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Original Review Article

EARTHWORMS AS A BIO INDICATOR FOR ASSESSING SOIL TOXICITY: A REVIEW ON IMPACT IN AGRICULTURE

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RECEIVED:
5/09/2024

ACCEPTED FOR PUBLICATION:
15/10/2024

PUBLISHED ON:
27/10/2024

Cite as: Jain et al., (2024) Earthworms as a Bio Indicator for Assessing Soil Toxicity: A Review on Impact in Agriculture JABAAS Vol 2(3) pp120-134

ABSTRACT

This review article is an attempt to study the impact of toxicity in the realm of insecticides and pesticides which is very dangerous to target (earthworm) and non-target (other than earthworm) organisms and lower LC50 leads to death of earthworms which is important for nutrients rotation, that's why overall productivity of agriculture crops may go down. Various pesticides directly or indirectly pollute air, water, soil, and the overall ecosystem, which causes serious health hazards to living beings.

We have discussed in this review article about understanding of different kinds of toxicity levels and their impact on targeted invertebrates. The author also discussed tools to assess heavy metal toxicity as a biomarker e.g. total protein assay, metallothionein (MT), catalase and lipoxigenase (LOX), and glutathione (GSH) assay. Other alternatives to control crop loss due to pest attacks may include the application of various biopesticides. Another way of avoiding pesticide use includes developing some crop varieties that are resistant to some pests through the use of transgenic approaches.

Keywords: Pesticides, Insecticides, Toxicity, LD50, LOX and GSH.

I. INTRODUCTION

Historians have identified the use of pesticides as far back as the time of Homer around 1000 B.C. Nevertheless, the earliest recorded cases of insecticides are linked to the use of burning brimstone (sulfur) as a fumigant. Insecticide options were scarce at the beginning of World War II, however, by the end of the war, a fresh approach to insect control had developed thanks to the introduction of modern chemical compounds. DDT was the first synthetic organic insecticide to be introduced. Insecticides, whether chemical or biological, have long been used to control insect populations. Attaining this control can be done through the elimination of insects or by stopping them from carrying out harmful actions. Insecticides can be classified as either naturally derived or synthetic and are applied to combat pests using a range of delivery methods, such as spraying, baiting, slow-release mechanisms, diffusion, and more (Stejskal et al. 2021).

Pesticides are categorized as naturally occurring toxic compounds that not only target harmful organisms but also impact beneficial ones. Typically, they possess high toxicity levels and have the potential to accumulate in the food chain. Pesticides undergo rigorous toxicological assessments, and those deemed highly harmful are regularly banned from usage. The utilization of pesticides is governed by regulations outlined in directives, acts, and national, EU, and international resolutions.

These substances must adhere to stringent standards to ensure the safety of animals, the environment, and most importantly, human beings (Izbicki et al. 2024). The widespread utilization of pesticides across various sectors of the economy has led to significant repercussions, including the emergence of toxic effects on living organisms, the eutrophication of water reservoirs, and the decline in soil fertility. The extensive application of pesticides has resulted in environmental pollution across all aspects.

The application of pesticides is now a fundamental component of our present-day society to cater to the requirements of a swiftly increasing population, estimated to hit 10 billion by 2050 (Saravi and Shokrza-deh 2011; Edwards and Bohlen 1992). Over the course of the previous decade, an estimated According to the Pan-Germany report in 2012, the global expenditure

on pesticides amounted to a staggering \$38 billion. A considerable amount of these pesticides builds up in the soil, and the continuous use of them may result in adverse effects. The presence of pesticides in the organo mineral components of complex structures has a significant impact on several processes, including mobilization, immobilization, bioavailability, and transport (Edwards and Bohlen 1992).

They conducted a study to examine the impact of different pesticides on earthworms. It was discovered that the pesticides had sub-lethal consequences, causing the earthworms' cuticle to rupture, the release of coelomic fluid, body swelling, paleness, and softening of tissues. Alternative research has also shown that these pesticides can lead to cellular autolysis (Edwards and Bohlen 1992), damage to the male reproductive system (Edwards and Bohlen 1992), swelling (Edwards and Bohlen 1992), and coiling of the tail (Wang et al., 2012). Both high and low doses of insecticides have been found to cause physiological harm, such as cellular dysfunction and protein catabolism, to earthworms (Edwards and Bohlen 1992).

Approximately 45% of the annual crop production is lost due to infestation of various pests; therefore, effective management of pests using a wide range of pesticides becomes important in confronting pests and increasing the production of crops. However, due to the heavy use of pesticides, soil-targeted organisms such as earthworms are going to die thus impacting overall crop productivity.

Earthworms are known to have numerous positive effects on soil quality, particularly for crop production such as improvement in soil aeration, structural change, nutrient cycling, water movement, and plant growth (Baskar et al. 2023). Earthworms are important decomposers of organic matter, in obtaining their nutrition from organic matter and soil material. As they move through the soil, earthworms create tubular channels or burrows that can persist for a long time.

Soil fertility refers to the ability of a specific type of soil to support plant and animal productivity, maintain water and air quality, and promote human health and habitation within the boundaries of natural or managed ecosystems (A and Entoori 2022). These burrows increase the soil porosity, allowing more air and water to percolate in the soil. The increased porosity also reduces the bulk density and thus promotes the root development. Furthermore, earthworm

excrement or casts contribute to soil fertility as they contain essential nutrients such as nitrogen, phosphorus, potassium, and magnesium. These casts also harbor microorganisms that multiply as they digest organic matter in the earthworms' intestines (Edwards and Arancon 2022).

Earthworms make up a significant portion (80% of soil) of the biomass of terrestrial invertebrates. As a result, they can serve as effective bio-indicators for chemical contamination in soil within terrestrial ecosystems. By acting as an early warning system, they are helpful in identifying any decline in soil quality. Furthermore, the soil polluted by toxic pesticides, namely Cu, Fe, Pb, Mn, etc. causes harmful impacts on different invertebrates (Miglani and Bisht 2019).

Research has indicated that the skin of earthworms absorbs heavy metal contaminants (Singh et al. 2024) and the extent of the contamination depends on the specific chemical makeup of the pesticides and the characteristics of the soil (Singh et al. 2024). As a result, the organisms' organs experience a sequence of chemical reactions, encompassing pathways, transportation, adsorption, and desorption that help in toxicity removal from the earth up to some extent.

2. MATERIALS AND METHODS

The author has utilized various web resources such as Science Direct, Web of Science, Google Scholar, and Mendeley for curating data on earthworm toxicity and their impacts, over the earthworm by curating LD 50 and LC 50 data, to access their impact on agriculture productivity.

3. RESULTS

1) The toxicity and lethal effects of pesticides on earthworms

The effect of pesticide is determined by the amount administered to an organism by the LC50 doses (also known as the Fifty percent lethal concentration, denotes the amount of pesticide that is discharged into the air and is quantified in milligrams per liter). A higher level of lethality for the pesticide is indicated by a lower LC50 value. In 2018, the Canadian Center

for Occupational Health and Safety stated that the LD50 values are measured in milligrams per kilogram of body weight (Hendriks et al. 2013).

Based on research (Edwards and Bohlen 1992), earthworms are extremely susceptible to pesticides, especially insecticides. For instance, Neonicotinoids, Strobilurins, Sulfonylureas, Triazoles, Carbamates, and Organophosphates, Earthworms are recognized to have negative effects, and thus mortality is high (Edwards and Bohlen 1992).

Pesticides can cause direct harm by changing their physiological functions (Kaka et al. 2021). At elevated levels of pesticides exceeding 25mg/kg Earthworm survival and reproduction are negatively impacted.

Factors such as species of earthworms, contaminant type, and pesticide concentration greatly impact earthworm's life (Rodriguez-Campos et al. 2014) (see fig. 1). Persistent nature of pesticides causes the death of micro and macro fauna. The water table is also affected.

Various classes of pesticides over *Eisenia fetida* experiences shown to have various adverse effects such as varying doses, which impact various stages of the reproduction cycle (including cocoon production, number of hatchlings per cocoon, and incubation period) in a dose-dependent manner (Arachchige et al. 2024; Yasmin and D'Souza 2010). For instance, the toxicity of di-methyldithiocarbamate on *Eisenia fetida* (Earthworms) was assessed by (Arachchige et al. 2024) at LC50 value of 12.636 mg/L in soil. In another earthworm, *Aporrectodea giardia* species has the potential to be a strong contender for assessing the Ecotoxicological risks of soils that have been contaminated with a combination of metals.

2) Ecological Group in Earthworms

There are four ecological groups in which earthworms are classified, and every group is characterized by unique characteristics present in the soil environment. These traits, as described by (Fründ et al. 2010) also encompass the earthworms' susceptibility to different types of pesticides.

Epigeic worms, such as *Lumbricus rubellus*, *Dendrobaena octedra*, and *Lumbricus castaneus*, are typically found in the upper 10-15 cm layer of

soil. They primarily consume decomposing organic material present in the leaf litter. They are extremely vulnerable to pesticides due to their susceptibility.

On the other hand, *Aporrectodea caliginosa*, *Allolobophora chlorotica*, and *Allolobophora icterica* are relatively large, measuring between 1 and 20 cm in size. These earthworms consume organic material that is combined with minerals in the soil, particularly in areas where pesticides have infiltrated and mingled with the soil.

Anecic earthworms such as *Lumbricus terrestris* and *Aporrectodea longa*, exist even larger colored, and pigmented. Their muscles are well-developed and are highly active burrowers. Certain species have the potential to reach sizes ranging from 10 to 110 cm. The worms predominantly consume organic matter found on the surface, particularly at night. They construct extensive sub-vertical tunnels (ranging from 1 to 6 meters) and consequently consume a greater quantity of soil. As a result, they are exposed to pesticides through the consumption of soil contaminated with harmful chemicals.

Compost worms, such as *Eisenia fetida* and *Dendrobaena veneta*, are frequently utilized in vermicomposting techniques. The worms possess a vibrant red hue accompanied by distinct stripes, hence earning them the moniker of “tiger worms.” They are typically kept in controlled soil pits, which reduces their exposure to soil toxicants.

3) Insecticide, Pesticide, and effect on earthworm

Global utilization of pesticides can be understood in Fig. 2 (a) where in the pie chart we can see that 60% of pesticides are globally utilized in the form of insecticides. **In Fig. 2 (b)** In the pie chart earthworm is affected mostly by Herbicides (47%) which is the highest, while the effect of Insecticide is (29%) and other insecticides containing 18% & 6% as fungicides and others respectively.

4) Pesticide categorization

Insecticides are classified into various groups derived from their chemical composition, based on the 2016 report by the Insecticide Resistance Action Committee (IRAC). **Pesticides are classified into** different categories including a) Insecticides, b) Fungicides, c) Herbicides, d) Rodenticides, e) Nem-

atocides, f) Molluscicides, and g) Plant growth regulators (Cortet et al. 2002). According to a global estimate, the annual expenditure on pesticides amounts to \$38 billion (2012). which has been increase to 3.5 million tonnes in 2020 (Sharma et al. 2019).

Unfortunately, the majority of pesticides lack specificity and ultimately kill all those innocent organisms that play a vital role in diverse soil ecosystems. During development, the utilization of sophisticated agricultural methods has led to the buildup of toxic substances in both humans and a variety of other animals.

Before the Industrial Revolution, farming practices were environmentally friendly, and there was a strong connection between agriculture and ecology. However, with the onset of the Industrial Revolution in the early to mid-1900s, this connection was disregarded, leading to increased productivity at the expense of environmental quality. As a result, ensuring the safety of agro-ecosystems has become a challenging task, with adverse effects on soil health.

The presence of soil biota, including earthworms, is vital for soil productivity and the sustainable functioning of ecosystems. Nutrient cycling, in particular, is a critical function that is essential for life on Earth (A and Entoori 2022). Soil fauna, including earthworms, play a crucial role in soil carbon storage, nutrient cycling, and hydrology, which ultimately affect soil quality and should be considered in land management strategies. Soil is an essential element of terrestrial ecosystems and is frequently acknowledged as the “Biological powerhouse of the planet” (Shaaban and Nunez-Delgado 2024).

By taking a zoological approach to soil quality assessment, we can gain a deeper understanding of the complex interactions between soil ecosystems and crop productivity. This knowledge can contribute to the development of sustainable agricultural and environmental practices (Vasseur and Bonnard 2014).

Earthworms are associated with increased carbon and nitrogen soil contents and provide a higher diversity of niches for microorganisms through bioturbation via their mucus secretion that enhances the metabolism of plant-growth-promoting soil microorganisms and soil biocontrol microbial agents.

Changes in soil microbial community composition due to earthworm presence influence soil chemical-physical parameters, decomposition rates, and plant growth, particularly in the root system.

5) Earthworm changes the structure of the soil bacterial community & Crop productivity

Earthworms had a significant impact on the structure of the soil bacterial community, increasing alpha and beta-diversity and 16S rRNA gene abundance. As a result, Earthworms enhanced microbial decomposition rates of organic matter in soil. They change community structure. Earthworms influenced soil chemico-physical parameters, such as soil macro porosity, soil water content, and soil pH Earthworms acted as natural fertilizers, enriching the soil with nutrients and microorganisms (Brown et al. 1999).

An earthworm biomass exceeding 30 g/m² was demonstrated to be essential to enhance grain yield by more than 40%, which is agriculturally significant. (Brown et al. 1999).

Farmers can achieve this by adopting practices that promote earthworm activity, such as reducing tillage, minimizing chemical inputs, and incorporating organic matter into the soil.

6) Role of Earthworms in plant defense

Earthworms play a crucial role in aiding plants to combat parasitic nematode attacks. They achieve this by reducing the density of nematode populations improving the plants' ability to withstand these parasites and promoting the growth of beneficial microbes that counteract root pathogens (Jana et al. 2010).

Earthworms were found to enhance plant growth by 20% and increase nitrogen content by 11%. However, their presence did not impact plant resistance against chewing herbivores, such as caterpillars, slugs, and rootworms. Earthworms were associated with a 22% decrease in plant resistance against phloem-feeding herbivores like aphids (Xiao et al. 2018).

The degree of pesticide toxicity is significantly affected by temperature. In a study conducted by (Edwards and Bohlen 1992), the toxicities of Abamec-

tin and Carbendazim were examined under freezing temperatures. (Bindesbøl et al. 2009) investigated the impact of the effects of temperature and soil composition on the toxicities of Chlorpyrifos and Carbofuran. (Bindesbøl et al. 2009) conducted a study on the impact of Carbaryl in varying temperature conditions, including both low and high temperatures.

On the other hand, (Bindesbøl et al. 2009) examined the avoidance behavior of three pesticides in temperate and tropical conditions. The conducted investigations have unveiled that alterations in temperature can have an impact on the toxicity of pesticides. However, it is important to note that the results obtained from these studies were not definitive and lacked support from other research (Fig. 3).

Numerous research studies have been conducted on the neurotoxic effects induced by different insecticides, specifically, the chemicals Neonicotinoidimidacloprid, Oxadiazineindoxacarb, and Pyrethroids alpha-cypermethrin and Lambda-cyhalothrin are commonly used in pest control. In addition to that combined impact of Organophosphate chlorpyrifos and Pyrethroidcypermethrin.

All of these insecticides primarily impact the nervous system. Neonicotinoids disrupt the transmission of stimuli in the nervous system by causing irreversible blockage of acetylcholine receptors. Oxadiazines act as blockers of voltage-gated sodium channels. Pyrethroids stimulate the sodium and potassium channels of neurons and delay the closing of these channels during depolarization. Organophosphates inhibit the action of the enzyme acetyl cholinesterase, leading to the accumulation of acetylcholine, excessive stimulation of cholinergic receptors, and disruption of neural activity. (Edwards and Bohlen 1992) conducted a study that found that higher concentrations of Carbaryl is a chemical compound commonly used as a pesticide. (50 kg/ha) decrease protein content and antioxidant enzymes glutathione-S-transferase (GST). Antibiotics, carbamates, and organophosphates induce an intermediate toxicity response in earthworms. (Bindesbøl et al. 2009) reported that neonicotinoids are the most toxic to *Eisenia fetida* among the six chemical classes, followed by pyrethroids, while IGRs exhibit the lowest toxicity. Organophosphates are not highly toxic to earthworms. Considering the high efficacy

of neonicotinoids against target organisms, environmental managers should carefully evaluate their use in integrated pest management (IPM) programs to avoid causing serious damage to earthworms.

7) Impact of insecticides on earthworm growth and reproduction

Different reproductive parameters, including maturation, cocoon production, viability, hatching, and sperm production, were investigated about genotox-

icity when exposed to various types of insecticides and other chemical classes (Wang et al. 2012; Edwards and Bohlen 1992) conducted a study on the effects of Chlorpyrifos, an organophosphate insecticide, was examined over a period of 7, 14, 21, 28, and 35 days.

It was found that dose concentrations of 0.1 and 0.2 had minimal effects on growth during the first 7 and 14 days of exposure but had a detrimental effect on earthworm progress is seen after a period exceeding two weeks.

Table 1 Type of pesticide has specific mechanisms of action that target the biological processes unique to the target pests. Understanding these actions is crucial for effective and safe pesticide use in agricultural industrial and residential settings.

S. No.	Types of Pesticides	Use and Action	Examples
01.	Insecticides	A substance employed to manage, eradicate, or avert the onslaught of insects that cause harm, exterminate, or alleviate damage to plants or animals.	DDT, methyl parathion, Phorate, Chlorpyrifos, Imidacloprid, Cypermethrin, and Dimethoate are all types of pesticides.
02.	Herbicides	Chemicals are employed to manage the harmful weeds and other plants that are growing alongside the desired species, resulting in inadequate plant development.	Acetochlor, Butachlor, Terbis, Glyphosate, 2,4-D, and 2,4,5-T are commonly used herbicides in agriculture.
03.	Fungicides	Chemicals are employed to eradicate or impede the proliferation of fungi/ailments that afflict vegetation/creatures.	Carbendazim, Ampropylfos, and Carboxin are three types of fungicides commonly used in agriculture.
04.	Rodenticides	Rodenticides are substances utilized to exterminate rodents such as mice and rats.	Warfarin and arsenic oxide.
05.	Nematicides	Substances are employed to deter or hinder the nematodes that harm different crops.	Aldicarb and Carbofuran are two types of pesticides.

Table 2. The impact of soil pesticides on earthworms can have indirect consequences on crop productivity as they play a crucial role in soil health and ecosystem functioning that directly support plant growth. Preserving the earthworm population can sustainably enhance crop productivity.

Pesticide	Concentration of pesticide/exposure	Test conditions	Species	Responses	Reference
Copper oxychloride (pure)	8.92, 15.92, 39.47, 108.72, 346.85 mg Cu/kg substrate	Substrate = Dried, ground, finely sieved cattle manure 56 days Temp = 25°C	<i>Eisenia fetida</i> (Freshly hatched earthworms)	Earthworm growth and cocoon production were significantly reduced	(Römbke, Garcia, and Scheffczyk 2007)
Malathion (pure)	80.150, 300, 600 mg/kg soil 1, 5, 15, 30 days	Soil like substrate pH = 6.5, Temp = °C, Moisture = 50%,	<i>Eisenia fetida</i> (Adults)	Substantial decrease in body mass lowered sperm quality.	(Römbke, Garcia, and Scheffczyk 2007)
Ace-tochlor	5, The soil was treated with doses of 10, 20, 40, and 80 mg/kg. The growth of the plants was observed at intervals of 7, 15, 30, 45, and 60 days. Additionally, the plants were monitored for 28 days. (Reproduction)	OECD artificial soil The artificial soil developed by the OECD has a pH level that is yet to be specified. It maintains a moisture content of 50% and is kept at a temperature of 20°C	<i>Eisenia fetida</i> [grown-ups]	Adverse impact on growth and reproduction	(Römbke, Garcia, and Scheffczyk 2007)
Cypermethrin (pure)	5, 10, 20, The soil was treated with 40 to 60 milligrams per kilogram, and the experiment lasted for 4 to 8 weeks.	The artificial soil developed by the OECD exhibits a pH level that is yet to be specified. Additionally, it maintains a moisture content of 50% and is subjected to a temperature of 20°C.	<i>Eisenia fetida</i> (Juveniles)	Significant reduction in cocoon production Juveniles more sensitive than adults	(Römbke, Garcia, and Scheffczyk 2007)
Benomyl (pure)	0.32, 1.0, 3.2, 10, 32 mg per kg soil 56 days	The artificial soil developed by the OECD Temp = °C The pH value is 6.1 Moisture = 56% LUFA 2.2	<i>Eisenia fetida</i> (Adults)	The toxicity of the concentration of benomyl was found to be lower in artificial soils of tropical regions compared to temperate regions. Furthermore, there was no reproduction of benomyl in natural tropical soil due to its low pH levels.	(Römbke, Garcia, and Scheffczyk 2007)

(Wang et al. 2012) observed a notable decline in the growth of *A. caliginosa* when subjected to two organophosphates, specifically diazinon, and chlorpyrifos, at a dosage of 60 and 28 kg/ha, respectively. Similarly, (Edwards and Bohlen 1992) noted that. Earthworms were highly susceptible to Methyl parathion and phorate, exhibiting the manifestation

of toxicity gradually presents various symptoms, including coiling, curling, excessive production of mucus, slow movements, enlargement of the clitellum, deterioration in the nervous system, and fading of pigmentation. These effects were induced using organophosphorus insecticides.

The use of **Malathion, an organophos-**

phate, resulted in a substantial decrease. The increase in body weight adversely affected the male reproductive organs (Raafat et al. 2012). The observation of changes in cell proliferation and DNA structure of spermatogonia in earthworms was documented by (Wang et al. 2012).

Additionally, the sensitivity of sperm counts as an indicator has been recognized by (Wang et al. 2012), and Not only can the sperm count be influenced by Malathion, but the quality of sperm can also be affected by its metabolites (Wang et al. 2012). (Wang et al. 2012) conducted the study suggested the reduction in body mass observed as a potential sign of decreased food consumption could be indicated by this. The capacity of earthworms to regulate their pesticide exposure enables them to effectively control their growth inhibition.

The Impact of various pesticides on the growth and reproduction of earthworms has been extensively studied by different researchers. In a study conducted by (Wang et al. 2012), it was found that Aldicarb, cypermethrin, profenofos, chlorfluazuron, atrazine, endosulfan, and metalaxyl all types of pesticides commonly used in agriculture all had a detrimental effect on the growth rate of *Aporrectodea caliginosa* and *Lumbricus terrestris*.

Similarly, (Edwards and Bohlen 1992) discovered Chlorpyrifos is a pesticide that negatively affected the growth of earthworms exposed to a concentration of 5 kilograms per hectare after eight weeks. Furthermore, it has been observed the proliferation of earthworms is significantly more pronounced during their adolescent phase compared to the mature phase.

Another study by (Edwards and Bohlen 1992) revealed that chlorpyrifos exposure also significantly affected the reproduction of earthworms, particularly in terms of fecundity. Additionally, (Edwards and Bohlen 1992) found that cypermethrin had a greater adverse impact on the earthworms' reproductive cycle during their juvenile stage compared to the adult stage, with the application rate is 20 kilograms per hectare causing substantial harmful impacts. These are just a few examples of the numerous researchers in this domain who have examined the impacts and reactions that have been explored.

4. ROLE OF BIOPESTICIDE AND TRANS-GENIC CROPS

Various types of Biopesticide can be a good replacement for pesticides.

Types of Biopesticides (See Fig 4)

1. Microbial Pesticides
2. Biochemical Pesticides
3. Insect Growth Regulators
4. Insect Pheromones
5. Essential Oils, and Plant-Based Extracts
6. GMO Products
7. Biopesticides from Cyanobacterial and Algal Sources
8. RNAi-based biopesticides

There are several types of biopesticides, (refer Table 3) which is discussed below.

Biopesticide can be classified based on their natural sources, the mode of extraction, and sometimes even molecular or componential applications in their production.

1. Microbial Pesticides

These bio-chemical substances originate from microorganisms such as fungi, viruses, and bacteria and are known as bio-insecticides. Bioinsecticides are used to destroy the insects that damage crops. Bioherbicides are secreted from several bacterial species like entomopathogens including *Paecilomyces*, *Hirsutella*, *Verticillium*, *Lecanicillium*, *Metarhizium*, and *Beauveria*.

The successful use of Bt and some other microbial species led to the discovery of many new microbial species and strains, as well as their valuable toxins and virulence factors that could advantage the biopesticide industry. Some such strains have also been translated as commercial products (Ruiu 2018; Ujváry 2010). The microbial pesticides will only affect specific species, and the reason behind their ability to infect is related to the crystalline occlusion bodies through which they are effective against Lepidopteran caterpillars, otherwise known as chewing insects. To increase the Bt insecticidal Cry I Ac toxin protein's pathogenicity and rate of action relative to its wild-type counterpart, more occlusion bodies containing the toxin were created. The occlusion body is a virion that has been combined with the Bt toxin to form ColorBtrus, a recombinant baculovirus. EPNs (Ento-

Table 3 Updated list of Bio-pesticide and target Insect and Crops

Source	Type	Organism	Pest Type	Target Crop	References
Virus	Insecticide	Plodia interpunctella	The Indian meal moth is a common pest found in stored food products.	Nuts, oilseeds, bran, and grains are essential components.	(Richards, 1932)
Bacteria	fungicide	Bacillus subtilis	Botrytis spp.	Produce, fruits, and decorative plants.	(Opender Koul, 2012)
	Insecticides	Bacillus thuringiensis var. tenebrionis	The alfalfa weevil and the elm leaf beetle are both common agricultural pests.	starchy tuber	(Saber, 2020)
Plant extracts	Herbicides	Essential oils derived from plants.	Numerous arthropods are attracted to Ragwort.	Grassland	(Isman, 2020)
	Nematicide	Quillaja saponaria	Nematodes that parasitize plants.	Agricultural land dedicated to the cultivation of field crops, vineyards, and orchards.	(Guerra, 2020)
Fungi	Herbicides	Chondrostereum purpureum	Remove the stumps of deciduous trees and shrubs.	Forestry	(Bailey, 2014)
	Fungicide	Trichoderma harzianum	Sclerotinia sclerotiorum	Non-consumable crops, strawberry cultivation, and sheltered crops.	(Dolatbadi, 2011)
Pheromones (Semi-chemical)	Attractant	Complex sex pheromones, like dodecagon-1-ol, consist of multiple components.	The codling moth, also known as Cydia pomonella, is a common pest of apple and pear trees.	Fruits like pears and apples.	(El-Sayed, 1999)
Nano-biopesticides	Sargassum muticum derived NPs	None	Ariadne merione, a Lepidopteran Pest		(Narware, 2019)
Insect growth regulators	Insecticides	Bemisia tabaci (GENNADIUS)	Pervading Bt Cotton.	Cotton, vegetables, fruit crops, and ornamentals.	(Kumar, 201)
RNAi-Based	Biopesticides	Diabrotica virgifera	Diabrotica virgifera	V-ATPase subunits and alpha-tubulin	(Baumberger & Baulcombe, 2005)
Cyanobacteria-algal	Biopesticides	Nostoc piscinale and	Chlorella vulgaris Cyanobacteria are filamentous organisms, while green algae are single-celled	Insects and pathogens are significant factors in agricultural ecosystems	(Ranglová, et al., 2021)

mopathogenic nematodes) on the other hand are advantageous biological control agents that do no harm to mammals or non-target organisms, and environment, they are primarily located in the species *Steinernema* and *Heterorhabditis* and have a connection with mutualistic symbiotic bacteria from the genera *Xenorhabdus* and *Photorhabdus* (Chang et al. 2003).

2. Biochemical Pesticides

Chemical pesticides use synthesized compounds that kill; on the other hand, biochemical pesticides are naturally occurring substances, that control pest problems through harmless approaches. Further divisions of biochemical pesticides include Insect growth regulators, Insect Pheromones, Essential Oils, and Plant-Based Extracts.

3. Insect Growth Regulators

These chemical substances exhibit great selectivity and little toxicity towards non-target species. This type of biopesticide kills insects by blocking several essential functions that are necessary for their life (Gurr et al. 1999). IGRs are effective against many insects, including mosquitoes, cockroaches, and fleas. They inhibit the reproduction, egg-hatching, and molting of young insects despite their low toxicity to humans. Even mature insects can be killed by combining IGRs with other pesticides (Gwinn 2018).

4. Insect Pheromones

These chemical substances work well at interfering with insect reproduction in order to decrease the amount of offspring produced. These are insect-produced compounds that are replicated and used in integrated pest management strategies to suppress insects. Insect pheromones, however, are not really “insecticides” because they alter an insect’s behavior by altering its olfactory system rather than killing it (Gonzalez-Coloma et al. 2013). The perceiving insect absorbs these pheromones by its antennae, and inside the sensilla, they are dispersed through microscopic pores in the cuticle. Once the pheromone binds to a specific receptor protein within the

cell, a second messenger system connected to the brain machinery transforms the chemical signal into a stronger electric signal (Gurr et al. 1999).

5. Essential Oils, and Plant-Based Extracts

These pesticides are naturally occurring since they are produced from plants and contain a variety of bioactive compounds. For the management of insect pests, plant-based extracts and essential oils have gained popularity as alternatives to synthetic pesticides (Magierowicz et al. 2019). These substances possess the capacity to behave as repellents, anti-feedants, and attractants.

Additionally, they may impair respiration, which makes it more challenging for insects to recognize their hosts, reduce adult emergence, and delay oviposition through ovicidal and larvicidal activities, plant extracts and essential oils (EOs) have varying degrees of effectiveness based on the physiological characteristics of the insect species and the kind of plant, against insects (Ali et al. 2017; Halder et al. 2012; Tripathi et al. 2009).

Lemongrass oil and neem oil are well-known examples, and they are widely available in herbal shops across the world. Furthermore, the entomopathogenic bacteria *Beauveria bassiana* and neem oil together shown to be highly effective against pests that feed on vegetables (Halder et al. 2012).

6. GMO Products

GMOs, or genetically modified organisms, are used to create these compounds. Cry proteins are, without a doubt, the most powerful insecticidal agent given to genetically modified crops (GM crops) that carry transgenes from the soil bacterium *Bt* (Parween and Jan 2019). These compounds work by integrating genetic information into the plant, which uses it as a source to create pesticidal chemicals often known as plant-incorporated protectants (PIPs). PIPs, however, also need the level of study required for the continuous evaluation of these compounds’ environmental destiny (Parker et al. 2019).

7. Biopesticides from Cyanobacterial and Algal Sources

Figure 1: Pesticides behavior within the soil system. Pesticide percolates inside the soil and deteriorates the property of the soil. That affects soil micro and macro fauna including non-target species

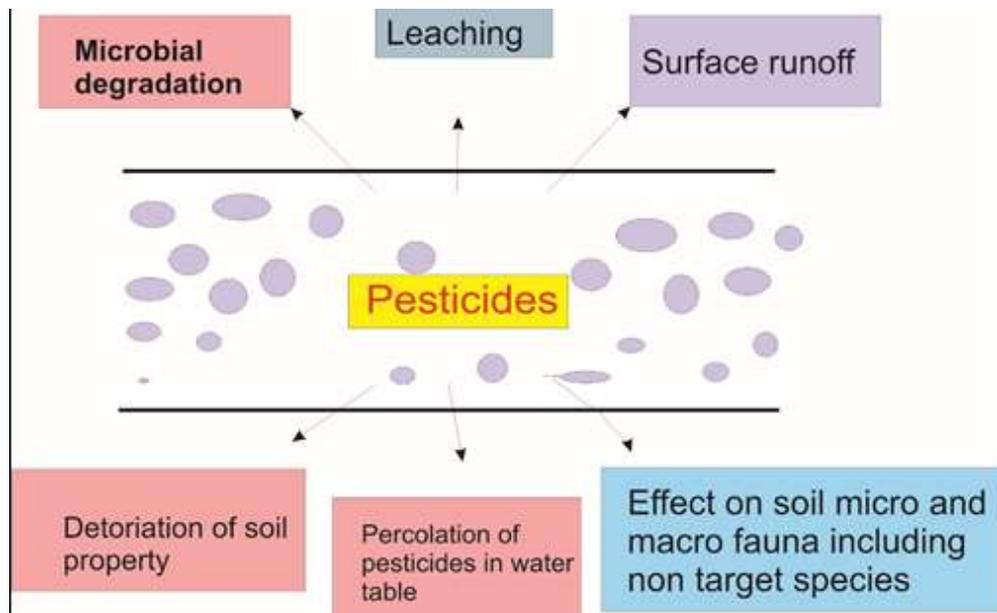
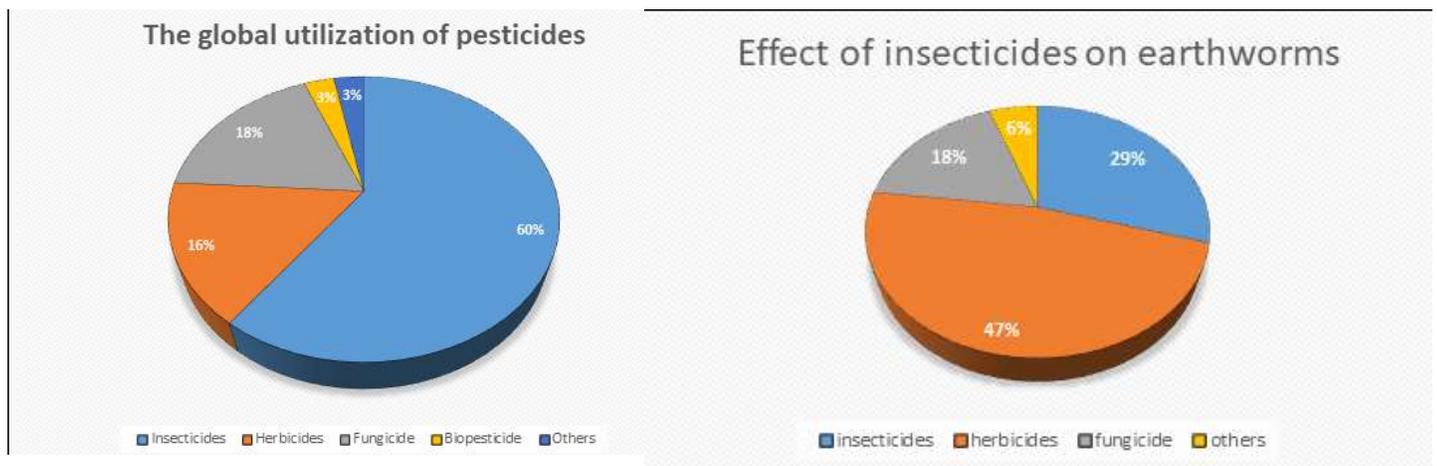


Fig 2. (a) The global utilization of pesticides. In the given pie chart. 60% of pesticides are globally utilized in the form of insecticides. The role of Herbicides is 16% in global utilization. Additional Pesticides are globally utilized as 18%, 3%, and 3% as fungicides, biopesticides, and others respectively. b) Effect of insecticides on earthworms. In the pie chart earthworms are affected mostly by Herbicides (47%) which is the highest, while the effect of insecticides is (29%) and other insecticides containing 18% & 6% as fungicides and others respectively.



Microalgae produces bioactive compounds, including antimicrobial substances that could serve as biopesticides (Gomiero 2018; Ranglová et al. 2021). In sustainable farming methods, microalgae can be utilized as a different approach to increase output. It has been observed that the single-celled green algae *Chlorella vulgaris*, a filamentous cyanobacteri-

um *Nostoc piscinale*, has biopesticide action against certain diseases. Wastewater contains all the necessary nutrients such as carbon, phosphorus, ammonium, and nitrogen in abundance, making it an ideal source of nitrogen for microalgae growth. The single-celled green algae *Chlorella vulgaris* is commonly employed for wastewater treatment due to its high

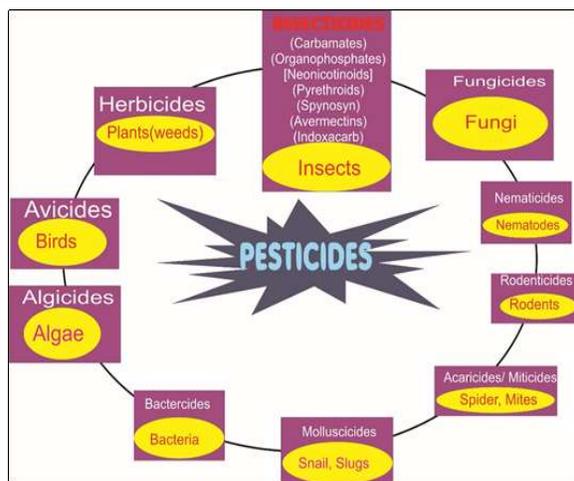


Fig. 3 Impacts of pesticides on various invertebrates may vary on the type of pesticide, its concentration, mode of application, and the specific characteristics and behaviors of the invertebrate species (Aktar et al. 2009; Ali 2023).

tolerance to ammonium levels (Ranglová et al. 2021).

8. Biopesticide Activity from RNAi-Based Treatments

Many transgenic crops have been developed to resist particular pests. Because RNA interference technology is more sensitive to infections and pests, it is employed in the creation of biopesticides (Parker et al. 2019). The production of transgenic dsRNA, which causes viral resistance and gene silencing in plants, is one of the mechanisms of RNA interference. (Baumberger and Baulcombe 2005), One potentially useful approach for combating the negative impacts of infections and pests is RNAi technology.

5. DISCUSSION

Effect of insecticides on earthworm gut bacteria and cast production

Earthworms are essential in soil ecosystems as they impact soil properties and control the biochemistry of terrestrial soil castings, or excrement, significantly contributing to the cyclic processes that occur in the soil ecosystem. These castings supply nutrients to plant roots and help maintain the soil's pedological characteristics. Earthworms are known for their voracious appetite and as they consume nutrient-rich organic matter, it passes through their gut, which acts as a straight tube bioreactor. The gut of earthworms also helps maintain a stable tempera-

ture through regulatory mechanisms (Edwards and Bohlen 1992) conducted the study. Interestingly, the interior of earthworms serves the environment and is considered to be a perfect habitat for numerous agriculturally significant microorganisms (Edwards and Bohlen 1992).

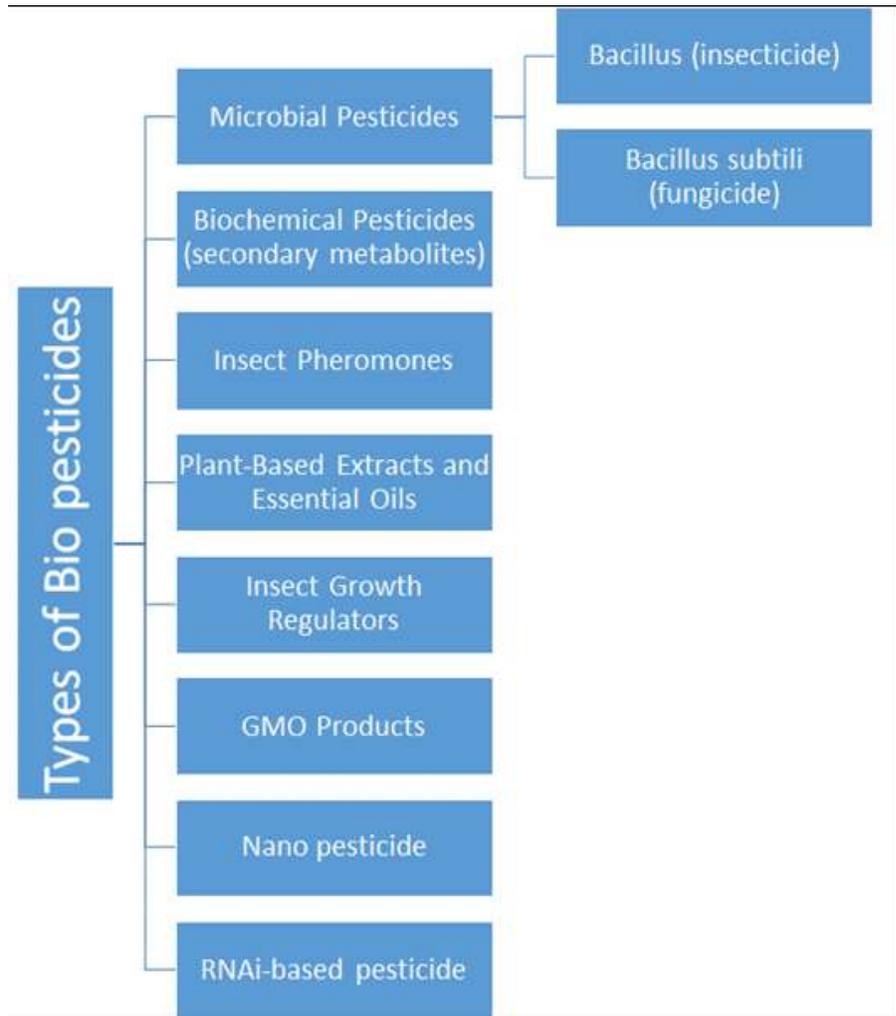
These earthworms primarily derive their energy and nutrients from the specific microbiota present in their gut, instead of deriving from the microorganisms within the soil that was consumed (Sampedro et al. 2006). In a study (Shi et al. 2007), a 14-day exposure to deltamethrin, to earthworm shows a toxic effect on the growth of earthworms was noted in a dose-dependent manner. Additionally (Shi et al. 2007) reported that the production of earthworm castings decreased when *L. terrestris* was subjected to methomyl, carbaryl, and imidacloprid for 7 days.

Research conducted in the laboratory investigated how earthworms respond to low levels of pesticides (Yasmin and D'Souza 2010).

6. CONCLUSIONS

The study emphasizes the utilization of pesticides in agriculture, leads to numerous ecological issues. There is compelling evidence indicating that the use of pesticides and fertilizers affects the population of earthworms and other soil organisms that are not the intended target resulting in a wide-ranging impact and an undesirable Change in the community.

Fig 4. Various types of Biopesticide can be a good replacement for pesticides.



At first, pesticides were employed to enhance farming efficiency and control communicable illnesses. Ignoring the harmful impacts on human health and the ecosystem. The environment and its components are being adversely affected by the widespread and varied utilization of pesticides. Negative impacts include an increase in pest that is resistant populations, there has been a decrease in advantageous soil microorganisms, predators, pollinators, and earthworms. Earthworms, being a crucial part of the soil fauna, are particularly vulnerable to pesticide exposure, especially insecticides, as the information has been recorded in the current analysis. The level of toxicity exhibited by insecticides towards earthworms differs depending on the classification of substances and their impact on the parameters of the life cycle of earthworms can be rewritten as follows. The environment has been significantly affected by the persistent nature of pesticides, as they have infiltrated various food chains and disrupted

the trophic levels, including those of humans and other large mammals. To mitigate the impact of pesticides, it is advisable to substitute chemical fertilizers with adequate organic manures, while minimizing soil disturbances, to enhance the beneficial activity of earthworms and maintain healthy and fertile soil. Although efforts have been made to comprehensively assess the toxicity of insecticides on earthworms, which are non-target species, these studies are limited in scope. Therefore, it is crucial to educate farmers about the valuable role of earthworms and reduce or minimize the use of pesticides to preserve a balanced ecosystem and promote biodiversity. In addition, Bio-pesticides use in agriculture is more beneficial to enhance crop productivity, and being biodegradable and environmentally friendly they don't cause such ecological disturbance (Rosell et al. 2008).

ACKNOWLEDGMENTS:

We offer our special thanks to Professor Dr. S.M. Bhatt IIMT UNIVERSITY for motivating us and editing the final manuscript.

Competing interests: The authors have no competing interests with any agency.

Availability of data and materials: Data collected from Pubmed, Google Scholar, and NCIM.

Consent for publication: Not applicable

Ethics approval consent to participate: Not applicable

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