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Aquatic Environment and Life Support Systems

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Learning Objectives

Students should demonstrate knowledge of the elements of life support systems and water found within these systems.

1.1 Introduction

For veterinarians to adequately evaluate the health or disease problems of aquatic animals, it is imperative that they understand the impact of, and assess the conditions in which the animal lives. Like terrestrial animals, the health of aquatic species is impacted by their environment. Suboptimal conditions can result in non-infectious diseases or create situations favorable for opportunistic pathogens, which can lead to decreased growth, increased morbidity and clinical disease. It is therefore imperative to include an assessment of water quality and a history of any changes in the environment as part of a clinical evaluation of any health problem.

Fish health management is a term used in aquaculture to describe management practices designed to prevent fish disease. Once fish get sick it can be difficult to salvage them. Successful fish health management begins with prevention of disease rather than treatment, accomplished through, among other considerations, good water quality management. Without this foundation it is impossible to prevent outbreaks of opportunistic diseases. Fish are constantly bathed in potential pathogens, including bacteria, fungi, and parasites. Suboptimal water quality generally associated with stressful conditions allows potential pathogens to cause disease.

The biotic integrity of an ecological system is often reflected by the health of organisms that reside in that system. Proper management of the aquatic environment therefore offers optimum environmental conditions for the growth and better health of the cultivated fish. It also strengthens the defense mechanism of the body to fight against invading disease-producing organisms. Eradicating any predatory or invasive fish species, disinfecting the pond, selecting quality and healthy seed for stocking, maintaining proper species ratios and stocking density, regulating water quality, as well as feeding and handling fish properly, are the various steps of these management measures.

With a very large number of cultured aquatic species, and hence different water systems in which they may live, the task of evaluating optimal conditions for every species may seem daunting. However, if considered on an ecosystem level for those that thrive naturally in freshwater, brackish or marine aquatic ecosystems, and evaluating these as life-support systems for different taxa, the task is simplified. Furthermore, many biotic and abiotic physicochemical features of fresh, brackish or marine water bodies are similar, even for very different taxa.

This chapter provides general information about basic features of aquatic environments and life-support systems important to the health of finfish, amphibians, mollusks, crustaceans and invertebrates that veterinarians might encounter. As aquatic mammals, birds and other homeothermic species are generally covered in veterinary curricula, they are not addressed here.

1.2 The Life-Support System in Aquaculture and Ornamental Fish Care

Aquaculture is the farming of aquatic organisms including fish, mollusks, crustaceans and aquatic plants, with some sort of intervention in the rearing process to enhance production, such as regular stocking, feeding, and protection from predators. Farming also implies individual or corporate ownership of the stock being cultivated. Aquatic ecosystems provide the basis for aquatic production. The worldwide practice of aquaculture runs the gamut from low-technology extensive methods to highly intensive systems. Although used for housing and maintaining ornamental fish, similar aquasystems are found in large public aquaria and private homes and are discussed in this chapter. The term 'life-support system' is understood as a complete system that satisfies the physiological needs of an aquatic organism in question. Diagrams and photographs of examples of life-support systems can be found in Chapter 3.

Managed ecosystems form the basis for aquaculture production, which has been widely offered as a means of complementing and ultimately supplementing traditional fisheries. Here, unlike fisheries, inputs, production processes and quality of output can be at least partially controlled, and ownership, care and environmental responsibility might be more easily established. By removing natural constraints to survival and productivity, and by husbandry and management, production need be limited only by availability of simple inputs such as land, water, seed (hatchling, spawn, fry or fingerling), fertilizers and feeds. The design of a life-support system is dependent on the type of aquatic system in question, being either a flow-through system or a closed system. In a flow-through system, the main elements include a spigot or waterfall that is adjustable for the amount of water flow necessary, a drain that is usually positioned at the opposite end of the system, potential for increased aeration, and, possibly, temperature controls.

A closed system may be a pond, tank or aquarium. In a pond, the most important elements are circulation and aeration, and it may also include a filtration system. Tanks and aquaria usually require a filtration system, aeration, and often temperature control. The source of the water is important when deciding on the type of aquatic system and the type of life-support system. A large well, river or lake is necessary for a flow-through system because of the high volume of water that is needed to maintain proper water quality. If city water is being used in a system, it is important to have a reservoir to dechlorinate any new water entering the system. In recirculating systems, water circulation and purification is accomplished with the use of powered pumps and filtration systems. The type of equipment used will depend on the type of fish, number of fish and the expected growth rate of those fish.

In aquaculture, high-intensity growth of fish leads to higher ammonia levels and waste production. The output of the pump should allow for a flow of the full volume of water in the system to

pass through the filter every two to three hours. This is referred to as the “turnover time”. It is calculated by dividing the water volume in the system by the volume per time rating of the circulation, measured at the return (the volume/time is variously described in gallons/minute or hour as GPM, GPH or metric m^3/hour). In tanks and ponds, flow rates are very variable, from less than five minutes in the smallest aquaria to more than 10 hours in larger ponds. Submersible pumps with low maintenance sealed impellers may be used in small ponds and aquaria, but larger aquasystems typically use external pumps. The filtration system should contain biological media with an appropriate surface area to allow for detoxification of ammonia based on the number of fish. On average, 1 m^2 of active surface area can process 1 gram of ammonia every 24 hours (Wildgoose, 2001).

Monitoring of the system may be fully automated or may be performed by individuals. The benefits of automated monitoring equipment include the ability to set alarms for power outages, changes in dissolved oxygen and large changes in ammonia. Some systems are able to calculate flow rates and water usage, which is beneficial for production calculations. Monitoring should include ammonia, nitrite, nitrate, dissolved oxygen, temperature, total alkalinity and pH to assure proper water quality parameters.

1.2.1 Extensive Culture System

The extensive culture system is characterized by low to no inputs (food, fertilizer, etc.) and low stocking densities. Extensive aquaculture is practiced in lakes, reservoirs, lagoons, ponds and tanks.

1.2.1.1 Ponds

The most common production system in use is the earthen pond. Earthen ponds are extremely popular among fish growers due in part to ease of construction, low maintenance, relatively small area requirements and ability to grow a wide variety of species. Additionally, because earthen ponds mimic nature, they may produce fish of an overall healthier appearance than other techniques. A natural supply of food is often available in earthen ponds, which may lead to better fish health. Pond culture can vary, from all life stages naturally occurring in a single pond to elaborate systems with discrete ponds for holding broodstock, spawning, rearing, growing and catch-out or harvest.

1.2.1.2 Tanks

Tanks essentially act as ponds but are generally constructed of concrete or fiberglass. Wood can also be used but must first be treated to prevent rotting. Concrete tanks have the advantage of being less expensive, easily constructed and formed into various shapes.

- Plastic, fiberglass or glass tanks are ordinary tanks which are either designed and constructed for rearing fish or used for storing water. They can be moved from one place to another.
- Wooden troughs are constructed with planks. They vary in size and depth. After construction, the tank is lined with nylon to prevent leakage. It can be moved from one place to other.

1.2.2 Semi-Intensive Culture System

The semi-intensive culture system is distinguished by increased stocking rates and the requirement for some level of input, such as food, fertilizer, chemicals, etc.

- Cage culture uses existing water resources (i.e. lakes or ponds) but encloses the fish in a cage or basket, which allows water to pass freely between the fish and the pond or lake. One of the main advantages is the ease of harvesting.

- Fish pens are enclosures installed in open waters that are not too deep for raising fish. The fish have access to the water bottom, unlike in cages.
- Rafts/trays or long lines are mostly used for oyster culture.
- Some improvised containers such as bathroom tubs, big plastic containers, abandoned plank canoe, earthen pots are used to culture fish where the farmer cannot afford the conventional rearing facilities. Some fish farmers in Lagos use these containers for breeding. Apart from breeding purposes, this type of rearing facility is not suitable for commercial fish farming.

1.2.3 Intensive Culture System

Intensive culture system is where animals are maintained in systems such as ponds, tanks and raceways, where the support parameters are carefully controlled and dependence on the natural environment is minimal. Such systems require a high degree of management and usually involve substantial investment and operating costs, resulting in high yields per unit area. They typically feature recirculating systems, which filter and reuse all or a portion of their water. Currently, this technique is in limited use due to its high start-up and maintenance costs. It has several advantages, however, in that this system is highly desirable in areas where a constant water supply may be questionable or a discharge is not appropriate. Other advantages of this type of system include the ability to incorporate the growth of a second product through hydroponics and the use of settled material as fertilizer. As the technology evolves and water withdrawal and discharge requirements become more stringent, this technique is bound to see an increase in use. Ponds are also very popular for housing ornamental fish, such as koi or goldfish. Typically lined with EPDM rubber, these can be fairly simple in having only a recirculating system and waterfall or extremely complex, with sophisticated filtration and water conditioning systems.

1.3 Physical Features of Life-Support Systems

Multiple designs from simple box filters to multistage proprietary units can be found in use for home aquaria. For freshwater systems, these typically include a mechanical filter to remove particulate and a biofilter placed afterwards in the return water flow. Often an ultraviolet (UV) light unit is installed to control planktonic algae. In saltwater systems, a protein skimmer is also used to remove dissolved organic matter. Adequate gas exchange is generally assured by the simple water turnover provided by the filter system, although additional aeration is often provided with the use of air pumps and air stones. Ponds will usually have the same components as freshwater aquaria, scaled up to meet the needs of larger fish. The most modern designs will typically feature a prefilter, mechanical filter, biofilter and UV light. Water is returned to the pond via a spillway or waterfall, which assures adequate gas exchange. Designs for these components vary, with some rudimentary systems having only a biofilter and waterfall. As this system relies on in-pond sedimentation, regular depopulation and cleanout is needed. Public aquaria differ only by size and scale from home aquarium systems. However, some large-scale water purification equipment is described below. Some examples of aquaculture open and closed husbandry systems (raceways, recirculating, ponds, net pens) are given in Chapter 3. Different systems for different life stages are described in Chapter 10.

1.3.1 Mechanical, Chemical and Other Types of Filtration

Recirculating systems can vary in complexity and design. Depending on the water source, varying types of water purification may be required or recommended to prevent pathogens at the point of

entry as well as within the system itself. Mechanical filters are often used to improve water quality and reduce pathogen loads by removing particulate matter in the system. Recirculating systems that are considered self-cleaning remove waste solids and provide some water purification prior to system reentry. Mechanical filter design based on water flow direction can be described as “down-flow”, where the water to be processed passes by gravity or under pressure down through the media or “upflow”, where it is forced under pressure up through the media. The advantage of this upflow design is to allow sediment to fall away from the media, making it somewhat self-cleaning (Tepper, 2000). Media used may be sand, polypropylene or fiberglass mesh or polypropylene brushes in downflow systems and polypropylene beads in upflow systems.

Prefilters can be used to separate settleable solids by rotating the water in an upflow direction. This strips off the heavier particulate, enabling more efficient water processing before entering the mechanical filter.

Industrial aquaculture facilities may use self-cleaning tanks in recirculating systems to remove waste solids and provide some water purification using large rotating drum filters. Also, water sources with high total solids prior to system entry may also require large screen filtration to remove solids. Other large facilities may employ pressurized canisters of upflow design for this purpose (floating bead filters). These canisters are also often seen in home pond filtration, although downflow box filters with filter mesh for particulate removal are also used, where they are the most common type of filter for small aquasystems like freshwater and marine home aquaria. Sand canister filters are sometimes used due to their high efficiency at removing solids and low cost. They have the disadvantage of being hard to maintain over time due to “caking” of the sand media.

1.3.1.1 Ultrasonic Cavitation

Life stages of larger organisms such as the salmon louse may be effectively removed from discharge water with ultrasonic cavitation (Svendson *et al.*, 2017). Anecdotally, some hatcheries have seen reduction in levels of other parasites with the installation of some types of low-head oxygenators, perhaps through a similar mechanism such as microbubbles.

1.3.1.2 Ultraviolet Filtration

Pathogens can be removed effectively from water by UV filtration. These filters cause DNA damage to microorganisms by radiating with light in the UV-C spectrum. The dose required to inactivate an organism varies, with some organisms proving more difficult and thus requiring a higher dose than others. Published values for specific pathogens can be found and may vary with the system used, the source of water, species being reared, and the potential pathogens of concern. These doses are calculated under ideal laboratory conditions and, in reality, the effective dose required may be much higher in a system with poor water quality. UV filters consist of a light source (bulbs) contained within a vessel that permits water to be passed through at varying rates. The amount of time the water is in contact with the light and the wattage of the bulb determines the dose. These systems are subject to impacts from turbidity of the water and require regular maintenance to remain effective. The glass must be cleaned of debris and silt and most manufacturers recommend bulb replacement at a minimum of once a year. Water with a high organic load will reflect the UV radiation and cause a decline in the effective dose. Systems with high total solid loads or otherwise high in organics may require prefiltration to assure reliable UV effectiveness. Flows must be monitored and not increased to exceed the capabilities of the UV filter.

Other factors that may impact the dose requirements for pathogen inactivation in a system are the bacterial repair mechanisms – photoreactivation and dark repair – that may allow bacteria to resist being killed by repairing the UV damage. Preventing exposure of the water to

natural light will help to inhibit photoreactivation of bacteria in the system, as will increasing the UV dose in the system beyond the standard published doses. Preventing reduction of lamp intensity, which can occur with poor water quality or increased flow, will also help to reduce photoreactivation in the system. In cold-water aquaculture with short water retention times, photoreactivation is more likely to become a problem than dark repair. Likewise, in recirculating systems that have long water retention times (many cycles of the same water or a high reuse percentage with very few additions of new water) dark repair may be more of a concern. UV filtration at lower doses is often employed in aquasystems for planktonic algae control (using a UV light unit).

1.3.1.3 Ozone Filtration

Another type of water treatment that can be used is ozone. This effective oxidizer is produced in an onsite system and requires a high voltage generation unit. It is usually reserved for larger industrial aquasystems and public aquaria. A large source of purified oxygen is required to produce the ozone, which can be linked to a low head oxygenator or other oxygen feed systems that may already be in use to increase production capacity of a facility. Ozone oxidizes cell membranes, making it effective at killing bacteria, but fish and humans should not be directly exposed to it. As with UV, contact time in the water is important, with considerations for an initial killing dose followed by a residual dose to meet the required contact time. The residual dose can be reached inside a residual holding tank, allowing for longer contact time. The ozone must be transferred to the water by some method of injection system and inactivated before reaching the fish holding tanks, as fish are highly sensitive to residual ozone. When used in seawater systems, additional byproducts may need to be managed.

1.3.1.4 Biological Filtration

The conversion of ammonia to nitrite and nitrate is critically important in aquasystems where new water exchange is minimal. These systems must be treated with care to ensure that adequate levels of beneficial bacteria are maintained to perform this conversion. In cases of high recirculating water reuse systems that recirculate water in the 90% range, a dedicated biofilter is used before the water reenters the main system. In the biofilter, the conversion is accomplished through the use of specialized media designed to maximize bacterial performance.

Features of a biofilter that impact its capacity to convert ammonia through to nitrate in any aquasystem are the type of media used and the surface area per unit volume for bacterial growth, as well as the water flow pattern, degree of oxygen exchange and adequacy of sediment removal. Most biofilters use plastic composite material, although older systems may employ lava rock. Sand can also be used in upflow-designed tanks, called fluidized bed bioreactors, after the water has been cleared of settleable solids. Many types of biological filters that use varying technologies are already in use in recirculating aquaculture. This field is rapidly growing and expanding. With increasing popularity and utility of recirculating systems, the capabilities of these units continue to improve and it is important that a system is designed to allow for upgrades and improvements. While biofiltration conserves water and increases production capacities, these filters require substantial floor space and add a high cost to initial system installation. In selecting this type of system, the long-term benefits such as water usage savings, increases in production capacity, and decreased effluent discharge should be weighed against the initial cost. The details of biofilters are complex. Additional information can be found in specialized texts such as *Recirculating Aquaculture* by Timmons *et al.* (2018). Further study is recommended prior to working with these types of complex systems.

1.3.2 Aquasystem Water

1.3.2.1 Abiotic (Chemical and Physical) Properties

Oceans, rivers, lakes, and even ponds, are various types of aquatic ecosystems. The abiotic components of aquatic ecosystems are the physical and/or chemical factors characteristic of the environment that act on the living organisms during any part of their life. These abiotic factors are also called ecological factors. They vary from ecosystem to ecosystem; for example, the abiotic factors in the marine biome differ from those of fresh water in terms of chemistry, light, current and temperature.

Some of the chemicophysical parameters of water have a direct influence on fish health. Any abrupt and wider than normal fluctuations of such values cause a state of stress in fish, sometimes resulting in widespread disease outbreaks. Dissolved oxygen content, pH, turbidity, temperature, introduction of some chemicals, detergents, pesticides and naturally produced toxic products like hydrogen sulfide, ammonia, dinoflagellate toxins are potential stress-related parameters.

1.3.2.2 Temperature

Fish are ectotherms, which means that they do not regulate their body temperature internally so it fluctuates with external environmental changes. Sudden change often causes stress and sometimes even death in extreme situations. Temperature affects the development and growth of fish more than any other single factor and also influences other variables. Metabolic rate either increases or decreases two-fold for every 8°C. Each species has an optimal temperature range for growth and reproduction. Cultured species should be selected according to the prevailing water temperature in each locality. This is because the temperature range at which an organism can survive depends on their temperature adaptation pattern over time.

Water temperature greatly influences water quality, because the growth and metabolism of phytoplankton, bacteria and other microorganisms increases with increasing temperature. Moreover, water holds less oxygen at higher temperatures. Dissolved oxygen depletion is much more likely to occur during hot weather than during cooler periods. Rates of chemical processes such as ionization, mineral dissolution, adsorption and ion exchange also increase in response to higher water temperature. The onset of potentially harmful water quality events is faster in warmer weather. A deep pond can also be subject to thermal stratification, with large temperature fluctuations among layers of the water column.

- Warm-water species grow best above 21°C
- Cold-water species grow best below 21°C
- Cool-water species grow best at mid-range from 21°C to 28°C
- Sudden temperature changes cause stress and even death.

1.3.2.3 pH

The term “pH” is a mathematical transformation of the hydrogen ion (H^+) concentration; it conveniently expresses the acidity or basicity of water. The lowercase letter “p” refers to “power” or exponent, and pH is defined as the negative logarithm of the hydrogen ion concentration. Each change of one pH unit represents a 10-fold change in hydrogen ion concentration. The pH scale is usually represented as ranging from 0 to 14, where lower number reflects higher acidity and the higher number reflects higher alkalinity. Water with a pH of 4.5 or lower has no measurable alkalinity. Water with a pH of 8.3 or higher has no measurable acidity. At 25°C, a pH value of 7 is neutral; this describes the neutral point of water at which the concentrations of hydrogen and hydroxyl ions (OH^-) are equal (each at 10^{-7} moles/l). The pH of freshwater ecosystems can fluctuate

considerably within daily and seasonal timeframes, and most freshwater animals have evolved to tolerate a relatively wide environmental pH range. Animals can, however, become stressed or die when exposed to pH extremes or when pH changes rapidly, even if the change occurs within a pH range that is normally tolerated. The pH of water in ponds often increases during the day and decreases at night. Fish and other vertebrates have an average blood pH of 7.4. Recommended pH range for cultured fish is 6.5–9.0. Acid death point is around 4; alkaline death point is around 11. The hydrogen ion concentration affects aqueous equilibria involving ammonia, hydrogen sulfide, chlorine and dissolved metals. Interactions of pH with these variables are often more important than the direct effects of pH on aquatic animals. For example, the toxicity of ammonia to fish increases with an increase in pH. Chemical interactions among carbon dioxide, hydrogen ions and the anions that produce alkalinity buffer the pH of most natural waters in a range of about 6–8.5.

1.3.2.4 Dissolved Oxygen

Dissolved oxygen monitoring is critical in aquaculture. Temperature, salinity and elevation affect dissolved oxygen. As these three factors increase, dissolved oxygen at saturation decreases. For example, the cold temperature of the Antarctic results in higher dissolved oxygen concentrations compared with warmer tropical waters. Freshwater at sea level holds 9.2 ppm at 20°C and 7.6 ppm at 30°C. Fish become more active and increase their metabolic oxygen needs as temperature increases. As temperature rises, fish also need more dissolved oxygen to grow muscle tissue. Minimum tolerable dissolved oxygen levels increase with a rise in temperature.

In general, most fish species will grow and thrive within a dissolved oxygen range of 5–12 mg/l (parts per million). However, if levels drop below 4 mg/l they may stop feeding, become stressed and begin to die. Dissolved oxygen ranges for cultured fish are as follows:

- 0–2 ppm – small fish may survive a short exposure, but lethal if exposure is prolonged. This range is lethal to larger fish.
- 2–5 ppm – most fish survive, but growth is slower if prolonged; may be stressful; aeration devices are often used below 3 ppm.
- 5 ppm to saturation – the desirable range for all.
- With rainbow trout, the minimum lethal limit is 1.6 ppm at lower temperatures and 2.5 ppm at higher temperatures.

Oxygen and pH are best measured in situ (with probes) or as soon as possible after collection (preferably before leaving the site) as, in most situations, levels will change during storage and transport.

Biological oxygen demand is a measure of the oxygen used by all organisms in an aquasystem.

1.3.2.5 Carbon Dioxide

Carbon dioxide (CO₂) is consumed during photosynthesis by plants and expired during respiration by animals, plants (at night) and bacteria in an aquasystem. When added to pond water by respiration or diffusion, it forms a weak acid (carbonic acid), which lowers the pH. Dissolved oxygen and pH cycles follow the same daily peaks and troughs. Carbon dioxide levels of below 10 mg/l are thought to be well tolerated by fish. While levels greater than 20 ppm often harm fish, especially if dissolved oxygen levels are low, sensitivity to CO₂ varies between species. The level of CO₂ in source water varies greatly, and is further affected by the respiratory and photosynthetic activity of animals and plants, and the level of decomposition of organic material in that water (a very significant contributor to CO₂ levels in some nutrient-rich waters). CO₂ can build up to significantly high levels in systems with large numbers of animals and relatively slow water turnover.

The effect of increased CO_2 in water is to reduce the rate at which CO_2 from the animal's own metabolism can be released from the blood through the gills. As a result, CO_2 in the blood also increases (known as hypercapnia), resulting in a drop in the blood pH and acidosis. At the same time, the oxygen-carrying ability of the hemoglobin in the blood is reduced. The animal can counteract the effect by balancing the acidosis with an exchange of ions, such as increasing the uptake of bicarbonate and losing hydrogen and phosphate ions and little harm is done. In the long term, this balancing act can have a more profound effect on the health of the animal. Carbon dioxide causes problems in recirculating aquaculture systems without aeration or degassing.

1.3.2.6 Nitrogen

The nitrogen (N_2) biogeochemistry of aquaculture ponds is dominated by biological transformations of nitrogen added to ponds in the form of inorganic or organic fertilizers and formulated feeds. Nitrogen application in excess of pond assimilatory capacity can lead to the deterioration of water quality through the accumulation of nitrogenous compounds (e.g., ammonia and nitrite) toxic to the fauna. Principal sources of nitrogen include animal excretion and sediment flux derived from the mineralization of organic matter and molecular diffusion from reduced sediment, although cyanobacterial nitrogen fixation and atmospheric deposition are occasionally important.

1.3.2.7 Hydrogen Sulfide

Hydrogen Sulfide (H_2S) is a poisonous gas with a “rotten egg” smell, produced by anaerobic decomposition of organics. Sulfur is an essential element for plants, animals and bacteria. H_2S is present in natural waters and in aquaculture systems, mainly as the sulfate ion. Sulfide can occur in water because it is a metabolite of *Desulfovibrio* species and certain other bacteria found in anaerobic zones, usually in sediment. These bacteria use oxygen from sulfate as an alternative to molecular oxygen in respiration. There are three forms of sulfide (H_2S , HS^- and S_2^-), and they exist in a pH and temperature-dependent equilibrium. As pH increases, the proportion of H_2S declines, and that of HS^- rises until the two forms have roughly equal proportions at pH 7. At greater pH, HS^- is the dominant form, and there is no S_2^- until the pH is above 11. Hydrogen sulfide is toxic to aquatic animals because it interferes with reoxidation of cytochrome a3 in respiration. This effect is caused almost entirely by H_2S , while HS^- is essentially non-toxic. Even if it is toxic, S_2^- is not an issue, because it does not occur at pH values found in aquaculture systems.

1.3.2.8 Chlorine

Chlorine is harmful/toxic to fish at values greater than 0.03 ppm. Tap water may range from 4.0 to 8.0 ppm. Sodium thiosulfate can be used to neutralize the chlorine. Chlorine may be used to disinfect equipment, tanks, countertops, and nets at 10 ppm for 24 hours or 200 ppm for 30–60 minutes. Effectiveness is reduced by organic material such as mud, slime and plant material. Sodium hypochlorite is available at concentrations of 15%, 50%, or 65% active.

1.3.2.9 Alkalinity

Alkalinity is the ability of the water to accept hydrogen ions and neutralize them and offers a buffering system to reduce pH swings. It is measured by the amount of acid (hydrogen ion) that water can absorb (buffer) before achieving a designated pH. Total alkalinity is expressed as milligrams per liter or parts per million calcium carbonate (mg/l or ppm CaCO_3). It consists of negatively charged bases – carbonates, bicarbonates, and hydroxides. Alkalinity is expressed in equivalent concentrations of calcium carbonate.

Carbonates and bicarbonates are sources of carbon for plants, which is used in photosynthesis to make carbohydrates. Carbonates and bicarbonates are the most common and most important components of alkalinity. A desirable range of total alkalinity for fish culture is CaCO_3 75–200 mg/l. Carbonate–bicarbonate alkalinity (and hardness) in surface and well waters is produced primarily through the interactions of CO_2 , water and limestone. Rainwater is acidic because of exposure to atmospheric carbon dioxide and sulfuric acid (H_2SO_4).

1.3.2.10 Hardness

Hardness refers to the concentration of divalent cations (calcium, magnesium, and sodium). Water hardness is important to fish culture and is a commonly reported aspect of water quality. It is also expressed as the calcium carbonate equivalent concentration. The same carbonate rocks responsible for most of the alkalinity are the main sources of calcium and magnesium cations for hardness. Hardness may be an index of potential pond productivity. The minimum hardness for fish is 20 ppm and optimum hardness is around 100 ppm.

Hardness is traditionally measured by chemical titration. The hardness of a water sample is reported in milligrams per liter as calcium carbonate (mg/l CaCO_3). Calcium carbonate hardness is a general term which indicates the total quantity of divalent salts present and does not specifically identify whether calcium, magnesium and/or some other divalent salt is causing water hardness. If limestone is responsible for both hardness and alkalinity, the concentrations will be similar, if not identical. However, where sodium bicarbonate (NaHCO_3) is responsible for alkalinity, it is possible to have low hardness and high alkalinity.

1.3.2.11 Conductivity, Salinity, Total Dissolved Solids

Conductivity is a measure of water's capability to pass an electrical current. This ability is directly related to the concentration of ions in the water (US Environmental Protection Agency, 2016). These conductive ions come from dissolved salts and inorganic materials such as alkalis, chlorides, sulfides and carbonate compounds (Miller *et al.*, 1988). Compounds that dissolve into ions are also known as electrolytes (Sommer and Spitzer, 2004). The more ions that are present, the higher the conductivity of water. Likewise, the fewer ions that are in the water, the less conductive it is. A sudden increase in pond conductivity can indicate pollution. Agricultural runoff or a sewage leak will increase conductivity due to the additional chloride, phosphate, and nitrate ions.

Salinity is a measure of the total concentration of dissolved solids, usually in parts per thousand (ppt). Anions (negatively charged) are chloride, sulfate, bicarbonate and bromide. Cations (positively charged) are sodium, magnesium, calcium, potassium, and strontium. Sodium and chloride are the major solids. Because the electrolytes form ionic particles as they dissolve, each with a positive and negative charge, salinity is a strong contributor to conductivity. The concentration of water is as follows:

- Freshwater: less than 2 ppt
- Brackish water: 2–16 ppt
- Saltwater: 35 ppt.

Most aquatic organisms can only tolerate a specific salinity range (Clean Water Team, 2002). The physiological adaptation of each species is determined by the salinity of its surrounding environment. Salinity tolerances depend on the osmotic processes within an organism. Fish and other aquatic life that live in fresh water (low salinity) are hyperosmotic (cells have a high ability to eliminate water and retain ions). On the other side of the spectrum, saltwater (high salinity) organisms are hypo-osmotic and maintain a lower internal ionic concentration than seawater. Most

species of fish are stenohaline, exclusively freshwater, or exclusively saltwater (Myers, 1949). However, there are a few organisms that can adapt to a range of salinities. These euryhaline organisms can be anadromous, catadromous or true euryhaline. Anadromous organisms live in saltwater but spawn in freshwater. Catadromous species are the opposite – they live in freshwater and migrate to saltwater to spawn (Myers, 1949). True euryhaline species can be found in saltwater or freshwater at any point in their life cycle (Myers, 1949). Estuarine organisms are true euryhaline.

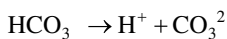
The sum of all ion particles that are smaller than 2 microns (0.0002 cm) is the total dissolved solids. This includes all the disassociated electrolytes that make up salinity concentrations, as well as other compounds such as dissolved organic matter. Freshwater aquasystems should have less than 2000 mg/l of total dissolved solids and most water sources should have much less than that (American Public Health Association *et al.*, 2017).

1.3.2.12 Light, Color, Vibration and Noise

Aquasystem water color influences microplankton growth, which can affect the stability of the water chemistry. Water clarity can affect fish. If fish that prefer turbid waters (e.g., bullhead, catfish, walleye) are cultured in relatively clear water they will experience stress; survival and growth will be adversely affected. Intensive aquaculture systems, particularly recirculating systems, utilize equipment such as aerators, air and water pumps, blowers, and filtration systems that inadvertently increase vibration and noise levels in fish culture tanks (see also vibration and noise in Chapter 10). Field and laboratory studies have shown that fish behavior and physiology can be negatively impacted by intense sound. Chronic exposure to aquaculture production noise could therefore cause increased stress, reduced growth rates, and cold feed conversion efficiency, and decreased survival. In the wild, geological and geophysical exploration, pile driving, drilling, dredging, and vessel traffic all produce manmade noise and vibrations, which may have negative effects on native fauna, which may range from physiological and behavioral effects to physical damage.

1.3.2.13 Greenhouse Gases and Climate Change

Elevation of carbon dioxide in the atmosphere leads to acidification of our water. This change affects all aquatic life. Millions of dollars are lost each year on decreased growth and increased mortality due to large fluctuations in the pH and temperatures of the water. Elevation in the CO₂ levels of the atmosphere push the chemical reaction:



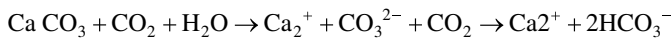
to move more toward a disassociation of water molecules and increase the HCO₃⁻. In the ocean, this fluctuation is well buffered by salt water, but in coastal areas and rivers the addition of runoff increases the nitrogen, carbon and Ca₂⁺, creating large fluctuations in the pH. Climate change has resulted in an increase of the ocean temperatures, changes in mixing and increased intensity of storms globally. These changes, together with large fluctuations in pH, result in large hypoxic zones and the inability of fish to acclimate to them.

Fish in estuaries and coastal areas are affected the most by acidification due to the increased fluctuation of the pH. This fluctuation is exaggerated by nutrient runoff, coastal upwelling, and atmospheric pollution. Fish have been shown to be able to regulate their pH within days, but they rely on the ability to move the CO₂ into the lower partial pressure of carbon dioxide (pCO₂) of the aquatic environment. As atmospheric CO₂ increases, fish can compensate metabolically and regulate their pH but still show elevated levels of the CO₂ in their extracellular fluids. Studies have shown that this elevated level of CO₂ can cause problems with osmoregulation. In addition, studies

have shown this increased level of CO₂ can result in increased otolith size in clownfish, Atlantic cod and pollock (Heuer and Grosell, 2014).

The increased CO₂ in the plasma has been shown to affect the metabolic rate and the formation of CaCO₃⁺. This occurs when HCO₃⁻ is secreted into the intestinal lumen. This secretion stimulates the secretion of CO₂. This combination precipitates the Ca²⁺ from the seawater that has been ingested. These then combine to form CaCO₃⁺. The higher PCO₂ in the plasma has been proven to increase these secretions in midshipmen and toadfish. CaCO₃⁺ is important for osmoregulation in fish by influencing the osmotic pressure in the luminal cavity. Changes in the amount of CaCO₃⁺ has the potential to decrease the osmotic balance (Heuer and Grosell, 2014).

Elevations in atmospheric CO₂ have been shown to decrease calcification rates in the carbonate equation:



Decreases in calcification rates affect coral reefs, planktonic microalgae, coccolithophores, phytoplankton, and all shellfish. Coral reef growth has been shown to decline due to the decrease of calcification, with a predicted decrease of 21–40% over the period of 1880–2065 (Turley *et al.*, 2006). This decline is also dependent on the temperature of the water and other available nutrients. Oysters and abalone have been the focus of many studies in regards to acidification. These organisms have been shown to have a decreased rate of survival at the larval stage due to lack of calcification of the shell and decreased ability to ingest nutrients. The shell of mollusks is produced throughout all its life stages. During the larval stages, the shell is made from deposition of amorphous calcium carbonate, which then forms into a crystalline aragonite. Both these stages of shell development are more soluble in a low pH. Studies have shown that certain levels of pH will cause holes to form in the shells (Wessel *et al.*, 2018).

Acidification of our oceans has a great effect on the ecosystem of the ocean. It is leading to changes in the phytoplankton that are available for ingestion by fish and cultivation. It is predicted to decrease the available fish population of our reefs, which will damage the animal protein consumption of many developing nations. The high fluctuations of pH in the estuaries and coastal areas have the potential to greatly affect fisheries. Mitigation of these fluctuations should include consistent monitoring of the water to enable fisheries to add buffering if necessary (Clements and Chopin, 2017). Options for buffering may include:

- Crushed shell hash in the sediment
- Sodium bicarbonate
- Movement of fish pens away from river mouths.

Further mitigation strategies should include breeding plants and animals that have a high tolerance for low pH. Integration of a multitrophic aquaculture system can help to mitigate pH fluctuations and other stress caused by run-off and increased population. An integrated system should include three levels of nutrient absorption (Clements and Chopin, 2017):

- Shell fish: small organic particle suspension extractive.
- Seaweed: dissolved nutrients suspension extractive.
- Invertebrates: large organic particle deposit extractive.

While acidification of natural aquasystems can only be mitigated by global reduction of atmospheric carbon dioxide, further research is necessary to fully understand the impacts of global climate change and the acidification of our oceans.

1.3.2.14 Ammonia, Nitrite and Nitrate

Ammonia is a colorless, odorless substance which can accumulate in aquasystems and cause direct mortality, a decrease in production and increased incidence of many diseases. In water, ammonia occurs in two forms, which together are called the total ammonia nitrogen. Ammonia gas is usually released from the gills or from decomposing organics dissolved in the water. Some reacts with the water to produce ammonia ions. The remainder is present as un-ionized, which is acutely toxic to aquatic life. Chemically, these two forms are represented as NH_4^+ and NH_3 . NH_4^+ is called ionized ammonia because it has a positive electrical charge, and NH_3 is called un-ionized ammonia (UIA), since it has no charge. This is important to know, since NH_3 , UIA is the form that is toxic to fish. The percentage of UIA in solution depends on the pH and temperature of the water; as both go higher, so does the toxicity.

A typical aquasystem has bacteria, which in the presence of dissolved oxygen converts (oxidizes) ammonia to the intermediate form of nitrite and then to nitrate. Ammonia is converted into nitrite (NO_2^-) due to the influence of *Nitrosomonas* bacteria. Nitrite (NO_2^-) is converted into nitrate (NO_3^-) due to the influence of *Nitrobacter* bacteria. Most test kits measure total ammonia nitrogen, so the aquarist must determine what percentage of the total is toxic.

Nitrite is an intermediate in the oxidation of ammonium to nitrate. Nitrite is less toxic to fish than ammonia. An elevated ambient nitrite concentration is a potential problem for freshwater fish since nitrite is actively taken up across the gills in competition with chloride. Nitrite is a well-known toxicant for fish, as well as a disrupter of multiple physiological functions including ion regulatory, respiratory, cardiovascular, endocrine and excretory processes. One critical consequence of nitrite accumulation is the oxidation of hemoglobin to methemoglobin, compromising blood oxygen transport (Kroupova *et al.*, 2005). Nitrite toxicity to fish varies considerably and depends on many external and internal factors. Among the most important ones are water quality (e.g. pH, temperature, cation, anion and oxygen concentration), length of exposure, fish species, fish size and age, and individual fish susceptibility.

Nitrate is relatively non-toxic; however, high concentrations reduce animal growth and can decrease survival. For example, concentrations over about 400 mg $\text{NO}_3^- \text{N/l}$ have been shown to depress the growth rate and survival of marine shrimp (*Litopenaeus vannamei*). In intensive bio-floc aquaculture systems used for shrimp culture, concentrations of 400 mg $\text{NO}_3^- \text{N/l}$ have been observed after using water for roughly three culture cycles. Salinity influences the toxicity of nitrate, as it does with ammonia and nitrite. At higher salinities, these compounds are less toxic to aquatic animals. Several options exist for dealing with nitrate, including water exchange, phytoremediation, denitrification, and heterotrophic assimilation (Svobodova and Kolarova, 2004; Svobodova *et al.*, 2005; Tepper, 2000).

It is recommended that un-ionized ammonia should be less than 0.02 ppm to prevent stress and reduced growth. Ammonia has been reported to be lethal to catfish at about 0.4 ppm (Jensen, 2003). Ammonia, nitrite and nitrate are labile ions and must be analyzed rapidly or samples frozen as soon as possible after filtration. Samples that cannot be submitted within a few hours should be filtered (0.45 mm) then frozen to reduce the loss of ammonia and changes in nitrite/nitrate concentrations. Plain blood collection tubes are suitable containers for freezing. Two 10-ml tubes will provide enough samples for all three tests. Do not fill completely; leave space for expansion during freezing.

1.3.2.15 The Nitrogen Cycle

In aquaculture, the nitrogen cycle eliminates ammonia from the water column by bioconversion to nitrite and then nitrate. Nitrate is used by plants, including algae, as a nutrient. This constant change from ammonia to nitrite to nitrate is called the nitrogen cycle (Figure 1.1). In ponds, this

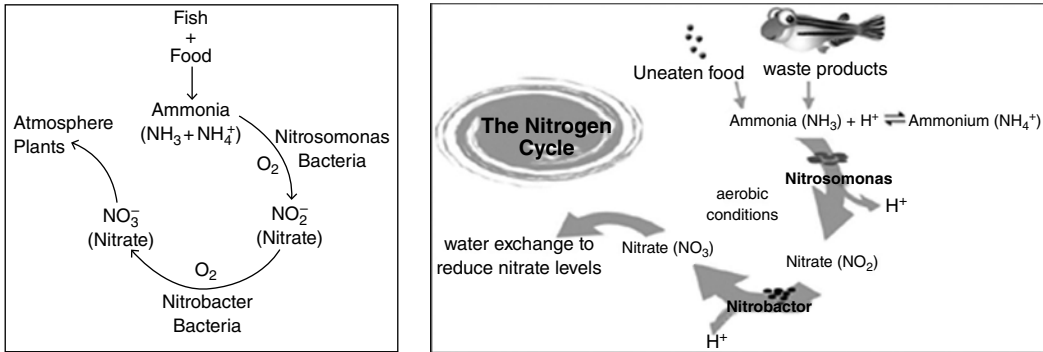


Figure 1.1 The nitrogen cycle (Francis-Floyd, 2003). *Source:* image on the right courtesy of Gillian Taylor.

process takes place in the surface layers of the mud, but in tanks or aquaria, a special place is provided for the bacteria to live and flourish. This is called a biological filter, or biofilter. One important point to mention about the nitrogen cycle is that both groups of nitrifying bacteria need oxygen to function. If oxygen levels are insufficient, the process can slow down and the levels of ammonia and nitrite will rise in the system.

1.3.2.16 Interaction of Chemical and Physical Properties of Water that Affect Aquatic Animal Health

Some chemicophysical parameters of water have a direct influence upon fish health. Any abrupt or large fluctuations of these parameters often cause a state of stress in fish, sometimes resulting in widespread disease outbreaks. Dissolved oxygen content, pH, turbidity, temperature, introduction of some chemicals, detergents, pesticides and naturally produced toxic compounds like hydrogen sulfide, ammonia, and dinoflagellate toxins are potential stress-related parameters. Carbon dioxide concentration up to 20–30 mg/l can be tolerated by fish provided that oxygen is near saturation. At lower levels of dissolved oxygen, the toxicity of carbon dioxide increases. The optimum pH range is between 6.7 and 8.6; liming agents may be applied to correct a low pH.

Ammonia concentration above 1 mg/l can indicate organic pollution. Hydrogen sulfide toxicity increases with decreasing pH and it is harmful even at a concentration of 1 mg/l. Making the aquasystem environment more congenial and hygienic reduces stress and promotes fish health. For example, excessive application of inorganic fertilizers and accumulation of organic matter in older aquasystems may cause an over production of phytoplankton, and the appearance of algal and bacterial blooms, leading to dissolved oxygen depletion to lethal levels. For health and optimum growth, the dissolved oxygen level should not drop below 2–5 mg/l. Carbon dioxide concentration up to 20–30 mg/l can be tolerated by fish provided oxygen is near saturation. Nephrocalcinosis in salmonids has long been recognized as a pathological entity related to high dissolved CO_2 , eventually leading to the formation of large mineralized deposits within the excretory tissue of the kidney and associated kidney pathology. The condition can result in poor condition and performance and occasional fish loss, particularly if other stressors are present.

At lower levels of dissolved oxygen, the toxicity of carbon dioxide increases. The optimum pH range is between 6.7 and 8.6; liming agents may be applied to correct low pH. Problems with high pH are common in fry nursery ponds and in ponds used to grow freshwater prawns (*Macrobrachium rosenbergii*). This is because fertilization practices used to prepare ponds for stocking are designed to promote fast-growing phytoplankton blooms that rapidly take up carbon dioxide. Unfortunately,

the early life stages of fish and crustaceans are particularly susceptible to pH toxicity and juveniles are less able than older animals to avoid areas of pH shift by moving to areas of stable pH in the pond (such as deeper waters).

Ammonia concentration above 1 mg/l indicates organic pollution. Ammonia is very important in intensive systems; in small amounts, ammonia causes stress and gill damage. Fish exposed to low levels of ammonia over time are more susceptible to bacterial infections, have poor growth and will not tolerate routine handling. Deformities and significant behavioral changes associated with chronic exposure to nitrates have been documented in rainbow trout (*Oncorhynchus mykiss*) raised in recirculating aquaculture systems with nitrate concentrations at levels less than one-tenth the recommended maximum nitrate nitrogen level of 1000 mg/l.

Hydrogen sulfide toxicity increases with decreasing pH and it is harmful even at 1 mg/l concentration level. Proper and timely management of soil and water by manipulating feeding, fertilization, liming, addition of water and aeration eliminates most of the environmental stressors and provides better and healthier environments for the growth of fish. Hydrogen sulfide has been referred to as a silent killer of shrimp, causing tissue corrosiveness by irritating soft tissues in the gills, gut, stomach walls and hepatopancreas. H_2S stresses shrimp, lowering their resistance to infection. A safe level for H_2S in giant tiger shrimp (*Penaeus monodon*) ponds has been reported as 0.033 ppm while white shrimp (*P. vannamei*) post larvae has been said to tolerate up to 0.0087 ppm and juveniles up to 0.0185 ppm (Panakorn, 2016).

1.3.3 Biotic Factors of Water

1.3.3.1 Biological Oxygen Demand

The biological oxygen demand is a measure of the oxygen used by all organisms in the aquasystem. Microbes (bacteria and fungi) use oxygen to decompose organics (may use 1–3 ppm dissolved oxygen in 24 hours). Phytoplankton respire at night to use oxygen (may use 5–15 ppm dissolved oxygen nightly). Fish respire day and night (may use 2–6 ppm dissolved oxygen in 24 hours). Dissolved oxygen levels fall at night, since all organisms are respiring, and rise during the day, since plants photosynthesize to use carbon dioxide and eliminate oxygen (may gain 5–20 ppm dissolved oxygen daily). Diffusion and wave/wind action add oxygen (may add 1–5 ppm dissolved oxygen).

The measurement of biological oxygen demand is a chemical procedure which determines the amount of dissolved oxygen needed by aerobic organisms in a body of water to break down organic material present in a given water sample at a certain temperature over a specific time period. It is not a precise quantitative test, although it is widely used as an indication of the organic quality of water. It is most commonly expressed in milligrams of oxygen consumed per liter of sample for five days of incubation at 20°C and is often used as a robust surrogate of the degree of organic pollution of water. Sources of biological oxygen demand include topsoil, leaves and woody debris; animal manure; effluents from pulp and paper mills, wastewater treatment plants, feedlots, and food-processing plants; failing septic systems; and urban rainwater runoff.

1.3.3.2 Plants and Algae

The type and amount of vegetation in an aquatic environment can have a positive or negative effect on the health of the aquasystem. Vegetation can improve the growth of fish by allowing for shade, shelter, and removal of nitrates and phosphates from the water. At night, excessive vegetation can also decrease the oxygen available to the fish, since plants will use oxygen for respiration and may impede movement of the fish and decrease their ability to catch prey or avoid predation. Studies performed on bass, sunfish and brill have shown that different species of fish react differently to

vegetation levels. In one study, the size and life stages of fish also affect the amount of vegetation that is most appropriate. In this study, bass were less tolerant of the plants filling the water column than brill or sunfish. Larger bass were negatively affected when 30% of the water column was filled with plants, while smaller bass were negatively affected when 50% of the water column was filled. These studies express the importance of knowing the species of plant and the species of fish for which the veterinarian is caring.

Floating beds of vegetation are a good answer to nitrate and phosphate removal. Studies performed on rye grass in floating beds has shown that it effectively removes a significant amount of the NO_3 (Bartucca *et al.*, 2016). By creating floating beds of vegetation, the nitrate and phosphate levels can be reduced without encumbering the water column. It is important to monitor the oxygen, nitrogen, and phosphorous levels to decide the best density of planting.

1.3.3.3 Algal Bloom

Overabundance of nutrients like phosphorus, particularly when the water is warm and the weather is calm, can lead to excessive growth of algae. According to the World Health Organization standard, when the population of algal cells exceeds 100 000 cells/ ml, the condition is termed an algal bloom. Excessive algal growth can lead to nocturnal hypoxic conditions where fish may start to gasp on the water surface, and mass mortality events may occur. Phytoplankton blooms can also cause large diurnal fluctuations in other water quality variables (e.g. very high pH due to excessive use of free CO_2 for photosynthesis) in mid-afternoon. Such conditions are stressful to fish. Blue-green algae can also produce toxic substances that are lethal to some fish (see also Harmful Algal Bloom below). They can also produce compounds that impart a strong off-flavor to fish. In addition, dead algae can accumulate at the pond bottom and create toxic gases from decomposition.

The best approach to controlling algae is to regulate nutrient inputs by moderate stocking and feeding rates. Phosphorus is one of the major factors responsible for increased levels of phytoplankton in an aquasystem. It is possible to precipitate phosphorus from pond water by applying sources of iron, aluminum, or calcium ions. These ions precipitate phosphorus as insoluble iron, aluminum or calcium phosphates. The use of alum (aluminum sulfate) 20–30 mg/l or gypsum (calcium sulfate) 100–200 mg/l to precipitate excessive concentrations of phosphorus may also be feasible. Alum is acidic and more suitable for use in waters of 500 mg/l total alkalinity and above. Gypsum is better for use in low alkalinity waters.

1.3.3.4 Larger Aquatic Plants

Larger aquatic plants or macrophytes include pondweed and milfoil. These plants are undesirable in aquasystems because they interfere with fish management (feeding and harvesting), compete with plankton for nutrients, provide shelter for undesirable fish, contribute to oxygen depletion and high ammonia levels when they decompose and contribute to water loss through transpiration. Drying and desilting of aquasystems every one or two years will keep the growth of aquatic plants in check.

1.3.3.5 Pests and Predators

Pests and predators are unwanted animals or plants that destroy fish or hinder the production of target fish species. Birds, fish, reptiles, amphibians, mammals and even certain invertebrates are known predators of cultured fish. Bird predation is the major source of fish loss at aquaculture facilities. Tadpoles feed on aquatic plants and animals, while frogs can eat fish fry. They lay eggs on the surface of ponds, which interferes with surface oxygenation, thereby restricting fish growth if not checked. Sea anemones can grow on boards, sluice gates and grooves, and on concrete dikes in high-salinity, brackish water areas, and are poisonous to fish.

Fish predators can be used in controlling an overpopulation of fish in ponds. Wild fish are one of the potential sources of fish pathogenic organisms. Some of these wild species are predatory in nature and prey upon the young of cultivated species. They can rapidly multiply and compete for food with the cultivated fish. Control of nuisance fish can be done by poisoning or dewatering the pond. Piscicides of plant origin are preferred over common chemical insecticides used in agriculture due to the long-lasting toxicity and residual effect of the latter. Moreover, fish killed by insecticides become unfit for human and animal consumption. Piscicides derived from plants such as mahua oil cake are most commonly used as their toxicity lasts for about two weeks and the fish killed are fit for human consumption. Mahua oil is applied at 200–250 ppm. However, the poisoning process kills only the unwanted fish species of the pond without affecting the pathogenic organisms they may carry. Disinfection of water is an effective means of disease control in fish culture by reducing the numbers of pathogens to minimum levels.

1.4 Monitoring and Regulation of Life-Support Systems

Proper and timely management of soil and water by manipulating feeding, fertilization, liming, addition of water, aeration, bottom raking, and so on, eliminates most of the environmental stressors and provides better, healthy environments for the production of fish and other aquatic animals. Proper management also increases the immune response against pathogens. Eradication of predatory and nuisance fish, disinfecting the pond, selection of quality and healthy seed for stocking, maintaining proper species ratio and stocking density, water quality regulation, proper feeding and proper handling are the important steps of this management exercise. Water quality parameters that are commonly monitored in the aquaculture industry include temperature, dissolved oxygen, pH, alkalinity, hardness, ammonia, nitrites and nitrates. Depending on the culture system, carbon dioxide, chlorides, and salinity may also be monitored. Some parameters such as alkalinity and hardness are fairly stable, but others like dissolved oxygen and pH can fluctuate daily. Most of these parameters are measured with physical reagents or meters. However, there are several classical methods of water analysis which are still used today. Atomic absorption analysis provides the atomization of a sample in flame or electrothermal plasma. Liquid sample is turned into an atomic gas through desolvation, evaporation, and volatilization. Electrochemical methods are based on an analysis of the processes occurring at the electrodes and in the inter-electrode space followed by the measurement of the potential and/or current in an electrochemical cell containing the analyte. Gravimetric analysis involves determining the amount of analyte through the measurement of mass (Table 1.1).

Relative concentration changes for dissolved oxygen, carbon dioxide and pH in ponds over 24 hours are shown in Table 1.2. Biological oxygen demand measurement requires taking two measurements. One is measured immediately for dissolved oxygen (initial), and the second is incubated in the laboratory for five days and then tested for dissolved oxygen remaining (final). This represents the amount of oxygen consumed by microorganisms to break down the organic matter present in the sample during the incubation period.

In ponds with moderate to high alkalinity (good buffering capacity) and similar hardness levels, pH is neutral or slightly basic (7.0–8.3) and does not fluctuate widely. Higher amounts of CO₂ (i.e. carbonic acid) or other acids are required to lower pH because more base is available to neutralize or buffer the acidity. The relationship of alkalinity, pH and CO₂ is shown in Table 1.3. The number (factor) found in the table which corresponds to the measured pH and water temperature is multiplied by the measured alkalinity value (mg/l as CaCO₃). The product of these numbers estimates CO₂ concentration (mg/l).

Table 1.1 Water quality factors, commonly used monitoring procedures, and preferred ranges for fish culture. Details for specific test procedures can be obtained from a commercial supplier or appropriate text (e.g. APHA 1989).

Water quality factor	Test procedure	Preferred ranges for fish culture
Temperature	Thermometer, telethermister	Species dependent
Dissolved oxygen	Titrimetric (modified Winkler) polarographic meter, calorimetric kits	> 4–5 ppm for most species
Total ammonia-nitrogen (ionized and un-ionized)	Calorimetric kits, (Nesslerization or salicylate), ion specific probes	NH < 0.02 ppm
Nitrite	Calorimetric kits (diazotization), ion specific probes	< 1 ppm; 0.1 ppm in soft water
pH	Electronic meter, calorimetric kits,	6–8 ppm
Alkalinity	Titrimetric with pH meter, titrimetric with chemical indicator	50–300 ppm calcium carbonate
Hardness	Titrimetric kit	> 50 ppm, preferably > 100 ppm calcium carbonate
Carbon dioxide	Titrimetric kit	< 10 ppm
Salinity	Conductivity meter	species dependent typically < 0.5–1.0 ppt for freshwater fish)
Hydrogen sulfide	Calorimetric kit	No detectable level
Clarity	Secchi disk, turbidimeter	Species dependent

Table 1.2 Relative concentration changes for dissolved oxygen, carbon dioxide, and pH in ponds over 24 hours (Tucker 1984).

	Change		
Time	Dissolved oxygen	Carbon dioxide	pH
Daylight	Increases	Decreases	Increases
Night	Decreases	Increases	Decreases

Since most test kits measure total ammonia nitrogen, it is important to determine what percentage of the total is toxic. In healthy ponds and tanks, ammonia levels should always be zero. Since the toxicity of UIA begins as low as 0.05 mg/l, a positive total ammonia nitrogen test needs to be followed by a test to find the actual concentration of UIA. Once the pH and temperature are known, the fraction of UIA present can be determined (Table 1.4), as presented in Table 1.5. Samples that cannot be submitted within a few hours should be filtered (0.45 mm) then frozen to reduce the loss of ammonia and changes in nitrate/nitrite concentrations.

Salinity is usually not measured directly, but is instead derived from the conductivity measurement (Wagner *et al.*, 2006). This is known as practical salinity. These derivations compare the specific conductance of the sample to a salinity standard such as seawater. Salinity measurements based on conductivity values are unitless but are often followed by the notation of practical salinity units (Nelson and Siegel, 2014).

Table 1.3 Factors for calculating carbon dioxide concentrations in water with known pH, temperature and alkalinity measurements.^a

pH	Temperatures (°C)						
	5	10	15	20	25	30	35
6.0	2.915	2.539	2.315	2.112	1.970	1.882	1.839
6.2	1.839	1.602	1.460	1.333	1.244	1.187	1.160
6.4	1.160	1.010	0.921	0.841	0.784	0.749	0.732
6.6	0.732	0.637	0.582	0.531	0.495	0.473	0.462
6.8	0.462	0.402	0.367	0.335	0.313	0.298	0.291
7.0	0.291	0.254	0.232	0.211	0.197	0.188	0.184
7.2	0.184	0.160	0.146	0.133	0.124	0.119	0.116
7.4	0.116	0.101	0.092	0.084	0.078	0.075	0.073
7.6	0.073	0.064	0.058	0.053	0.050	0.047	0.046
7.8	0.046	0.040	0.037	0.034	0.031	0.030	0.030
8.0	0.029	0.025	0.023	0.021	0.020	0.019	0.018
8.2	0.018	0.016	0.015	0.013	0.012	0.012	0.011
8.4	0.012	0.010	0.009	0.008	0.008	0.008	0.007

Source: Tucker, 1984.

^a Factors should be multiplied by total alkalinity (mg/l) to get carbon dioxide (mg/l). For practical purposes, CO₂ concentrations are negligible above pH 8.4.

Total dissolved solids are reported in mg/l and can be measured by gravimetry or calculated by multiplying a conductivity value by an empirical factor; standard methods for the examination of water and wastewater accepts a total dissolved solids constant of 0.55–0.7 mg/l, although if the water source is known to be high in calcium or sulfate ions, a constant of 0.8 mg/l may be used (American Public Health Association *et al.*, 2017). Depending on the ionic properties, excessive total dissolved solids can produce toxic effects on fish and fish eggs. Salmonids exposed to higher than average levels of CaSO₄ at various life stages experienced reduced survival and reproductive rates. When total dissolved solids ranged above 2200–3600 mg/l, salmonids, perch and pike all showed reduced hatching and egg survival rates (Scannel and Jacob, 2001).

It is important for the clinician to be familiar with the types of life-support system monitoring reports used in modern aquatic facilities. Each facility will have its own template or digital form tailored to the specific monitoring procedures.

1.5 Maintaining Optimal Conditions

Various processes, both natural and artificial, as well as varied physical components, are all involved with creating an aquasystem that provides the healthiest environment for maintaining its inhabitants.

Table 1.4 Fraction of un-ionized ammonia in aqueous solution at different pH values and temperatures. *Source:* calculated from data in Emmerson *et al.* (1975).

pH	Fraction at temperature															
	(°F)	42.0	46.4	50.0	53.6	57.2	60.8	64.4	68.9	71.6	75.2	78.8	82.4	86.0	89.6	
(°C)	6	8	10	12	14	16	18	20	22	24	26	28	30	32		
7.0	0.0013	0.0016	0.0018	0.0022	0.0025	0.0029	0.0034	0.0039	0.0046	0.0052	0.0060	0.0069	0.0080	0.0093		
7.2	0.0021	0.0025	0.0029	0.0034	0.0040	0.0046	0.0054	0.0062	0.0072	0.0083	0.0096	0.0110	0.0126	0.0150		
7.4	0.0034	0.0040	0.0046	0.0054	0.0063	0.0073	0.0085	0.0096	0.0114	0.0131	0.0150	0.0173	0.0198	0.0236		
7.6	0.0053	0.0063	0.0073	0.0086	0.0100	0.0116	0.0134	0.0155	0.0179	0.0206	0.0236	0.0271	0.0310	0.0369		
7.8	0.0084	0.0099	0.0116	0.0135	0.0157	0.0182	0.0211	0.0244	0.0281	0.0322	0.0370	0.0423	0.0482	0.0572		
8.0	0.0133	0.0156	0.0182	0.0212	0.0247	0.0286	0.0330	0.0381	0.0438	0.0502	0.0574	0.0654	0.0743	0.0877		
8.2	0.0210	0.0245	0.0286	0.0332	0.0385	0.0445	0.0514	0.0590	0.0676	0.0772	0.0880	0.0998	0.1129	0.1322		
8.4	0.0328	0.0383	0.0445	0.0517	0.0597	0.0688	0.0790	0.0904	0.1031	0.1171	0.1326	0.1495	0.1678	0.1948		
8.6	0.0510	0.0593	0.0688	0.0795	0.0914	0.1048	0.1197	0.1361	0.1541	0.1737	0.1950	0.2178	0.2422	0.2768		
8.8	0.0785	0.0909	0.1048	0.1204	0.1376	0.1566	0.1773	0.1998	0.2241	0.2500	0.2774	0.3062	0.3362	0.3776		
9.0	0.1190	0.1368	0.1565	0.1782	0.2018	0.2273	0.2546	0.2836	0.3140	0.3456	0.3783	0.4116	0.4453	0.4902		
9.2	0.1763	0.2006	0.2273	0.2558	0.2861	0.3180	0.3512	0.3855	0.4204	0.4557	0.4909	0.5258	0.5599	0.6038		
9.4	0.2533	0.2847	0.3180	0.3526	0.3884	0.4249	0.4618	0.4985	0.5348	0.5702	0.6045	0.6373	0.6685	0.7072		
9.6	0.3496	0.3868	0.4249	0.4633	0.5016	0.5394	0.5762	0.6117	0.6456	0.6777	0.7078	0.7358	0.7617	0.7929		
9.8	0.4600	0.5000	0.5394	0.5778	0.6147	0.6499	0.6831	0.7140	0.7428	0.7692	0.7933	0.8153	0.8351	0.8585		
10.0	0.5745	0.6131	0.6496	0.6844	0.7166	0.7463	0.7735	0.7963	0.8207	0.8408	0.8588	0.8749	0.8892	0.9058		
10.2	0.6815	0.7152	0.7463	0.7746	0.8003	0.8234	0.8441	0.8625	0.8788	0.8933	0.9060	0.9173	0.9271	0.9389		

Table 1.5 Stepwise determination of un-ionized ammonia.

STEP	ACTIVITY
1	Measure pond TAN with commercial test kit
2	Measure Pond temperature
3	Measure Pond pH
4	Find the multiplication factor in Table 1.1 using the pond temperature and pH
5	Multiply TAN and factor from step 4 to determine un-ionized ammonia nitrogen

TAN, total ammonia nitrogen.

1.5.1 Biofilms

Millions of bacteria and viruses are found in every milliliter of water in a healthy aquasystem and on every surface, and they comprise very complex ecosystems called biofilms. The relationship between beneficial and pathogenic organisms is very complex. Culturing bacteria only identifies less than 1% of the bacteria in water. Current technologies such as quantitative polymerase chain reaction are demonstrating a vastly diverse microbial ecosystem. A managed system demonstrated over 890 genera and over 2500 species of bacteria which only accounted for 69% of the bacterial DNA; the rest were unidentified (Stamper, 2015).

Aquatic microorganisms occur in two realms: planktonic and sessile. The planktonic state is fairly straightforward, whereas the sessile biofilm ecology is extremely complex. Biofilms include organisms beyond bacteria and comprise fungi, algae, protozoans and metazoans. An intricate ecological process occurs as various environmental parameters change. These organisms secrete an assortment of chemicals known as extracellular polymeric substances, which add to the total organic carbon within the water column and are critical to the health of the organisms and water environment (Stamper, 2015; Tepper, 2019).

An important consideration in any aquatic system is the potential for pathogenic bacteria to create a biofilm that will allow it to persist in a tank or other culture system. Biofilm formation and disinfection can be especially problematic in recirculating aquaculture systems. Once a highly pathogenic organism is introduced, it can persist in the system indefinitely and continue to cause recurrent disease outbreaks.

Secreting a protective coating can allow bacteria to survive desiccation and standard cleaning techniques. If recurrent bacterial disease becomes an issue in cultured species, additional disinfection of the system may be indicated. This may require depopulation while strong disinfectants are used, following a period of dewatering and drying of the system. Physical methods such as heat and pressure washing can be used and a combination of physical and chemical treatment techniques may increase the effectiveness of the disinfection.

1.5.2 Chemical Flocculation and Bioflocculation

As urban populations grow and the science surrounding water quality improves, regulatory changes in discharge limits for large-scale aquaculture operations have led to a need for more intense handling of solids. One sophisticated and large-scale method of processing biological solid wastes uses the theory of biological flocculation or “bioflocculation”. While the veterinarian may not typically be called upon to manage such systems, it is important to understand that these

systems might be in use, how they operate at a basic level, and how they may impact animal health. Perhaps the most common concern for aquatic veterinarians is how chemicals used for disease treatment in a system may impact the function of these solids treatment systems.

Flocculation involves the clumping of suspended particles together into a settleable “floc”. Chemical flocculation is used in wastewater treatment plants to remove solids from water on a large scale, where surfactant chemicals such as alum or ferric chloride are used. More recently, polyacrylamide has also been used.

Large-scale aquaculture systems with large biological loads (and thus a high volume of fecal wastes) may use this chemical flocculation method. Home aquaria are also often treated with chemical flocculants to clear cloudy water.

Another method that may be used in small-scale aquaculture systems is bioflocculation, where microorganisms cause particles to clump together when natural chemicals such as polysaccharides are secreted as a result of their normal biological functions. This naturally occurring process is important for the proper functioning of all filter systems. Small-scale aquaculture facilities may employ an enhanced system of bioflocculation. While fine suspended organic particles from animal wastes will settle out on their own in a stagnant tank or pond over time, organic particles such as phosphorus never quite settle out. Microorganism activity speeds up the process and leads to a more effective solids removal by clumping particles together into a floc which can be removed from the water. This type of system can be used to treat wastewater and reduces the total dissolved solids and phosphorus following prefiltration to remove larger particles. The intensity of the wastewater treatment required will depend on many factors, and the discharge of such chemicals as nitrogen and phosphorus may be limited by a water jurisdiction agency to reduce eutrophication impacts on the environment. Flocculation is commonly used in combination with coagulation, which creates microflocs by decreasing the negative charges that repel particles and cause them to remain in suspension.

The budget, amount of land area available for this application, biological load in the system, and the regulatory limits of the discharge permit will determine the type and size of system used. Typically these systems require a long time to “turn over” and are limited in volume. These methods are well suited to a settling basin or belt filter.

As with a biological filter, the type and amount of chemicals or microorganisms present in the system will impact the speed and effectiveness of this water treatment method. Treatment chemical residues in the wastewater may interact with chemical flocculants. High concentrations of toxic chemicals containing formaldehyde or chlorine used to treat diseases in fish may negatively impact the microorganism population of the system and treatment may need to be altered to a lower concentration over a longer period to reduce the lethal dose in the water treatment system. In some cases, bypassing the settling basin or belt filter may be considered.

The principle of bioflocculation has other applications and is currently a growing area of research for aquatic animal feeds, pathogen reduction, and biofuel production.

1.5.2.1 Effect of Therapeutants and Disinfectants on Biofilters

Certain therapeutants and disinfectants, such as iodine and ozonation can have a detrimental effect on the functioning of active biofilters. Always consult product information before using these products in an aquasystem.

1.5.3 Environmental Toxins and Pollutants

1.5.3.1 Heavy Metals

Heavy metals occur naturally in the environment due to geologic weathering but can also occur from human use. It is important to survey the immediate environment as well as the areas upstream

to the water source used in an aquasystem. Atmospheric pollution from coal combustion is a major cause of many heavy metals being found in source water. Residential and agricultural use of pesticides and fertilizer may contribute to several types of heavy metals in groundwater, well water, lakes, ponds and estuaries. Other types of industrial waste to consider are leather tanneries, textile industry, battery storage or production, plastic production, metal finishing, mining, and pigment production.

There are essential and non-essential heavy metals. Essential metals include copper, zinc, chromium, nickel, cobalt, molybdenum and iron. Nonessential metals include cadmium, mercury, tin and lead (Sfakianiakis *et al.*, 2015).

Essential metals can cause disease if they are deficient and if the levels are too concentrated. The most bioavailable form of these metals is the dissolved ionic form. These metals help during larval development of the neurologic and gastrointestinal systems (Sfakianakis *et al.*, 2015; Authman *et al.*, 2015).

Essential and nonessential heavy metals can cause damage at high concentrations. Knowing that there is potential for heavy metal in the water source is an important part of diagnosing decreased reproductive health, slow growth, and acute mortalities. The periods during which fish are most susceptible to heavy metal toxicity are the embryonic and larval stages. They can cause lordosis, increased mortality, hatching delays and other anomalies. According to Sfankianakis *et al.* (2015), morphological deformities are commonly used as a biomarker to study the effects of contamination.

Fish are susceptible to heavy metals through their gills, gastrointestinal tract and through skin absorption. These metals will accumulate in the liver, kidney, and gills. Arsenic has also been found to accumulate in the retina and cadmium has been found in the heart muscle (Authman, 2015).

Although there are differences in the mechanism of action, most heavy metals cause damage to the liver, gills, reproductive, immune, and neurologic systems. They cause damage to the gills through hyperplasia and necrosis. Aluminum causes lamellar fusion which leads to necrosis. Iron accumulates in the gills as a precipitate causing vascular occlusion. At chronic doses, many heavy metals negatively affect leukocytes, erythrocytes, and antibodies. They can also affect the endocrine system, decreasing reproductive health. Iron can precipitate on eggs causing a reduction of oxygen reaching the larvae. Other heavy metals decrease growth through increased biological stress, disruption of osmotic balance, and developmental changes. Many heavy metals cause neurologic damage, including mercury, which causes behavior changes, cognitive changes, ataxia, and convulsions (Kennedy, 2002).

Studies on cadmium show that it is primarily absorbed via the gastrointestinal system and the gills. It interrupts the absorption of calcium, which can cause problems with reproduction, growth, and development. It can also cause acute death from hypocalcemia. Cadmium levels as low as 0.001 ppm affect the hatching and larval survival of many species of fish.

Studies have been performed showing that excessive copper can affect the gills, gastrointestinal system, and the sensory system. These studies showed that zebra fish larvae had difficulty orienting in the water column, reduced hatching and impairment of growth (Authman, 2015).

1.5.3.2 Insecticides

Insecticides may be used in the local environment for the control of agricultural pests or insects that cause vector borne diseases. These compounds can affect fish through runoff and atmospheric accumulation on pond surfaces. There are many types of insecticides on the market. Most insecticides affect the neurologic system using different mechanisms of action. They may affect acetylcholinesterase activity, block sodium channels, bind nicotinic receptors, affect the peripheral nervous system and directly degrade neurotransmitters in several other ways. They can also cause

chromosomal aberrations, decrease protein levels, affect the immune system, and mimic or block reproductive hormones (Sabra and Mehana, 2015). Compared with insects, they differ in their specificity for vertebrates, their toxicity, their bioaccumulation, and their environmental persistence.

Organochlorines have been banned in most countries due to their highly toxic nature. They are toxic to most animals, have a high level of bioaccumulation, and persist in the environment for long periods of time (Fulton *et al.*, 2013).

Organophosphates have a lower level of bioaccumulation and a shorter half-life in the environment. They may be used as a treatment for several fish parasites, but can be easily overdosed. Some fish are more sensitive to organophosphates than others. If using organophosphates for treatment of disease, always determine the species sensitivity.

Carbamates are frequently used for agriculture and residential insect control. They also have minimal bioaccumulation and environmental persistence. They are less toxic to fish than organophosphates, but still can cause chronic toxicity if overused around an aquasystem (Fulton *et al.*, 2013).

Pyrethroids and nicotinoids are also commonly used in the environment. Nicotinoids cause irreversible block of the post synaptic nicotinoid receptors. Pyrethroids cause damage to the sodium channel in the neurons leading to hyperexcitability, tremors, ataxia and paralysis (Sabra and Mehana, 2015).

There are many other types of insecticides to consider. Although most will not persist in the environment more than one or two weeks, if used consistently, they may lead to chronic disease. This may appear as decreased growth, low reproduction, and slow increase in mortality.

1.5.3.3 Herbicides

There are many different classes of herbicides. Most herbicides do not directly affect fish; however, different species of fish respond to varied levels and classes of herbicides. Studies performed on forestry herbicides in regard to fish development showed no evidence of toxicity in zebra fish to picloram, clopyralid, imazapic, glyphosate, imazapyr, and triclopyr (Stehr *et al.*, 2009). However, studies evaluating *Cyprinus carpio* and rohu showed an increased mortality and histologic changes in the kidney, liver, and gills when exposed to glyphosate at 86 mg/l. This same study showed increased mortality in response to the use of the salt formulation of 2, 4-D at 100 mg/l and paraquat at 26 mg/l.

Atrazine has been shown to affect the hypothalamus–pituitary–gonadal axis in vertebrates. One study has shown that this herbicide negatively affects the egg production of fathead minnows (Tillitt, 2010).

Pretilachlor is frequently used in rice fields. Studies performed on *Clarias batrachus* exposed to this herbicide had significant behavioral changes, including decreased feeding and increased buccal activity (Soni, 2018).

At high levels, triazine has been shown to cause acute toxicity in fish. This herbicide can also cause negative affects regarding reproduction and reproductive development (Hostovsky *et al.*, 2014).

When diagnosing possible toxic exposure in a fish population, herbicides should be considered if they have been used in the water or in the area nearby due to potential runoff. When developing a farm management program that includes herbicides, it is important to consider the following:

- The sensitivity of the species you are working with.
- The potential exposure to wild fish in the area and their sensitivity.
- The bioaccumulation of the herbicide directly into the water or in the runoff.
- The half-life of the chemical if it is used in an unpopulated pond.

1.5.3.4 Harmful Algal Blooms

Micro- and macroalgae have been implicated in millions of dollars of loss in aquaculture over the past decade (Hallegraeff *et al.*, 2017) and large numbers of deaths of aquatic animals in the wild and in aquariums. These harmful algal blooms have recently increased in our environment due to increased migration, climate change, CO₂ levels, and nutrients in the water. Harmful algal blooms are caused by microalgae species including diatoms, cyanobacteria, raphidophytes, prymnesiophytes, pelagophytes, and silicoflagellates (Landsberg, 2002). The most common freshwater algal blooms are caused by cyanobacteria.

Increased water traffic has allowed for migration of algal blooms from continent to continent, allowing the microalgae to find the most conducive environment for growth. Changes in water temperature, pH, and nutrient upwelling have allowed many of these microalgae species to bloom in new areas where animals are not adapted to their presence (Anderson *et al.*, 2012). Climate change has also affected the percentage of toxic compared with nontoxic strains such as *Microcystis* and a decreased overwintering period as seen in *Cylindrospermopsis* (Wood *et al.*, 2015).

The effects of harmful algal blooms are very difficult to anticipate due to the variety of microalgae, the toxins that each species contain, and the sensitivity of aquatic animals to those toxins. Each species of microalgae has a different environmental niche. Some are benthic and require limited sunlight. Others must remain near the surface. There are some microalgae that can adjust their buoyancy to move to the most effective area where both photosynthesis and nutrient absorption are greatest. Many adaptations of microalgae allow for them to have an advantage over other algae. These adaptations also tend to give the nontoxic forms of the species less of an advantage over the toxic forms.

The toxins of algal blooms include hepatotoxins, neurotoxins, and dermatotoxins. Each bloom may occur without the presence of toxins in the water and the toxins may only be found in the water before the mortality is detected. This adds to the difficulty in the diagnosis of algal bloom toxicity. The Centers for Disease Control and Prevention list the majority of toxins that are known at this time (Roberts *et al.*, 2020). A more complete explanation of the individual events that have shown proven effects of the toxins in aquatic animals can be found in Jan Landsberg's (2002) article "The effects of harmful algal blooms on aquatic organisms".

Cyanobacteria can cause fresh water algal blooms. They are found in ponds, lakes and estuaries. When fresh water incursion occurs and nutrient levels are high, these blooms can reach marine life on the coast. Cyanobacteria most commonly produce microcystins, anatoxins, and cylindrospermopsin. Most aquatic species appear to be resistant to anatoxins and cylindrospermopsin. There have been bird deaths with anatoxin-a(s) and a possible alligator mortality with cylindrospermopsin. Microcystins have been shown to cause both acute disease and chronic disease. The route of uptake is through the gastrointestinal tract, gills, and skin. There have been many field observations and laboratory experiments with fish regarding microcystin-LR. These studies have shown that microcystins affect the leukocytes, liver enzymes, growth rate, and ionic stability. The histopathological changes are dose dependent and include degenerative changes to the kidney, liver, and gills (Malbrouck, 2009).

Fish appear to be more resistant than mammals to many neurotoxins, such as brevetoxins, domoic acid, and saxitoxins. These toxins bioaccumulate in fish and cause high mortality rates in birds and marine mammals. Saxitoxins cause paralytic shellfish poisoning (PSP) in mammals due to sodium channel blockage. There are 21 derivatives of saxitoxins involved with PSP. Recent studies show a direct effect on mollusks causing increased mortality and poor growth. Herring exposed to *Alexandrium tamarense* show increased mortality due to asphyxiation (Landsberg, 2002). Brevetoxins include nine types of neurotoxins, several of which are found in

sea spray and act as a respiratory irritant. The neurotoxins alter membrane properties in the neurologic cells. Manatee deaths caused by brevetoxins is postulated to be caused by the ingestion of tunicates found on the surface of sea grass and through inhalation when traveling through large blooms (Landsberg, 2002).

Domoic acid is an excitatory neurotransmitter binding glutamate receptors. There are some copepods that are very sensitive to domoic acid, others are less sensitive and act as a vector. Domoic acid has not been identified in fish mortality events, but it has caused mortalities in brown pelicans after ingestion of anchovies (Lansberg, 2002).

Harmful algal blooms can also cause a reduction in the available oxygen during respiration, exaggerating the toxic effects. Many of the species involved in these blooms are more tolerant to the low oxygen levels and will thrive on the nutrients created by the increased mortality of other organisms. Harmful algal blooms also contain superoxide radicals, phycotoxins and fatty acids that have been acknowledged to play a role in aquatic species mortality through gill damage (Dorantes-Aranda, 2015; Hallegraeff *et al.*, 2017). Studies performed by Hallegraeff show that reactive oxygen species do not cause damage to fish independently, but when free fatty acids are present they work in synergy to increase the potency of the fatty acid. This results in damage to the gills and to osmoregulation. Microalgal blooms such as barbed diatoms can also cause damage to the gills through mechanical stress by lodging in fish gills (Hallegraeff *et al.*, 2017).

There is no effective treatment for the toxicity that occurs due to harmful algal blooms. Avoidance of the blooms is best achieved by decreasing runoff of high nutrient materials, planting grasses and other plants near waterways to help reduce nutrient runoff and use the nutrients in the water, and careful monitoring. If a large bloom occurs in an aquaculture facility, there are several considerations that need to be made before action is taken:

- Is the toxin present a danger to humans, aquatic species, or both?
- Is an early harvest an option?
- Will killing the algae cause more toxin to be suddenly released?

With blooms that are specifically ichthyotoxic due to fatty acids and reactive oxygen species, the fish may still be safe for harvest. If so, it is important to prevent histamine build-up by keeping the fish alive long enough to allow the system to be flushed. This can be accomplished by airlift upwelling and targeted clay applications to remove the ichthyotoxins (such as bentonite clay at 0.05–0.25 g/l for *Prymnesium*, *Karenia*, *Karlodinium*, *Chattonella*, *Heterosigma*, and *Alexandrium*). The last three listed have a lower removal percentage than the first three (Hallegraeff *et al.*, 2017). If an early harvest is not an option, then means for movement of the fish and decreased nutrient load should be considered. The feed should be stopped to decrease nutrient levels. Cages can be moved to unaffected areas if possible. To reduce concentration, surrounding the fish with perimeter skirts and increased aeration or airlift upwelling can be used. Clay flocculation should be carefully considered based on the area the fish are in and the type of algal blooms present. Most clays can cause damage to benthic fauna present, especially mollusks (Hallegraeff *et al.*, 2017)

1.6 Diseases Associated with Suboptimal Water Quality

Many diseases or conditions have been wholly, or in part, associated with suboptimal water quality. Examples may be found in Chapter 4.

Supplementary materials available online
<ul style="list-style-type: none"> • Bibliographic references. • Further Resources.

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