ASTRAL

*A*ll Atlantic Ocean **S**ustainable, Profi**T**able and **R**esilient **A**quacu**L**ture

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Evidence of accomplishment

Report

Contents

Acronyms

AI	artificial intelligence
AIDAP	artificial intelligence data analytics platform
ASTRAL	All Atlantic Ocean Sustainable, Profitable and Resilient Aquaculture
Chl-a	Chlorophyll *a*
CSIR	Counsil of Scientific and Industrial Research
DO	dissolved oxygen
EC	electrical conductivity
EU	European Union
FAN	free ammonia nitrogen
FBU	formazin backscatter units
FNU	formazin nephelometric units
FURG	Federal University of Rio Grande
H2020	Horizon 2020
HABs	harmful algal blooms
ICT	information and communications technology
IHHNV	infectious hypodermal and hematopoietic necrosis virus
IMTA	integrated multi-trophic aquaculture
IOCCG	International Ocean Colour Coordinating Group
IoT	internet of things
IR	infrared
ISFET	ion-sensitive field-effect transistor
LoRaWAN	long range wide area network
MEMS	micro-electro-mechanical systems
MI	Marine Institute
ML	machine learning
NORCE	Norwegian Research Centre
OIE	International Organisation for Animal Health
OWIs	operational welfare indices
PAR	photosynthetically active radiation
PCB	polychlorinated biphenyls
PCR	polymerase chain reaction
PMW	passive microwave
ppt	parts per thousand
psu	practical salinity units
RFID	radio-frequency identification
RTD	resistance temperature devices
SAR	synthetic aperture radar
SPM	suspended particulate matter
SST	sea surface temperature
TAN	total ammonia nitrogen
TSM	total suspended matter
TSS	total suspended solids
UAV	unmanned aerial/aquatic vehicle
UCT	University of Cape Town
WQ	water quality

1 Summary

This document was created as part of the H2020 All Atlantic Ocean Sustainable, Profitable and Resilient Aquaculture (ASTRAL) project, funded as part of technology development efforts in Work Package 3, in support of the human capital development plan in Work Package 1. It provides a broad overview of the state of the technology landscape available for aquaculture, touching briefly on the relevant parameters, sensors, associated analytics, and considerations associated with their use and application. Examples are provided of relevant commercial solutions, in addition to pertinent ASTRAL-specific technological developments and research topics.

Although the ASTRAL project focuses primarily on integrated multi-trophic aquaculture (IMTA) production, this document provides an approachable and informative guide with a much broader scope. This guide is split into several sections including: the sensors and technology used for four different monitoring topics, namely physico-chemical water quality parameters and sensing methods, aquaculture stock and biomass estimation sensors, threat detection, and environmental variables; and the considerations in terms of instrument choice, operational environment, and system design.

2 Introduction

Aquaculture is one of the fastest growing industries in the world today, and is increasingly recognised for its enormous – even critical – role in sustainable food production for the global population[1]. But it is also acknowledged that it can be vulnerable to significant environmental risks as well as potentially resulting in negative environmental impacts. The integrated multi-trophic aquaculture (IMTA) concept is gaining traction as a way to increase the sustainability of intensive aquaculture systems with an ecosystem-based approach.

In IMTA, as illustrated in Figure 1, multiple aquatic species from different trophic levels are farmed in an integrated system that optimises the benefits of complementary ecosystem functions. Farmers combine the cultivation of fed species such as fish or shrimp, with extractive species such as seaweeds and aquatic plants, which recapture dissolved nutrients, and shellfish and other invertebrates, which recapture organic particulate nutrients for their growth. Ecosystems are engineered for environmental sustainability, economic stability and ultimately increased societal acceptance and trust in aquaculture as a reliable, safe and healthy food source. It is an ancient concept[2] which can now be applied in the modern context of sophisticated engineering and optimised through the use of now readily available technologies.

[1] Correia M, Azevedo IC, Peres H, Magalhães R, Oliva-Teles A, Almeida CMR and Guimarães L (2020). Integrated Multi-Trophic Aquaculture: A Laboratory and Hands-on Experimental Activity to Promote Environmental Sustainability Awareness and Value of Aquaculture Products. *Frontiers in Marine Science* 7:156. doi: 10.3389/fmars.2020.00156

[2] Barrington, K., Chopin, T. and Robinson, S., (2009). Integrated multi-trophic aquaculture (IMTA) in marine temperate waters. *Integrated mariculture: a global review. FAO Fisheries and Aquaculture Technical Paper*, *529*, pp.7-46.

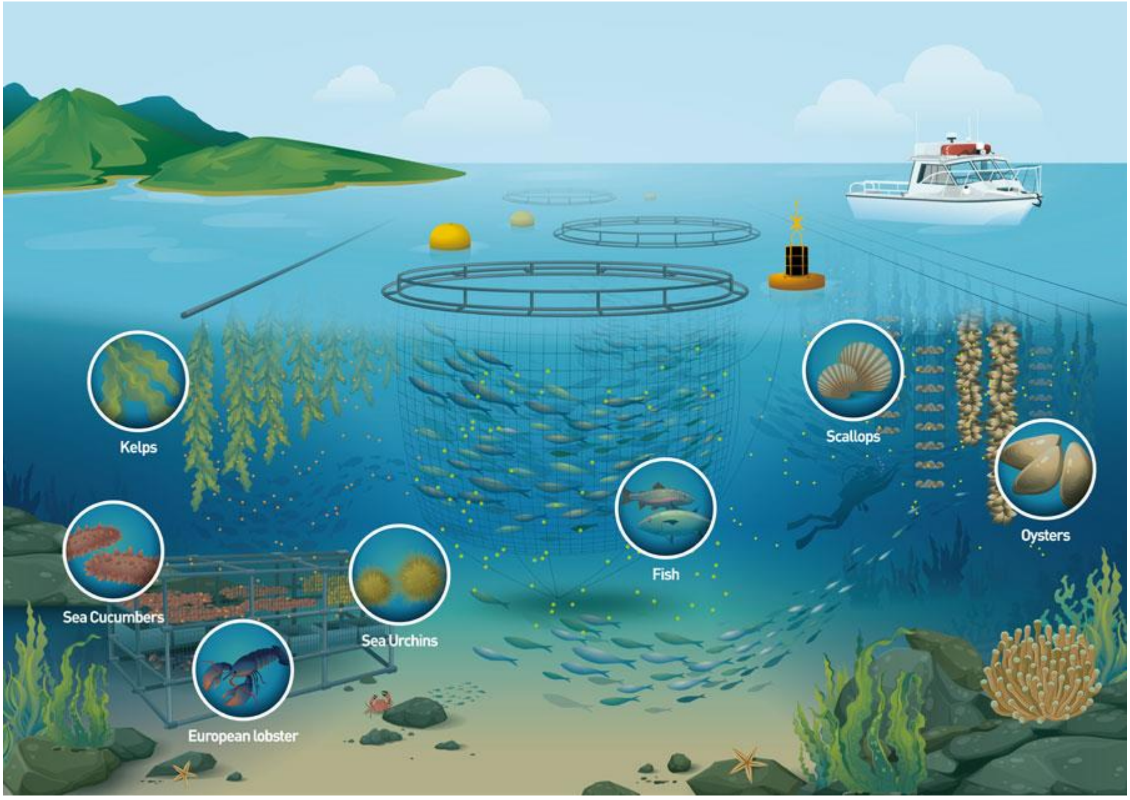

Figure 1. An illustration of the integrated multitrophic aquaculture concept in open systems (image obtained from Marine Institute website[3])

To achieve stability in an engineered ecosystem, a thorough understanding of its physico-chemical and biological dynamics and interactions is essential. Each IMTA system is unique in terms of siting, structure (flow-through, land-based, open-water), species cultivated, nutrient inputs and uptakes. Each system will have a different risk profile and different vulnerabilities to variable environmental conditions (including climate change), as well as to the introduction of biological hazards (biosecurity risks). Sites may be remote, or lack energy supply and communications infrastructure: each will have individual requirements and constraints for achieving the necessary insight into the operation of the system. Regardless of the size of the operation or its location, however, sensor device robustness and stable performance at the appropriate measurement sensitivity and frequency should always be a priority.

As interest in the industry grows and targeted technology becomes more readily available, physico-chemical and biological water measurement and monitoring solutions can be designed and built to meet the specific needs of each IMTA farm. The advantages of automated systems and the development of consistent datasets for identifying/predicting trends and change are clear – and the learning from these data to identify risk patterns and create early warning alerts can be invaluable.

While there are technology applications in many aspects of IMTA farming (feed automation and optimisation is another area of rapid technological development in mono-specie farming), this guide focuses on the use of technology for water quality, biomass measurements and monitoring. Bearing in mind the unique requirements of each site, farming system, and business model, this guide provides

[3] https://www.marine.ie/Home/site-area/infrastructure-facilities/lehanagh-pool-marine-research-site?language=en

an overview of principal water quality parameters, biomass estimation methods, and sensor technologies commonly used to measure and monitor these variables. The need to understand and meet site- and purpose-specific requirements is emphasised, and aspects such as sensor placement, measurement frequency, instrument robustness requirements and differing record-keeping needs are beyond the scope of this document. However, these are critical aspects to the successful design and implementation of appropriate monitoring, data management, visualisation and communications solutions.

The current report aims to provide information on a range of technologies suited to all levels of IMTA farming, from cost-effective solutions for small scale farms to more advanced and data-intensive systems appropriate for larger commercial and research operations wanting to increase efficiency in farm management and optimise production.

For a comprehensive overview of the monitoring recommendations on the most appropriate sensors, observing platforms, sampling strategies, and approaches at different IMTA sites, see ASTRAL Deliverable 5.5

3 Physico-Chemical Water Quality Parameters

Good water quality is vital for maintaining the health and performance of farmed organisms. Water quality needs to be managed by monitoring and maintaining the chemical and the physical properties of the water in the aquaculture facility within the tolerance limits, and preferably within the optimal ranges, of the farmed organisms. As demonstrated in Figure 2, farmed organisms have different tolerance limits and optimal ranges for the different water quality parameters. Outside of these ranges the organisms may become stressed, which can lead to poor growth, behavioural changes, diseases and mortalities. Water quality requirements and ranges may also be slightly different for reproduction or spawning, or during different life stages of the organisms. Aquafarmers need to understand the different parameters and interactions that contribute to poor water quality. Some parameters can be stable, whereas others (e.g. pH, dissolved oxygen) can fluctuate hourly.

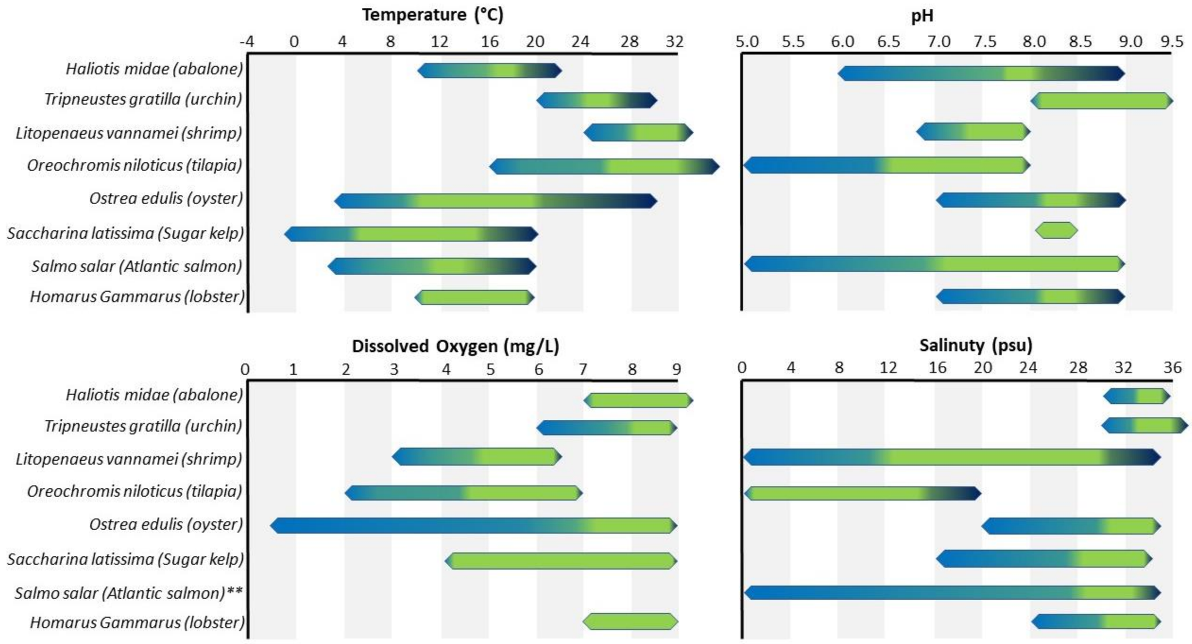

Figure 2. Optimal (in green) and tolerance ranges (blue arrow ends) of some of the water quality parameters for key ASTRAL cultivated species. ***Salmo salar* dissolved oxygen tolerance range is 70-100% relative to temperature and pH

3.1 Temperature

Optimal temperature and tolerance ranges of the ambient water temperature depend on the cultivated species. Generally, it is important that the temperature does not fluctuate significantly, and that it is maintained within the optimum ranges of the cultivated species. Temperature affects the physiology and metabolic rates of cultured organisms, and resultantly also the growth, respiration rates, reproduction, and consumption of feed. Biological reactions and systems tend to speed up with increasing temperature in a certain range to a point where further increase rapidly causes damage. For example, if you are growing South African abalone at 15 degrees, an increase to 17 degrees is positive, but if you are growing at 20 degrees an increase to 22 degrees can have severely negative consequences. Water temperature can also affect plant respiration and photosynthesis, and different species will have optimum temperatures where peak photosynthetic activity can take place. Often, short term cooling is less harmful than short-term warming, within a critical range. In land-based IMTA systems, water passing through a series of systems containing different organisms will also change its temperature. In particular, seaweed ponds or raceways tend to increase water temperature. This can be positive, where ambient seawater temperature is below optimum for animal growth[4 5].

In such sensitive and complex systems, temperature monitoring and control is of fundamental importance. In addition to its own effects, temperature can also influence the chemical and physical properties of water; for example, an increase in water temperature leads to a decrease in the solubility of gases (such as dissolved oxygen), but also might result in an increase of the water pH.

[4] Nobre, A. M., Robertson-Andersson, D., Neori, A., & Sankar, K. (2010). Ecological–economic assessment of aquaculture options: comparison between abalone monoculture and integrated multi-trophic aquaculture of abalone and seaweeds. *Aquaculture*, *306*(1-4), 116-126.

[5] Bolton, J. J., Robertson-Andersson, D. V., Shuuluka, D., & Kandjengo, L. (2009). Growing Ulva (Chlorophyta) in integrated systems as a commercial crop for abalone feed in South Africa: a SWOT analysis. *Journal of Applied Phycology*, *21*(5), 575-583.

In many cases land-based ponds or tanks are exposed to the elements, and are directly affected by seasonal and daily weather changes, including day-night warming and cooling. Open ocean-based aquafarms are directly impacted by the physical conditions of the ocean water column, with no realistic human intervention options aside from perhaps changing the depth of cages, or the selection of seasonal farming only. In these cases, siting the location of the farm, in physical environmental conditions conducive to the cultured species, is of vital importance. It should be noted that turbidity can increase the water temperature in both land-based or ocean-based aquafarms, as the additional suspended substances absorb solar radiation more efficiently than pure water.

Temperatures are measured by contact thermometers, either the traditional non-electric (e.g. based on thermal expansion of a substance) or by a variety of electronic devices such as thermocouples, resistance temperature devices (RTD), and thermistors (RTD made of platinum). Further details on each of these types of sensors can be found in Parra et al (2018)[6], who recommends RTD or thermistors for aquaculture applications due to their high working range, adequate accuracy, and low cost.

Sea surface temperature (SST) can also be measured using infrared (IR) or passive microwave (PMW) radiometers on board satellites. IR emissions provide information of the <0.1 mm thin skin layer of the ocean surface; while these IR emissions cannot be detected through cloud, the data are obtained at a spatial resolution of 700 – 4000 meters with approximately once to twice daily coverage. PMW emissions originate from a thicker ocean surface skin layer (several millimetres thick) and can generally be measured through cloud; however, these measurements cannot be derived within ~100 km from the coast due to thermal interference from land masses, and have a low spatial resolution of typically 50-75 km. SST data from IR and PMW sensors can provide useful information of synoptic scale changes to surface conditions, and are freely available from most global environmental monitoring missions. However, some uncertainty in SST products should be accounted for, and the coarse spatial resolution is unlikely to capture variations in temperature across a single aquaculture facility, and does not provide an indication of temperature changes with water column depth.

3.2 Salinity

Salinity refers to the total concentrations of dissolved salts in water; it is usually derived from measurements of electrical conductivity (EC) - a measure of a liquid's capacity to conduct an electrical flow - which is directly related to the amount of dissolved salts (ions) in the water. Salinity based on conductivity measurements are unitless, but usually have the notation of practical salinity units (psu) or parts per thousand (ppt). Temperature and pressure affect conductivity values, and the EC reading needs to be related to salinity accordingly. As with other parameters, different cultivated organisms have preferred optimum ranges of salinity, outside of which they become stressed or can die. Salinity can affect the water clarity; salt ions tend to aggregate and bind suspended solids together, increasing their settling/sinking rates.

[6] Parra, L., Lloret, G., Lloret, J. and Rodilla, M., (2018). Physical sensors for precision aquaculture: A Review. *IEEE Sensors Journal*, *18*(10), pp.3915-3923.

In many aquaculture operations, salinity does not vary significantly with respect to the physiological tolerances of the organisms. Small fluctuations of a few units from typical sea water salinity do not generally have an impact. Salinity is, however, critical in estuarine aquaculture, where salinity can vary throughout the tidal cycle on an hourly basis, and can be important where heavy rain affects land-based systems, or occasional large rainfall events affect inshore coastal systems.

EC sensors are usually either electrode-based or electrodeless (inductive). The first method measures the ability of a solution to conduct an electric current between two electrodes; this is a simple sensor that is easily calibrated, but the electrodes may be subject to corrosion[7]. Inductive sensors consist of a generating coil (current transformer), which generates a magnetic field; this magnetic field is modified by the salinity of the water, inducing a proportional electric field in a receiving coil[8]. While inductive sensors are more robust and easily maintained, they need to be placed a certain distance away from other sensors due to the use of an inductive field, while sensor calibration should be done when assembled to account for the magnetic fields of other sensors and platforms[9].

ASTRAL technology under development includes a low-cost Internet of Things kit, which will incorporate a variety of probes to measure key water quality parameters (e.g. dissolved oxygen, pH, turbidity, conductivity)

3.3 pH

The "power of Hydrogen", or pH, is a value between 0 and 14 that defines how acidic or basic a water sample is on a logarithmic scale; this is calculated from the number of hydrogen ions (H^+) in solution, and has a unit of $-\log[H^+]$. A pH of 7 is considered neutral, with a lower pH being more acidic and representing a higher concentration of hydrogen ions (H^+), while a higher pH means that a solution is more basic. Carbon dioxide (CO_2) levels also affect the pH of water, and fluctuate daily as a result of photosynthesis, respiration and decomposition.

CO_2 can exist either in its dissolved state or react with water to form carbonic acid (H_2CO_3):

$$CO_2 + H_2O \rightarrow H_2CO_3$$

Carbonic acid can then dissociate into bicarbonate (HCO_3):

$$H_2CO_3 \rightarrow H^+ + HCO_3^-$$

[7] Souza Dias, Francisco (2020): Salinity sensors. Available from http://www.coastalwiki.org/wiki/Salinity_sensors [accessed on 17-11-2021]

[8] Parra, L., Lloret, G., Lloret, J. and Rodilla, M., (2018). Physical sensors for precision aquaculture: A Review. *IEEE Sensors Journal*, *18*(10), pp.3915-3923.

[9] Souza Dias, Francisco (2020): Salinity sensors. Available from http://www.coastalwiki.org/wiki/Salinity_sensors [accessed on 17-11-2021]

Which can then dissociate into carbonate ions:

$$HCO_3 \rightarrow H^+ + {CO_3}^{2-}$$

These processes all contribute to further decreasing the pH of the water.

Saltwater has a higher pH than freshwater. pH affects the solubility and toxicity of some chemicals and heavy metals in water. For instance, low pH reduces the solubility of calcium carbonate, which is required for shell production and growth in aquatic organisms; on the other hand, higher pH levels tend to increase the amount of toxic free ammonia nitrogen in the water (see section 3.4). The inclusion of algae in an aquaculture system helps to raise the pH during the day when the system/seaweed can take up CO_2. In full or partial recirculating systems the pH tends to be lowest at sunrise.

Increasing ocean acidification and the associated drop in pH presents a risk for shell-producing organisms such as molluscs (e.g. abalone, oysters, mussels). There is a relationship between pH, free- and total ammonia nitrogen (see section 3.4), which requires that the balance between these parameters is carefully observed. In the South African IMTA lab for example, consisting of abalone tanks integrated with *Ulva* raceways (Figure 3), there is a complex interaction of these parameters as daytime photosynthesis by the *Ulva* (seaweed) oxygenates the water and raises the pH by absorbing CO_2. This does not happen at night when the *Ulva* is a net respirer, i.e. producing more CO_2 than it takes up. As abalone tend to feed and are most active at night, there is a quite a complicated daily rhythm of ammonia and pH in these integrated systems.

Figure 3. The Buffeljags abalone farm IMTA system, part of the South African IMTA lab, with seaweed (Ulva) raceways in the center and the abalone tanks on the sides. The Ulva oxygenates the water, absorbs CO_2, and takes up ammonia so that the water can be safely recirculated back to the abalone raceways (credit: Viking aquaculture)

Whilst ion-sensitive glass electrodes (potentiometric sensors) have historically been the most popular pH sensors for seawater monitoring, they can be unstable and fragile. Other technologies have included ion sensitive field effect transistors (ISFETs), spectrophotometric systems, and optodes. The development of solid-state sensors based on inorganic materials (e.g. metals) and carbon-based materials (e.g. polymers), as well as nano-engineered materials and composites offer promising future applications (see review in Avolio et al 2020)[10].

3.4 Aquatic Nutrients

A variety of nutrients can end up in an aquaculture system, whether naturally from coastal ocean upwelling or river water, from feeds, or from aquatic animal waste and effluent. The most abundant of these nutrients are nitrate, nitrite and ammonium, (all nitrogen-based, N) and phosphorus (P). N and P are the primary limiting macronutrients of plants in general, including both phytoplankton and seaweeds.

Cultured marine organisms excrete nitrogenous waste comprising particulate as well as ammonia nitrogen, a portion of which can occur in the form of free ammonia nitrogen (FAN, NH_3). FAN is a toxic compound which occurs in water in a temperature, pH, and salinity dependent equilibrium with ammonium (NH_4^+):

$$NH_4^+ \leftrightarrow NH_3 + H^+$$

When the pH and/or the temperature increases, the proportion of the toxic compound NH_3 increases. Total ammonia nitrogen (TAN), the sum of NH_4^+ and NH_3, is usually measured in aquaculture systems; the proportion of FAN is subsequently calculated with respect to temperature, pH, and salinity, with the use of look-up-tables or online converters[11,12].

There are many laboratory-based techniques (e.g. spectroscopy and chromatography) and devices to analyse nutrient sample concentrations which, while highly sensitive, are laborious and not suitable for continuous in-water measurements, if they are required – particularly when calculating concentrations of FAN, which can vary hourly in response to temperature and pH changes. Nutrient sensors that are used in aquatic environments usually fall into the category of either electrochemical or optical, with a high degree of variation in fabrication and operating methods. Essentially electrochemical sensors rely on the electrochemical reactions at the surface of an electrode, and depending on the signal transduction method used can be classified into potentiometric (measurement of voltage), amperometric/voltammetric (measurement of current), and impedimetric/conductimetric (measurement of conductivity or resistivity) systems. Optical approaches rely on the UV-visible absorbance of either the nutrients or of a resulting colorimetric change after interaction with a reagent; optical fiber sensors use nanoparticles or dyes that interact with, for example, dissolved ammonia to give either intensity or wavelength variations in the optical fiber probe[13]. A comprehensive overview on the recent progress of these sensors can be found in Mahmud et al (2020)[14], whereas an ammonia nitrogen specific review is provided by Li et al (2020)[15].

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[10] Avolio, R., Grozdanov, A., Avella, M., Barton, J., Cocca, M., De Falco, F., Dimitrov, A.T., Errico, M.E., Fanjul-Bolado, P., Gentile, G. and Paunovic, P., (2020). Review of pH sensing materials from macro-to nano-scale: Recent developments and examples of seawater applications. *Critical Reviews in Environmental Science and Technology*, pp.1-43.

[11] e.g. Free Ammonia Calculator: https://www.engineering.iastate.edu/~jea/w3-research/free-ammonia/nh3.html

[12] e.g. Ammonia look-up tables: https://www.handymath.com/cgi-bin/nh4no3tble.cgi?submit=Entry

[13] Leal-Junior, A.G., Frizera, A. and Marques, C., (2020). High sensitive ammonia detection in water with Fabry-Perot interferometers. *IEEE Photonics Technology Letters*, *32*(14), pp.863-866.

[14] Mahmud, M., Ejeian, F., Azadi, S., Myers, M., Pejcic, B., Abbasi, R., Razmjou, A. and Asadnia, M., (2020). Recent progress in sensing nitrate, nitrite, phosphate, and ammonium in aquatic environment. *Chemosphere*, p.127492.

[15] Li, D., Xu, X., Li, Z., Wang, T. and Wang, C., (2020). Detection methods of ammonia nitrogen in water: A review. *TrAC Trends in Analytical Chemistry*, *127*, p.115890.

ASTRAL technology under development includes a MEMS-based sensor which will measure spectral absorbance in the visible, spectral fluorescence at 4 illumination wavelengths, and use a machine learning model to infer physico-chemical and biological parameters from these spectra.

3.5 Turbidity, Total Suspended Solids and Water Clarity

Turbidity and water clarity relates to the optical characteristic of a water body, i.e. how much it scatters and attenuates light. Turbidity is dependent on the amount, type, size, and colour of total suspended solids (TSS) and dissolved organic matter in the water, which scatters and absorbs the light entering the water making it appear cloudy or murky. Water clarity is defined by how clear or transparent the water is, specifically relating to how far sunlight can penetrate, which in turn can influence the photosynthetically active radiation that is available for submerged cultured algal species. Several things can contribute to increase turbidity, including fish waste, uneaten feed, suspended sediments (silt or clay) or organic material (plankton, micro-organisms – including Biofloc) in the water column. Excessive amounts of small algae may also increase water turbidity - the risks associated with harmful algae are discussed in section 6.1. Macroalgae are generally not included in the term “turbidity” despite contributing to reduced light levels in the water column.

TSS are a measure of the mass of solids larger than 2 microns in diameter per volume of water (e.g. g/m^3). TSS are usually filtered out of the water and weighed, a method that is time consuming. Turbidity is usually used as an indicator that provides information on the qualitative changes in TSS concentrations.

The use of Biofloc can be a major contributor to TSS. Biofloc technology utilises the action of microorganisms aggregated in flocs and has the potential to revolutionise IMTA aquaculture production. The microorganisms improve water quality but also represent a complementary food source to the produced aquatic organisms. This bio-based technology allows super intensive production (high density) in small spaces with minimal or no water renewal, increasing circularity and potentially reducing the aquaculture waste production to almost zero. However, it may provide a hazardous environment to some species (e.g. bivalve molluscs) without proper management. The control of total suspended particles within the aquatic environment plays an important role to enable proper Biofloc management and high-density production with minimal risks to animal welfare and health.

Turbidity is usually measured by optical methods using turbidity meters, (submersible) turbidity sensors or other optical scatter-detection techniques, the simplest of which consist of a light beam, a detector placed at 90° incident to the beam angle, inside a narrow detector window with an acceptance angle of no more than 30°. More complex sensor designs and measurement capabilities exist, which can include one or more light sources at different wavelengths, and multiple detectors at different angles, and the choice of sensor might depend on the colour or level of turbidity and backscattering (see Anderson 2005[16] for more information); the units of the measurements depend on the instrument design and light source, and the data from different turbidity instruments designs are not directly comparable. In order to compare the raw data of turbidity measurements across different locations and times, it is important to use instruments with identical optical configurations and processing capabilities for a proper data synchronisation. Most turbidity sensors on the market use an infrared LED light source, which means that it would have units of formazin nephelometric units (FNU)[i] or formazin backscatter units (FBU). An extensive and informative online guide to measuring turbidity, TSS and Water Clarity can be found at the Fondriest Environmental Learning Center[17].

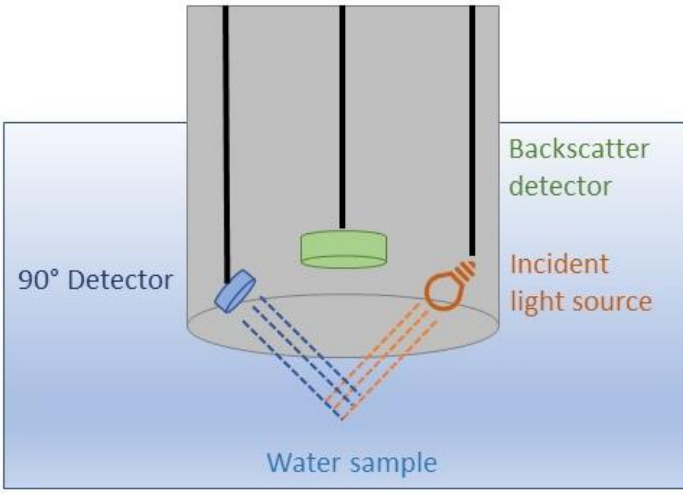

Figure 4. An illustration of a turbidity sensor that includes a 90-degree angle measurement of scattered light

3.6 Dissolved Oxygen (DO)

Dissolved oxygen refers to the amount of free oxygen molecules (i.e. not bonded to other elements) that is present in a liquid. Apart from some bacteria, dissolved oxygen is needed for the survival and development of all aquatic organisms. Low levels of DO can affect feeding behaviour, lead to animal stress, low growth rates, less feeding, or even death. DO requirements are different depending on factors such as species, organism size, feeding rate, activity level and temperature. The amount of DO in the water is also dependent on water temperature and salinity, with the solubility of oxygen decreasing with both (either) temperature and salinity increases. In partially or fully closed systems there is generally a cyclical behaviour in DO; during the day photosynthetic bacteria, phytoplankton and macroalgae can make use of sunlight to photosynthesize and produce oxygen, while at night DO is reduced by all marine animals and plants during respiration and decomposition. Mechanical aeration, commonly used in land-based aquaculture systems (shown in Figure 5), can assist in avoiding very low dissolved oxygen concentrations. Some farms may even have a specialised oxygenator for this purpose.

[16] Anderson, C. W. (2005). Turbidity 6.7. In USGS National Field Manual for the Collection of Water-Quality Data. U S Geological Survey.

[17] https://www.fondriest.com/environmental-measurements/measurements/measuring-water-quality/turbidity-sensors-meters-and-methods/

Figure 5. A mechanical aeration system in operation in the shrimp ponds at FURG, Brazil

DO can be measured by the electrochemical or optical methods, or by the traditional laboratory-based technique of iodometric (Winkler) titration. Electrochemical sensors, also called Clark-type or amperometric sensors, consists of an anode and a cathode in an electrolyte solution, covered by an oxygen-permeable membrane; the reduction of the oxygen produces an electrical current proportional to the partial pressure of oxygen in the sample. These sensors require frequent maintenance, cleaning and membrane replacement, and because they consume oxygen during measurements, they require either water or sensor movement for accurate readings. Optical sensors measure the interaction between oxygen and specific luminescent dyes based on the fluorescence quenching principle; the sensor consists of excitation light sources, a substrate film attached to a fluorescence-sensitive substance, and an optoelectronic detection element; the oxygen molecules interact with the dye and interfere with its fluorescence, and as a consequence the measured effect is inversely proportional to the partial pressure of oxygen. Optical sensors are relatively expensive, require more power, and are influenced by ambient temperature (although usually automatically corrected), however their low maintenance and low (if any) calibration drift often make them preferable for long-term and continuous in situ monitoring applications. A comprehensive review of DO detection technology can be found in Wei et al (2019)[18].

[18] Wei, Y., Jiao, Y., An, D., Li, D., Li, W. and Wei, Q., (2019). Review of dissolved oxygen detection technology: from laboratory analysis to online intelligent detection. *Sensors*, *19*(18), p.3995.

4 Sensing methods for Water Quality Parameters

4.1 Water Quality Biosensors

Bivalve molluscs, such as mussels or oysters, are resilient animals capable of reacting to environmental proxies. These filter-feeding animals open their shells to perform biological activities, such as respiratory gas exchange and collection of food, and are able to close them in resting periods or to protect themselves in response to water property variation e.g. temperature, pH, salinity, dissolved oxygen, food availability[19], and possibly high levels of particulate (e.g. algal cells) which may irritate the gill lamellae. Given that shell closure will also occur in response to external or physical stimuli (e.g. touch, vibrations), these movements need to be distinguished from those of water quality responses. Continuous measurement of valve movements can identify non-typical patterns of behaviour and may be a fast and sensitive holistic method to assess water quality that provide early warning of deteriorating conditions. In aquaculture these methods can be used both in culture systems that harvest bivalves applied to the same species, as well as in culture systems that do not harvest bivalves as an independent biosensor.

Methodologies to measure the valve opening amplitude are named valvometry techniques and can be based on different physical phenomena and a variety of detection devices and techniques. Previously validated methodologies include inductor pairs, electrodes, load cells and the most popular Hall effect sensor[20, 21, 22] (based on the magnetic field generated by a small neodymium magnet); an illustration of the Hall effect sensor is shown in Figure 6. Within ASTRAL micro-electro-mechanical systems (MEMS) accelerometer-based sensors are being developed and assessed. These valvometry techniques usually need devices and cables connected to the animals' shells, which may cause impairment in their behaviour. Commercial products for bivalve early monitoring do exist. For instance, MOSSELMONITOR® [23] (by Aquadect) consists of an integrated data acquisition hardware (based on inductor pairs) with an alarm system and a logger.

[19] Resgalla Jr, C., Brasil, E. D. S., & Salomão, L. C. (2007). The effect of temperature and salinity on the physiological rates of the mussel *Perna perna* (Linnaeus 1758). *Brazilian archives of biology and technology*, *50*, 543-556.

[20] Hartmann, J. T., Beggel, S., Auerswald, K., Stoeckle, B. C., & Geist, J. (2016). Establishing mussel behavior as a biomarker in ecotoxicology. *Aquatic Toxicology*, 170, 279-288.

[21] Tran, D., Haberkorn, H., Soudant, P., Ciret, P., & Massabuau, J. C. (2010). Behavioral responses of Crassostrea gigas exposed to the harmful algae *Alexandrium minutum*. *Aquaculture*, *298*(3-4), 338-345.

[22] Liao, C. M., Jau, S. F., Lin, C. M., Jou, L. J., Liu, C. W., Liao, V. H. C., & Chang, F. J. (2009). Valve movement response of the freshwater clam *Corbicula fluminea* following exposure to waterborne arsenic. *Ecotoxicology*, *18*(5), 567-576.

[23] https://www.mosselmonitor.nl/

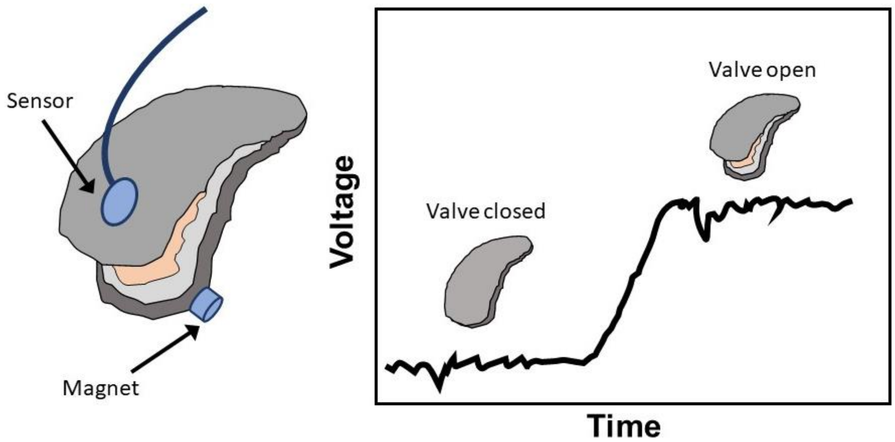

Figure 6. Illustration of a Hall effect biosensor attached to an oyster to assess valve gape (based on figure 3 from Clements & Comeau 2019 [24])

A computer vision based valvometry technique has been previously developed and validated by FURG[25] in a lab setup using mussels. This methodology eliminates the need for instrumentation attached to the bivalves' shells while still performing a direct measurement of their valve gape amplitude. The promising validation results in the laboratory setup and the innovative advances in this valvometry technique suggest that further investment in developing such a sensor commercially would be justified. Furthermore, the availability of an alarm system may be a fast and useful tool to understand the animals‘ behaviour over time. Through statistical analysis, this system may be able to identify behavioural variations caused by environmental proxies while still being robust to natural opening fluctuations. By analysing the bivalves opening, the alarm program also informs on the filtration rate activity of each animal inside the IMTA system[26]. This methodology lends itself well to sites already farming bivalves, but the introduction of biosecurity risks should be considered if the use of this method requires the introduction of new species - even if only used as water quality indicators - into a farming system.

ASTRAL partners are assessing two different valvometry techniques for assessing biological response (i.e. valve gape amplitude) of oysters to environmental proxies (e.g. water quality parameters); one is a computer vision-based sensor, while the other is a micro-electro-mechanical systems (MEMS) accelerometer-based sensor.

[24] Clements, J. C., & Comeau, L. A. (2019). Use of high-frequency noninvasive electromagnetic biosensors to detect ocean acidification effects on shellfish behavior. *Journal of Shellfish Research*, *38*(3), 811-818.

[25] de Vargas Guterres, B., da Silveira Guerreiro, A., Sandrini, J. Z., & da Costa Botelho, S. S. (2020). Feasibility of visual signals on the construction of biosensors based on behavioral analysis of *Perna perna* mussels. *Ecological Informatics*, 59, 101118.

[26] Riisgård, H. U., Kittner, C., & Seerup, D. F. (2003). Regulation of opening state and filtration rate in filter-feeding bivalves (*Cardium edule, Mytilus edulis, Mya arenaria*) in response to low algal concentration. *Journal of experimental marine biology and ecology*, *284*(1-2), 105-127.

4.2 Remote Sensing of Water Quality

Particles in the water can either absorb or scatter light, and the resulting effect (or colour) of these interactions depend on their composition, size and concentration. Ocean colour remote sensing uses optical radiometers on board satellites to measure light at specific wavelengths emerging from the water surface, which can be used to detect certain WQ-related parameters that have a measurable and known effects on the colour of the water. It should be noted that not all WQ parameters can be derived using satellite-based information (e.g. DO, pH) and that remote sensing measurements are not meant to substitute for *in situ* WQ monitoring, but should rather be used to supplement the limited temporal and geographic scales of field-based measurements. The advantage of satellite datatsets is that they can cover a much larger area than any in situ sampling, and repeatedly over a few days or weeks. When successfully ground-truthed with local field measurements, satellite data become a powerful tool for understanding regional and temporal environmental variability.

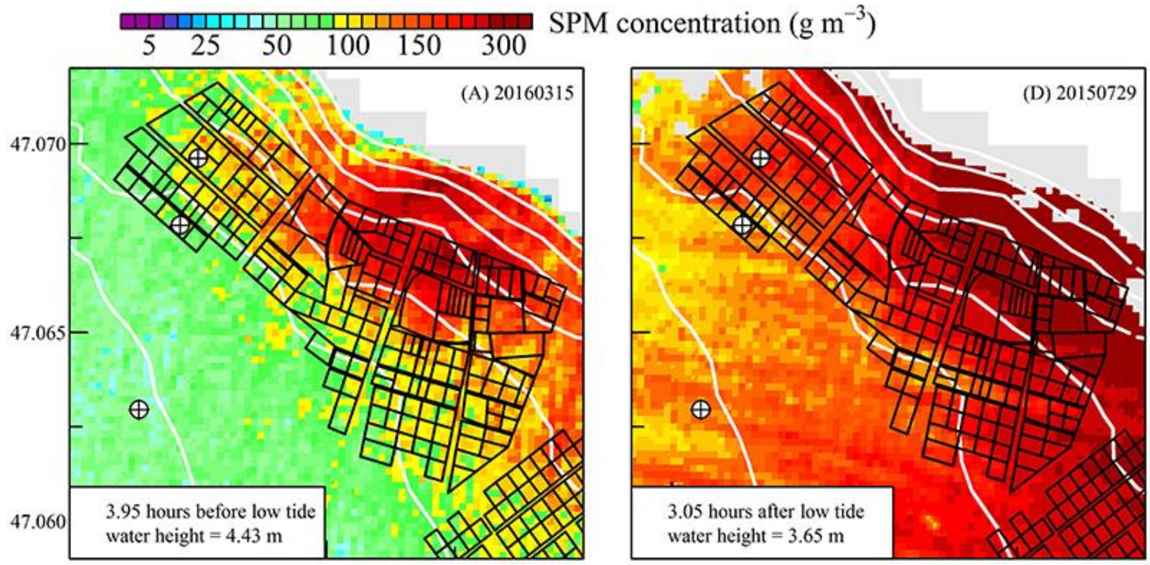

Figure 7. An example showing the use of Sentinel 2 ocean colour data to monitor suspended particulate matter (SPM) concentration at an oyster farming site in the intertidal zone of Bourgneuf Bay along the French Atlantic coast (adapted from Gernez et al, 2017 [27]). This study formed part of the H2020 Tools for Assessment and Planning of Aquaculture Sustainability (TAPAS) project[28].

Algorithms have been developed to quantify parameters such as suspended particulate matter, turbidity, light attenuation, dissolved organic matter, and phytoplankton pigments. The simplest of detection techniques relate waveband ratios empirically to constituent concentrations; this is achieved with varying degrees of error depending on how the calculation is made, and the environmental conditions at the time of measurement. A popular ocean colour parameter is the concentration of the photosynthetic pigment chlorophyll a (Chl-a), which is generally used as a proxy for phytoplankton biomass; Chl-a products are freely available from most environmental monitoring satellites at spatial

[27] Gernez, P., Doxaran, D., & Barillé, L. (2017). Shellfish aquaculture from space: potential of Sentinel2 to monitor tide-driven changes in turbidity, chlorophyll concentration and oyster physiological response at the scale of an oyster farm. *Frontiers in Marine Science*, *4*, 137.
[28] https://www.researchgate.net/project/TAPAS-EU-h2020

resolutions of 300 –1200 meters at near-daily time scales. While free 10-60 meter resolution ocean colour data are available from the Sentinel 2A and 2B satellite constellation, these data require users to apply appropriate atmospheric correction and biogeochemical algorithms themselves, and only provide satellite coverage every 3-5 days. A brief review of the use of remote sensing for phytoplankton and harmful algal bloom detection can be found in section 6.1 of this document, whereas a comprehensive review on the use of earth observation in support of WQ monitoring can be found in IOCCG report 17[29].

CoastObs[30] was an EU H2020 project aimed at using satellite remote sensing to monitor coastal water environments; the consortium is transitioning into a commercial product and service provider for coastal water monitoring, offering useful parameters for aquaculture operations such as turbidity, eutrophication, phytoplankton size classes, primary production, and the presence of harmful algae.

5 Stock Monitoring and Biomass Estimation

Abundance, biomass, or stock estimation are necessary to maintain an optimal yet balanced healthy ecosystem; these data determine optimal stocking densities, average daily growth, harvest timelines, and the optimal feed application to promote growth without undue waste or environmental impacts. Traditional biomass estimations usually involve taking subsamples of the cultivated organisms and performing visual inspection (e.g. clinical signs of disease, parasites, shell damage/quality) and manual measurements (e.g. weight, length). These methods are laborious, time consuming and prone to human error; they can also cause stress and physical damage, increasing organisms' susceptibility to disease. Some intermediate systems have automated processes for weighing and sorting individuals. However, any mechanical handling of the animals can potentially damage or stress them. As a result, there has been a move toward the development of techniques and technologies that offer rapid, reliable and repeatable non-invasive monitoring based on machine vision, acoustics, and remote sensing, among others.

5.1 Machine Vision and Artificial Intelligence

Machine vision technology uses a combination of cameras or other sensors and machine learning to acquire, process and analyse imagery of cultured animals; in the case of biomass estimations this technique mainly utilises monocular cameras or stereovision systems detecting visible and infrared light[31]. Infrared optical methods generally involve getting fish to swim through a scanning unit where they are counted as they break the lattice of infrared light beams; however, the efficiency and accuracy of this method decreases for small fish, or in turbid water[32]. The relationship between size (i.e. width, length, area) and mass is species-specific and models still need to be determined individually; for

[29] IOCCG (2018). Earth Observations in Support of Global Water Quality Monitoring. Greb, S., Dekker, A. and Binding, C. (eds.), IOCCG Report Series, No. 17, International Ocean Colour Coordinating Group, Dartmouth, Canada.
[30] https://coastobs.eu/
[31] Li, D., Hao, Y. and Duan, Y., (2020). Nonintrusive methods for biomass estimation in aquaculture with emphasis on fish: a review. *Reviews in Aquaculture*, *12*(3), pp.1390-1411.
[32] Baumgartner, L.J., Bettanin, M., McPherson, J., Jones, M., Zampatti, B. and Beyer, K., (2012). Influence of turbidity and passage rate on the efficiency of an infrared counter to enumerate and measure riverine fish. *Journal of Applied Ichthyology*, *28*(4), pp.531-536.

example, machine vision technology has been applied for estimating shrimp biomass based on eyeball length detection[33].

Machine vision technology does not only rely on accurate cameras with sufficient resolution and capture speed, but also the AI-based deep learning algorithms that are used to translate the captured images into useful information for decision making. For highly mobile species, such as fish, counting using machine vision requires various additional algorithms to account for animal overlap and movement, including tracking algorithms[34]. Cutting-edge commercial examples feature combined camera and embedded artificial intelligence in a single unit[35]. Other commercial examples include the XpertSea's XpertCount[36], a portable imaging device that enables the user to quickly and automatically count shrimp larvae inside a container; and the Aquabyte[37] software platform and computer vision technology, which provide lice recognition and counting, biomass estimation, appetite detection, and feed optimisation.

ASTRAL will develop machine vision technology using cutting-edge cameras and artificial intelligence applied to the identification of harmful phytoplankton, the determination of sea urchin biomass, and holistic water quality monitoring through bivalve behavioural response (biosensors)

5.2 Acoustics

Acoustic-based methods use either sonar cameras (imaging sonar) or echosounders to capture data on biomass, and are divided into active and passive acoustics. Unlike optical devices, these sensors can operate in the dark and in turbid environments. Passive acoustics use hydrophones to detect sounds and do not emit signals into water, while active acoustics consists of transmitter units that emit sound frequencies into the water, such as echosounders and sonar cameras. Echosounders emit a frequency that is reflected back to the sensor when it encounters a substance of different density to the water; the scattered echo signals are measured by the receiver and converted to voltage parameters. While frequently used in fish finders to locate shoals of fish for commercial and recreational fishing, its application in fish farming and aquaculture can be more difficult; the EU-funded PerformFISH[38] project is addressing challenges such as accurately determining the density and biomass of fish in a net pen[39]. Commercial options based on split beam active acoustic technology are also available[40]. While echosounders are capable of sampling large volumes of water, they have difficulty in restricted study

[33] Chen, F., Xu, J., Wei, Y. and Sun, J., (2019). Establishing an eyeball-weight relationship for *Litopenaeus vannamei* using machine vision technology. *Aquacultural Engineering*, *87*, p.102014.
[34] Pérez-Escudero, A., Vicente-Page, J., Hinz, R.C., Arganda, S. and De Polavieja, G.G., (2014). idTracker: tracking individuals in a group by automatic identification of unmarked animals. *Nature methods*, *11*(7), pp.743-748.
[35] https://www.innovasea.com/aquaculture-intelligence/biomass-estimation/
[36] https://xpertsea.com/valuable-insights
[37] http://www.aquabyte.ai/index.html
[38] http://performfish.eu/
[39] https://thefishsite.com/articles/using-echo-sound-to-estimate-biomass-in-aquaculture
[40] https://www.biosonicsinc.com/products/aquaculture-biomass-monitor/

areas (tanks) and species identification[41]. Imaging sonar converts sound from acoustic sensors into video images, but have complex processing requirements and are data and computationally intensive and expensive. The feasibility of hydro-acoustic techniques[42] as well as combined optical and acoustic systems[43] have been assessed for estimating cultivated macroalgae biomass, with promising results.

5.3 Biosensors for Physiological Monitoring

While some of these techniques have been applied as part of holistic water quality monitoring approaches mentioned in section 4.1, here the sensors are applied to "sentinel animals" that live in a common environment while acting as representatives of the cultivated stock, providing real time monitoring of physiological and behavioural indicators of animal health and well-being. Examples include measurements of body temperature, orientation, heart rate, and movements which are related to both physiology (e.g. metabolism, energy expenditure, and growth rates) and external stressors (e.g. water quality, handling, light) through rigorous calibration experiments[44].

The AE-FishBit[45], a small implanted biosensor for fish consisting of a tri-axial accelerometer, a microprocessor, a battery and a RFID tagging system, was developed as part of the AquaExcel H2020[46] project; this device monitors the respiratory frequency of individual fish, but needs to be recovered from the fish to download the data.

Acoustic telemetry is a method where individual animals are tagged with electronic transmitters containing sensors that measure different variables (e.g. depth, 3D positioning, acceleration, orientation); the data are sent to submerged receivers using acoustic signals. While this method provides detailed and valuable information on individual animals instead of entire populations, it requires handing and performing surgery which has inherent risks and can affect animal survival and behaviour[47].

5.4 Remote Sensing of Biomass

Remote sensing is usually applied in the field of aquaculture as part of water quality monitoring (usually in terms of harmful algal bloom detection), site selection, and production monitoring[48].

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[41] Li, D., Hao, Y. and Duan, Y., (2020). Nonintrusive methods for biomass estimation in aquaculture with emphasis on fish: a review. *Reviews in Aquaculture*, *12*(3), pp.1390-1411.

[42] Lubsch, A., Burggraaf, D., & Lansbergen, R. (2020). *Feasibility study on remote estimation of biomass in a seaweed cultivation farm applying sonar: technical report* (No. C110/20). Wageningen Marine Research.

[43] Tang, G., Li, Y., Wills, P. S., Hanisak, D., & Ouyang, B. (2021, April). Development of a macroalgal biomass sensor for an integrated multi-trophic aquaculture (IMTA) system. In *Big Data III: Learning, Analytics, and Applications* (Vol. 11730, p. 1173007). International Society for Optics and Photonics.

[44] Andrewartha, S. J., Elliott, N. G., McCulloch, J. W., & Frappell, P. B. (2015). Aquaculture sentinels: smart-farming with biosensor equipped stock. *J. Aquac. Res. Dev*, 7, 1-4.

[45] https://www.aquaexcel2020.eu/news/ae-fishbit

[46] https://www.aquaexcel2020.eu/

[47] Føre, M., Frank, K., Norton, T., Svendsen, E., Alfredsen, J. A., Dempster, T., ... & Berckmans, D. (2018). Precision fish farming: a new framework to improve production in aquaculture. *Biosystems engineering*, *173*, 176-193.

[48] Gernez, P., Palmer, S. C., Thomas, Y., & Forster, R. (2021). Remote Sensing for Aquaculture. *Frontiers in Marine Science*, *7*, 1258.

Remote sensing techniques based on reflectance from either satellite[49, 50] or unmanned aerial platforms (e.g. Che et al 2021[51]) have been used for seaweed biomass estimations. Bell et al (2020)[52] compared the performance of several remote sensing platforms (including satellite sensors, UAV-mounted optical sensors, underwater imagery and side-sonar scanning) for offshore kelp production, and their ability to provide information on canopy area, density, and tissue nitrogen content using imaging spectroscopy to estimate the physiological condition of the plants.

Conventional satellite-based methods have a number of limitations with the quality of the images being influenced by local climatic conditions[53]. Moreover, the use of satellite images with high spatial, temporal and spectral resolutions that may meet the needs of local applications is commonly unfeasible for most long-term monitoring programs[54]. Unmanned aerial vehicles have flexibility in flight scheduling, low cost and good spatial resolution. They have been increasingly used in environmental monitoring applications (*ibid.*) and may provide quick responses for short-term monitoring of HABs[55].

Remote sensing techniques can also include in-water measurements: the use of a flipper-propelled underwater robot (U-CAT) for the surveillance of fish and sea-based aquaculture cages was assessed in the AquaExcel H2020 project; part of the research involved assessing the influence of the robot on the behaviour and welfare of the fish (compared to using divers or thruster-driven robots) [56].

6 Threat Detection

6.1 Phytoplankton (Harmful Algal Blooms)

Phytoplankton are photosynthetic micro-organisms that are present in all natural water bodies. Their presence cannot be detected with the naked eye unless they are in sufficient concentrations as to discolour the water and/or affect its turbidity; but even then, individual algal cells cannot be distinguished without magnification. All phytoplankton contain chlorophyll a (Chl-a) as their primary photosynthetic pigment, while different species also include a variety of accessory pigments. It is the presence of these pigments that can discolour the water when they occur in sufficient concentrations

[49] Hu, L., Zeng, K., Hu, C., & He, M. X. (2019). On the remote estimation of Ulva prolifera areal coverage and biomass. *Remote Sensing of Environment*, 223, 194-207.

[50] Lu, T., Lu, Y., Hu, L., Jiao, J., Zhang, M., & Liu, Y. (2019). Uncertainty in the optical remote estimation of the biomass of *Ulva prolifera* macroalgae using MODIS imagery in the Yellow Sea. *Optics express*, 27(13), 18620-18627.

[51] Che, S., Du, G., Wang, N., He, K., Mo, Z., Sun, B., ... & Mao, Y. (2021). Biomass estimation of cultivated red algae Pyropia using unmanned aerial platform based multispectral imaging. *Plant Methods*, *17*(1), 1-13.

[52] Bell, T. W., Nidzieko, N. J., Siegel, D. A., Miller, R. J., Cavanaugh, K. C., Nelson, N. B., ... & Griffith, M. (2020). The utility of satellites and autonomous remote sensing platforms for monitoring offshore aquaculture farms: A case study for canopy forming kelps. *Frontiers in Marine Science*, *7*, 1083.

[53] Zohdi, E. and Abbaspour, M. (2019). Harmful algal blooms (red tide): a review of causes, impacts and approaches to monitoring and prediction. *International Journal of Environmental Science and Technology*, 16(3):1789–1806.

[54] Wu, D., Li, R., Zhang, F., and Liu, J. (2019). A review on drone-based harmful algae blooms monitoring. *Environmental monitoring and assessment*, 191(4):1–11.

[55] Dugdale, S. J. (2007). An evaluation of imagery from an unmanned aerial vehicle (UAV) for the mapping of intertidal macroalgae on Seal Sands, Tees Estuary, UK. PhD thesis, Durham University.

[56] Kruusmaa, M., Gkliva, R., Tuhtan, J. A., Tuvikene, A., & Alfredsen, J. A. (2020). Salmon behavioural response to robots in an aquaculture sea cage. *Royal Society open science*, 7(3), 191220.

(it should be noted that even non-photosynthetic phytoplankton can discolour the water, as in the case of *Noctiluca scintillans*). A healthy population of these micro-organisms can be interpreted as a good indicator of the health of the foundational ecosystem of a water body. Their presence is essential, as they form the base of aquatic food webs and contribute about half the earth's photosynthetic capacity[57].

Although phytoplankton are generally benign, some can have potentially devastating negative effects on animal and ecosystem health; these are referred to as harmful algal blooms (HABs) and include phytoplankton which are harmful due to their overabundance (e.g. resulting in a lack of dissolved oxygen after bloom collapse and decay), their ability to cause physical damage (e.g. blocking fish gills), or their ability to produce toxins that can either harm the animals or the people who consume the contaminated animals. It is not unusual to find HAB species in mixed phytoplankton assemblages. Thus, is it important that aquaculture management includes a good understanding of regional harmful algal species, the local environmental conditions under which they have a tendency to bloom, the concentrations thresholds at which blooms become harmful to cultured stock, and appropriate monitoring and mitigation strategies.

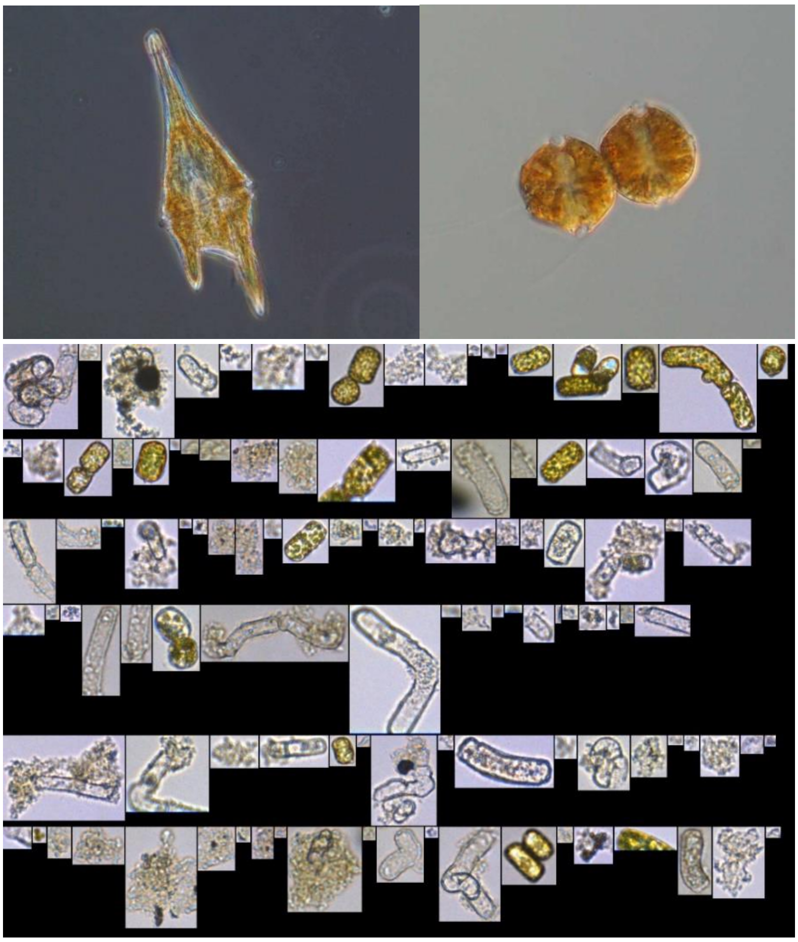

Figure 8. *Ceratium furca* (top left) and *Alexandrium catenella* (top right) as seen under a microscope (magnification 100x; credit: G. Pitcher). Both are bloom-forming species along the South African coastline. Bottom: a variety of Flow Cam image captures (magnification 40x; credit: SAMS).

[57] Behrenfeld, M. J., Randerson, J. T., McClain, C. R., Feldman, G. C., Los, S. O., Tucker, C. J., ... & Pollack, N. H. (2001). Biospheric primary production during an ENSO transition. *Science*, *291*(5513), 2594-2597.

The most accurate, accessible, and widely used phytoplankton species identification is cell microscopy. However, this method is time consuming and generally requires specialised knowledge and training in the preparation of samples and phytoplankton taxonomy, or the services of an appropriately skilled consultant. Unfortunately there are not currently many easily available, cost effective, automated phytoplankton identification platforms. Aquaculture farms generally rely upon flow cytometry, which requires highly specialised and expensive equipment such as a FLOWCYTOBOT[58] or FlowCam[59], and expert training and experience in the identification of individual phytoplankton cells. There is increased interest in Artificial Intelligence (AI) as a means to automate the identification process and some excellent progress has been made in various algal identification projects using AI and machine learning techniques [60,61,62].

ASTRAL will develop a prototype low-cost vision-based sensor with built-in Artificial Intelligence Deep Learning models for phytoplankton (>10 µm) classification and identification, representing a significant advance in the accessibility of phytoplankton identification technology.

Phytoplankton concentrations can also be approximated by determining the Chl-a concentration in water, a frequently used proxy for gross algal biomass. It is worth noting that while the emphasis is often on identifying high algal biomass, concentrations that are too low can also be a threat. Filter feeders like mussels depend on sufficient algal biomass to thrive in open aquaculture systems, and threshold low biomass concentrations require management action, for example the supplementation of appropriate feed. Additionally, there are toxic species that do not need to be present in large quantities to be problematic, so a simple Chl-a concentration product may not be adequate for every farm, and a good understanding of regional phytoplankton dynamics is needed.

Methods to measure phytoplankton biomass include the use of fluorometers. The Chl-a pigments in phytoplankton have the ability to fluoresce, meaning they can take in sunlight energy across the wavelength spectrum and re-emit a portion of it at a specific wavelength. Instrumentation to measure this response typically illuminates a water sample at the maximal excitation wavelength via the use of an LED, and reads the emission at the wavelength of maximal response. The strength of this signal is approximately proportional to the algal biomass. While the fluorescence response of individual cells may vary as a result of many physiological and environmental factors, the use of fluorometers is generally considered to be a sufficiently reliable method for measuring algal biomass indirectly for

[58] https://www2.whoi.edu/site/andersonlab/instrumentation/imaging-flowcytobot-ifcb/
[59] https://www.fluidimaging.com/
[60] Li, C., Ma, P., Rahaman, M.M., Yao, Y., Zhang, J., Zou, S., Zhao, X. and Grzegorzek, M., 2021. A State-of-the-art Survey of Object Detection Techniques in Microorganism Image Analysis: from Traditional Image Processing and Classical Machine Learning to Current Deep Convolutional Neural Networks and Potential Visual Transformers. *arXiv preprint arXiv:2105.03148*.
[61] Pastore, V.P., Zimmerman, T.G., Biswas, S.K. *et al.* Annotation-free learning of plankton for classification and anomaly detection. *Sci Rep* 10, 12142 (2020). https://doi.org/10.1038/s41598-020-68662-3
[62] Kaichang Cheng, Xuemin Cheng, Yuqi Wang, Hongsheng Bi, and Mark C. Benfield. 2019. Enhanced convolutional neural network for plankton identification and enumeration. *PLoS One* 14, 7 (July 2019), e0219570.

water quality purposes. These sensors can take the form of benchtop units (e.g. where pigments in a water sample are first extracted and then measured) or in water optical units.

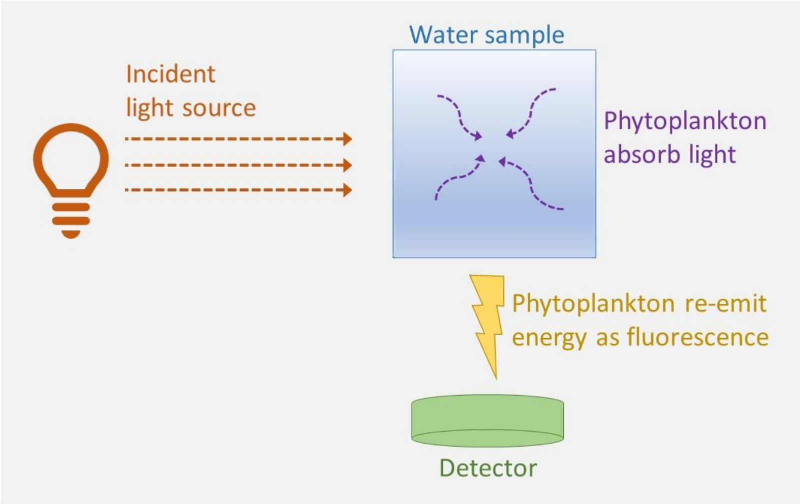

Figure 9. An illustration of the fluorometer concept

Sensors on board satellites can assist in the monitoring of HABs directly using ocean colour (e.g. Chl-a concentration, fluorescence, particle size) or indirectly through the environmental conditions that might facilitate HABs (e.g. sea surface temperature or wind speed and direction). Ocean colour remote sensing uses optical radiometers to measure light at specific wavelengths (mostly over a selection of wavelength bands) emerging from the water surface. The simplest of Chl-a detection techniques relate waveband ratios empirically to Chl-a concentrations with varying degrees of error depending on how the calculation is made and the environmental conditions at the time of measurement, both in-water and atmospheric. Most international space agencies' environmental monitoring satellites provide Chl-a data products as a part of their freely available product suites; these data are usually available at near-daily time scales and spatial resolutions of between 300 and 1200 meters. Apart from a handful of species that display distinctive spectral signatures at sufficiently high biomass, species-level identification and detection from satellites is inherently elusive; *in situ* identification is still required to determine the species composition of most blooms. The true value of satellite-based HAB monitoring is for the early detection of potentially harmful high biomass phytoplankton blooms, which can actuate *in situ* sampling; once the species composition of a bloom has been determined, satellite products can be used for continued synoptic-scale monitoring of the movement, spatial extent, and biomass. A comprehensive review on the use of remote sensing for HAB observation can be found in IOCCG report 20[63].

[63] IOCCG (2021). Observation of Harmful Algal Blooms with Ocean Colour Radiometry. Bernard, S., Kudela, R., Robertson Lain, L. and Pitcher, G.C. (eds.), IOCCG Report Series, No. 20, International Ocean Colour Coordinating Group, Dartmouth, Canada. http://dx.doi.org/10.25607/OBP-1042

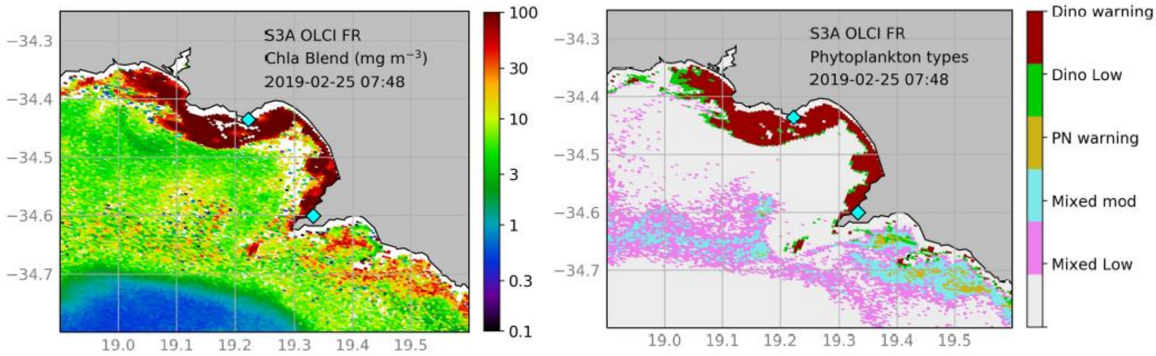

Figure 10. Satellite products derived from full resolution Sentinel 3A Ocean Colour and Land Imager for the 25th of February 2019, showing a yessotoxin producing dinoflagellate bloom (dominated by *Gonyaulax spinifera*) in Walker Bay on the southwestern coast of South Africa. The left panel shows the chlorophyll a concentration, whereas the right panel shows the probabilistic phytoplankton community concentration (from Smith and Bernard, 2020[64]).

Cyanobacteria present a particular challenge to freshwater aquafarming, notably in inland water bodies which tend towards eutrophication[65, 66]. The unique chemical character of cyanotoxins has led to the development of a number of methodologies to identify the presence of toxic cyanobacteria in water, including Polymerase Chain Reaction (PCR) tests. Results from chemical analyses vary widely and it is largely agreed that there is no industry standard for testing for cyanobacteria[67]. Bickman et al.[68] recently reported a portable biosensor system for rapid detection of freshwater cyanotoxins. It is a planar waveguide optical sensor that delivers quantitative fluorescent competitive immunoassay results in a disposable cartridge. More of this type of portable biosensor is in great demand in the aquatic industry. ASTRAL is developing a camera-based device coupled with a microfluid pump and microscope lens that enables high-quality image/video capture. Trained AI models carry out phytoplankton classification over new image samples.

There are a number of satellite-based HAB monitoring and prediction services available online for varying coastal and inland waters. In the EU, the PRIMROSE project[69] for shellfish safety provides HAB warnings for coastal aquaculture using satellite remote sensing. Likewise, NOAA's HAB Operational Forecast System[70] provides monitoring and forecasts for the Gulf of Mexico. In South Africa, the OCIMS programme[71] provides web-based HAB and aquaculture Decision Support Tools[72]. There are many other regional examples.

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[64] Smith, M. E., & Bernard, S. (2020). Satellite ocean color based harmful algal bloom indicators for aquaculture decision support in the southern Benguela. *Frontiers in Marine Science*, *7*, 61.

[65] Onyango, D.M., Orina, P.S., Ramkat, R.C., Kowenje, C., Githukia, C.M., Lusweti, D. and Lung'ayia, H.B., 2020. Review of current state of knowledge of microcystin and its impacts on fish in Lake Victoria. *Lakes & Reservoirs: Research & Management*, *25*(3), pp.350-361.

[66] Amit Kumar Sinha, Michael A. Eggleton, Rebecca T. Lochmann, An environmentally friendly approach for mitigating cyanobacterial bloom and their toxins in hypereutrophic ponds: Potentiality of a newly developed granular hydrogen peroxide-based compound, Science of The Total Environment, Volumes 637–638, 2018, Pages 524-537, ISSN 0048-9697, https://doi.org/10.1016/j.scitotenv.2018.05.023.

[67] Su X, Sutarlie L, Loh XJ. Sensors, biosensors, and analytical technologies for aquaculture water quality. *Research*. 2020 Feb 17;2020.

[68] Bickman, S.R., Campbell, K., Elliott, C., Murphy, C., O'Kennedy, R., Papst, P. and Lochhead, M.J., 2018. An innovative portable biosensor system for the rapid detection of freshwater cyanobacterial algal bloom toxins. *Environmental science & technology*, *52*(20), pp.11691-11698.

[69] https://www.shellfish-safety.eu

[70] https://oceanservice.noaa.gov/facts/hab-forecast.html

[71] https://www.ocims.gov.za/

[72] https://www.ocims.gov.za/hab/app/

6.2 Pathogens

This category of threats to water quality encompasses living organisms in water that cause disease. Pathogens can be bacterial, viral, fungal or fungal-like (oomycetes) and other eukaryotes or animal parasites. Pathogens present a serious threat to the health of aquaculture systems and the successful production of the cultivated species.

Every aquaculture farm should have a comprehensive Biosecurity Programme in place detailing the Welfare Indicators for each farmed species. The OIE (International Organisation for Animal Health) lists a number of reportable diseases in accordance with the Aquatic Code[73], and ongoing testing for the relevant pathogens should form part of the Biosecurity Programme. The presence of a pathogen in a farming system and/or animal is often first identified as stress or a change in behaviour of the animal. At the first signs of distress or disease, animals should be closely examined and monitored, and written records kept of their condition until clinical observations can be made by the attending veterinarian. Observations on the farm are of critical importance in the detection and identification of pathogens, and it is essential that all staff are aware of the huge advantages of early detection of disease in the system. Operational Welfare Indices (OWI's) play a role in the ongoing monitoring of the health statue of stocks.

Known behavioural changes and certain clinical signs can be the first step towards the identification of stress and disease cause by specific pathogens. For example, the appearance of ulcerative lesions (open dermal ulcers) in certain freshwater and estuarine fishes are indicative of contaminated or stressed environments and possible infection with *Aphanomyces invadans*, the causative agent of Epizootic Ulcerative Syndrome (EUS), whereas the appearance of small white nodules on the foot, epipodium and mantle of abalone are indicative of tubercle mycosis, a disease caused by the marine oomycete *Halioticida noduliformans* (Figure 11). Characteristic clinical signs of vibriosis in certain fish species include red spots on the ventral and lateral areas and swollen, dark skin lesions that ulcerate and fish scratching against objects and flashing is often a sign of parasitic infections. In the absence of a pathogen that can be detected by visual inspection (like a macroparasite), a veterinarian will confirm the initial diagnosis using histopathology, microbiology (especially for bacteria, fungi, oomycetes) and/or microscopy methods to confirm the initial diagnosis. Histopathology is the detection of change in animal tissue(s) resulting from physiological stress or infection by a disease agent, and requires biopsy, chemical fixation of the tissues, and examination under a microscope. This method works well for identifying known pathogens and it is a commonly used route for detecting emerging (new to the farm) pathogens.

[73] Oidtmann, B., Johnston, C., Klotins, K., Mylrea, G., Van, P.T., Cabot, S., Martin, P.R., Ababouch, L. and Berthe, F., 2013. Assessment of the safety of aquatic animal commodities for international trade: the OIE aquatic animal health code. *Transboundary and emerging diseases*, *60*(1), pp.27-38.

Figure 11. *Haliotis midae* exhibiting typical clinical lesions of tubercle mycosis caused by the marine oomycete *Halioticida noduliformans.* Infected abalone were characterized by multifocal areas of necrosis of the epithelium, underlying muscle fibres and connective tissues of the foot, epipodium and mantle. The lesions were typically 2–3 mm in diameter. (credit: Brett Macey)

A common and serious genus of pathogen in fish and shellfish aquaculture worldwide is *Vibrio*. Many Vibrio species, such as *Vibrio vulnificus*, *Vibrio harveyi*, *Vibrio anguillarum*, *Vibrio alginolyticus*, *Vibrio parahaemolyticus* and *Vibrio salmonicida*, can cause mass mortality and vibriosis (marked by infection on skin and other organs) in many cultured fish, shrimps, and shellfish. About a dozen *Vibrio spp.* cause infections in humans, one of which causes cholera (*Vibrio cholerae)* and several are zoonotic (for example *V. parahaemolytics, V. vulnificus*) causing infection in both humans and animals.

Analytical methods for quantitative and qualitative pathogen detection in aquaculture include culture-based methods, molecular methods (e.g. PCR, real-time PCR, ELISA, immunohistochemistry), biosensors, and microscopy observation methods. Culture-based methods are considered the current standard[74], but polymerase chain reaction PCR and more recently real-time PCR technologies are much faster, more sensitive and precise, and provide results within hours rather than days. The OIE Manual of Diagnostic Tests for Aquatic Animals provides a comprehensive guide to the formal identification of, and proof of freedom from, common pathogens, detailing PCR and real-time PCR as the preferred test for quantification and identification of many pathogens, and a requirement for proof of freedom from disease when exporting animals. PCR is very sensitive but identifies only the presence of the targeted pathogen. For assessing the concentration of pathogen, a real-time PCR assay can be used. Real-time technologies make use of fluorescent dyes that can be detected during each step of the PCR process, thus allowing for real-time quantification of the pathogen. Since several dyes are available, one can also multiplex, allowing for simultaneous detection of more than one pathogen or gene.

[74] Chatterjee, S. and Haldar, S., 2012. Vibrio related diseases in aquaculture and development of rapid and accurate identification methods. *Journal of Marine Science Research and Development S*, *1*(1), pp.1-7.

Nucleic acid probes (DNA & RNA) are very sensitive, and are useful for discerning between closely related pathogens of which one may be harmful and the other not, thereby avoiding unnecessary generalised disinfection (PCR also has this capability). These methodologies are typically designed to detect known bacteria or viruses and are less well suited for identification of new, unknown or previously unidentified pathogens[75].

Figure 12. From left to right: PCR technology requires sophisticated laboratory equipment and expertise; DNA/RNA probe instrument; Lateral flow test kit. Photo credits: CDC

The PCR group of analyses, now commonly understood as the benchmark in detecting the SARS-COV-2 virus causing COVID-19, require microbiological laboratory equipment and expertise, and technological efforts are focusing on rapid-result probes for use in situ by farmers. LAMP (Loop mediated isothermal AMPlification) technology enables DNA amplification, and portable instrumentation is increasingly available as a low cost method of detecting certain pathogens (see Fig. 12 left and centre, full PCR vs. a probe instrument). Commercially available molecular probes have been developed for IHHNV and type-A baculovirus which are common concerns in shrimp farming.

Lateral flow tests (“strip tests”; Fig. 12, right) can be considered the most portable form of sensor that combines sample separation, interaction, and detection in one chromatographic strip. In the case of COVID-19, they detect proteins (antigens) associated with the virus. They can be set up to target either viral RNA or bacterial cells[76], but lack sensitivity when compared with the development of low-cost portable sensors for organic chemical, biochemical and biological hazards is still somewhat behind those designed for physical water parameters, but the high demand from the aquaculture industry is driving concentrated effort towards their improvement and increased availability.

It should be noted that, as for harmful algal species, many pathogenic organisms are ubiquitous in marine and aquaculture environments, and that the management of water quality, biosecurity and animal husbandry can in large part determine whether they become problematic. That is to say, efforts at detecting pathogens should not only be concentrated on intake water, and strict biosecurity can not necessarily prevent them. Many pathogen types can be considered opportunistic in the sense that they may regularly be present in small quantities but thrive when water quality is not optimally maintained or when environmental conditions become favourable for them. So there is integrated feedback between management practices, the maintaining of environmental and health standards, and the proliferation of pathogens.

[75] Subasinghe, R.P., Curry, D., McGladdery, S.E. and Bartley, D. (2003). Recent technological innovations in aquaculture. *FAO Fisheries Circular*, *886*.
[76] Su, X., Sutarlie, L., & Loh, X. J. (2020). Sensors, biosensors, and analytical technologies for aquaculture water quality. *Research*, 2020.

The ASTRAL project addresses the presence and effects of pathogens on IMTA through literature review and analytical data collection, and will provide recommendations for future pathogen monitoring programmes

6.3 Microplastics

Microplastics are a contaminant of natural waters that have been identified as an increasing threat to ecosystem health. These are small pieces of plastic less than 5mm in length and can be so small as not to be visible with the naked eye. They are a man-made pollutant, entering water bodies via processes such as storms, water runoff and wind - as well as directly by littering. Microplastics are the result of the photochemical and mechanical driven breakdown of larger plastic items but are also the result of certain manufacturing processes. Regardless of the origin or means of arrival in water bodies, they present an increasing threat to both human and animal life.

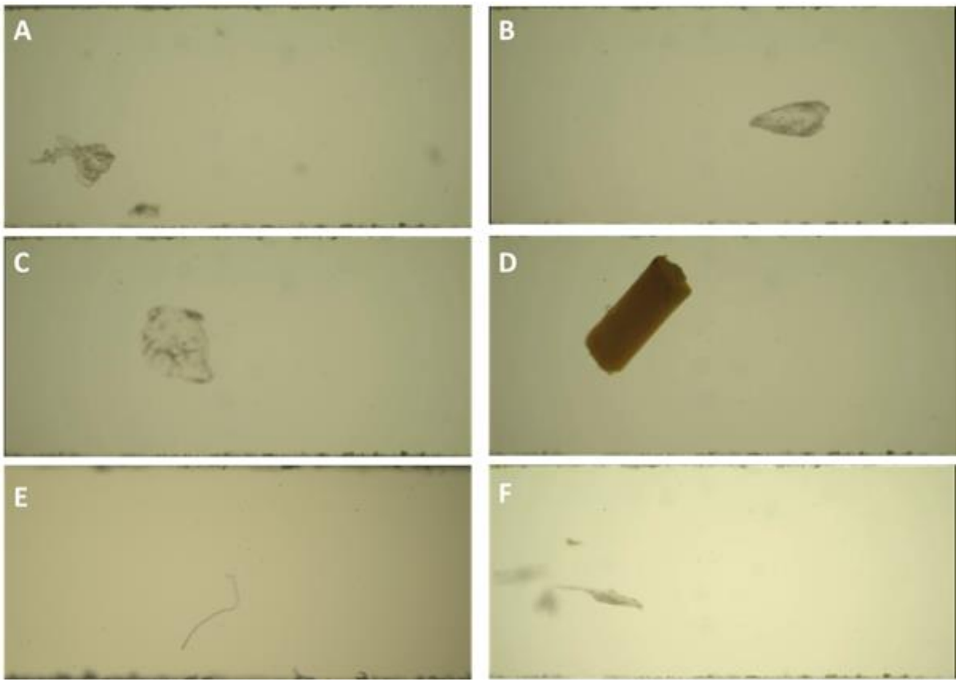

Figure 13. Images taken during the validation of the imaging block of the microplastic sensor prototype. A,B,C) Polystyrene microplastics with sizes of 0.5 to 1 mm. D) Opaque plastic cylinder of 0.7 x 2.5 mm^2 used in validation of the particle detection block. E) Plastic microfiber. F) Example of focused and non-focused particles on the same frame.

It has been established that microplastics represent a substrate for biofilm formation (a thin layer of bacterial cells). The presence of biofilms allows the rapid coagulation with biogenic particles, including phytoplankton[77]. So, filter-feeding organisms can easily ingest microplastic aggregations along with

[77] Michels, J., Stippkugel, A., Lenz, M., Wirtz, K. and Engel, A., 2018. Rapid aggregation of biofilm-covered microplastics with marine biogenic particles. *Proceedings of the Royal Society B*, *285*(1885), p.20181203.

their phytoplankton food source. Microbial biofilms associated with microplastics have been shown to promote pathogen transmission[78], compounding that category of threat to water quality. Many plastics are inherently toxic, containing a variety of chemicals like phthalates and bisphenol A. Furthermore, they can absorb harmful chemicals co-occurring in the environment, increasing their toxicity – this happens for example when microplastics are found together with polychlorinated biphenyls (PCB's), also produced in manufacturing processes and which have been linked to cancer and other serious human and animal health problems. It has recently been discovered[79] that sunlight can not only break plastics down into microplastics, but can alter the chemical composition of these micro particles, producing tens of thousands of water-soluble compounds. It is still currently unclear exactly how these compounds may affect the environment.

A number of techniques and technologies have been developed to identify microplastics in water[80]. The simplest of these just identifies the presence of polluting particles, while more complex processes can identify the polymers involved. The question of microplastics is very much still under research, and operational systems for detecting them in commercial environments are not yet readily available. With access to a microscope laboratory, some microplastics can be identified and quantified by basic magnification.

ASTRAL aims to characterize the occurrence, size range, and composition of microplastics as an emerging pollutant, assess the impacts of microplastics on IMTA production, and provide recommendations for regional microplastics monitoring programmes

A range of chemical techniques is available for the characterisation of microplastics, all requiring sophisticated laboratory facilities and expertise. Some of these methods include Raman spectroscopy (using particulate light scattering to identify composition), infra-red Fourier Transformed spectroscopy (using absorption properties to identify composition), and thermo-analytical gas chromatography (during thermal decomposition large molecules cleave at their weakest bonds, producing smaller, more volatile fragments, which are then identified using mass spectrometer). With the growing applicability of AI and ML to visual identification techniques, it is anticipated that these methods will attract increasing attention and investment.

It is worth noting that aquaculture activities themselves also carry the risk of contributing to pollution in marine and freshwater environments. The EU AQUA-LIT[81] project aims to provide a sustainable toolbox of innovative ideas and methodologies to: prevent marine littering from aquaculture activities;

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[78] Harrison, J.P., Sapp, M., Schratzberger, M. and Osborn, A.M., 2011. Interactions between microorganisms and marine microplastics: a call for research. *Marine Technology Society Journal*, *45*(2), pp.12-20.
[79] Walsh, A.N., Reddy, C.M., Niles, S.F., McKenna, A.M., Hansel, C.M. and Ward, C.P., 2021. Plastic Formulation is an Emerging Control of Its Photochemical Fate in the Ocean. *Environmental Science & Technology*, *55*(18), pp.12383-12392.
[80] Prata, J.C., da Costa, J.P., Duarte, A.C. and Rocha-Santos, T., 2019. Methods for sampling and detection of microplastics in water and sediment: A critical review. *TrAC Trends in Analytical Chemistry*, *110*, pp.150-159.
[81] https://aqua-lit.eu/

have better monitoring schemes in place; and remove and recycle litter from the aquaculture facilities both before litter enters the sea and for litter already existing at sea.

The ASTRAL project is developing a low-cost benchtop microscopic imaging sensor for the detection of microplastics in the range of 0.5 mm to 5 mm in diameter from a discrete water sample

7 Environmental Variables

7.1 Light (Photosynthetically Active Radiation)

Micro- and macro-algae photosynthesise primarily using light in the visible part of the wavelength spectrum (blue to red, 400 to approximately 700 nm). Incoming solar radiation from the sun spans a vast wavelength range and many different radiation types, but the proportion of incoming solar radiation that is available for photosynthesis, and hence in large part determines the potential for growth in algae, is photosynthetically active radiation (PAR).

Measuring PAR can be a useful way of monitoring light conditions in an IMTA setup. For water quality considerations, high light levels can accelerate photosynthesis and therefore algal production, which may not be desirable. Low light levels inhibit photosynthesis in many organisms and can be an indication to monitor dissolved oxygen levels closely at that time – oxygen in water is produced by algal photosynthesis, and if this is inhibited, dissolved oxygen levels may fall, with great risk to higher trophic level organisms. Likewise, fluctuating carbon dioxide levels will affect pH: photosynthesis consumes carbon dioxide, and if this process is inhibited, increased levels of dissolved CO_2 may lead to acidification.

PAR also directly affects water temperature as well as the air temperature (although not as much as infra-red radiation). Farmed aquaculture species often have an optimal temperature range for their well-being and maximal production. With IMTA systems using outdoor tanks, raceways or other water containing infrastructure, prolonged exposure to high levels of PAR (and/or UV light) can directly affect animal well-being. This is something requiring consideration in IMTA site positioning, design and the selection of farmed species. Mitigation strategies such as shade structures for sensitive animals can be considered in such conditions. For seaweed, seasonal growth patterns are governed by daylength and seasonal irradiance, and these should be well understood in the context of observed variability in light availability.

It is important that PAR is measured in quantum units, which can be added up over an hour, day or year according to monitoring needs. This requires the measurement of the photon flux density, the number of photons passing through a particular area per unit time. It is usually expressed in micromoles of photons per square meter per second (μmol/m^2/s). Measuring PAR can be done from the ground, with an *in situ* mooring such as a buoy, or by satellite. There are a number of near-real time satellite PAR standard data products freely available from the major scientific satellite operators across the globe. These are appropriate for regional PAR estimates and trend analysis, with reasonable accuracy at varying spatial resolutions depending on the sensor. For unique site-specific conditions, for example monitoring light levels at different times of day, different sun angles, across shaded areas and so on, a ground based or hand-held PAR sensor would be better.

A variety of inexpensive hand-held PAR sensors and probes are available commercially for both above-water and underwater measurements. Some give a single integrated PAR value across the visible spectrum, some give spectral information as well, at selected wavelengths or (approximately) continuously across a larger wavelength range. Many are available as part of an integrated sensor measuring a range of different air and/or water quality parameters.

It can be useful to monitor PAR from a mooring if farming in IMTA systems with large ponds/tanks or in nets/ponds at sea. Again, there is a variety of available sensors, the additional considerations with a mooring being the physical robustness of the instrument as well as biofouling. It should also be remembered that having to collect data manually from a mooring is not generally operationally efficient, and moored instruments should have some kind of telecommunications facility in order to be able to receive data and communicate with the instrument remotely, particularly with regards health and functioning of the sensor.

7.2 Weather

While variability in PAR is largely driven by region and season, there are many other changeable environmental variables which can be generally included under the umbrella term of “weather”. Variability in air temperature, wind conditions, precipitation forecasting and cloud cover trends and forecasts are all potentially valuable information depending on the exposure of the IMTA system to outside elements.

An understanding of localised seasonal weather conditions and variability within seasons is usually a requirement for the site selection process. One important weather-related consideration is the site’s vulnerability to extreme events. A trend of heavy rainfall may lead to flooding, and extreme rainfall events can impact upon water salinity. Likewise, lack of rainfall can affect surface run-off patterns, with potentially devastating effects on seaweed raft farms, for example. Large temperature fluctuations (very hot days and cold nights, or hot summers and freezing winters) need to be addressed in structural design, and the possibility of extreme upwelling events should be assessed in terms of the potential for a dramatic drop in the temperature of intake water. Vulnerability to high winds is another consideration which can inform appropriate design and orientation of physical IMTA infrastructure. Heavy fog can cause operational delays if good visibility is necessary for water body sampling with a small craft. Large storms and the resulting swells can damage land-based infrastructure too close to the coast, and sea-based infrastructure is often damaged by storms or other extreme events.

Fortunately, there is a large range of easily available technology and applications which make it straightforward to gather historical weather data, record current weather conditions, and acquire accurate forecasts from local or international service providers, many of them at no cost.

In the absence of an internet connection, or for monitoring very localised weather conditions, an off-the-shelf weather station can be very useful. These are easily and inexpensively available, with a wide choice of instruments available for integration into these units.

7.3 Sea state

As with weather conditions, the main aim of monitoring sea state conditions is to be able to forecast or predict conditions in terms of whether they are going to be favourable or unfavourable for targeted IMTA activities. Vulnerability to sea state (or surface water state in freshwater bodies) is relevant mainly to IMTA farming either completely open to a large water body or adjacent to one, although it can be relevant for adjunct IMTA activities such as water sampling from outside the farm, where surface roughness may prohibit the use of a small boat, for example.

Sea state conditions can include current strength and direction predictions as well as waves and surface roughness monitoring. There are a number of instruments and systems available to do this. For powerful currents and/or those with large spatial extent, data products using satellite altimetry are increasingly accurate. Likewise for surface roughness and wave heights, satellite-borne Synthetic Aperture Radar (SAR) products are very useful. Mitigation of such risks can be built into farm design: for example, offshore seaweed systems tend to be situated deeper than onshore systems, to survive large swells at offshore sites.

In situ systems can include instruments like wave buoys moored at a fixed point which measure vertical motion as the buoy is buffeted by ocean conditions. If vulnerability to increased wave height is an identified risk at an IMTA farm, wave forecasts are accessible in much the same manner as weather forecasts, from online service providers. These generally make use of both satellite and in situ monitoring, and some systems allow integration of data from privately owned local instrumentation which can assist in forecasting accuracy.

Figure 14. The Mobilis DB2000 data buoy deployed at the Marine Institute, Lehanagh Pool, Ireland.

8 Technology in Support of IMTA: Considerations

8.1 Instrumentation

8.1.1 A Cost-Benefit Approach

It is important to invest some effort in determining an individual IMTA system's instrumentation requirements, both in terms of variables to monitor/measure but also in terms of the accuracy and precision required. Accuracy refers to the proximity of the measurement to the "true" quantity, and precision refers to the stability and repeatability of the measurement (by how much two measurements of the same quantity differ). The most obvious reason for determining threshold requirements is instrument cost. Very highly accurate and precise instrumentation is generally more expensive than lower cost instrumentation for less demanding requirements, and it may be the case that great accuracy and precision is not needed for a specific IMTA application. In complex systems where technology costs can be significant, a full cost-benefit analysis is recommended to determine the commercial advantages for risk identification and mitigation, versus the life cycle cost of the system from acquisition and installation to retirement, as well as operational and maintenance costs.

With aquaculture on the rise globally, there are an increasing number of so called "low cost" Water Quality monitoring platforms available. The term "low cost" is of course entirely relative. At one end of this scale of innovation there is the PROTEUS H2020[82] project, representing dramatic (10x) reductions in both unit function cost and unit size, compared with the state-of-the-art systems currently in use. The sensor integration on PROTEUS is extremely advanced, with a microfluidic sensing chip of carbon nanotubes and MEMS physical and rheological resistive sensors included.
Meanwhile, the WAZIUP[83] project acknowledges the need for very low-cost instrumentation for under-resourced farms, and has designed an integrated system for use in African fish farms with minimal financial resources. It simply measures pH, dissolved O2, and water temperature.

The development of the ASTRAL low-cost IoT Kit aims to find a balance between IMTA user requirements in terms of functionality, technology, and affordability

Affordability, efficiency and effectiveness are three valuable guiding principles when considering costs and benefits of expenditure on instrumentation. The development of the ASTRAL IoT kit, with its intent to provide a cost-effective yet flexible and interoperable communications solution (see section 8.2.2), represents the careful – and challenging - balance of these sometimes opposing principles.

[82] http://www.proteus-sensor.eu/
[83] https://www.waziup.eu/

8.1.2 Operational Environment

Aquaculture farming environments are generally not ideally suited for instrumentation designed to be housed in a laboratory. When used in the field, the primary consideration for instrument selection is usually portability, but sensitivity to wetness and salt (metal corrosion) is also a primary concern. Instruments designed for air temperature measurements, for example, may malfunction if immersed in water.

Instrumentation robustness or ruggedness should be considered when intending to operate them in the field. Shock cases (preventing damage when dropped), proper protection for transportation, waterproof housings – these are all additional investments but may represent vulnerability to an expensive risk if overlooked. If instruments are intended for deployment for any length of time within the IMTA system, they need to be sufficiently physically robust in order not to degrade over time by rust, constant motion, salt exposure, and other environmental hazards.

Securing a consistent and reliable power supply is another challenge faced by many aquaculture farms as they tend to be apart from city infrastructure and sometimes entirely off-grid. If power supply is a concern, this must be taken into consideration when designing any kind of monitoring system. Battery-operated sensors are one potential option, acknowledging that there is a cost involved in replacing and/or recharging batteries. A solar power installation is another potential solution, and these are increasingly available as plug-and-play units bought off the shelf. An understanding of local climatic and cloud cover conditions is of course a requirement here.

It takes at the very least, some level of knowledge to initiate (calibrate), operate and maintain even simple hand-held probes and other direct measurement instruments, and certainly a significant level of specialised knowledge when dealing with more complex integrated systems, or more specialised laboratory equipment. Personnel costs incurred by training and maintaining instruments should be included in the cost-benefit approach outlined above.

Figure 15. Examples of two types of operational aquaculture environments: the open ocean-based aquaculture system of the Ireland IMTA Lab at the Lehanagh Pool Marine Institute (left), and the land-based flow-through aquaculture system of the South African IMTA Lab at Viking Aquaculture's Buffeljags farm (right).

8.1.3 Calibration, Maintenance, Biofouling

Once an instrument has been selected for its appropriateness in terms of accuracy, precision, cost and robustness, it usually needs to be calibrated before use. Some instruments are shipped already calibrated; others will include instructions on how to do so on site. With consistent usage, or just over time, it is to be expected that instrumentation will suffer from measurement “drift”. Drift is the deviation of measurements from the accuracy and precision determined at the point of calibration. It is commonly caused by thermal fluctuations affecting delicate internal electronics and can inadvertently be accelerated by incorrect handling and storage of the instrument. A slow rate of drift is mostly considered unavoidable, but it is recommended that close attention be paid to the instrument’s calibration requirements as stipulated by the manufacturer. The process of calibration, whether performed in-house or by the instrument manufacturer, is a significant consideration in terms of direct expense, but also in terms of interruptions to operational usage especially if the instrument has to be shipped far afield for regular calibration.

Other forms of instrumentation maintenance should also be accommodated for in the planning and implementation of a monitoring system. Sensors require cleaning with specialised chemicals and lint-free wipes at regular intervals, sometimes before and after each measurement. Instrumentation with moving parts also requires specialist attention in order to maintain optimal functioning and avoid measurement interruptions.

Biofouling is a primary concern when deploying instruments in aquatic environments. Biofouling can result from algal growth, colonisation by floating larval filter feeders e.g. mussels, or simply from being entangled in seaweed or seagrasses. Sudden and dramatic incidents such as entanglement may be easily noticeable in the measurements, but this is often not the case. Algal growth on a turbidity sensor might block incoming light leading to a negative measurement bias, but this will be incremental and not necessarily identifiable from one day to the next. Vulnerability to biofouling is dependent on the unique aquatic environment and can generally only be established once an instrument has actually been deployed, with an appropriate cleaning and maintenance schedule implemented following close monitoring of the instruments and their measurements.

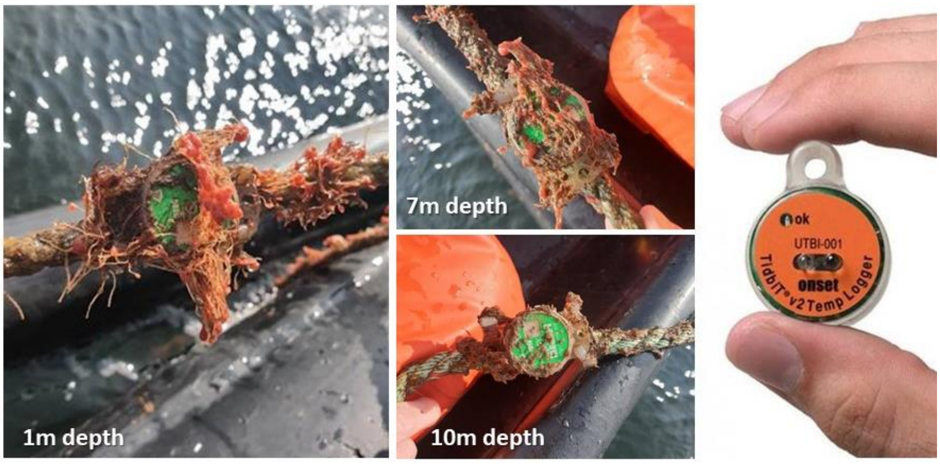

Figure 16. Examples of biofouling of temperature sensors (Tidbits) at different depths in an open water aquaculture system, with a clean sensor for reference on the right (credit: Marine Institute)

8.2 System Design

8.2.1 Automation and Efficiency

Apart from finding a balance between the appropriate sensor at a suitable cost and measurement sensitivity, some thought must be given to the most efficient use of the product. Sensors have different methods of displaying or capturing measurement information. The simplest of probe and sensor options may simply display the measurement on a small screen to be recorded by the user, or save the information to internal memory to be downloaded by the user at a later stage. While it is possible to do multiple manual measurements with a single probe and note down the values, this might be too inefficient and time-consuming for most commercial aquaculture operations; this is also not always a viable option for ocean-based farms which might require multiple measurements across different locations and depths.

Aquafarms differ in their monitoring frequency requirements: some parameters might be monitored at lower frequencies (e.g. daily, weekly) in open ocean settings, while closed and semi-closed aquaculture systems might require more frequent (e.g. hourly) or continuous measurements to ensure that the physicochemical water properties remain within the optimum ranges of the farmed organisms. Depending on the scale and intensity of the operation, a farm might have multiple sensor arrays operating across the facility. The ability to automate these measurements allows the user to consistently and efficiently capture information at the appropriate scales, while reducing the costs associated with personnel. The minimisation of human error and potentially increasing the quantity and quality of data collected can also be significant improvements to the overall monitoring system. It is important to bear in mind, however, that while personnel costs associated with making measurements may be reduced, there is little utility in gathering frequent measurements if they are not also frequently scrutinised. Large datasets require specialised skill in data handling and analysis. The application of Artificial Intelligence in monitoring software and systems (see section 8.2.4. Data Visualisation and Analytics) is a developing approach towards addressing this need.

Automation can be considered at any or all of the various levels of a measurement and monitoring system. It could simply form part of the measurement and recording, or part of the monitoring in terms of setting off system alerts if certain criteria are met, but could go as far as actively reacting and intervening, e.g. supplying feed once a set of criteria are met (e.g. SICA[84], Arvotec[85], Feed Smart), aerating the water, or lowering fish cages down the water column under specific environmental conditions.

Below are some of the main considerations for the development of automated monitoring systems:

- Energy-efficiency: Automated measurements, particularly in remote environments, are often dependent on energy consumption and frequently employ energy harvesting techniques such as solar panels. Often protocols are developed that allow sensors to enter “sleep-mode” in between measurements in order to save power.
- Cost: Lower cost systems may be more desirable and affordable for smaller farms.

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[84] http://www.ctninnova.com/en/proyectos/acoustic-sensor/
[85] https://www.arvotec.fi/

- Autonomous operation: The system may need to run autonomously for a long time without any maintenance, helping to reduce the cost of operations.
- Intelligence: Dynamic solutions for conserving energy, data predictions and algorithms to prevent data loss, improving the overall efficiency of system.
- Usability: Needs to be simple to understand with intuitive interfaces and quick to operate by non-specialist user.
- Data volumes: Frequent measurements can result in large volumes of data which are surplus to the needs of the farmer although they may be valuable for research purposes. Software that allows for the requirements of different user interfaces can be valuable. Personnel requirements for data analysis and curation should also be considered.
- Interoperability: The system must be compatible with different types of sensors and communication technologies depending on the requirements and available resources of specific farms.
- Data analytics: sensor data need to be transformed into valuable insights for the non-specialist user. Data processing relies on AI machine learning data pipelines to deliver tailored information.
- Real-time control: cloud-based and edge computing technologies enable real-time sensing and feedback control loops to intervene in the water environment when needed.

8.2.2 ICT Infrastructure and Connectivity

Automatic data collection systems can be taken a step further to form part of an Internet of Things (IoT) system with cloud-based data management. Depending on the scale and intensity of an aquaculture operation, a farm might have multiple sensor arrays deployed across the facility. With recent advances in sensor technologies and IoT approaches it is now possible to monitor and control sensors remotely through mobile computing and wireless communication technology.

An example IoT system applicable to the marine aquaculture landscape is illustrated in Figure 17; such a system could consist of one or more sensors, which are directly connected to an end-node containing a processing unit (micro controller and sensor software), power unit (controlling a battery, solar panel, and/or other power harvester), communication unit, and optional on-board data storage. There could be one or more of these end-nodes on a farm. An end-node could send data directly to an external server or the cloud; however, in the case of multiple end-nodes or power limitations, the system might include an additional local coordinator node which acts as a gateway to the external server or the cloud, and facilitates both local and secure remote data transfer, monitoring and management of all the end-nodes and their sensors.

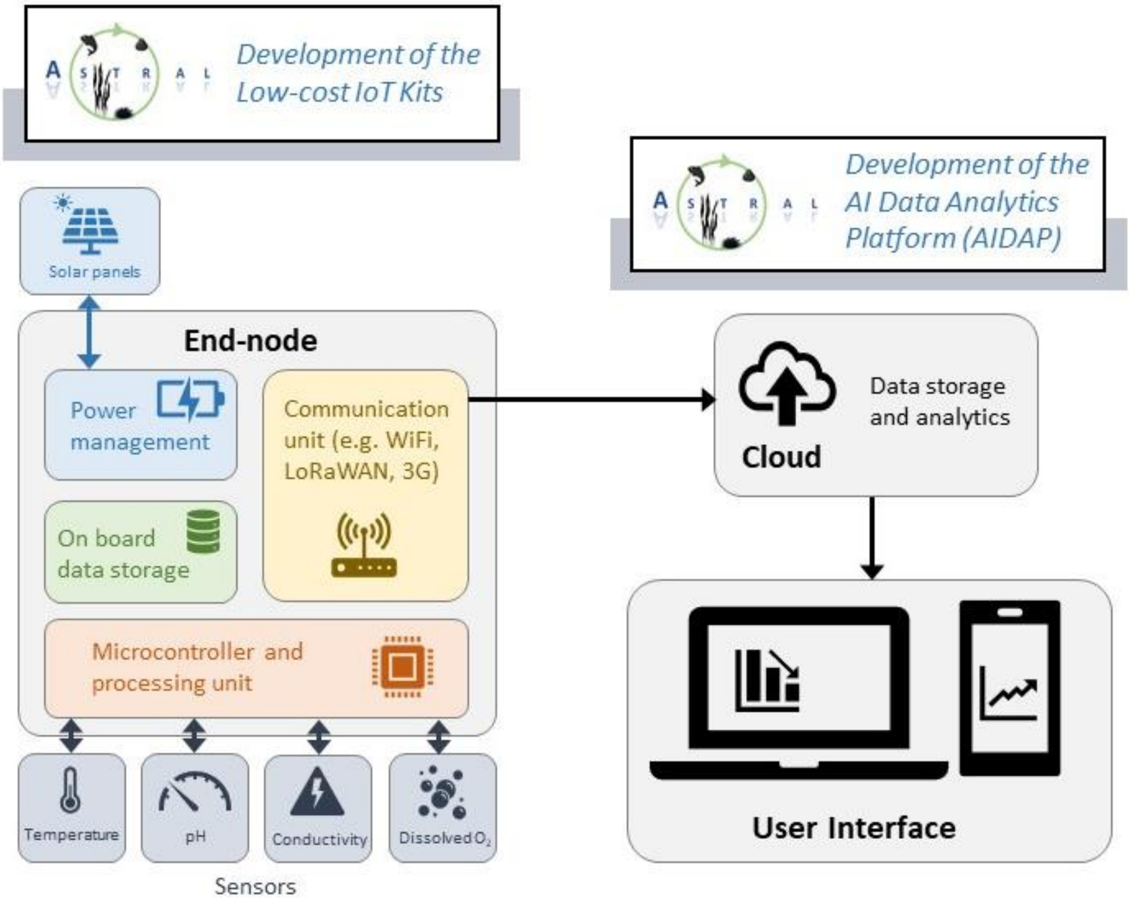

Figure 17. Illustration of an IoT system to support aquaculture water quality monitoring. Areas of relevant ASTRAL technological developments are shown on the figure.

There are a variety of different wireless communication technologies, which have different power consumption requirements, costs, data transfer speeds, and transmission distances: it is important to select a suitable technology for the specific application; some of these include 4G (1000-3000m); Zigbee[86] (10-100m); Bluetooth (20-200m); EnOcean[87] (50-300m); long range wide area network LoRaWAN; Wi-Fi or wireless local area network (20-100m). Faults can occur in the various layers of an IoT aquaculture network, from the acquisitions (sensors), power supply, communications, to data storage or processing; having models to diagnose and address such faults in a timely manner should form part of the ICT infrastructure (e.g. Chen et al 2017[88]).

In support of marine aquaculture the IMPAQT[89] project have developed an Underwater Acoustic Telemetry Platform, which features miniaturized low-power, low-cost acoustic Underwater Transmitter Nodes and a Gateway buoy; the transmitters can be attached to seaweeds or cages, and can be connected to external sensors to transmit their data.

[86] https://csa-iot.org/all-solutions/zigbee/
[87] https://www.enocean.com/en/
[88] Chen, Y., Zhen, Z., Yu, H., & Xu, J. (2017). Application of fault tree analysis and fuzzy neural networks to fault diagnosis in the internet of things (IoT) for aquaculture. *Sensors*, *17*(1), 153.
[89] https://impaqtproject.eu/

ASTRAL will develop a low-cost IoT Kit, equipped with several probes for key water quality parameters, long-range communication protocols, data storage functionality, and optimized self-powering capabilities for remote deployment.

8.2.3 Integrated Monitoring Systems

The concept of an "integrated monitoring system" involves the combination of several discrete monitoring processes into a single computer-controlled system. This allows for centralised digital control of the connected devices and sensors, but also for the potential of remote system control by personnel not physically on the farm. Integrated monitoring systems in combination with IoT enables farms to automate many processes and reduce labour costs, making it less expensive to produce and deliver stock. A further advantage is that operations are inherently traceable, thereby providing transparency in terms of recordkeeping and decision-making.

Various "smart" systems for aquaculture are available commercially, many mainly focusing on water quality. They generally comprise two components – an instrumentation module containing the sensors themselves, and the digital interface for sensor communications and control, which is frequently in the form of a web-based application and/or a smartphone app. Even in relatively simple smart systems, functionality allows for the setting of alarms when threshold measurement values are reached. More sophisticated systems may allow for the devices/sensors to communicate with each other and even act on information received from one another. Commercial aquaculture offerings include YSI Multiparameter or DO monitors, with conditional feed timers, communications, and network control through the YSI AquaManager[90]® software, together with real-time viewing software (AquaViewerII).

Automatically integrating the data from measurements made on site allows for greatly improved ease of data access, paving the way for the benefits of rigorous data analysis: better understanding of the relationship between farmed organisms and their environment and opening the door to predictive capabilities[91, 92]. Augmented by AI or machine learning to optimise data collecting processes and parameterise integrated water quality or animal/plant stress indicators, integrated systems can be quite powerful in terms of alerting farm management to potential issues at an early stage of development – a critical advantage in aquaculture[93]. Part of the H2020 iFishIENCi[94] Project includes the development of a flexible Biology Online Steering System called iBOSS, which will monitor fish behaviour, physiology, and environmental conditions in real-time; it includes an Artificial Intelligence

[90] https://www.ysi.com/aquamanager

[91] Lafont, M., Vallauri, A.,Dupont, S., Dupont, C. Digitization of aquaculture: real-time monitoring and prediction for a sustainable future, 2019, *Aqua Europe* 2019.

[92] Lafont, M., Dupont, S., Cousin, P., Vallauri, A., & Dupont, C. (2019, June). Back to the future: IoT to improve aquaculture: Real-time monitoring and algorithmic prediction of water parameters for aquaculture needs. In 2019 *Global IoT Summit (GIoTS)* (pp. 1-6). IEEE.

[93] O'Donncha, F. and Grant, J., 2019. Precision aquaculture. *IEEE Internet of Things Magazine*, *2*(4), pp.26-30.

[94] http://ifishienci.eu/

system that acts as a digital representation of the fish, which can either inform the farmer or make automatic changes to the environment.

8.2.4 Data Visualisation and Analytics

Users need to be able to access, view, and interact with sensor measurement data in a meaningful manner. Although raw data may be required for research or calibration purposes, displaying measurement data in this format is not user-friendly and can be quite forbidding. Operational aquaculture applications tend to require more intelligent solutions where the measurements are transformed into knowledge and actionable information that are displayed in intuitive ways.

User-interfaces include computer software or mobile applications that fetch data from a local device, server, or the cloud. Most commercial sensors come with proprietary software solutions that only operate with sensors from the same company and the level of sophistication and functionality of these software vary with sensor type, brand, and cost. Interoperable solutions between different sensors or brands are not generally available. The most basic functionality may include sensor control and configuration, potentially with static or interactive graphs of the relevant parameter(s) being measured. Additional functionality might include statistical summaries of information, or the ability to access and query historical data, input notes and ancillary information, and export graphics for reporting purposes.

Most operational farms require software solutions that are scalable from a single sensor to several sensor nodes, able to monitor parameters, organisms, and sensor/system performance in real time. Commercial solutions like INNOVASEA[95] cloud-based solution offers monitoring software with real-time data and updates, powerful analytics and tools accessible from a user's personal computer or mobile devices. Others, like AquaManager[96] aquafarming software, include capabilities for production management, increasing the efficiency, performance, and profitability of a farm. AquaCloud[97] takes a collaborative approach and encourages co-creation of solutions for common challenges.

Smart solutions include predictive models and AI that input data from sensors and even external sources (e.g. web-based weather or remote sensing information) to generate predictions or recommendations that support optimal farm management and decision making; for example the XpertSea Growth Platform for shrimp aquaculture does not only display the information from sensors (e.g. water quality metrics), but includes models for recommended feeding rates, predicted growth rates, and harvest times. Advances in the specialised application of artificial intelligence and machine learning techniques to environmental and biological data can provide valuable insight into the functioning of dynamic environmental systems, as well as the prediction of environmental conditions and the responses of animals and plants.

[95] https://www.innovasea.com/aquaculture-intelligence/cloud-based-software/
[96] https://www.aqua-manager.com/
[97] www.aquacloud.ai

Machine learning is readily employed for data analytics capabilities, and enormously successful in relatively constrained environments such as aquaculture farms. Scalability – monitoring a single tank and extrapolating to an entire farming system – is another promising area of AI application.

AI can also be applied to the internal workings of the data collection/management system itself by assessing system performance and responding appropriately within the system, for example securing data storage and managing interactions with associated instrumentation and operating systems. Other common applications are the sending of alerts intended the reach the user, like traffic-light visuals e.g. green light for all systems running normally, or all parameters within optimal range. Conditional thresholds, alerts and communications can be programmed at any level of system operation, and learning improves as databases grow - drawing in information from external sources e.g. web-based weather or remote sensing information can contextualise the ongoing learning of the platform very effectively. User-focused AI such as chatbots and natural language processing for voice-activated/responsive systems are another application area of AI under rapid development[98].

In a wide-reaching project designed to be upscaled eventually to industry-wide application, DataBio H2020[99] focuses on using "big data" to contribute to the successful production of food, energy and biomaterials. Big Data from three identified sectors - agriculture, forestry and fisheries – are aggregated into a state-of-the-art big data platform which is designed to exist on top of partners' existing infrastructure and data collection and management solutions. The software has the capability to aggregate data from the different sectors, intelligently process them, and analyse and visualise them. It draws on existing datasets, facilitating their optimally effective usage, and independent developers are able to design tools, services and applications based on the DataBio platform, for specific user requirements. The DataBio objectives[100] are met through the emphasis on interoperability between computing systems and platforms, intuitive ease of use and commitment to the sustainability of the industries themselves.

9 Summary and Conclusions

This guide has described the primary water quality and environmental variables important for IMTA aquaculture and the most commonly used sensor types in the industry, as well as new approaches for measuring and monitoring these variables. There is rapid movement in the development of technology appropriate for aquaculture farming in general, and no less in terms of water quality and

[98] Al Rasyid, M.U.H., Sukaridhoto, S., Dzulqornain, M.I. and Rifa'i, A., 2020. Integration of IoT and chatbot for aquaculture with natural language processing. *Telkomnika (Telecommunication Comput. Electron. Control*, *18*(2), pp.640-648.

[99] https://www.databio.eu/en/

[100] https://www.databio.eu/en/about-databio/summary/

environmental monitoring sensors and systems. The implementation of sophisticated systems in terms of sensor inter-communication (IoT) and specialised user interfaces (AIDAP) can facilitate a seamless experience for users with different profiles and requirements. Remote access, automated data analysis and reporting, and intelligent alert/alarm mechanisms play a valuable role in reducing water quality-related and environmental risks, and optimising aquaculture production.

The guide emphasises the need for a thorough understanding and analysis of each farm's individual needs and requirements when selecting and implementing a monitoring system of this sort. As with all technologies, trade-offs exist between cost, practicality, complexity and sophistication. It is critical to be informed about the different available sensors and system design options. With cost effective smart technology targeted at smaller-scale commercial aquaculture farms on all sides of the Atlantic, ASTRAL aims to elevate farmers' access to intelligent technology and smart sensor systems that reduce labour costs and facilitate improved decision-making regarding farm management.

ASTRAL is firmly in support of economically viable and environmentally sustainable aquaculture and the technological solutions enabling it.

Further reading can be found at the following links:

https://www.astral-project.eu/

https://www.fondriest.com/environmental-measurements/parameters/water-quality/

https://freshwater-aquaculture.extension.org/water-quality-in-aquaculture/ (and links to resources therein)

https://ec.europa.eu/programmes/horizon2020/en/what-horizon-2020

H2020 IMPAQT project, IMTA and Precision Aquaculture training course:

https://www.open.edu/openlearncreate/course/view.php?id=7116

[i] *Pertains to instruments that comply with ISO 7027, the European drinking-water protocol. This includes many of the most commonly used submersible turbidimeters.*