevent the effect of antibiotics and therefore makes the traditional methods of treatment useless. AMR has now emerged as one of the most pressing global challenges in the 21st century due to the rapid rise in the incidence rates of AMR infections and the dearth of new antimicrobial drugs (Tang et al., 2023). This bacterial resistance can be transmitted from animals to humans through direct contact, ingestion of food and via the environment hence complicating the existing public health issues (CDC, 2021; Gilbert et al., 2021).

Based on the challenges elicited by antibiotic usage and the AMR issue, it becomes worthwhile to look for other strategies of controlling diseases among the livestock. The traditional antibiotics have emerged as a promising option called the antimicrobial peptides (AMPs). These are the endogenous molecules that exist in most of the living organisms. They have antimicrobial characteristics having broad-spectrum activity ranging from bacteria fungi, viruses as well as cancerous cells (Erdem Büyükkiraz and Kesmen, 2022). Most AMPs function and interfere with microbial cells’ membrane causing quick pathogen elimination and decreased chances of resistance (Lei et al., 2019). This book chapter aims to give an introduction into the use of antimicrobial peptides that may replace antibiotics in the farming of livestock and poultry. Innovative solutions like the AMPs in the fight against the global menace of AMR, and the safe and sustainable farming practices will be further explained in this chapter.

29.2 Antibiotic Use in Livestock and Poultry Farming

Administration of antibiotics in livestock and poultry farming originated in the middle of the 20th century for purposes of promoting animal growth and preventing the diseases (Tang et al., 2017). Initially, antibiotics were embraced as some kind of revolutionary tool that could improve the rate of animal growth and feed efficiency, hence significantly improving the productivity. The repetitive use of sub-therapeutic doses of antibiotics became extensive, as preventive measure against potential outbreaks. This practice became contributory in sustaining the rapid growth of industrial-scale farming set-ups. (Rogers, 2018).

Although there were initial benefits of use of antibiotics in animal farming, the long-term use has led to several major challenges. The most serious issue is the development of antibiotic resistance. The continuous exposure to antibiotics creates selective pressure on microbes. This encourages the survival and proliferation of resistant strains (Van Boeckel et al., 2017). This resistance spread among animals, to humans, and throughout the environment, developing a complex and difficult-to-manage public health threat. In addition, inappropriate use and overuse of the antibiotics in farming have reduced the efficiency of these drugs, raising a serious concern on the availability of these drugs in future for veterinary and human medicine (O'Neill, 2016).

Administration of antibiotics in the livestock and poultry industries despite being for animal consumption also affects directly the human beings as well. The residues of antibiotic can also be administered into the human food chain through the consumption of the treated animals' meat, milk, or eggs (Koutsoumanis et al. , 2021). These contribute to the emergence of AMR in the human body, making common infections hard to treat and, therefore, increases the costs incurred in healthcare (Founou et al., 2016). Secondly, the effects of this chemical compound on the environment are also significantly significant. By applying manure and having agricultural run-off some of the antibiotic residues and the resistant bacteria can get into the soil and water systems. Hence, it destroys the subsystems of local economies and stimulates the growth of the environmental resistance (Kumar et al. , 2019).

29.3 Antimicrobial Peptides: Nature, Classification, and Mechanisms of Action

AMPs are bioactive molecules derived from natural sources and significantly smaller compared to most proteins. These are the components of the natural immunity of almost all types of organisms, including people, animals, plants, and microorganisms. Normally, it consists of 10-50 amino acids and exhibits the qualitative differences in extending the antimicrobial spectrum against bacteria, fungi, viruses, and some cancer cells (Lei et al., 2019). Based on the features of these peptides, one of the main characteristics of AMPs is the amphipathicity. They both have moieties that are capable of non-polar and polar interactions with liquids. This aspect enables the AMPs to interface with microbial lipopolymers, which results in their breakdown. This feature makes it central to their antimicrobial function (Mishra et al., 2018). Due to their fast action, AMPs have the potential to quickly destroy a pathogen and are capable of doing so within minutes. This eliminated the development of resistance chances as compared to the normal antibiotics (Mookherjee et al. , 2020). Moreover, most of the isolated AMPs have multiple functions and show immunomodulatory properties including chemotaxis, wound healing, and regulation of the host's immune response (Dijksteel et al., 2021).

29.3.1 Classification of AMPs Based on Structure and Function:

According to the structure and functions, AMPs can be classified into several classes. On the basis of their secondary structures, the different categories of AMPs are alpha-helical peptides, beta-sheet forming peptides, extended peptides and looped peptides (Fjell et al. , 2012). Magainins that are a type of alpha-helical peptides have the capacity to form helices in a membrane environment. Beta-sheet peptides like defensins contain disulfide bonds in them with an account of their stabilization. As for indolicidin, extended peptides lack many secondary structures and have a number of precisely proline and arginine. Denatured peptides, for instance bactenecins, are further stabilized by one or more loops concerned by disulfide bonds (Huan et al., 2020).

Functionally, AMPs may be grouped according to the way they function in the body. Some of the AMPs are selective to microbial membranes which in turn destroys the membranes and leads to cell lysis. While others have intracellular targets like nucleic acids, proteins or a metabolic pathway (Zhang and Gallo, 2016). Also, some AMPs show anti-biofilm functionality, and thus can combat the biofilm-related infections that are normally impermeable to traditional antibiotics (de la Fuente-Núñez et al., 2016).

29.3.2 Mechanisms of Action of AMPs:

One of the most studied and well-documented modes of action of most AMPs is the disruption of microbial membrane. AMPs become embedded with the negatively charged parts of bacterial membranes. These negatively charged components are lipopolysaccharides in Gram-negative and teichoic acids in Gram-positive bacteria. This interaction can lead to poration – essentially the creation of pores, decrease in membrane thickness, and ultimately cell lysis (Kumar et al., 2018). For example, the AMP melittin depolarizes the lipid bilayers by forming pores on them. These pores cause the specific cells to die at a very fast rate (Raghuraman and Chattopadhyay, 2007).

Apart from disruption of the membrane, several AMPs permeabilize the microbial cell membrane and effect intracellular components. For instance, the AMP buforin II is capable of passing through the membranes with minimal dep picture and works almost as an antiseptic in that it binds to DNA and RNA to prevent transcription and translation. A second example of an AMP is indolicidin, which gets between the base pairs of the DNA thus disrupting the normal functioning of the DNA and this kills the bacterial cell (Lei et al., 2019). It has also been reported that most of the AMPs inhibit the biofilm formation, degrade the formed biofilms or enhance the effect of conventional antibiotics against biofilms (Batoni et al., 2016). This is particularly relevant as biofilms are generally considered to be more resilient to a scenario of antibiotic treatment and generally associated with chronic infections (Pletzer et al., 2016). The comparative analysis of mechanism of action of AMPs and conventional antibiotics is given in the Fig-I.

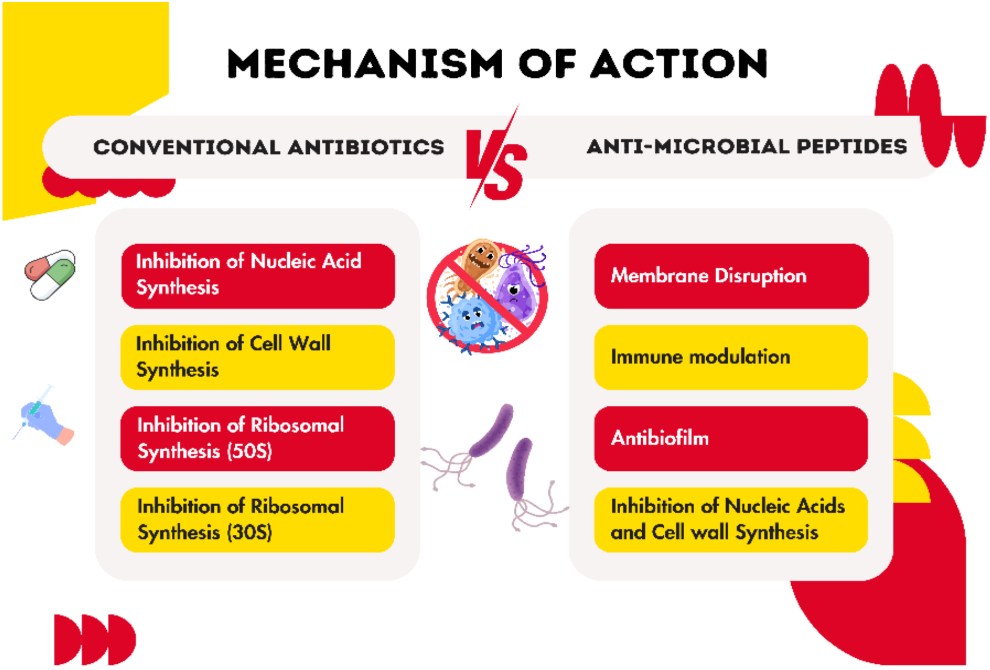

Figure 101

Comparison of MOA of AMPs and Conventional Antibiotics

29.3.3 Importance of AMPs in Innate Immune Defense in Animals:

Coupled with the natural antibodies, AMPs play an irreplaceable role in the innate immune protection of animals. They are formed by the epithelial cells of the target sites, immune cells, and other tissues after invasion by microorganisms (Ganz, 2003). The AMPs play the first barrier function to neutralize the pathogens in a short time to avoid infection. For instance, in mammals cathelicidins, defensins are major categories of AMPs these seem to have important role in host defense against bacterial, viral, fungal pathogens (Brown & Hancock, 2006). In addition to the antimicrobial properties, AMPs mobilizes immune cells during infection, knock out pathogens by promoting phagocytosis, increase cytokine and chemokine production to organize the immune response (Haney et al., 2019). They also participate in the process of wound healing by inducing cell migration, and proliferation and angiogenesis (Ramos et al., 2011).

29.4 Sources and Diversity of Antimicrobial Peptides

With respect to their structure, the AMPs are a heterogeneous group of molecules present in almost all organisms and thus constitute an extremely vast reservoir of molecules that can be considered as therapeutic targets against microbial infections. They have a very significant role to protect the innate immunity of animals against pathogens in their bodies. For instance, cathelicidins and defensins in mammals, have broad-spectrum antimicrobial activity (Hancock and Sahl, 2016). Plants also synthesize their own kind of AMPs known as the plant defensins which are present in seeds, leaves and roots and which shield the plants from fungal and bacterial attacks (Parisi et al., 2019). Microorganisms, like Gram +ve bacteria *Bacillus subtilis*, produce subtilosin, which play a role in microbial competition and defense (Huang et al., 2009).

New advancements in synthetic biology and peptide synthesis have enabled the design of new and enhanced AMPs with enhanced killing effect and stability. These synthetic AMPs are derived from the natural form but have been engineered to have enhanced effectiveness together with minimized toxicity (Mookherjee et al., 2020). For instance, the change of the peptide sequence or coupling it with nanoparticles will improve the circulation time of the compound and improve the targeting capability (Mahlapuu et al., 2016). However, several difficulties have been encountered in order to further enhance the synthetic AMPs for application in a clinical practice. These are challenges such as the issue of efficiency/safety, the issue of scale up from the laboratory, and the issue of regulatory approval (Wang et al., 2016).

AMPs exhibit structural heterogeneity that influence their ability as antimicrobial peptides and their mode of action. Depending on their structure, they are alpha-helical peptides like magainin, beta-sheet peptides like defensin, extended peptides like indolicidin, and looped peptides like bactenecin (Fjell et al., 2012). All the types of structures of the agents under consideration affect the microbial membranes or intracellular targets in different ways, which defines their antimicrobial efficiency and activity profile (Huan et al., 2020). The variation in the sequence among the AMPs also affects their functions due to the differences in the arrangements. Such changes in the amino acid sequences of the peptides influences the stability of the peptides, their mode of action as well as the resistance mechanisms exhibited by microbes. This is because knowing these sequence-structure-function correlations is highly useful in designing AMPs for certain functions in the antimicrobial systems (Mishra et al., 2018).

29.5 In vitro and In vivo Efficacy of AMPs in Livestock and Poultry Farming

Antimicrobial peptides (AMPs) represents the potential antimicrobial action against the prevalent pathogens in the livestock and poultry farming based on the in vitro and in vivo studies. The peptides may thus transform into the new antibiotics as the difficulties associated with antibiotic resistance and food security are considered horrible. The efficacy of AMPs against several bacterial strains, that are commonly implicated in livestock and poultry infections, was evidenced through in vitro experiments. For example, nisin and pediocin have been reported to effectively inhibit *Staphylococcus aureus* and *Escherichia coli*. These peptides either damage bacterial membranes or inhibit bacterial cell functions causing death of the microbial cells (Wang et al., 2016). The in vivo studies support the research results which indicate that the AMPs help in minimizing the bacterial infections and enhance the quality of live of animals. For instance, the AMP cecropin has been claimed to possess possibilities of controlling salmonellas in chickens whether as a prophylactic or therapeutic weapon. Therefore, the presented AMPs are effective under real farming circumstances, where bacterial infections are risky from the economic and health points of view (Xie et al., 2019).

The ability of AMPs to act under various farming conditions could be attributed to the numerous ways through which they function. In general, AMPs interact with the microbial membranes and can either create pores in the membranes or cause their destabilization. Moreover, some peptides get into the bacterial cells and interfere with the internal structures like DNA or specific enzymes, which are vital for bacteria's existence. For example, lactoferricin interferes with the bacteria ability to build biofilms and enhances the vulnerabilities of the bacteria to the host immune system (Wang et al., 2019).

Such factors as delivery methods and dosing schedules are, however, commonly involved in modulating in vivo efficacy. The delivery of used AMPs through either an encapsulation technique or formulation with a stabilizing agent allows for optimal bioavailability and controlled release of the peptides within the gastrointestinal tract of the animal (Kang et al., 2019). The dosage optimization is essential so that therapeutic levels of the active compounds in the form of AMPs can be achieved, but with minimal toxicity or resistance to the AMPs. Moreover, the given frequency and time of administration of AMP also affect the antimicrobic ability of AMP against certain pathogens (Xie et al., 2019).

29.6 Economic and Practical Implications of Adopting Antimicrobial Peptides in Farming

Introduction of AMP in agriculture has a viable solution to one of the biggest problems of AMR hence the adoption of antimicrobial peptides in agriculture stands as a solution to a major issue on one side of its argument. However, the successful implementation of these practices in the agricultural field, not only entails a sound economic sense, but effective practical knowledge as well. The major factors that would influence the possibility of using these AMPs as antibiotic substitute in farming is largely with cost. Despite the fact that the initial cost of synthesizing AMPs is comparatively higher than that of the traditional antibiotics, the cost has significantly decreased over the years due to advancement in techniques in producing peptides and the biotechnology processes involved (Gomes et al., 2017). On the other hand, some studies pointed to a decrease in disease frequency and severity as a result of the use of AMPs, thus reducing treatment expenditure through increased production in the livestock and poultry farms (Park et al., 2018). Furthermore, the chance to reduce or even drop reliance on traditional antibiotics may partially help minimize costs of antibiotic resistance that is thought to be in the billions per year in terms of treatment, hospitalization, and lost output (O'Neill, 2016).

The use of AMPs as part of some conventional farming practices is not an easy feat, as it entails several aspects. In one of the most challenging obstacles to date, the problem relates to the synthesis of appropriate delivery systems that maintain the stability and bioavailability of AMPs throughout the gastrointestinal tract of the animals. Researchers have also tried to formulate and deliver AMPs using various carriers like liposomes and polymers, this would help in reducing enzymatic degradation of AMPs and increase their absorption (Kang et al., 2019). The employment of AMPs under the new paradigms of farm management practices, such as dosage regimens and monitoring schedules, has to be done differently. Training of the farmers on proper utilization of the AMPs is recommended and the close control by the specialists in veterinary is very essential. This also includes the specification of the regulatory frameworks whereby when AMPs are employed, then they have to meet all the high safety and efficacy standards set by the various regulatory bodies which may in one way or the other slow down the rate of uptake of the particular technology (Mookherjee et al., 2020).

The generalisation of the AMPs in the farming sector can generate important socio-economic effects. Internationally, decreased application of conventional antibiotics in farming can possibly help in enhancing the quality of human life by decreasing the impacts of antibiotic-resistant organisms. Thus, it would lessen the strain on the healthcare systems, contributing to the enhancement of human health (van Harten et al., 2018). From the economic aspect the expanded production of AMPs might contribute to the growth of biotechnology and pharmaceutical sectors which might provide job opportunities and stimulate innovations in the agriculture industry. But this may not be possible with small scale farmers who may not afford to invest in these technologies apart from the fact that it may take a lot of resources to train individuals for such technologies (Tang et al., 2018).

29.7 Future Directions and Challenges

Innovations in research and development of AMPs are considered for enhancement of their therapeutic effect and use in agriculture. In recent years, due to developments in the fields of synthetic biology and bioengineering new designer forms of AMPs have been developed that are more stable, selective and less toxic (Mookherjee et al., 2020). Also, the application of the nanotechnology together with AMPs has been proven to have potential in delivery and effectiveness (Kang et al., 2019). However, there are some of the issues that still exist causing a rein on the total implementation of AMPs. These are; High production costs, some regulations, and strictly enforced safety measures (Wang et al., 2016). Secondly, it cannot be overlooked that there may be cases of resistance against the AMPs similar to those observed against antibiotics (Mishra et al., 2018). The various ways of surmounting these challenges include embracing the appropriate production models that are cheap and have the scalability, nurturing of research collaborations between the private and public sectors to share the costs of research and development, as well as fostering adequate regulatory measures that may foster innovation while addressing the risks at the same time (Hancock et al., 2016). Thirdly, it is important to investigate the factors that contribute to resistance and derive effective ways of minimizing the problem. Education and training of farmers and veterinary personnel on the correct methods of deploying the AMPs will enhance their adoption into farming practices effectively and sustainably (Tang et al., 2018).

29.8 Conclusion

The problem of antimicrobial resistance (AMR) has led change in the scientific approaches for controlling diseases in the livestock and poultry farming. Antimicrobial peptides or AMPs are another class of molecules that can be considered as potential substitutes for the antibiotics since they are effective against the broad range of microorganisms and there is significantly lower chance of the pathogen to develop the resistance to the AMPs. Nonetheless, in spite of having many sources and ways of action, they show substantial efficacy against a set of some important pathogens that threaten livestock and poultry. These two methods have narrowed down the AMPs with the potential of decreasing infections and enhancing animal health hence acting as substitutes to antibiotics. However, the employed, real, and the economic impacts of the application of AMPs in farm practices have to be taken into consideration. To address the existing barriers of adopting AMPs; enhancement in peptide synthesis, delivery systems, and the enhancement of regulatory conditions are the future challenges. The prospects of the continued use of AMPs in agriculture settings in the future would, therefore, require continued research and development on efficient methodologies to enhance the stability, specificity, and costs of production of these peptides. Such achievements will involve decision-making and investment, public-private partnerships, and ensuring broad training of farmers and veterinary practitioners for ensuring transition to this form of disease control strategy. AMPs' continued implementation is especially crucial to support sustainable and safe farm practices, to help combat the development of AMR on a global level and to protect public health.

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Chapter 30 : Unlocking The Potential Of Adeno-Associated Viruses In Neuroscience

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Abstract

In modern neuroscience, Adenoviruses are playing a crucial role due to their adaptable behavior and efficient transferring of genetic information in various nerve cells. They are considered as an excellent choice for vector-mediated gene delivery because of their non-cytotoxic nature and prolonged expression in neurons. They have multiple applications including the treatment of various nervous disorders, control of brain activity and neuron tracing. Ongoing attempts to engineer AVV Capsid, for efficient transduction, decreased immunogenicity and improved tropism have increased their utilization in, gene therapeutics and neuroscience research. Although AAV vectors provide extraordinary advantages, their limitations i.e., transduction abilities must be taken into consideration. This chapter highlights the revolutionary role of AAV in modern neuroscience by highlighting its diverse application in transferring genetic material, control of nervous system activities and revealing the intricacies of brain biology. By providing general overview of novel methodologies and innovations using AAV vectors, it aims to demonstrate the revolutionary role of AAV in modern neuroscience.

Keywords: Capsid engineering, AAV, Gene delivery, Neuroscience, Viral vectors

30.1 Introduction:

Adeno-associated viruses play a key role in neuroscience, as they can transfer genetic information to multiple types of nerve cells, and have outstanding adaptability. These viruses are mostly used for research in neuroscience (Haery et al., 2019), and gene therapy of neurological disorders (Deverman et al., 2018, Hudry & Vandenberghe, 2019). Initially they were detected in 1960 from the stocks of adenovirus culture. Currently, AAV are mostly used as a vector for transferring genes to nervous system due to their prolonged expression in nerve cells and non-cytotoxic nature (Bulcha et al., 2021). Additionally, AAV can cause infection in humans that makes them a desirable choice for the creation of gene therapy vectors (Lundstorm, 2023). Remarkably, for gene therapy AAV is a prime candidate as it causes a moderate immune system activation with a very low chances of effects on non-target tissues. AAV has a single stranded linear DNA with a length of about 4.7kb. For replication, the genome of AAV relies on cellular polymerase as it lacks its own polymerase. Despite the widespread distribution of AAV, they are almost non-cytotoxic as they lack the ability of independent replication. Rather they require a virus that have the ability to replicate for the initiation of co-infection. Only capsid and replication genes are present in its genome that are responsible for capsid structure and regulation of viral replication (Zengel & Carette, 2020). AAV enters the cell by endocytosis during infection cycle and then by using vesicle bounded with clathrin it moves towards nucleus. After moving towards nucleus, the capsid of virion is lost releasing the genetic material, where depending on the absence or presence of co-infected virus, the viral genome either enters lysogenic cycle or proceed to lytic cycle (Wang et al., 2019).

30.2 AAV Mediated Gene Delivery:

In neuroscience, AAV are used for delivering genetic material efficiently within the cell or even within a specific type of organism. Depending on the purpose of experiment, the administration of viral particles is divided into two categories, either direct injection for delivering genetic material into specific region of brain (Ravindra et al.,2020, Rocchi et al., 2022) or widespread route for viral expression globally (Chan et al., 2017). Infection of an accurate and defined region of nervous system, thereby lowering its effects on non-target tissues has been possible by stereotaxic inoculation while retro-orbital and intravenous injections enable noninvasive, consistent delivery of genes to the central and peripheral nervous system (Li & Samulski, 2020). The accurate dispensation and targeting of adeno associated viruses in CNS has been revolutionized by using techniques such as intrathecal administration and convection enhanced delivery (Naidoo et al., 2019, Ajeeb & Clegg, 2023).

30.3 AAV for Neuronal Tracing:

AAV vectors are primary tools for the brain circuits manipulations and to study the functions of genes because of their efficient transduction abilities and prolonged gene expression in glial and nerve cells.

There are two major classes of viral tracers based on their ability to transverse synapses i.e. transsynaptic and non-transsynaptic. Tran-synaptic viruses can cross over synapses and spread to interlinked neurons on the other hand non-transsynaptic neurons cannot propagate through synapse to surrounding neurons. Both classes contain viruses that are transferred either retrogradely or anterogradely along axons. Notably, AAV1 and AAV9 among all the variants of AAV can be propagated transynaptically (Zingg et al., 2020; Zingg et al., 2022).

Knowing the region of virus injection, following the beginning of infection and mapping of viral propagation and expression area is essential for the identification and tracking of neuronal circuits. So viral vectors are broadly classified as anterograde and retrograde tracers based on the direction of viral propagation in cell. Anterograde tracers have the ability of neuron visualization and their respective targets by infecting soma of neurons and then moving towards axons. On the other hand, retrograde tracers make entry at the pre-synaptic terminal level and allows the visualization of projecting neurons as shown in Fig. 1(Zingg et al., 2017) by moving towards cell body of infected neurons retrogradely through axonal transport. Many serotypes of AAV have anterograde transport abilities such as AAV13(Han et al., 2022). On the other hand, AAV-DJ/9(During et al., 2020), AAV11 (Han et al., 2023), AAV9-retro (Lin et al., 2020) and AAV2-retro (Tervo et al., 2016) have retrograde transport abilities.

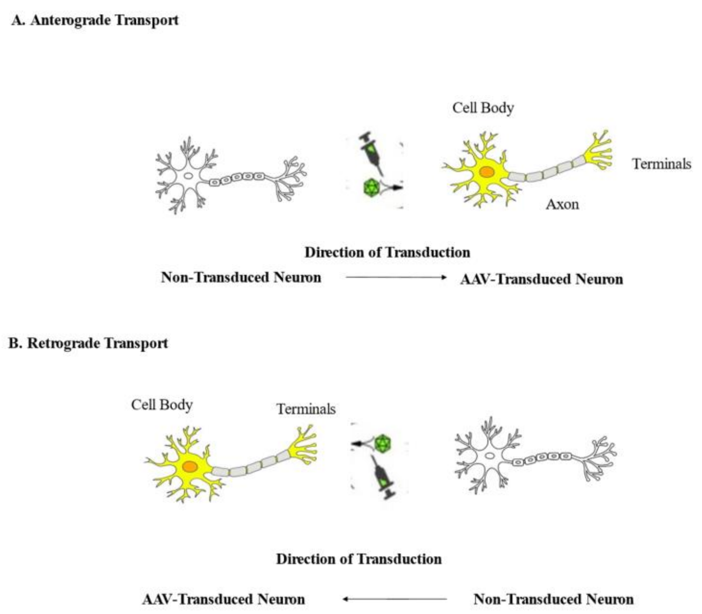

Figure 102: Viral vectors Classification

30.4 AAV for Controlling Brain Activity:

Gene delivery by using AAV in chemogenetic and optogenetic tools allows the accurate manipulation of activities related to neurons, facilitating inspection of complexities of brain circuits, and behavior in normal and diseased condition. These genetic strategies include the transportation of photoactivatable ion channels known as ligand-gated G -coupled ion channels named designer receptors activated by designer drugs (DREADDs) for chemo genetics (Atasoy& sternson, 2018; Vlasov et al., 2018; Campbell & Marchant, 2018) and opsins for optogenetics (Takao et al., 2022; Altahini et al., 2024). Cellular access by using stimulation of light facilitates the activation or blockage of activity related to neurons inside particular cells is accomplished by the expression of halorhodopsin (HR) or channelrhodopsins (ChR) in optogenetic applications (Govorunova et al., 2022) and in case of chemogenetics, the activation or blockage by the inoculation of particular Clozapine N-oxide drug is achieved by the engineered Gi or Gq-coupled human muscarinic acetylcholine receptors (hM4Di or hM3Dq) expression (Zhu & Roth, 2015). However, the mechanism of shared drug activation obstructs the combination of these two systems. To resolve this issue, Vardy et al. (2015) innovated a unique Gi DREADD based on human k-opioid receptor named as KORD, initiated by specific ligand, Salvinorin B, thus making multiple application of chemo genetic systems possible. With the advancement of chemo genetics and optogenetics, delivery of ligand responsive proteins and photosensitive mediated by AAV enables scientists to manipulate activity of neurons with spatial resolution and remarkable temporal offering significant insights into neural function and malfunction.

30.5 AAV vectors for Targeting CNS abnormalities:

rAAV vectors having the abilities to cross membrane between the blood and interstitium of brain when delivered systemically provide the best option for treating multiple CNS abnormalities. Systemic administration offers widespread CNS transduction in contrast to the approaches that rely on site specific vector administration, such as intracerebroventricular (Galvan et al., 2021), intrathecal (Bey et al., 2020) and intraparenchymal (Rosenberg et al., 2020) injections, that confined transduction primarily to the site of injection. To attain sufficient level of transduction in central nervous system, systemic inoculation requires higher dosage of vector, that increases the chances of immunotoxicity (Hinderer et al., 2018). Systemic administration of Adeno associated viral vectors has been demonstrated to be generally reliable in many clinical experiments addressing conditions related to peripheral nervous system such as Duchenne muscular dystrophy (NCT03368742), spinal muscular atrophy type 1 (NCT02122952) and mucopolysaccharidosis (NCT03315182) (Parez et al., 2020). Specifically, for targeting CNS conditions in adults, that require higher vector dosage of rAAV in comparison to dosage provided to children, it is essential to consider that transgene or AAV capsid may cause immune response related issues.

It is important to introduce changes in rAAV vector capsid to reduce safety concerns, to minimize unnecessary transduction of peripheral organs and to enhance transgene expression level as shown in Fig. 2. The fact that approximately 50% of human population have pre-existing natural antibodies from exposure to naturally existing AAVs poses a significant problem for systemic AAV injection as these antibodies significantly reduce transduction efficiency due to their cross reactivity across serotypes (Govindasamy et al., 2006). Therefore, modifying the capsid of rAAV vectors to avoid neutralizing antibodies is an essential methodology to increase the effectiveness of delivering AAV vectors systemically and antibody delivery mediated by rAAV-vector in central nervous system.

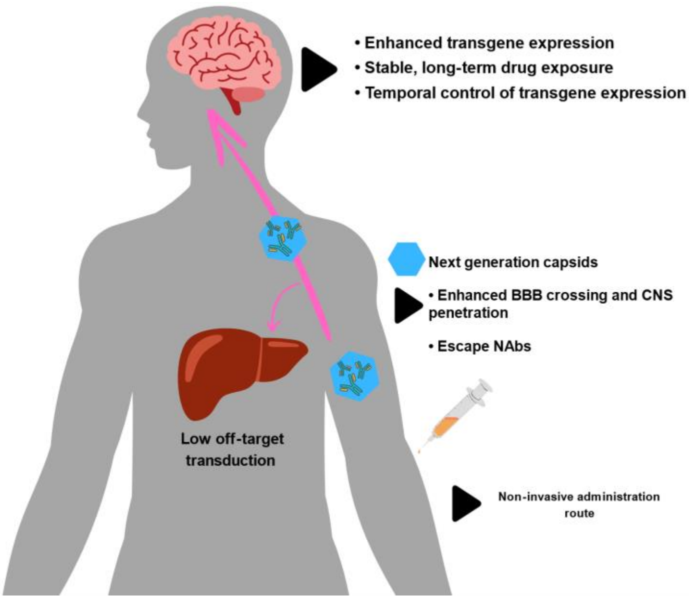

Figure 103: Desirable attributes of rAAV vectors for systemic delivery in brain

30.6 AAV serotypes for Central Nervous System:

Due to low cytotoxicity, efficient transduction, persistent gene expression and decreased immune response in host, recombinant AAV are extensively used as a vector for gene delivery in CNS. Currently numerous serotypes of AAV have been identified. The most widely used serotypes for central nervous system include AAV1, AAV2, AAV4, AAV5, AAV7, AAV8, AAV9 and AAVrh10 (Murlidharan et al., 2014; Lisowski et al., 2015; Tamrazian et al., 2019). Summary of these serotypes and their outcome is given in Table 1.

Table 36: AAV Serotypes for Central Nervous System

Serotype	Outcomes	References
AAV1	Efficient transduction of glial cells and neurons	Griffin et al., 2019
AAV2	Strong Neural tropism	Grey, 2013
AAV4	Preferentially cause transduction of ependymal cells by striatum injection	Thomas 2011
AAV5	Cause Astrocyte tropism and transduction In non-human primates and rats AAV5 effectively transduce glial cells and neurons in multiple sites of brain.	Griffin et al., 2019 Mandel and Burger, 2004

AAV7 & AAV8	By direct injection into the brain, they mainly transduce neurons	Cearley and Wolfe, 2006
AAV9 & AAVrh10	Can spread widely, move along axons either retrogradely or anterogradely, have the ability to cross the membrane between the blood and interstitium of brain making non-invasive treatment of nervous disorders possible	Hordeaux et al.,2015 Zhang et al., 2011

30.6 Promoter and Enhancers Effect on Transgene Expression:

For transgene expression various promoters and enhancers specific to brain cell types have been identified. AAV expression in various brains cell types such as vascular cells, glia and neurons can be achieved by using promoters. Gene expression specific to brain cell types can be derived by promoters for cerebellum Purkinje cells (de Leeuw et al., 2016; Nitta et al., 2017), catecholaminergic cells (Chan et al., 2017), GABAergic interneurons expressed by parvalbumin (Hoshino et al., 2021) and serotonergic cells (de Leeuw et al., 2016).

Enhancers (short regions of genes for regulation of transcription) have the ability to drive expression of transgene through AAV vectors. Enhancers when combined with nominal promoters, allow labelling of specific cell types and brain regions in various vertebrate specie. For instance, expression to GABAergic interneurons in the forebrain of rodents, human cells, and zebra finches is limited by enhancer named as mDlx (Dimidschstein et al., 2016).

Expression of transgene from AAV can be increased by using minimal promoter and multiple copies of enhancer (Graybuck et al., 2021; Mich et al., 2021). Moreover, for the compensation of low-level expression the transgene itself can be modified. Effectors (Gong et al., 2020), reporters (Campbell et al., 2020) and sensors (Unger et al., 2020) continue to be altered for enhanced expression, targeting and sensitivity.

30.7 Engineering and optimization of AAV capsid:

The strains of AAV are divided into multiple serotypes on the basis of their distinctive antigenicity of capsid proteins. These variants are specific in infecting various tissues due to their different tissue tropism mechanism.AAV vectors can acquire multiple serotypes by using distinctive proteins for capsid during packaging (Weinmann &Grimm, 2017).

Comprehensive research has been directed for the improvement of AAV tropism. Various studies have identified insertion and substitution sites for random and deliberate amino acids (Adachi et al., 2014; Ogden et al., 2019). Many engineered capsids with unique tropism have numerous variants that have broad or limited transduction abilities within peripheral nervous system or central nervous system, retrograde transport in neurons and specificity for cell type.

Stereotaxic inoculation allows infections within specific and confined area of brain , reducing effects on non-target tissues while retro-orbital and intravenous inoculation by using serotypes with ability to cross blood brain barrier allow non-invasive and compatible gene delivery within peripheral and central nervous systems (Li & Samulski, 2020).Many serotypes have broad glial and neuronal transduction inside brain regions due to direct intraparenchymal inoculation (Watakabe et al., 2017).

Specific cell type infection can be facilitated by combining AAV delivery with gene expression and recombinase system driven by promoters that are tissue specific. Precise activation of gene in both temporal and spatial manner may be achieved by directing Cre-dependent AAV vectors to a desired direction during developmental stage (Ravindra et al., 2020). AAV can transfer desired gene by using constituent promoters such as EF1a promoter and cytomegalovirus promoter (Fenno et al., 2014) or for the identification of gene functioning AAV vector can be specifically expressed in a more precise area of cells by utilizing tissue specific promoters (Koh et al., 2017; Le et al., 2022).

Targeting the cell body of neuron projections is essential for the selection of manipulating nerve cells terminating at specific regions. Attaining this accuracy imply transferring an AVV vectors having retrograde transport abilities like AVV2 (Tervo et al., 2016) AVV11 (Han et al., 2023), AVV-DJ/9 (During et al., 2020) to the projection areas.

Ongoing attempts to engineer AVV Capsid for efficient transduction, improved tropism and decreased immunogenicity are increasing their utilization in gene therapeutics and neuroscience research.

30.8 Limitations:

Although AAV vectors have many advantages such as low cytotoxicity and persistent gene expression, it is essential to know their limitations specifically regarding efficient transduction. The transduction level of AAV vectors is relatively low in comparison to vectors of other viruses. For example, AAV vectors need larger number of virions per cell during infection cycle to achieve transduction level similar to lentiviral vectors (Colella et al., 2018, Bulcha et al., 2021). The efficient transduction of AAV has been limited by many factors such as natural tropism of AAV serotypes (Aschauer et al., 2013, Yang et al., 2023) which is not according to particular cells as well as entry of vector is inhibited by neutralizing antibodies that already exist in the host (Mingozzi and High, 2017, Rossi et al., 2019, Zengel et al., 2023). Moreover, the usage of AAV vectors for many purposes that require transgene to express in a desirable way is limited by very low packaging ability of AAV vectors. So, the researchers must take into account these limitations while choosing vector system.

30.9 Conclusion:

AAV vectors have revolutionary role in neuroscience as they are capable of efficient gene delivery, neuronal tracing, revealing intricacies of brain and provide many therapeutic options for neurological disorders. Continuous improvements in vectors design, delivery techniques and capsid optimization will increase their efficiency of transgene expression in neurons. By expanding the utilization of AAV vectors, researchers can achieve enhanced gene delivery system with low immunogenicity that will be very helpful in future.

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Chapter 31: Holistic Approaches to Animal Health: Integrating Preventive Medicine

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Abstract

Holistic approaches mean to give stability that looks at the whole animal not just their mental health. Holistic medicines intend to cause disease, treat symptoms, and increase overall animal health and production. Animals, like humans, may face various environmental factors such as pain, stress, anxiety, poor digestion, and some other factors, which may affect animals' health and reduce production at an economical level. Holistic approaches as integrative medicine can be as an alternative to different antibiotics and other medicines. The consumption of preventive medicines may decrease risk factors which enhance animal health. In this book chapter, we explore how holistic approaches ensure their benefits in livestock by inhibiting different factors including; stress, pain, anxiety, nutrition, and some biosecurity measures. We learn about various holistic approaches to preventive medicines and how they work in animals. We also discuss some disease and their control in holistic approaches at an early age. Overall, this chapter highlights the significance of holistic approaches and integrative medicines in animals' lives to stimulate good health and production which have good effects at an economic level, also has some strategies to help clients and veterinarians accumulate staff training, client communication, and education about holistic approaches which enhance animal welfare as well as farm production.

Keywords: Holistic approaches, Holistic therapies, livestock, preventive medicines, Veterinarian.

31.1 Introduction:

31.1.1 Holistic Animal Health:

Holistic health is a perspective of life that contemplates multidimensional aspects of fitness. In animals' holistic health is considered medication for the whole pet, Considering nutrition, genetics, environmental behavior, and medical history. (Nordenfelt, L.2006) Furthermore, holistic veterinary care is defined as “Using all diagnostic and medication techniques that are accessible and effective”. It is an integrative approach to animal health, focusing on animals' bodies, minds, and spirit. It surpasses traditional veterinary medicines by concentrating on preventive care and natural medicament rather than treating manifestation with drugs. (Nordenfelt, L.2006) A veterinarian emphasizes the coordination between the nature of emotional, physical, and environmental aspects that inclusively impact animal well-being.

31.1.2 Holistic Medicines:

Holistic deals to whole of something” so to grasp a holistic approach to medicines means that the whole animal is not only physically, but also mentally emotionally, and spiritually. According to the American Veterinary Medical Association, Holistic veterinary medicine means “Alternative medicines, such as utilizing naturopathy and homeopathy in place of traditional medicines based on a credence system not circumscribed in science. (Dimpfel, W. 2015)

There are various methodologies for treatment, and the owners look to be well-informed. The international interest in the consumption of veterinary naturopathy and complementary medicine increasing day by day. (Nordenfelt, L.2006) The medicine practices, that odd hybrid of art and science that affects all lives confidentially have been especially influenced by science and technology and pressures toward reductions and specialization. In recent eras, it has also become a most productive ground for the development of a holistic perspective and practices. (Dimpfel, W. 2015)

Holistic medicine is an alternative for this type of medical approach. It doesn’t abandon the need for swift and polished medical or surgical action but does focus health promotions in animals for their well-being. Holistic medicines comfy all of the procedures that have been appearing in other cultures and at other times. After examination and proper testing, a holistic veterinarian will select the best combination of conventional and alternative therapies for animals. (Fox, M. W. 2016)

31.1.3 Importance of Holistic Approaches in Animal Health:

Veterinary medicines instead of traditional medicines, which usually focus just on treating symptoms, holistic approaches intend to root cause of health issues while increasing overall wellness. One key factor of holistic animal health is conceding the repercussions of lifestyle aspects on animals' health, this consists of diet, exercise, and environmental flourishment. A balanced diet customized to animals' certain nutritional needs can sustain immune function, digestion, and overall life. (Fox, M. W. 2016) Moreover, proper exercise helps to perpetuate a healthy weight, encourages cardiovascular health, and decreases stress.

Animals, like humans, can encounter anxiety, stress, and behavioral issues that affect their overall health. Holistic approaches to integrate techniques such as behavioral tutoring, environmental improvement, and stress reduction policies to enhance emotional viability and mental regulation. Holistic approaches classify encouraging a strong bond between animals and their caretakers. Pragmatic interconnection, trust, and communication improve the overall well-being of animals and provide a more positive veterinary experience. (Irvin, A. D. 1997)

31.1.4 Overview of Preventive Medicine in Veterinary Practice:

Preventive healthcare, as well as vaccination, is an important part of every day for animals' health. However, in a veterinary context, little curiosity has been afforded to the factors of preventive health care considered linked to small animals and factors that may enhance the uptake of these measures. (Stanossek, I., et al.,2022) Veterinary remedies are used rationally or irrationally in the animal sector for ameliorative, prophylactic, and growth-stimulating drugs, processing and preserving food, Stress control in abattoir proceeding slaughter, and progenitive control. Rational utilization of drugs is the use of accurate drugs, in the right amount, at the accurate time. Irrational use of veterinary remedies is the major difficulty when utilized in food-producing animals. Irrational medication can badly impact animals' health including; reducing the quality of medicine therapy leading to an increased death rate and anguish. (Qian, J et al., 2023)

Veterinary drugs play a vital role in livestock production and are usually used in animals for therapeutic and preventive purposes, development promotions, processing, and feed preservation, and can be controlled in feed or drinking water. (Yitbarek, T. 2024) The consumption of antimicrobials in livestock suggested a risk of infection or earlier onset of clinical infection insinuating disease-preventing prophylactic use. It also consists prevention of infections that have not been clinically recognized or their use to inhibit infections in a group of animals, the use of antimicrobials for treating animals is referred to as the therapeutic use of antimicrobials. (Stanossek, I., et al.,2022)

Any antimicrobial agent that is administered as a growth promoter in low doses can destroy or inhibit the development of microbes that reduce animal production as infectious pathogens. A preservative is a stuff that can inhibit, hinder, or prevent the development of microbes or other disastrous effects caused by their presence. (Bekoff, M. 2000).

31.2 Principles of Preventive Medicine

Livestock plays an essential role in the socioeconomic tasks of many smallholder farmers all over the world. These farmers depend upon livestock for their livelihood in their region. (Thornton et al., 2003, Donadeu et al., 2019). Although disease significantly impacts the production and death rate of animals, it affects the income and well-being of millions of people. Furthermore, backward zones are inclined to zoonotic pathogens that originate disease in livestock promoting morbidity and increasing death rate. (Yitbarek, T. 2024)

The use of preventive medicines against disease offers to minimize or terminate the risk for users, and in some instances increase animal productivity. The consumption of vaccines for animal diseases is crucial for decreasing mortality, enhancing health, and increasing productivity, which may good impact on the overall cost ratio. The development of animal vaccines is also important for animal health due to the economic importance of the disease, the challenges related to chemotherapy treatment, and the emergence of antibiotic resistance. The consumption of vaccines is an operative strategy for controlling the disease challenges. (Bekoff, M. 2000).

Vaccination in livestock turns around the principle of reducing disease occurrence and regulating health through dynamic measures, rather than reacting to disease after occurrence. These are some principles of preventive medicines which enhance animal wellness.

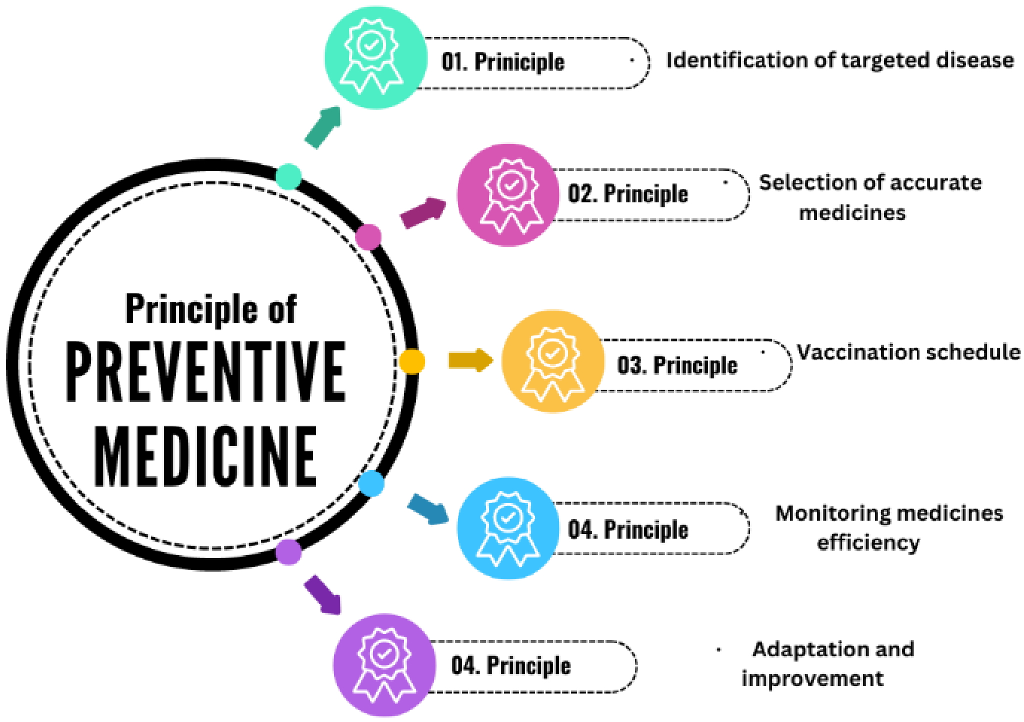

Figure 104 (Principles of Preventive Medicines Which regulate animal production)

31.3 Understanding Disease Pathogenesis

Livestock infectious disease causes major losses to relevant industries and also may cause the spreading of some zoonotic diseases that become a threat to public health. In recent, the epidemic modes of animal disease have become more complex, and new infections regularly emerging, thus increasing standards for prevention and control of these infections. This topic aims to spotlight the recent research, related to the pathogenesis and immune mechanism of livestock diseases which may provide prevention and control against specific diseases. (Robi, D. T, 2024)

Disease pathogenesis in animals implies a complex interrelation between the animal's body and overwhelmed pathogens. Pathogens may be introduced to animals' environments through direct exposure to infected animals, ingestion of contaminated feed additives, or contact with contaminated areas. After exposure to animal pathogens, a mechanism to evade and unsettle the animal's immune system, allowing them to grow inside the body. As pathogens grow rapidly, can destroy the host tissues through different mechanisms. (Pal, A, 2020) For example, bacterial pathogens may assemble toxins that directly destroy cells and intrude into cellular functions, while viral pathogens may expropriate host cells, leading to tissue damage or cell death. Overall understanding the pathogenesis of animal disease is crucial for executing preventive and control strategies, and for developing effective medication for animal health. (Ponamarev, V, 2024)

31.3.1 Risk Assessment and Management

Increasing worldwide development and international dealings contribute to the quick augmentation of animal disease. Initiation of animal disease in immature livestock populations can result in a huge-scale outbreak with serious economic and socio-ethical effects. Most risk assessments executed over the last decennium concentrate on a single disease and single introduction route, commence and label specific risk questions. The risk questions frequently emerge in response to new disease incidents to assess the increased incursion risk from such incidents. (Radostits, O. M, 2007)

The incursion risk of animal diseases is largely intent on by the administration in the world and interrelation of disease-free territory with these areas. These interrelations are the so-called introduction course and either be traded in livestock or their products, trade in non-native animals, relocate wildlife, or movement of people if the disease is zoonotic, or origination of vector if the disease is vector-borne. Data on the worldwide distribution of livestock diseases and the data on introduction routes is hugely accessible from worldwide databases such as WAHIS (World Animal Health Information System). (Smith, B. P, 2014)

Understanding the progressive structure of livestock risk allows the stakeholders to know the intensity of each risk across the supply chain. Therefore, to deal with these situations, it is important to investigate the livestock supply chain-related prospects to decrease their impact on the supply chain and other industry partners. Although this study supervises to addresses the risk management issue for the animal supply chain. (De Vos,, 2020) Risk management has four steps as shown in figure 02:

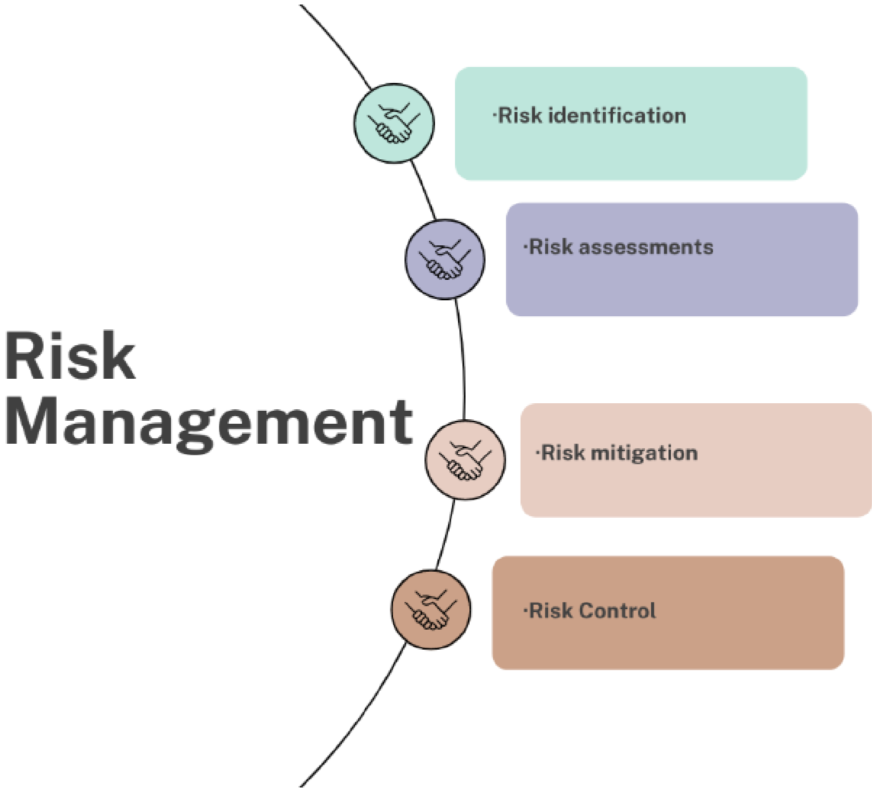

Figure 105 **(Steps of Risk Management)**

Risk identification and assessments are initial and important factors of risk management for the livestock supply chain and the specific objectives are as These. Recognize the serious risks associated with the animal supply chain. Judgment of determined risks using the multiple-criteria decision-making (MCDM) method. Impart the risk control rate based on the risk assessments.

31.3.2 Importance of Nutrition and Diet

Clinical nutrition can be explained as the dietary administration of clinically infirm animals that are targeted as correction of nutritional deficiencies or surpluses, replacement to feed to improve a disease or a disorder that does not have a nutritive cause, and feeding of medicines or nutrients to help the obstruction of disease. Disease and nutrition are closely interrelated in nutritional moderation become an important part of livestock management of the cases assist with an articulation of "Therapeutic diets", that impart optimal health, production, and animal well-being. (Sahoo, A. 2020).

Table 37: Importance of nutrition and diets

Aspects	Description	Benefits

Growth and Development	A proper diet strengthens animal growth and development.	Achieved aimed weight more rapidly, and reduced veterinary costs, enhance overall production.
Immune System	A balanced diet supports the immune system.	Reducing the risk of disease, and lowering the cost help to reduce the economy.
Reproductive Performance	Feed additives improve reproductive performance.	High rate of fertility, Higher production, and decreased calving difficulties.
Disease Resistance	Some nutrients can aid in preventing specific diseases.	Lower the number of diseases correlate to nutrition such as ketosis or milk fever.
Economic Efficiency	Efficient feed consumption reduces feed costs and increases animal efficiency.	Enhance milk production, egg productivity, and development rates. Higher profit margin, and better returns on investing money
Energy and Productivity	Enough energy intake helps a high level of production.	Increase overall production and enhance animal welfare.

This table highlights how well-managed feed and proper nutrition can be provided as preventive medicine, influencing the overall health, production, and welfare of animals health while also benefiting the environment ecologically and economically.

31.4 Environmental Factors and Their Impact on Health

Environmental factors play a vital role in the health and well-being of animals. Some factors are given below which may impact on animal's health. (Humboldt-Dachroeden, 2021)

Space and Housing:

Populated or imperfectly designed housing facilities can enhance stress levels among animals and assist the spread of disease. Providing adequate space, conventional flooring, and comfortable shelter regions for improving good health and welfare.

Social Environment:

Social interaction within the herd or group can affect the stress level and etiquette of animals. Pugnacity, Social order disputations, and overcrowding can assist in injury which decreases overall livestock production.

Temperature and Humidity:

Animals are sensitive to harsh conditions such as high temperature and humidity. High temperatures can assist in heat stress, which decreases feed increment, impairs reproductive functions, and enhances vulnerability to disease.

Air Quality:

Low air quality in animal facilities, usually due to poor ventilation, can result to respiratory damage to animals such as pneumonia and bronchitis, which may decrease overall livestock production.

Water Quality:

Clean and sufficient amounts of water are crucial for animals' health. Contaminated water may spread diseases and other toxins to animals, leading to gastrointestinal issues, dehydration and increased mortality.

Biosecurity Measures:

Applying strict biosecurity measures is important for preventing the interconnection and spread of disease within the animal population. Proper sanitation, insulation, and control of outsiders and equipment can assist in reducing disease risk and maintaining animal growth. (Humboldt-Dachroeden, 2021)

31.5 Integrative Veterinary Care

Integrative veterinary care is defined as a coalition of compatible therapies with conventional care, which is instructed by the best research available. With growing interest in complementary therapies by men has induced inquiries and use of these medications in animals. In addition, IVC includes rehabilitation, laser and ultrasound therapies, acupuncture, integrative feeds, and herbal therapies. (Memon, M. A. 2023)

31.5.1 Traditional Veterinary Medicine vs. Holistic Approaches

Traditional veterinary Medicines and Holistic approaches indicate two different models in healthcare, with their own principles, procedures, and techniques. Traditional veterinary medicines highly depend upon scientific reports, diagnostic tests, and pharmaceutical intermediation. It generally gives attention to treating certain signs or diseases with remedies, surgeries, and other methods in animals. Veterinarians usually specialize in some areas such as internal medicine, surgery, and pathology. They give attention to preventive measures such as vaccination, deworming, drenching, dehelmintization, and proper check to maintain animal health for better production. TVM can equipped to face emergencies and acute medical conditions with modern diagnostic tools and techniques. (Pun, J. K. 2020)

As above we already discussed that holistic veterinary medicines are suggested for the whole animal, and consist of physical, mental, emotional, and environmental factors. It contains natural therapies such as acupuncture, herbal medicines, chiropractic adaptation, and feed supplements. It also focuses on decreasing stress and promoting animal health, through procedures like; massage, aromatherapy, and behavior modification. Treatments are modified according to the animal's needs to maintain optimal health. (Memon, M. A. 2023)

Both have their own merits, and the selection between them usually depends on components such as the general health conditions of the animal, the priority of the owner, and the accessibility of resources. In some cases, both TVM and HVM may be used against some diseases to provide comprehensive care, ultimately both promote good animal health and increase production which may reduce economic losses. (Khan, A. 2023)

31.5.2 Role of Alternative Medicine in Preventive Care

Alternative medicines in preventive animal care are sophisticated approaches that prefer holistic well-being and natural medications over conventional pharmaceutical complexes. (Khan, A. 2023) As shown in figure 03.

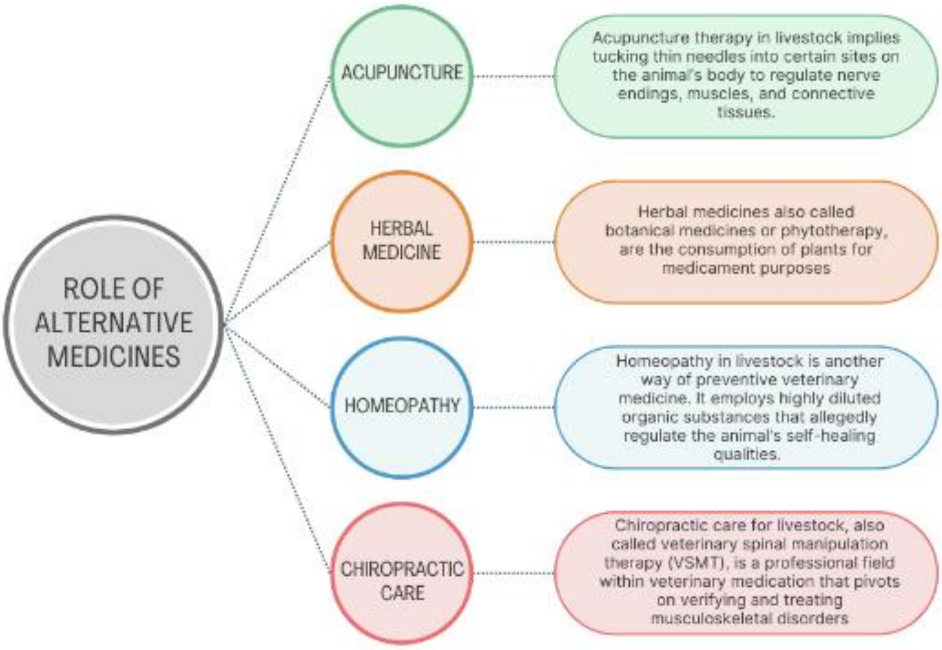

Figure 106 Role of Alternative Medicine in Preventive care in livestock

31.5.3 Acupuncture

Acupuncture therapy in livestock implies tucking thin needles into certain sites on the animal's body to regulate nerve endings, muscles, and connective tissues. (Medina, C et al,. 2024) The earlier practice, Handled by Traditional Chinese medicines, has attained vogue in veterinary medicines as an alternative therapy for different conditions such as Pain Management, Reproductive health, Digestive disorders, Immune support, and Performance enhancement. (Gong, J et al,. 2024)

31.5.4 Herbal Medicine

Herbal medicines also called botanical medicines or phytotherapy, are the consumption of plants for medicament purposes. It's one of the earliest forms of healthcare known to humans, back thousands of years, Herbal medicines use different components of plants, including leaves, roots stems, flowers, fruits, and seeds for their therapeutic abilities. Many plants contain a vast assemblage of chemical compounds that can have medical effects on animal health. Some famous herbs used in herbal medicines are Echinacea, Ginseng, chamomile, ginger, turmeric, and garlic. (To, K. L. 2024).

31.5.5 Homeopathy

Homeopathy in livestock is another way of preventive veterinary medicine. It employs highly diluted organic substances that allegedly regulate the animal's self-healing qualities. It increases overall health and production without any other side effects. Some farmers merge homeopathic medication with holistic livestock health care, improving treatment and prevention procedures. (To, K. L. 2024).

31.5.6 Chiropractic Care

Chiropractic care for livestock, also called veterinary spinal manipulation therapy (VSMT), is a professional field within veterinary medication that pivots on verifying and treating musculoskeletal disorders in livestock. while it's mostly linked with pets and horses, chiropractic medication can also be given to livestock such as cattle and sheep. (Vredegoor, et al,. 2016)

31.6 Holistic Approaches to Common Health Issues

Holistic approaches to common health issues in livestock enclose a vast range of approaches focusing on enhancing animal health, averting disease, and managing ailments using natural ways whenever possible. We discussed the above holistic approaches identified as usually associated with and influenced by many factors, including nutrition, genetics, management practices, and environmental conditions. Slightly than entirely focused on handling symptoms or common diseases, a holistic approach targets recognizing underlying causes and contributing elements. (Haussler, et al., 2007) Farmers and veterinarians can make targets and effective strategies to prevent and control health issues by taking a comprehensive and unified view. For example, as a substitute for simply treating a cow with antibiotics for puerperal sepsis, A holistic approach may examine the cow’s diet, milking techniques, and living conditions to recognize potential contributing elements, such as poor nutrition, inadequate sanitation, or excessive stress. (LeBlanc, et al., 2006)

31.6.1 Allergies and Skin Conditions

Skin infections or allergies in animals may arise with the extra usage of antibiotics, The consumption of antimicrobial agents enhances the pervasiveness of resistant bacterial strains. The quick emergence and proliferation of resistant bacteria is a worldwide issue. Allergies and skin conditions in animals are types of inflammations expressed after some foreign particles interact with animals' bodies, Due to its large area, the skin offers many injuries possibly attained by the introduction of infective agents to the animal’s body. A holistic approach can manage different allergic or skin conditions to promote good health or production. (Sharma, A., & Gupta, S.2024) There are numerous factors contributing good health of animals which are the following,

- Identify triggers

- Dietary changes
- Supplements
- Stress management
- Skin care routinely
- Natural remedies
- Less consumption of Antibiotics
- Professional guidance

31.6.2 Digestive Disorders

Animals rely on their Gastrointestinal tract, and a healthy GIT can describe a healthy animal. Gut health is associated with feed additives that animals digest for their metabolic functions. Animals' function and production are highly dependent on healthy GIT. Some disorders such as Diarrhea including clogged heart failure may be treated with holistic approaches for promoting good health in livestock. (Loy, J. D et al., 2023)

31.6.3 Stress and Anxiety Management

Identification of pain or stress or their severeness is hard to determine in livestock, which is a major challenge in dairy cattle because cattle have grown as prey animals. There are two types of pain in livestock which are: Surgical pain or disease pain. Both types of pain can potent a bad effect on animal health. (Edwards, et al., 2023) Stress can decrease the resistance level against many pathogens which cause many other diseases, both pain and stress may be treated with the help of holistic medicines to stimulate health and increase the overall production of animals. (Bomzon, A. (2011).

31.7 Preventive Healthcare Protocols

The biggest advance in livestock health in the last few years has been ideally the switch from treatment of clinical disease to preventive measures. Some preventive healthcare protocols are included**.** Vaccination Programs, Parasitic Control, Behavioral Management, Exercise and Physical Activity. All these factors may enhance animal health which increases animal production as well as increasing the economy. (Youssef, F. S et al., 2023) We discussed all these preventive healthcare protocols broadly.

31.7.1 Vaccination Programs

By vaccination, many diseases can be controlled at an earlier stage in livestock. A sheep, goat, and bovine are protected by the process of vaccine against hydatid disease. These diseases can be caused by infection at the larval stage of Echinococcus granuloses the major cause of subclinical mastitis, ketosis, rumen acidosis, and endometritis. These infections increase the risk factor of causing disease, through this the success of milk and meat production in livestock is reduced. (Blair, S. N et al., 1985) The extent of milk fever, clinical respiratory infections, clinical parasitism, and bovine virus diarrhea have been treated and increased production through the process of vaccination. Major or common diseases can be controlled through vaccination programs. Vaccines are a potent tool for the control of infectious diseases in livestock.

31.7.2 Parasite Control and Prevention

Parasitism is a contagious disease caused by external agents such as ticks, flies, lice, protozoa, and helminths. Furthermore, Gastrointestinal parasites can influence animal husbandry, and, health triggering discomfort pain, suffering affliction, in drastic cases, death. Consequences that lead to significant economic losses in the grazing system. The control depends on a complex interaction of environmental factors, animal and human. Helminth infections are pervasive in grazing ruminants and cause remarkable costs due to production losses. In addition, anthelmintic resistance in parasites is now extensive throughout Europe and a major threat to ruminant livestock farming. The Targeted Selective treatments were present to reduce the stock treated within a flock. They are classified into pathophysiological markers (such as anemia and Dag score) and parasite-based markers. Breeding livestock have greater immunity against different parasite tenders a green strategy. (Ahmad S, et al., 2023)

31.7.3 Exercise and Physical Activity

Exercise and physical activity may incidentally affect health behaviors such as overgrazing, overeating, stuff abuse, risk-taking, rest management, and some other factors. Considerable evidence specifies that physical activity positively impacts weight control and calorie intake, enhancing overall animal growth and production. (Kim S.O et al., 2024)

31.8 Implementing Holistic Practices in Veterinary Clinics:

Implementing holistic practices in Veterinary clinics, first veterinary workers must undergo coaching substitute therapies as we discussed above. Next, merge these practices into extant medication protocols, providing them alongside conventional remedies. Evolve panoramic health judgment considering emotional, physical, and environmental factors infecting animals. Motivate open communication with pet holders to understand their priorities and concerns, providing a cooperative approach to animal care, for the best possible outcomes for animal patients. (Lakshmi, K. 2023)

31.8.1 Staff Training and Education

Staff training and education in holistic approaches to animal administration to relate to comprehensive strategies designed to furnish individuals with knowledge and skill in viable and integrating. These projects accentuate the interrelation between animal health, environmental supervision, and socio-economic factors influencing practices such as rotatory grazing, biodiversity maintenance, and natural treatments for livestock health. Training may include hands-on practice, theoretical workshops, and field courses to secure practical comprehension and application. By encouraging holistic possibilities, this education aims to enhance animal welfare, increase farm productivity, and bestow to an unending agriculture ecosystem. (S Shaw, J. R., & Coe, J. B. 2024)

31.8.2 Client Communication and Education

Client communication in holistic livestock management entails informing clients about beneficial practices and their procedures through newsletters, hands-on practices, workshops, and discussions. These attempts may enhance the benefits of methods like rotatory grazing and natural health remedies for increasing animal health and farm production. Educational content, consisting of case learns and practical helping material, points out the economic and ecological advantages. By aiding transparent, associated relationships, these enterprises certify clients to embrace and support holistic approaches, providing for more adaptable and sustainable agricultural systems. (Shaw, J. R., & Coe, J. B.2024)

31.9 Conclusion:

The future direction in holistic veterinary medicine is likely to be described by an extensive integration of various modalities targeted at addressing the health and well-being of the livestock from a comprehensive possibility, The assuming of telemedicine and telehealth manifesto can ease the controlled consultation and observation-making the holistic approach more approachable to pet owners. Through a growing interest in holistic medicines, research grows the efficacy and safety of complementary therapies for livestock, giving evidence-based support for their consumption. Educational opportunities for veterinary students may expand to ensure that future generations may have broader aspects of health and well-being.

There are many challenging barriers in holistic adoption care including the insufficiency of standardized protocols, Limited research on cost, efficiency, and resistance from typical practitioners. There is also a lack of information about performing livestock procedures and techniques. Specialized training for farmers in rural areas may also pose a challenge. Furthermore, economic pressure and industry highlight production over holistic well-being and impede widespread adoption.

Advances in holistic animal therapies consist of pure herbal formulation, comprehension of acupuncture’s efficiency, and initiating implementation of chiropractic care, Nutritional approaches as nutrigenomics be revealed. Veterinarians are inspecting holistic methods to stimulate livestock health and production while decreasing dependence on traditional medicines.

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