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REVIEW ARTICLE

Toxicity, physiological, histopathological, handling, growth and antiparasitic effects of the sodium chloride (salt) in the freshwater fish aquaculture

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Abstract

This paper provides the currently available knowledge in the literature regarding the use of sodium chloride (NaCl) in freshwater fish aquaculture and aquaria as related to toxicity, growth performance, transportation, physiology, immunity, histomorphology and antiparasitic treatment. This review assessed and discussed all of these factors, as well as the potential strategies available to be used in fish farming. Acute toxicity to NaCl varies widely among fish species (3.5–150.0 g L^{-1}) depending on some environment factors, and many fish species are sensitive to concentrations near those required for their development, growth or for controlling and treating parasites. Acute exposure to NaCl may lead to mortality in different fish species, cause changes in feeding and swimming behaviour, as well as in growth performance and histomorphology of gills, while sublethal concentrations are less harmful. To reduce stress during transport of freshwater fish, concentrations of 1.0–10.0 g L^{-1} of NaCl in water have been tested for some fish species. Data have shown that survival, body growth of fish, food intake and stimulation of food conversion are affected by the NaCl concentration. Moreover, the effects of NaCl on the immune system, physiology and behaviour must also be considered. Sodium chloride may be a chemotherapeutic for controlling and treating ectoparasite diseases in farmed freshwater fish because of its effectiveness and low cost, but this compound may not be used in high or extreme toxic concentrations, since the maximum tolerance may be near the therapeutic doses. Therefore, this chemical product should be used with parsimony in fish farming.

KEYWORDS
aquaculture, blood, fish, growth, parasites, treatment

1 | INTRODUCTION

Aquaculture has emerged as an important protein source to feed the growing human population, of which this industry should explore potential resources and utilize them for the sustainable production of nutritive foods. The utilization of the resources of aquaculture has become necessary due to the pressure of increasing populations in developing countries. Fish are an important protein source and other important nutrients for human consumption. Aquaculture plays an important role in global efforts towards eliminating hunger and malnutrition since this is supplying aquatic products rich in protein, essential fatty acids, vitamins and minerals. The food fish consumption has increased in recent years, and this increase in per capita fish consumption has been largely attributed to the rapid development of aquaculture. Global aquaculture currently supplies 52% of food fish for direct human consumption (FAO, 2020). Different species of finfish play different roles in food and nutritional security for human consumption around the world. Moreover, some groups of farmed fish are important commodities in international trade. The ornamental fish industry is also particularly important for the economy of various regions. It has been estimated that freshwater aquarists represent 90% of the global market of the ornamental fish industry, which is equivalent to USD$ 4 billion (Puello-Cruz et al., 2010). Aquarium fish is a popular hobby of growing interest worldwide, which has resulted in the expansion of trade of diverse ornamental fish species in more than 125 countries (Dey, 2016). It was estimated that over 2 billion live ornamental fish are transported annually, with trade shown for over 50 exporting countries and over 75 importing countries (Vanderzwalmen et al., 2021). More than 4500 species of freshwater ornamental fish and 1450 species of marine fish were recorded for trade, with a value of US$ 800 million to US$ 30 billion per year (Stevens et al., 2017). Therefore, ornamental fish aquaculture is an important sector in modern society.

Recognizing the capacity of fish aquaculture for further growth and the environmental challenge of this growth demands new sustainable production strategies. Such strategies should include the technical developments regarding feeds, genetic selection, biosecurity and disease controls, with business developments in investment and trade (FAO, 2020), among other technical developments. As a part of sustainable culture, aquaculture is a promising activity when compared with other livestock production industries. However, aquaculture encounters challenges such as inadequate water quality, pond contamination and chronic and acute diseases. Currently, fish welfare during cultivation has become an increasingly relevant issue in various countries, and as a consequence, the importance of animal welfare in the fish aquaculture industry has grown. Animal welfare is particularly applicable to the fish aquaculture industry given the existing relationship between stressful situations and immune system responses in farmed fish (González et al., 2016), which provoke increased rates of parasitic infections and the need for treatments.

Sodium chloride (common salt or NaCl) was among the first nutrients to be identified as essential to life of vertebrate animals and is currently one of the most commonly used therapeutic in fish aquaculture. Sodium (Na^+) is an essential mineral for fish metabolism. Sodium (Na^+) and potassium (K^+) are responsible for the ionic balance in the body fluids of an animal and is also important in the absorption of nutrients in the digestive tract of fish. Sodium has an important role in the intestinal absorption of numerous dietary nutrients, mediated by several transporters that have Na^+/- dependent activity. Hence, NaCl is used in various protocols during the different culture phases of freshwater fish species because this salt is widely available, inexpensive, easy to handle and safer than other chemical products (Carneiro et al., 2005; Fathollahi et al., 2020; Peixoto et al., 2014; Selosse & Rowland, 1990). It has been hypothesized that salinity affects the energy budget in fish. If the salinity is too high or too low in the external environment than the fish body fluid, the fish will spend more energy to regulate osmotic balance (Boeuf & Payan, 2001; Salman, 2009). In addition, salinity also affects fish hormones (McCormick, 2001). It has been established that growth responses in fish adapted to freshwater is different from those adapted to brackish and seawater. Dietary salts in dilute waters may increase the absorption of amino acids and satisfy other metabolic requirements by providing ions that the fish are unable to extract from hypotonic environments for Na^+ and Cl^- homeostasis, and thus, save energy expenditure (Salman, 2009). Fish reared in freshwater may spend more energy than those in seawater for ionic and acid-base regulation. Therefore, this study focused only on the use of NaCl in fish aquaculture because the salinity is defined as the total concentrations of all ions in water or diet of the fish, then it is not just the concentration of NaCl in water. Several ions such as calcium, magnesium, sodium, potassium, bicarbonate, chloride and sulphate contribute to salinity.

NaCl has been used in fish aquaculture for various purposes, including treatment of fungus in fish and eggs (El-Gawad et al., 2016; Khodabandeh & Abtah, 2006; Singhal et al., 1986; Weirich & Tiersch, 1997), bacteria (Akinola & Olakunbi, 2019; Fathollahi et al., 2020), proliferative kidney disease in fish caused by the myxozoan *Tetracapsuloides bryosalmonae* (Enevova et al., 2018), fertilization, reproduction and early developmental stages of fish (Mahrosh et al., 2014; Neves et al., 2019), and in combination with anaesthetics (Davis et al., 1982) because it reduces stress and facilitates osmoregulation (Burgdorf-Moisuk et al., 2011). NaCl has also been used for several other purposes in fish aquaculture. Studies on the effects of salinity in euryhaline fish species have been well discussed (Boeuf & Payan, 2001; Gonzalez, 2012; Kültz, 2015; Salman, 2009) in contrast to studies on stenohaline fish species. The aim of this review was to gather information from published research that focused on the use of NaCl in freshwater fish. Focus was also given to acute toxicity and potential harm to the physiology, immunity, transport, growth, larviculture, histomorphology, control and treatment of ectoparasites in freshwater fish species.

2 | ACUTE TOXICITY OF NaCl IN FRESHWATER FISH SPECIES

Knowledge on the tolerance limits of freshwater fish to NaCl is of ecological significance in assessing fish distribution and their impact on ecosystems, as well as for management practices in fish farms. The most common laboratory measure of NaCl tolerance is the concentration that is lethal to 50% of individuals (LC_{50}) over periods of 24–96 h. Such determinations are experimental and establish a causal link between the salinity and mortality. Some species of freshwater fish tolerate high concentrations of NaCl for longer periods when compared with other freshwater fish species (Table 1), and concentrations may be near the lethal concentration for the fish species. However, they lack consideration of other changes in the water quality that may occur with an increased salinity. Thus, it is necessary to know beforehand the mean lethal concentration (LC_{50}) for each species of fish as well as the environmental conditions for exposure to NaCl.

TABLE 1 Lethal concentrations ($LC_{50\text{-}96\,h}$) of NaCl for different freshwater fish species

Fish species	LC (g L^{-1})	References
Trichogaster lalius	3.5	Zuanon et al. (2015)
Trichogaster labiosa	4.1	Zuanon et al. (2015)
Pethia ticto	6.1	Dubey et al. (2014)
Amblypharyngodon mola	6.2	Dubey et al. (2014)
Betta splendens	7.1	Fabregat et al. (2017)
Betta splendens	11.9	Zuanon et al. (2009)
Cyprinus carpio	7.7–10.6	Mubarik et al. (2018)
Acipenser persicus	7.7	Fathollahi et al. (2020)
Lophiosilurus alexandri	8.9	Luz and Santos (2008a)
Pterygoplichthys spp.	10.6	Brion et al. (2013)
Metynnis orinocensis	10.5–10.8	Velasco-Santamaría |and Cruz-Casallas (2008)
Danio rerio	10.4	Dolezelova et al. (2009)
Rhamdia quelen	10.8	Zeni et al. (2017)
Metynnis maculatus	11.4	Zeni et al. (2017)
Gambusia holbrooki	11.5	Newman and Aplin (1992)
Channa punctata	13.0	Dubey et al. (2016)
Barbodes gonionotus	13.9	Akther et al. (2009)
Pterophyllum scalare	14.2	Moreira et al. (2011)
Xiphophorus maculatus	16.5	Valente et al. (2021)
Lepomis gibbosus	21.3	Venâncio et al. (2019)
Poecilia reticulata	21.7	Dolezelova et al. (2009)
Pylodictis olivaris	100.0	Bringolf et al. (2005)
Ctenopharyngodon idella	151.0	Maceina and Shireman (1979)

The acute toxicity of NaCl has been determined for some freshwater fish species (Table 1). Toxic effects of NaCl include mortality, which is directly proportional to the concentration because the tolerance to this chemical varies among fish depending on the species, temperature and other environmental conditions (Al-Dahaman & Atti, 1977; Burgdorf-Moisuk et al., 2011; Fontaínhas-Fernandes et al., 2000; Ghosh & Pal, 1969; Grizzle & Mauldin II, 1995; Hoque et al., 2020; Piazza et al., 2006; Zeni et al., 2017).

For *Calla catla*, *Labeo rohita* and *Cirrhinus mrigala*, the $LC_{50\text{-}48\,h}$ of NaCl was of 6.0 g L^{-1} (Ghosh & Pal, 1969). For *Carassius auratus*, the $LC_{50\text{-}200\,h}$ of NaCl was of 7.3 g L^{-1} and for *Pimephales promela* was of 7.6 g L^{-1} (Alderman et al., 1976). The $LC_{50\text{-}24\,h}$ for the yolk sac and fry of *Clarias gariepinus* were 0.61 and 0.7 g L^{-1} of NaCl respectively (Magondu et al., 2011). For larvae of *Morone saxatilis*, the $LC_{50\text{-}24\,h}$ was of 1.6 g L^{-1} of NaCl and for juveniles varied between 1.4 and 18.2 g L^{-1} (Grizzle & Mauldin II, 1995), and for *Cyprinus carpio*, the $LC_{50\text{-}24\,h}$ was 10.9 g L^{-1} (Al-Mahmood & Farhan, 2020).

Carassius auratus tolerated a therapeutic bath of 12 h with 5 and 10 g L^{-1} of NaCl (Burgdorf-Moisuk et al., 2011). *Cyprinus carpio* tolerated baths with 2–4 g L^{-1} of NaCl (Kadhim & Al-Faragi, 2018). *Poecilia reticulata* tolerated a therapeutic bath of 24 h with 10 g L^{-1} of NaCl (Andrade et al., 2005). *Rhamdia quelen* tolerated 8.0 g L^{-1} of NaCl during 30 days of exposure (Camargo et al., 2006). *Leporinus macrocephalus* tolerated an exposure of up to 4.0 g L^{-1} of NaCl (Jomori et al., 2013).

Therapeutic baths of *Oncorhynchus tshawytscha* of 0.005–0.01 g L^{-1} showed 100% survival (Long et al., 1977). Similarly, *R. quelen* in baths for 30 days with 2.0–8.0 g L^{-1} of NaCl showed 100% survival (Carmargo et al., 2006). However, in *Gambusia affinis*, exposure of 96 h to 15.0 g L^{-1} of NaCl caused 50% of mortality, and in *Heteropneustes fossilis* exposure of 46 h caused 100% mortality (Al-Dahaman & Atti, 1977). Concentrations of 20.0 g L^{-1} showed no mortality of *G. affinis* for the first 8 h, but was 23% mortality at 24 h, 85% at 48 h and at 72 h was 100%. *Heteropneustes fossilis* showed mortality of 33.0% at 8 h with 15.0 g L^{-1} of NaCl, 93% at 24 h and 100% at 48 h (Al-Daham & Bhatti, 1977). Exposure to 3.0 and 5.0 g L^{-1} of NaCl for 5 and 1 min, respectively, caused 5% mortality in *Silurus glanis* (Krasteva et al., 2020). *Lophiosilurus alexandri* exposed to 7.5 and 10.0 mg L^{-1} of NaCl for 28 days showed 78.0 and 100% mortality respectively (Takata et al., 2021).

Piaractus mesopotamicus kept at 30, 40 and 50 g L^{-1} of NaCl showed 66, 100 and 100% of mortality at 15, 30 and 45 min of exposure respectively. However, 30, 40 and 50 g L^{-1} of NaCl for 4, 8 and 12 min showed no mortality when the fish were transferred to freshwater (Barbosa-Junior et al., 2010). In *Astyanax altiparanae* exposed to 3–15 g L^{-1} of NaCl for 96 h, fish mortality was 0% at the levels of 3.0 and 6.0 g L^{-1}, 75% at 9.0 g L^{-1} and 100% at 12.0 and 15.0 g L^{-1} (Salaro et al., 2015). *Danio rerio* exposed to 1 and 3 g L^{-1} of NaCl showed mortality of 18–30%, whereas the survival of *Gymnocorymbus ternetzi*, *Hemigrammus caudovittatus* and *Trichogaster trichopterus* was unaffected by these concentrations of NaCl (Rothen et al., 2002). These results indicate that *D. rerio* is more sensitive to NaCl than other fish species.

Clinical signs of NaCl toxicity include mortality, alteration in eye colour, erratic swimming, hyperactivity, increased opercular activity, jumping out of the test media, spasms, convulsions, swimming more actively, loss of equilibrium and swimming at the surface of water to obtain oxygen and oedema (Alderman et al., 1976; Al-Daham & Bhatti, 1977; Andrade et al., 2005; Akther et al., 2009; Al-Taee & Al-Hamdani, 2014; Burgdorf-Moisuk et al., 2011; Brion et al., 2013; Dewi et al., 2018; Demska-Zakęs et al., 2021; Mubarik et al., 2018; Parween et al., 2015; Velasco-Santamaría & Cruz-Casallas, 2008; Zeni et al.,. 2017). Therefore, fish farmers should verify these clinical signals to avoid losses of fish when using NaCl.

In aquaculture fish, in general, effective chemotherapeutic agents used are limited in their clinical use by the toxic effects. The ratio of the concentrations that the therapeutic effect and toxicity occur is referred to as the therapeutic index. Hence, it is needed to have at his disposal drugs that have a higher therapeutic index, that is, maximum therapeutic benefit with little or no toxicity for fish.

However, since this ideal situation is not achieved, we must remain cognizant of the toxicity of chemotherapeutic drugs as NaCl so that this agent can be used as effectively as possible. NaCl toxicity varies with water quality, mainly the electrical conductivity that increase their values of exposure (Camargo et al., 2006; Mubarik et al., 2018; Silva et al., 2009; Souza et al., 2006). For *M. saxatilis*, the lethal ratio of NaCl varied depending on the Ca^{2+} concentration in water (Grizzle & Mauldin II, 1995).

In *Poecillia reticulata* submitted to temperature stress tests, there is a beneficial effect of adding NaCl at concentrations of 2.0–5.0 g L^{-1}. The mortality was 43% without the addition of salt and, with the use of NaCl, there was a decrease in the mortality of 19% (1 g L^{-1}), 24% (3.0 g L^{-1}) and 28% (5 g L^{-1}) (Peixoto et al., 2014). Therefore, the addition of NaCl reduces stress in fish and this product is easily available for fish farmers. The use of NaCl represents an economically viable and safe option for many farms that produce food fish and ornamental fish farms but should be used with caution in certain stenohaline freshwater fish species.

3 | CONCENTRATIONS OF NaCl DURING AND POST-HANDLING PROCEDURES IN THE FRESHWATER FISH FARMING

Various fish farming operations involve transportation of fish from one facility to another for restocking practices, from a hatchery to ponds or from ponds to ponds, which frequently results in high mortality in fish farming. Transportation, confinement and handling are known to cause stress in fish and are related to a number of physiological responses such as the release of catecholamines and corticosteroids as well as increase in the plasma glucose and blood cortisol levels (Barton & Peter, 1982; Biswal et al., 2021; Brandão et al., 2008; Burgdorf-Moisuk et al., 2011; Burgdorf-Moisuk et al., 2011; Carneiro & Urbinatti, 2001; Gomes et al., 2003, 2006b; Mirghaed & Ghelichpour, 2019; Ramírez-Duarte et al., 2011). Fish respond to stressors in a proportional way that reflects the severity and duration of the stress (Gomes et al., 2006b; Mirghaed & Ghelichpour, 2019). Physiological mechanisms responsible for adapting to a stressor include nervous, immunological and hormonal mechanisms. The immediate mortality associated with transport stress is presumably blood ion disturbances. Therefore, the greatest challenge with any transport of live fish is to minimize the stress on the fish. In general, 1.0–10.0 g L^{-1} of NaCl solutions have been used to reduce stress during transportation of freshwater fish, and of 2.0–20.0 g L^{-1} in larviculture (Table 2). However, tilapia species have a high tolerance to salinity due to their adaptation evolutive (Kang'ombe and Brown, 2008).

Live transportation of fish is a major source of stress and can lead to poor welfare and mortality within the fish aquaculture industry (Vanderzwalmen et al., 2021). Transportation stress and loss of mucus increase salt losses from the blood, placing higher energy demands on fish. Excessive loss of salts can cause heart failure as well as nerve and muscle spasms (tetany). NaCl may be used for reducing the stress in freshwater fish, serving as an osmoregulatory agent in the mitigation of stress (Burgdorf-Moisuk et al., 2011; Camargo et al., 2006). Depending on the concentration, the addition of NaCl limits or prevents the loss of salts during transportation (Gomes et al., 1999; Wurts, 1995). For *Cyprinus carpio*, 3.0 g L^{-1} of NaCl mitigated stress responses and inhibited changes in cortisol, glucose and calcium levels, and decreased serum lactate, sodium, chloride, lysozyme, alternative complement (ACH_{50}) and total immunoglobulins, thus helping in hydromineral disturbance and immunosuppression after 5 h of transportation (Mirghaed & Ghelichpour, 2019). Transportation of *Salmo trutta* in water without NaCl caused increases in blood haemoglobin and glycaemia concentrations and decreases in haemoglobin concentration, plasma osmolality, muscle fat and liver glycogen contents. Although these blood alterations are present, they were markedly lower in the fish transported in 6.0 g L^{-1} of NaCl (Nikinmaa et al., 1983). Therefore, addition of NaCl in the transportation water of fish or after handling in fish farming can reduce the osmotic changes between the external environment and the plasma of the fish, reducing the stress in various fish species.

The use of 5.0 g L^{-1} NaCl during 4 h of transport was stressful for *Oncorhynchus mykiss* (Barton & Peter, 1982). Favero et al. (2019) demonstrated that the use of 2.0–8.0 g L^{-1} of NaCl in water during transport of juvenile *Lophiosilurus alexandri* in plastic bags was ineffective to mitigate stress. *Colossoma macropomum* transported during 24 h in plastic bags with 1.0–3.0 g L^{-1} of NaCl had low survival (Gomes et al., 2006a). *Rhamdia quelen* transported in water containing 1–6 g L^{-1} of NaCl had 100% survival (Gomes et al., 1999).

Morone saxatilis in confinement exposed to 4.0–5.0 g L^{-1} of NaCl had a survival of 84.8%. Moreover, the addition of NaCl to water in these concentrations increased the survival of *M. saxatilis* larvae during 24 h of confinement after transport. Larvae exposed to 4.0–5.0 g L^{-1} of NaCl during 24 h of confinement after transport showed no difference in survival (Grizzle et al., 1992). *R. quelen* exposed to 8.0–12.0 g L^{-1} of NaCl had no effects on the serum cortisol, potassium and sodium levels, plasma glucose levels and glycogen after transportation (Rosa et al., 2019), indicating a mitigation of stress. Transportation of *Brycon amazonicus* for 4 h in water with 0.1–0.6 g L^{-1} of NaCl showed no changes with the levels of plasma cortisol, glucose, total protein and haemoglobin, haematocrit and erythrocytes values. Moreover, lipid levels in liver and muscles were maintained (Urbinati & Carneiro, 2006), indicating a conservation of energy.

4 | HISTOMORPHOLOGICAL ALTERATIONS ON GILLS OF FRESHWATER FISH EXPOSED TO NaCl

Salinity is an abiotic factor that influences water quality, fish development and physiological status (Takata et al., 2021). Freshwater fish maintain their body fluids hyperosmotic in relation to the external environment. To solve this difference in osmotic concentration,

TABLE 2 Recommendation of NaCl concentrations in procedures of fish farming

Fish species	Procedures	Concentrations (g L^{-1})	Time	References
Brycon amazonicus	Transportation	6.0	4 h	Carneiro and Urbinatti (2001), Urbinati and Carneiro (2006)
Colossoma macropomum	Transportation	8.0	1.5–3 h	Gomes et al. (2003)
Colossoma macropomum	Transportation	2.0	14 h	Anjos et al. (2011)
Labeo rohita	Transportation	4.0	168 h	Biswal et al. (2021)
Oreochromis niloticus	Transportation	4.0–8.0	5 h	Oliveira et al. (2009)
Oreochromis niloticus	Transportation	5.0–10.0	5 h	Bizarro et al. (2018)
Oreochromis niloticus	Transportation	7.0	4 h	Uehara et al. (2021)
Brycon amazonicus	Transportation	6.0	4 h	Carneiro et al. (2002)
Oncorhynchus mykiss	Transportation	5.0	5 h	Tacchi et al. (2015)
Arapaima gigas	Transportation	5.0	3 h	Gomes et al. (2006b)
Arapaima gigas	Transportation	3.0	5 h	Souza et al. (2006)
Arapaima gigas	Transportation	3.0–6.0	48 h	Brandão et al. (2008)
Labeo capensis	Transportation	7.0	1–3 days	Coetzee and Hattingh (1977)
Cyprinus carpio	Transportation	3.0	5 h	Mirghaed and Ghelichpour (2019)
Morone saxatilis	Transportation	1.0	5 h	Mazik et al. (1991)
Alosa sapidissima	Transportation	10.0	—	Murai et al. (1979)
Ancistrus triradiatus	Transportation	1.0	48 h	Ramírez-Duarte et al. (2011)
Astyanax altiparanae	Transportation	6.0	—	Salaro et al. (2015)
Ancistrus triradiatus	Confinement	1.0	48 h	Ramírez-Duarte et al. (2013)
Oreochromis niloticus	Larviculture	2.0	28 days	Luz et al. (2012)
Oreochromis niloticus	Larviculture	2.0	10–20 days	Luz et al. (2013)
Pseudoplatystoma corruscans	Larviculture	2.0	5 days	Santos and Luz (2009)
Pseudoplatystoma corruscans	Larviculture	17.0	—	Beux and Zaniboni-Filho (2007)
Prochilodus costatus	Larviculture	2.0	5 days	Santos and Luz (2009)
Rhinelepis aspera	Larviculture	6.0	7 days	Luz and Santos (2010)
Betta splendens	Larviculture	2.0	96 h	Dias et al. (2016)
Bycon amazonicus	Larviculture	2.0	5 days	Jomori et al. (2013)
Bycon amazonicus	Larviculture	20.0	15 days	Luz et al. (2004)
Colossoma macropomum	Larviculture	2.0	10 days	Jomori et al. (2013)
Colossoma macropomum	Larviculture	2.0	10 days	Santos et al. (2021)
Astronotus ocellatus	Larviculture	2.0	10 days	Jomori et al. (2013)
Leporinus macrocephalus	Larviculture	2.0	12 days	Jomori et al. (2013)
Lophiosilurus alexandri	Larviculture	2.0	10 days	Luz and Santos (2008b), Cordeiro et al. (2015)
Hypsolebias radiseriatus	Larviculture	2.0	12 days	Araújo et al. (2021)
Hoplias lacerdae	Feed training	4.0–5.1	—	Salaro et al. (2012)

water enters through osmosis and a passive loss of ions occurs, producing diluted urine and absorption of monovalent ions through the gills. The main organs that participate in this regulation are the gills, kidney and intestine (Camargo et al., 2006). Hence, the gills of fish show several structural characteristics that are related to salt absorption.

Fish gills have multiple physiological functions. They are the predominant site for osmotic and ionic regulation, acid-base regulation, the removal of nitrogenous wastes and are the major site for gas exchange. The entire cardiac output perfuses the gill vasculature before re-entering the circulation system. The afferent arteriole feeds the posterior side of each filament and supplies the lamellae, which are the site of gas exchange. The oxygenated blood is carried by the efferent arterioles, which supply a central sinus and filamental vessels with blood that supplies the ion-transporting cells of the filament epithelium (Laurent et al., 1985;

Salman, 2009; Salman & Eddy, 1987; Sathorn et al., 2021). Thus, the gills of freshwater teleost fish are the major site of ion uptake in a dilute environment.

The gill epithelium in teleost fish contains several cell types that play an important role in ionic regulation. The branchial epithelium in marine fish contains at least four cell types, including chloride or mitochondria-rich cells that are responsible for salt secretion. Similar cell types are found in freshwater fish, although their chloride cells lack interdigitation and leaky junctions, which characterize the chloride cells of seawater fish and its role in salt absorption has been structurally implicated. The chloride cells of the gill epithelium are the major site for osmoregulation of salt uptake in freshwater fish (Laurent et al., 1985; Salman & Eddy, 1987). *Oncorhynchus mykiss* supplemented with 100 g of NaCl in the diet had increase in chloride cells number (Trombetti et al., 1996). Therefore, the fish gills respond to exposure to NaCl (Table 3).

5 | PHYSIOLOGICAL AND IMMUNE ALTERATIONS CAUSED BY NaCl IN EXPOSED FRESHWATER FISH

Blood parameters have been considered an important measurement for the health status of fish. Fish exposed to NaCl may suffer physiological changes to preserve the consistence and body stability. An increase in the haematocrit may result from losing too much water because of the difference between the internal and external environmental ion concentrations, which led to water loss as a result of osmotic pressure, thus increasing the haematocrit (Al-Hilali & Al-Khshali, 2016; O'Neal et al., 2006). Moreover, the number of erythrocytes and leukocytes may also vary in response to stressful situations such as exposure to NaCl (Table 4). This suggests that fish exposed to NaCl attempt to adjust their physiology to restore homeostasis. Osmotic sensitivity and salinity in fish

TABLE 3 Histopathological effects of exposure to NaCl on gills of different freshwater fish species

Fish species	Dose ($g\ L^{-1}$)	Exposure	Tissue alterations	References
Rhamdia quelen	10.0	120 h	Hyperaemia and epithelial hyperplasia in secondary lamellae	Carneiro et al. (2006)
Oreochromis niloticus	8.0	160 h	Transmission electron microscopy of gills revealed mitochondria-rich cells in fish of water with NaCl, with mitochondria developed and tubular system arising from the basolateral membrane.	Fontaínhas-Fernandes et al. (2001)
Oreochromis niloticus	10.0–15.0	5–10 days	Adhesion of the secondary lamellae accompanied by a marked increase in the number of mitochondria-rich cells	Mohamed et al. (2021)
Labeo rohita	8.0	48 h	Mild lesions on gills	Murmu et al. (2020)
Oncorhynchus mykiss	8.0–12.0	—	Increase in chloride cells	Salman and Eddy (1987)
Metynnis orinocensis	5.0–40.0	96 h	Lamellar congestion, hyperplasia and fusion, and gill congestion severity increased with salt concentration	Velasco-Santamaría and Cruz-Casallas (2008)
Rhamdia quelen	9.0–13.8	96 h	Pigment accumulation, vascular congestion, epithelial lifting, oedema, epithelial desquamation, lamellae under regeneration, lamellar fusion and epithelial hyperplasia and hypertrophy	Zeni et al. (2017)
Metynnis maculatus	9.0–13.8	96 h	Pigment accumulation, aneurism vascular congestion, epithelial lifting, oedema, lamellar fusion and epithelial hyperplasia and hypertrophy	Zeni et al. (2017)
Poecilia mexicana	10.0–50.0		Marked difference in overall appearance, and the number of chloride cells was dramatically increased as salinity increased	Sathorn et al. (2021)
Acipenser persicus	6.3–10.2	96 h	Haemorrhage, elongation of secondary lamellae, adhesion of secondary lamellae, hypertrophy of supporter cartilage, mucus coagulation and secretion, hyperplasia, lamellar necrosis and clubbing of gill lamellae	Fathollahi et al. (2020)
Lophiosilurus alexan	2.5–10.0	28 days	Vascular congestion, hyperplasia of gill filament epithelium, lamellar fusion, increase in mucosal cell hyperplasia, loss of structural integrity of the pillar cells and chloride cell	Takata et al. (2021)

TABLE 4 Haematological, biochemical and immunological effects of NaCl for different freshwater fish species

Fish species	Doses (g L^{-1} or g k^{-1})–Application	Alterations	References
Oreochromis niloticus	60.0 (bath)	Increase in plasma glucose levels, uric acid, creatinine, erythrocytes, lymphocytes and neutrophils number. Decrease in plasma alanine aminotransferase (ALT) levels	Elrahman et al. (2016)
Oreochromis niloticus	1.5 (bath)	Increase in plasma glucose levels	Abdelrhman et al. (2020)
Oreochromis niloticus	8.8–12.0 (bath)	Decrease in serum superoxide dismutase, serum catalase, total protein, erythrocytes number, haemoglobin and haematocrit, and increase in serum potassium and thrombocytes number	Elarabany et al. (2017)
Oreochromis niloticus	80.0 (diet)	Increase in osmolality, chloride and cortisol levels, and gill Na^+, K^+-ATPase activity	Fontaínhas-Fernandes et al. (2001)
Oreochromis niloticus	1.5–3.0 (bath)	Increase in serum total protein levels, haematocrit and osmolality	O'Neal et al. (2006)
Oreochromis niloticus	10.0–15.0 (bath)	Increase in plasma cortisol, sodium, potassium, calcium, triiodothyronine, malondialdehyde and haemoglobin levels and haematocrit, and decrease in plasma lactate levels	Mohamed et al. (2021)
Cyprinus carpio	5.0–15.0 (bath)	Increase in haematocrit, haemoglobin, number of erythrocytes and leukocytes	Al-Hilali and Al-Khshali (2016)
Cyprinus carpio	1.0 (bath)	Decrease in haematocrit, haemoglobin, serum alanine aminotransferase and creatinine phosphokinase levels	Al-Taee and Al-Hamdani (2014)
Cyprinus carpio	3.0 (bath)	Increase in total erythrocytes number, haematocrit, MCV and total protein, and decrease in haemoglobin and MCHC	Mubarik et al. (2018)
Cyprinus carpio	6.0–12.0 (bath)	Increase in plasma sodium and chloride levels, and decrease in urine formation rate	Salati et al. (2011)
Colossoma macropomum	2.0 (bath)	Decrease in plasma glucose levels	Anjos et al. (2011)
Carassius auratus	5.0–10.0 (bath)	Increase in plasma glucose, sodium, chloride and alanine aminotransferase, and decrease in potassium	Burgdorf-Moisuk et al. (2011)
Colossoma macropomum	2.0–4.0 (bath)	Increase in plasma glucose and chloride levels	Chagas et al. (2012)
Morone saxatilis	10.0 (bath)	Increase in plasma corticosteroids levels	Davis et al. (1982)
Ictalurus punctatus	10.0–12.0 (bath)	Increase in plasma sodium and chloride levels, and decrease in haematocrit	Davis and Simco (1976)
Sander lucioperca	10.0–20.0 (bath)	Increase in total leukocytes and erythrocytes number,	Demska-Zakęs et al. (2021)
Clarias gariepinus x *C. macrocephalus*	1.0 (bath)	Increase in serum cortisol, plasma sodium and chloride levels	Koeypudsa and Jongjareanjai (2011)
Pseudoplatystoma spp.	20.0 (bath)	Increase in haematocrit, total erythrocytes and lymphocytes number	Rodrigues et al. (2019)
Labeo rohita	8.0 (bath)	Increase in total erythrocytes and thrombocytes number and haematocrit, and decrease in haemoglobin	Murmu et al. (2020)
Rhamdia quelen	12.0 (bath)	Increase in serum calcium levels and decrease in serum chloride levels	Rosa et al. (2019)
Rhamdia quelen	25.0 (bath)	Increase in glucose and sodium levels	Souza-Bastos and Freire (2009)
	8.0–12.0 (bath)	Increase in Na^+/K^+ ATPase activity	Salman and Eddy (1987)
Odontesthes bonariensis	30.0 (bath)	Increase in osmolarity, levels of sodium, chloride and cortisol	Tsuzuki et al. (2000)
Odontesthes bonariensis	0.5 (bath)	Increase in cortisol, and decrease in haematocrit, glucose, chloride and sodium levels	Tsuzuki et al. (2001)

(Continues)

TABLE 4 (Continued)

Fish species	Doses (g L^{-1} or g k^{-1})–Application	Alterations	References
Odontesthes hatcheri	30.0 (bath)	Increase in osmolarity, levels of sodium and chloride, and decrease in cortisol levels	Tsuzuki et al. (2000)
Catostomus commersoni	9.0 (baht)	Increase in plasma osmolarity, chloride and sodium levels, and decrease in plasma lactate and total protein	Walker et al. (1989)
Lophiosilurus alexandri	10.0 (baht)	Decrease in levels of plasma alkaline phosphatase, total protein and number of erythrocytes and leukocytes, and increase in levels of cortisol, glucose and aspartate aminotransferase	Mattioli et al. (2020)
Carassius auratus	15.0 (bath)	Increase in cortisol and glucose levels	Tarkhani and Imanpoor (2012)
Xiphophorus maculatus	10.0–15.0 (bath)	Decrease in glucose levels	Valente et al. (2021)

detection are of the highest physiological interest. Fish have prolactin cells that are osmosensitive, in addition to chemoreceptors situated in the pseudobranch providing information on water salts (Boeuf & Payan, 2001).

Environmental salinity is a key factor for the survival of freshwater fish that are hyperosmotic. Any changes in salinity levels may affect various physiological processes of the metabolism of the animals. The involvement of Na^+, K^+-ATPase in many ion transport systems is well understood. Na^+, K^+ -ATPase activity is present in the crude homogenates of gills from seawater and freshwater teleost fish. When fish are moved from freshwater to a higher salinity, the blood becomes temporarily more concentrated, causing cell shrinkage and regulatory volume increase by uptake of solutes, mostly N^+ and Cl^- by rapid activation of salt uptake by the $N^+/K^+/2Cl^-$ cotransporter and slower organic osmolytes uptake or synthesis. These cell volume responses are limited and only help the fish cope temporarily with salinity changes until the main osmoregulatory systems respond to the new environmental conditions. Thus, blood Na^+ and Cl^- concentrations are important indicators of effective ion transfer through the gills. Changes in the concentrations of these electrolytes in intercellular space can be viewed as the first response of fish to a disturbance in respiratory regulation and disruption in the acid-base balance of the body (Demska-Zakęs et al., 2021; González et al., 2016; Perry & Gilmour, 2006).

Fish blood is brought into close contact with the environment as it flows through the small blood capillaries of the gills and skin surface. Salts diffuse from areas of high concentration in blood to areas of low concentration in freshwater. Therefore, the salts sodium and chloride are slowly and continuously lost to the environment. The gills and skin of fish are coated with a thin layer of mucus that helps reduce the loss of salts to the surrounding fresh water. Lost salts are replaced by reabsorbing them from the water or during food consumption. Body energy is used to replace lost salts (Wurts, 1995). The gills, kidney and intestine are the major osmoregulatory organs and play a role in hydromineral homeostasis, but the gills are the main site in ion exchange.

Rhamdia quelen exposed for 30 days to 2.0–8.0 g L^{-1} of NaCl showed no alterations in haemoglobin, haematocrit, VCM, CHCM, number of erythrocytes and perceptual of leukocytes and thrombocytes (Camargo et al., 2006). Copatti et al. (2011) suggested that dietary NaCl supplementation of 0.5–2.0% protects against the impact of acidic water. However, based on the results of net Na^+ fluxes, dietary NaCl supplementation of 1.0–2.0% may be used because the Na^+ imbalance is less pronounced. Immersion of *Sander lucioperca* in water with 10.0–20.0 g L^{-1} of NaCl had no alterations in haemoglobin, haematocrit and plasma levels of sodium, chloride, potassium, glucose, lactate, total protein, albumin and globulin (Demska-Zakęs et al., 2021). *Oreochromis niloticus* supplemented with 60.0 g of NaCl in diet for 6 weeks showed no alterations in plasma blood glucose, serum osmolality, serum cortisol and haematocrit (Lim et al., 2006). *Peckoltia oligospila* exposed to 15.0 g L^{-1} of NaCl for 15 min showed no alterations in levels of plasma glucose and total protein, haemoglobin, haematocrit, mean corpuscular volume (MCV), mean corpuscular haemoglobin concentration (MCHC), number of erythrocytes, leucocytes and thrombocytes, perceptual of lymphocytes, neutrophils and monocytes (Santos et al., 2020). Therefore, the physiological response of freshwater fish to NaCl may vary according to the species and concentrations used.

For *Salvelinus fontinalis*, it was shown that with between 0.9 and 1.8 g of NaCl kg^{-1} of body weight, the ability of body to handle excessive amounts of salt depends on the concentration in the intestinal tract. When the entrance of salt becomes too rapid, blood chlorides increase. Absorbed salt is readily excreted at lower levels. Large doses of NaCl result in a high increase in blood chloride, which requires a longer period after feeding to reach a normal level again. A 30 min bath of *S. fontinalis* with 30.0 g L^{-1} of NaCl or 10 min bath with 50.0 g L^{-1} caused an increase in blood salinity that quickly returned to normal after the fish were removed to fresh water. A 60 min bath with 30 g L^{-1} resulted in an extremely high chloride levels of the blood that required about 48 h to return to normal. A 15 min bath with 50 g L^{-1} resulted in the loss of the majority of the fish. Those alive after 48 h still had a high chloride level of the blood which was showing a tendency to reach a normal level (Phillips Jr., 1947).

Therefore, NaCl concentrations for freshwater fish should be used with caution to avoid causing extreme physiological disturbance.

6 | GROWTH PERFORMANCE ALTERATIONS OF FRESHWATER AND MARINE FISH EXPOSED TO NaCl

Development and growth in teleost fish are similar among fish species and are influenced by environmental factors. In fish, the continuous development and growth are controlled by 'internal factors', including central nervous, endocrinological and neuroendocrinological systems (Boeuf & Payan, 2001). Freshwater fish are hyperosmotic to the surrounding environment and present physiological problems of solute loss. To compensate for this loss, they resort to active uptake of salt ions from the environment. Diet is an important source of salt that can satisfy the osmoregulatory requirements of the fish and supplemental salt spares energy used in osmoregulation, thereby leaving more energy available for growth (Gangadhara et al., 2004; Gatlin et al., 1992). The dietary requirements for sodium and chloride in freshwater fish can be determined according to the amount needed for growth. Hence, these requirements of NaCl in freshwater fish have received the attention of various studies (Table 5).

Information on the effects of dietary supplementation and exposure to NaCl for stenohaline fish species has been reported. In general, fish survival, development and growth may be influenced by this mineral. Dietary salt has improved growth of stenohaline fish species raised at different concentrations, but some results have been controversial (Table 4). Nevertheless, Boeuf and Payan (2001) hypothesized that marine fish species present the best development at lower salinity, while freshwater fish present the best development at a higher salinity.

The explanation of such growth enhancement potential for dietary supplementation of NaCl or baths in stenohaline fish may be related to the osmoregulatory advantages. Ion losses in salinity can impair growth if not compensated by sodium and chloride ions. Hence, supplementation of diet and/or baths with moderate levels of NaCl may also spare energy used in osmoregulation, thereby leaving more energy available for growth. Several studies have shown that 20 to >50% of the total fish energy budget is dedicated to osmoregulation. However, recent studies indicate that the osmotic cost may be near 10%. The salinity affects the energy budget in fish. If salinity is too high or too low in the external environment than the fish body fluid, the fish spends more energy to regulate osmotic balance. Therefore, less energy remains for growth in these environments because of the high consumption of energy for active ion transport. Fish use nearly 10% of total energy for osmoregulation (Boeuf & Payan, 2001). Salinity affects fish hormonal activity as well. The hormones of gonadotropin, cortisol, insulin-like growth factor-1 and thyroid hormones play a role in osmotic regulation (McCormick, 2001) as well as thyroid hormones levels (Fontaínhas-Fernandes et al., 2000). *Labeo rohita*, *Cirrhinus mrigala* and *Cyprinus carpio* received diets containing 5.0–15.0 g L^{-1} and showed increases in levels of protease, amylase and lipase in both the intestine and hepatopancreas (Keshavanath et al., 2003). Dietary supplementation with 5.0–20.0 g of NaCl increased the digestibility of dry matter and protein in *Colossoma macropomum* (Keshavanath et al., 2012).

Feeding of dietary NaCl with 10.0 g L^{-1} enhanced the survival rates of *Oreochromis mossambicus* up to 84%, and the *Oreochromis aureus* x *O. niloticus* hybrid and *Oreochromis spilurus* up to 62% and 50% respectively. Best survival rates occurred after 2 weeks of feeding for *O. mossambicus* and *O. aureus* x *O. niloticus* hybrid, whereas in *O. spilurus* best survival was achieved at 3 weeks (Al-Amoudi, 1987). Tilapias are freshwater fish but are believed to have evolved from marine ancestors; thus, most tilapia species are able to tolerate a wide range of salinity. *Oncorhynchus mykiss* supplemented with 100 g of NaCl showed no changes in body weight, condition factor and survival (Trombetti et al., 1996). Feeding of *Brycon amazonicus* with *Artemia* and/or artificial feed during exposure to 20.0 g L^{-1} of NaCl had no influence on the weight and length (Luz et al., 2004). *Ictalurus punctatus* subjected to baths with 15.0 g L^{-1} of NaCl had 100% survival, whereas 30.0 g L^{-1} caused 40% mortality (Hubert & Warner, 1975). However, *O. niloticus* supplemented with 60 g of NaCl in the diet for 6 weeks showed no alterations in survival, weight gain, feed intake and protein efficiency ratio (Lim et al., 2006). Similarly, feed supplemented with 0.2–0.6 g of NaCl for *Cyprinus carpio* over for 28 days had no influence on survival (Singh et al., 2019).

Ictalurus punctatus were fed diets supplemented with NaCl at 0, 10, 20 or 40 g kg^{-1} for 10 weeks and exposed to nitrite. Mortality of fish from nitrite exposure tended to decrease with increasing NaCl in the diet at 6 weeks and was lower in the 40 g kg^{-1} (Welker et al., 2011). *Colossoma macropomum* that received diet supplemented with 80.0 g of NaCl, and fish that received this diet and were submitted to abrupt change from freshwater to brackish water of 15, 20 and 25% presented variations in tolerance and body growth. Growth was better in the second condition of the assay in relation to first condition. The estimated values of abrupt salinity change (LS_{50}) at 12 h was 11.0 in the first condition and 18.0 in the second condition. The estimated value of time of survival to the salinity (LT_{50}) in the first condition was 16 h, while in the second condition was 30 h (Carraro et al., 2007). Dietary supplementation of NaCl provoked a transitory increase in whole-body Na^+, K^+ and Cl in *R. quelen* fingerlings, but the best stabilization of these ion levels was observed in specimens exposed to salt in the water (Garcia et al., 2007; Garcia, Becker, Copatti, Baldisserotto, & Radünz-Neto, 2007).

Exposure of fish to different environmental salinities in later developmental stages, juveniles and adults is suggested to induce changes in osmoregulation and metabolism, which can affect fish survival and growth (Boeuf & Payan, 2001; Takata et al., 2021). However, the incorporation of NaCl in freshwater fish feed has been explored with various results (Table 5). The growth performance and health status of fish are strongly associated with the environmental conditions in the rearing ponds, including salinity, which can lead to acute or chronic stress. Stress is a general and non-specific response to any factor disturbing homeostasis.

TABLE 5 Body growth performance effects for different freshwater species after feeding or exposure to NaCl

Fish species	Doses ($g\ L^{-1}$)–application	Alterations	References
Morone saxatilis	1.0 (bath)	Increase in survival	Mazik et al. (1991)
Morone saxatilis	4.0–5.0 (bath)	Increase in survival	Grizzle et al. (1992)
Brycon amazonicus	20.0 (bath)	Decrease in survival	Luz et al. (2004)
Labeo rohita	0.002 (bath)	Increase in survival	Murmu et al. (2020)
Clarias gariepinus	50.0 (bath)	Increase in survival	Pruszynski et al. (1997)
Lophiosilurus alexandri	8.0–10.0 (diet)	Decrease in survival	Luz and Santos (2008a)
Lophiosilurus alexandri	40.0 (diet)	Decrease in weight and specific growth rate	Santos and Luz (2009)
Lophiosilurus alexandri	4.0 (bath)	Decrease in weight and specific growth rate	Santos and Luz (2009)
Icralurus punctatus	1.0 (bath)	Increase in weight gain and survival	Weirich and Tiersch (1997)
Perca flavescens	0.5 (bath)	Increase in survival	El-Gawad et al. (2016)
Oreochromis niloticus	30.0–60.0 (bath)	Increase in weight gain, relative growth rate and specific growth rate	Elrahman et al. (2016)
Oreochromis niloticus	10.0–20.0 (diet)	Increase in final weight, weight gain and specific growth rate	Debnath et al. (2017)
Oreochromis niloticus	0.015 (diet)	Increase in final weight and feed conversion, and decrease in specific growth rate, feed intake and weight gain	Abdelrhman et al. (2020)
Oreochromis niloticus	80.0 (diet)	Increase in final weight and feed intake	Fontaínhas-Fernandes et al. (2000)
Oreochromis niloticus	100.0 (diet)	Increase in final weight and feed conversion ratio	Fontaínhas-Fernandes et al. (2002)
Oreochromis niloticus	0.3 (bath)	Increase in final weight	Liti et al. (2005)
Tilapia rendalli	100.0 (bath)	Increase in final weight, diary weight gain and specific growth ratio, and decrease in survival	Kang'ombe and Brown, 2008
Tilapia rendalli	150.0 (bath)	Decrease in final weight, diary weight gain, specific growth ratio and in survival	Kang'ombe and Brown, 2008
Cyprinus carpio	10.0–15.0 (diet)	Increase in final weight, protein efficiency ratio and weight gain	Keshavanath et al. (2003)
Cyprinus carpio	15.0 (diet)	Increase in weight gain, feed conversion ratio and protein efficiency ratio	Nandeesha et al. (2000)
Cyprinus carpio	3.0 (bath)	Increase in body weight and survival and decrease in body length	Mubarik et al. (2018)
Cyprinus carpio	0.2–0.8 (diet)	Decrease in weight gain, total length and specific growth rate	Singh et al. (2019)
Cyprinus carpio	5.0–15.0 (diet)	Increase in body weight, growth rate, relative growth rate, food conversion efficiency and specific growth rate, and decrease in food conversion ratio	Nasir and Hamed (2016)
Cirrhinus mrigala	15.0 (diet)	Increase in weight gain, feed conversion ratio and protein efficiency ratio	Nandeesha et al. (2000)
Cirrhinus mrigala	3.0–8.0 (bath)	Decrease in weight gain and increase in survival	Hoque et al. (2020)
Cirrhinus mrigala	10.0–15.0 (diet)	Final mean weight, protein efficiency ratio (PER) and weight gain, and decrease in feed conversion rate	Keshavanath et al. (2003)
Mystus vittatus	40.0–100.0 (bath)	Increase in total food consumed and daily intake, decrease in diary growth, specific growth rate (SGR) and food conversion	Arunachalam and Reddy (1979)
Ramdia quelen	4.0 (bath)	Increase in body length and weight	Camargo et al. (2006)
Ramdia quelen	5.0 (diet)	Increase in weight, length and biomass per tank, and decrease in specific growth rate	Copatti et al. (2011)

TABLE 5 (Continued)

Fish species	Doses ($g\ L^{-1}$)–application	Alterations	References
Rhamdia quelen	12.0 (diet)	Increase in weight and biomass	Garcia et al. (2007)
Betta splendens	2.0 (bath)	Increase in weight gain, specific growth rate and survival	Dias et al. (2016)
Betta splendens	0.5 (diet)	Increase in survival	Puello-Cruz et al. (2010)
Betta splendens	6.0–10.0 (bath)	Decrease in survival	Fabregat et al. (2017)
Labeo rohita	20.0 (diet)	Increase in weight, weight gain, specific growth rate and biomass	Gangadhar et al. (2014)
Labeo rohita	10.0 (diet)	Increase in final weight, protein efficiency ratio and weight gain, and decrease in feed conversion rate	Keshavanath et al. (2003)
Labeo rohita	5.0–10.0 (diet)	Increase in weight, specific growth rate, protein efficiency ratio and net protein retention	Gangadhara et al. (2004)
Labeo rohita	2.0–4.0 (bath)	Increase in survival	Murmu et al. (2020)
Labeo rohita	2.0–8.0 (bath)	Decrease in survival and specific growth rate, and increase in condition factor and feed conversion ratio	Islam et al. (2014)
Labeo rohita	4.0–8.0 (bath)	Decrease in weight gain and increase in survival	Hoque et al. (2020)
Catla	5.0–8.0 (bath)	Decrease in weight gain and survival	Hoque et al. (2020)
Channa punctata	10.0 (bath)	Decrease in final weight and survival	Dubey et al. (2016)
Piaractus mesopotamicus	4.0–8. (bath)	Increase in total length, weight, specific growth rate and survival	Jomori et al. (2012)
Colossoma macropomum	20.0 (diet)	Increase in final length and weight, weight gain, specific growth rate and protein efficiency ratio, and decrease in feed conversion ratio	Keshavanath et al. (2012)
Rhinelepis aspera	6.0 (diet)	Decrease in total length, weight and specific growth rate	Luz and Santos (2010)
Pterophyllum scalare	7.5 (diet)	Increased in survival, specific growth rate, feed efficiency and final weight, and decrease in food coefficient ratio (FCR)	Motlagh et al. (2012)
Oreochromis shiranus	15.0 (diet)	Increase in body weight, weight gain and specific growth rate, and decrease in feed conversion ratio	Mzengereza and Kang'ombe (2015)
Oncorhynchus mykiss	116.0 (diet)	Decrease in weight gain and food conversion efficiency, and increase in instantaneous growth ratio	Salman and Eddy (1988)
Hypsolebias radiseriatus	6.0–8.0 (bath)	Decrease in survival	Araújo et al. (2021)
Pseudoplatystoma corruscans	20.0 (diet)	Increase in weight, total length, specific growth rate and survival	Santos and Luz (2009)
Prochilodus costatus	20.0 (diet)	Increase in weight, specific growth rate and survival	Santos and Luz (2009)
Chrysichthys nigrodigitatus	0.02 (diet)	Decrease in weight gain and daily weight gain, and increase in feed conversion efficiency	Udoh and Otoh (2017)

The primary responses of stress are neuroendocrine and tertiary responses that may affect the growth rate of fish. However, feeding of *Cyprinus carpio* for 60 days with diets supplemented with 0.0015–0.012 g of NaCl led to 100% survival (Mangat & Hundal, 2014). Feeding of *Rhinelepis aspera* larvae for 7 days with diets supplemented with 2.0–6.0 g of NaCl and transferred to water with salt showed no difference in survival when compared with specimens that were not transferred (Luz & Santos, 2010). Feeding of *Hoplias lacerdae* for 15 days with diets supplemented with 20.0–40.0 g kg^{-1} of NaCl had no influence on survival, body size and specific growth rate (Luz & Portela, 2002). The addition of up to 85 g kg^{-1} of NaCl to the diet of *O. mykiss* had no effect on food intake and feed conversion efficiency (MacLeod, 1978). The quantitative dietary requirements for sodium and chloride in fish are determined by growth, of which these nutrients are generally lost by the animals through gut, kidney and by passive diffusion across the gills and body surface (Salman & Eddy, 1988). Consequently, efforts have been made to quantify the relative importance of dietary and non-dietary sources of sodium and chloride in freshwater fish.

Diets supplemented with various levels of NaCl have been used in the fish aquaculture industry, mainly for adaptation to freshwater (Table 5). Nutritional effects of such diets are little understood, with different conclusions resulting from differences in diet preparation methods, salt content, nutrient balance and feeding Levels. The dietary requirements for sodium and chloride in freshwater fish can be determined according to the amount needed for growth, reproduction and that lost through the gut, kidney as well as passive diffusion across the gills and body surface. However, requirements of these minerals in freshwater fish have received little attention when compared with other nutrients.

7 | ANTIPARASITIC EFFICACY OF NaCl IN TREATED FRESHWATER FISH SPECIES

Intensification of fish aquaculture has led to the development of favourable conditions for a variety of fish diseases, including parasites (García-Magaña et al., 2019; Hakalahti-Sirén et al., 2008). Prophylaxis and therapy are important biosecurity measures in fish aquaculture. The application of these procedures in the cultivation must be prioritized in intensive aquaculture systems since high fish densities may facilitate the development of outbreaks of various parasites, especially ectoparasites. Sanitary handling must be applied to avoid diseases being introduced in the cultures of ornamental or food fish. Diseases have become the main constraint to the expansion of fish aquaculture, negatively affecting both economic and socioeconomic development of many countries. The annual economic losses due to parasitic diseases are estimated to be US$ 1.05–9.58 billion (Shinn et al., 2015). Among the parasitic agents, ectoparasites may survive in the aquatic environment independently of their hosts and became the major obstacles to the cultivation of freshwater fish species.

Most chemotherapeutics used in fish aquaculture may have toxic effects. Hence, environmentally friendly agents such as NaCl have been used to replace the toxic chemicals as antiparasitic agents in fish aquaculture. In general, parasitic diseases can be controlled by feeding infected fish with medicated feed or therapeutic baths, depending on the species. However, this feeding practice may be ineffective as sick fish show loss of appetite and low feed uptake. Furthermore, frequent use of antiparasitic chemotherapeutics has led to the development of parasite resistance to various compounds, posing serious challenges to both aquatic animal health and human health. Adequate use of antiparasitic agents may decrease the recovery time of infected fish populations, improve the welfare of treated fish and prevent the spread of the infection to fish farming. As fish aquaculture evolves from extensive pond culture to intensive tank and net-cage systems, chemotherapeutic dips and baths are increasing in use to treat a concomitant increase in ectoparasitic infections.

Many of the problems that occur in fish aquaculture and the aquarium industry are related to ectoparasites, especially the monogeneans and protozoans, which cause high economic loss. There has been an increase in diseases caused by monogeneans and protozoans in recent years in the aquarium and aquaculture industries due to their rapid growth (Tavares-Dias & Martins, 2017; Thoney & Hargis Jr., 1991). NaCl dips have been used to control and treat infections of monogeneans in both aquaculture and the aquarium industry (Table 6). Monogeneans of the gills show high resistance to NaCl treatments when compared with the monogeneans of the skin, probably because they are protected by the surrounding gill tissue within the opercular cavity (Thoney & Hargis Jr., 1991). NaCl is the second most commonly used product for the treatment of *I. multifiliis* infection because it is effective in the control and treatment of this protozoan (Table 6), which is one of the most virulent ectoparasites of freshwater fish and has a global distribution. However, treatment against parasites may be stressful to the host fish (Thoney & Hargis Jr., 1991). Furthermore, improperly administered NaCl treatments may cause toxicity in aquaculture fish (Foissner et al., 1985).

It has been suggested that 60–100 mg L^{-1} of NaCl baths for 8–12 h for ornamental fish maintained in ponds may reduce stress and dissemination of several ectoparasites (Piazza et al., 2006). Concentrations of 10.0–50.0 g L^{-1} of NaCl for 3 h were shown as the most effective treatment against *Heteropolaria colisarum* in *Lepomis cyanellus* when compared with other time periods, but did not achieve complete control (Foissner et al., 1985). *In vitro* exposure of *Lepidotrema bidyana* to 10.0 g L^{-1} of NaCl was effective (Forwood et al., 2013). In *Betta splendens*, the use of 0.5 g L^{-1} of NaCl in the diet led to the absence of infection by *Piscinoodinium pilullare*, which was present at body surface of fish reared at 0 g L^{-1} of NaCl (Puello-Cruz et al., 2010). Marchiori et al. (2015) demonstrated that 9.0 g L^{-1} of NaCl in the water impaired the viability of the eggs of the monogenean *Aphanoblastella mastigatus*. In addition, these treatments may avoid the problem of increased mucus production in the fish hosts, a physiological response triggered in response to adverse environmental conditions.

It is a common practice in fish medicine to manipulate salinity as a therapeutic measure against ectoparasites and osmotic stress. Therapeutic treatments using environmental salinity changes through a short- or long-term bath of NaCl have been used. The concentration of 0.025 g L^{-1} of NaCl had a detrimental effect on survival, growth and reproduction of the snail *Phanorbela trivolvis*, the intermediate host of the digenean *Bolbophonrs confusus*. However, this salinity had no negative effect on the survival of cercariae and digenean life cycles. In ponds of *Ictulurus punctatus* maintained at three salinities (0.0025, 0.00125 and 0.025 g L^{-1}), the snail densities in water at 0.025 g L^{-1} were lower than in the other treatments and no trematode infection was noted among snails or *I. punctatus* (Venable et al., 2000). Zeni et al. (2017) reported reduction in *I. multifiliis* in *Metynis maculatus* exposed to 9–13.0 g L^{-1} of NaCl when compared with the control salinity. This result was not observed in *R. quelen*. Therefore, NaCl may be used in therapeutic baths to treat infected fish. The concentration is determined by the time that the fish species are subjected to the baths and the tolerance of the fish. Nevertheless, the adequate concentration of NaCl for a prolonged or short-term bath depends on certain factors already discussed previously. Nevertheless, treatment doses may not be based on empirical and anecdotal information; because toxicity may, therefore, occur

TABLE 6 Management strategies of therapeutic baths with NaCl to control and treatment of freshwater ectoparasite species

Parasite species	Doses (g L^{-1})	Exposure time	Results	References
Piscinoodinium pillulare	6.0	4 h	High efficacy	Carneiro et al. (2002)
Trichodina indica	0.03	10 min	High efficacy	Singh et al. (1986)
Trichodina sp.	30.0	10 min	High efficacy	Vargas et al. (2003)
Trichodina sp.	5.0	Continuous	High efficacy	Németh et al. (2013)
Ictiophitirius multifiliis	5.0	7 days	High efficacy	Selosse and Rowland (1990)
Ictiophitirius multifiliis	10.0	96 h	High efficacy	Carneiro et al. (2005)
Ictiophitirius multifiliis	10.0	24-120 h	High efficacy	Carneiro et al. (2006)
Ictiophitirius multifiliis	7.5	-	High efficacy	Aihua and Buchmann (2001)
Ictiophitirius multifiliis	4.0	30 days	Ineffective	Garcia et al. (2007)
Ictiophitirius multifiliis	0.02	10-20 h	High efficacy	Lahnsteiner and Weismann (2007)
Ictiophitirius multifiliis	30.0	10 min	Ineffective	Klein et al. (2004)
Ictiophitirius multifiliis	1.5	72	Ineffective	Rahanandeh and Rahanandeh (2020)
Ictiophitirius multifiliis	2.0–3.0	6 days	High efficacy	Mifsud and Rowland (2008)
Ictiophitirius multifiliis	4.0	45 days	High efficacy	Miron et al. (2003)
Ictiophitirius multifiliis	0.003	10 days	High efficacy	Zaikov et al. (2006)
Ictiophitirius multifiliis	5.0	Continuous	High efficacy	Németh et al. (2013)
Trichodina spp.	10.0	3 min	High efficacy	García-Magaña et al. (2019)
Chilodonella spp.	7.0	21 h	High efficacy	Willomitzer (1980)
Pseudodactylogyrys bini and *Pseudodactylogyrys anguillae*	15.0–34.0	5 h	Moderate efficacy	Umeda et al. (2006)
Lepidotrema bidyana	0.5–10.0	1 h	Ineffective	Forwood et al. (2013)
Aphanoblastella mastigatus	9.0	24 h	High efficacy	Marchiori et al. (2015)
Dactylogyrus sp.	30.0	10 min	Ineffective	Vargas et al. (2003)
Gyrodactylus sp.	5.0	Continuous	Ineffective	Németh et al. (2013)
Monogenea gen sp.	10.0	-	High efficacy	Silva et al. (2009)
Monogenea gen sp.	2.0–8.0	2 h	Ineffective	Chagas et al. (2012)
Hemiclepsis marginata	0.03	15 min	High efficacy	Singh et al. (1986)
Myzobdella lugubris	10.0	48	Ineffective	Morrison et al. (1993)
Batracobdela sp.	15.0	15 min	High efficacy	Santos et al. (2020)
Argulus indicus	0.03	2–5 min	High efficacy	Singh et al. (1986)
Argulus foliaceus	15.0	15 min	High efficacy	Vasilean et al. (2012)
Argulus sp.	0.09	10 days	High efficacy	Dewi et al. (2018)
Argulus coregoni	15.0–50.0	15 h	Ineffective	Hakalahti-Sirén et al. (2008)
Argulus japonicus	30.0	5 min	High efficacy	Singh et al. (2018)
Argulus japonicus	20.0	5–10 min	Moderate efficacy	Kumar et al. (2018)
Lernaea cyprinacea	3.0	5 min	High efficacy	El-Deen et al. (2013)
Ergasilus intermedius	10.0	1 h	Ineffective	Ingram and Philbey (1999)
Epistylis sp.	15.0–30.0	1 h	High efficacy	Hubert and Warner (1975)
Epistylis sp.	15.0–20.0	48 h	High efficacy	Rodrigues et al. (2019)

with the use of concentration used. Pharmacokinetic data are seldom available for agents used to treat aquaculture fish. Therefore, there is a necessity of having safe and effective antiparasitic drugs in fish medicine and has high importance in the aquaculture industry for sanitary management of fish farmed for human consumption and ornamental fish.

8 | CONCLUSIONS AND PERSPECTIVES

Salinity is an important abiotic factor that can invoke a response in the fish. Freshwater fish present a variation in NaCl tolerance, which is lower than that of euryhaline fish. When a fish population is exposed to water with varying NaCl, their ability to tolerate the concentration will have a direct correlation with their level of fitness and will lead to a physiological adaptation to environment that may be varied. However, there have been few studies on the NaCl tolerance of freshwater fish regarding salinity exposure and lethal concentrations stress for determining the lethal dose.

The beneficial effects of dietary supplementation of NaCl on growth and physiology of euryhaline fish have been well discussed. However, the effects in freshwater fish have been little discussed. The effects of dietary NaCl on freshwater fish adaptation, osmoregulatory mechanisms, growth and feeding efficiency have been little investigated. Thus, many aspects still need studies and discussions. These include determination of dietary NaCl levels that show satisfactory freshwater adaptation without affecting feeding efficiency and growth. Responses of digestive enzymes are of special interest to understand nutritional impacts of NaCl. The acclimation process in NaCl involves several hormonal and osmoregulatory adjusts.

Freshwater fish should be monitored closely for clinical signs that suggest toxicosis of NaCl. While there is greater risk with higher concentrations, there is also a better chance for successful treatment. In order to optimize culture conditions of freshwater fish, it is desirable to understand the optimal NaCl concentration for growth and energy conversion efficiency. Nutritional effects of NaCl in freshwater fish are not fully understood, with different assessments resulting from differences in diet preparation methods, salt contents, nutrients balance, feeding levels and other practices. Hence, results of studies remain controversial, especially regarding the nutritional responses and NaCl adaptation for freshwater fish. So, more research is needed to clarify the growth of freshwater fish with the supplementation of NaCl. A dietary supplementation of NaCl may satisfy the osmoregulatory requirements of fish kept in freshwater while leaving more energy available for body growth. Therefore, fish reared at an optimum salinity would have reduced metabolic activity, which ultimately reserves energy for growth. While the use of common salt appears to represent an economically viable and safe treatment option for fish farms, it should be used with caution in certain stenohaline freshwater fish species.

The application of NaCl in freshwater fish farms is widely practiced due to its ectoparasiticide effects and its facilitation of osmoregulation. It is used as a preventive treatment for diseases in a fast bath with concentrations that vary according to the species of ectoparasite. This substance offers an alternative for controlling and treating fish infected and has led to a significant increase in survival of parasitized fish. For helminth and protozoan ectoparasite species, NaCl baths are often practiced since a high-dose, short duration treatment acts aggressively against the parasites. This salt not only causes direct osmotic problems to the parasites but also aid the fish with its protective mucus layer. Ectoparasites of stenohaline fish species are more likely to succumb at higher concentrations of NaCl than at the lower concentrations. Thus, this study identified the NaCl sensitivity range for freshwater fish species and therapeutic concentrations necessary to develop therapeutic treatments for various species of ectoparasites. Thus, more studies are being performed regarding the effects of NaCl in stenohaline freshwater fish for controlling and treating diseases caused by ectoparasites. The concentration is determined according to the time that the fish are subjected to the baths and the tolerance of the fish. It is noteworthy that keeping the fish in their rearing system during treatment may mitigate added stress from handling. Nevertheless, the adequate concentration of NaCl for a prolonged or short-term bath depends on certain factors discussed here.

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CONFLICT OF INTEREST

The authors declare that they did not have any conflict of interest.

AUTHOR CONTRIBUTIONS

Marcos Tavares-Dias elaboration of data and redaction of manuscript.

DATA AVAILABILITY STATEMENT

The data that support the findings of this study are available from the corresponding author upon reasonable request.

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