

Review

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Review

Addressing Phosphorus Waste in Open Flow Freshwater Fish Farms: Challenges and Solutions

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Abstract: Legislation and interest to protect and restore freshwater and marine ecosystems from aquaculture's environmental impact is global. However, aquaculture induced eutrophication continues to be a major environmental issue. Open freshwater fish farms in particular, providing fish with phosphorus-rich feeds pollute aquatic ecosystems since water soluble phosphorus, uneaten feed, feces, and metabolic waste from farmed fish increase phosphorus concentration in the adjacent waters. Several intestinal enzymes, transporters, and regulating factors are implicated in dietary phosphorus retention of farmed fish. For example, alkaline phosphatase and other transporters help the anterior intestine absorb phosphorus, while pH, calcium, and vitamin D affect these enzymes and transporters. Intestinal morphology and gut microbiome may also affect this process. Reducing phosphorus pollution from open-flow fish farms requires a thorough understanding of processes that affect nutrient retention and absorption as well as of the impact of dietary factors, anti-nutritional substances, and intestinal morphology. Optimizing feed composition, adding functional feed ingredients, and managing gut health can reduce phosphorus release and improve aquaculture sustainability. Processing and functional feed additives can mitigate anti-nutritional factors and, addressing these issues will reduce aquaculture's environmental impact, ensuring aquatic ecosystem health and global food security.

Keywords: aquaculture nutrition; phosphorus pollution; sustainability; eutrophication

1. Introduction

Aquaculture, like other forms of agriculture, has environmental impact. Farmed fish release nitrogen and phosphorus, which, if left untreated in water effluents, enter surface water bodies and cause eutrophication (Ahmed et al., 2019). Fresh water fish farms may also discharge veterinary drugs and antibiotics, harming aquatic biodiversity, accumulating antibiotics, and increasing antibiotic resistance (Lulijwa et al., 2020; Mavraganis et al., 2020).

Aquaculture has experienced rapid growth throughout history, driven by the increasing demand for seafood and the overexploitation of fish stocks. As the industry strives to address food security concerns, fish nutrition plays a vital role in ensuring the sustainability of the sector. To achieve this, researchers are exploring new feed compositions and ingredients that can optimize fish health and performance. Phosphorus holds a dual significance in fish nutrition. Firstly, it is an essential ingredient in fish feeds, since it is required for various physiological processes, including bone formation, energy metabolism, and cellular functions. Adequate phosphorus levels in fish diets are crucial for promoting growth and overall well-being (Fontagne et al., 2009; Sugiure, 2015; Lall & Kaushik, 2021). However, phosphorus also presents a potential challenge in terms of environmental pollution. Excessive phosphorus discharge from aquaculture operations can lead to water eutrophication, algal blooms, and other negative impacts on aquatic ecosystems. Phosphorus runoff from fish farms contributes to the nutrient load in surrounding water bodies, which can have detrimental effects on water quality and biodiversity (Varol & Balci, 2020; Mavraganis et al., 2021). To

mitigate these environmental concerns, aquaculture endeavours to optimize phosphorus utilization and minimize its environmental footprint. This involves developing innovative feed formulations that enhance phosphorus digestibility and absorption in farmed fish, thereby reducing phosphorus excretion into the environment. Additionally, techniques such as precision feeding, which aim to match feed supply with the nutritional requirements of fish, help prevent excessive phosphorus discharge (Glencross et al., 2023). By addressing the dual role of phosphorus as an essential ingredient and a potential parameter of pollution, the aquaculture industry can achieve sustainable growth while minimizing its environmental impact. The phosphorus retention efficiency or absorption in the fish intestine can be influenced by an interaction between dietary, intestinal anatomical and physiological parameters. Figure 1, illustrates the various factors which are implicated in the waste of phosphorus in aquacultured species. Several anatomical and physiological parameters related to the efficiency of dietary phosphorus absorption, such as active transporters, intestinal alkaline phosphatases, and villi morphology, are implicated in affecting the waste of P of farmed fish (Aslaksen et al., 2007; Dai et al., 2021; Denstadli et al., 2006). However, these parameters can be improved through the use of probiotics, which directly or indirectly enhance intestinal phosphorus absorption. Probiotics exert their positive effects on phosphorus absorption in fish by modulating gut health and competing with harmful bacteria (Jahangiri & Esteban, 2018; Nathanailides et al., 2021). By influencing the composition and balance of the gut microbiota, probiotics create a favorable environment for nutrient digestion and absorption, including phosphorus (Adeoye et al., 2016; Maas et al., 2021). This leads to improved functionality of the intestinal epithelium and enhanced nutrient transport mechanisms, including active transporters responsible for phosphorus uptake (Elsabagh et al., 2018; Islam et al., 2021). Moreover, probiotics outcompete pathogenic bacteria for nutrients and adhesion sites in the fish gut, reducing their presence and maintaining a healthier gut environment (Camara-Ruiz et al., 2020). This competitive exclusion contributes to optimal nutrient absorption, including phosphorus, which can positively impact the waste of phosphorus in farmed fish. As a result, probiotics not only improve the efficiency of dietary phosphorus absorption by enhancing various anatomical and physiological parameters but also indirectly reduce the waste of phosphorus in farmed fish (Elsabagh et al., 2018). By promoting gut health and mitigating the negative influence of harmful bacteria, probiotics contribute to improved intestinal phosphorus absorption and subsequently help reduce the environmental impact of phosphorus waste in aquaculture systems (Hlondzi et al., 2020; Nathanailides et al., 2021).

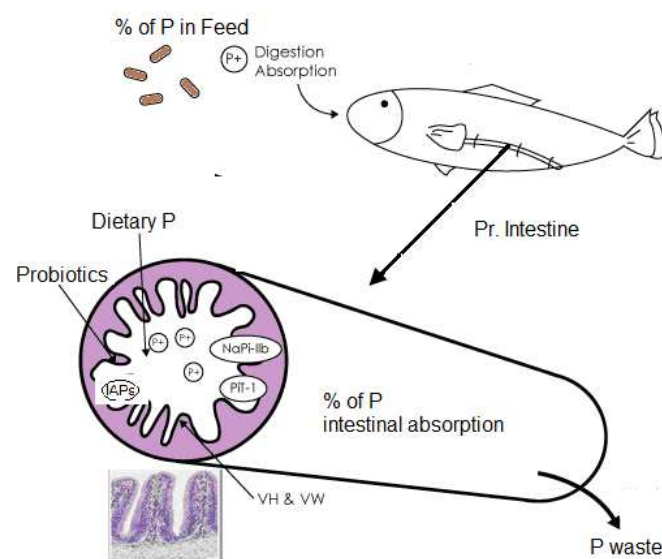


Figure 1. Gut microbiome (epithelium, mucus layer), Active transporters (e.g. PiT-1, NaPi-IIb), Intestinal alkaline phosphatases (IAPs), and Intestinal vilus morphology (V. Height V.H. and V. width, VW) are implicated in the efficiency of dietary phosphorus (P+) absorption in the proximal intestinal segment (Pr. Intestine), affecting the waste of P of farmed fish.

1.1. Phosphorus Requirements of Fish Farmed in Open Flow Aquaculture Systems

Phosphorus is an essential nutrient for fish, playing a crucial role in various physiological functions, including bone formation, tissue growth, acid-base balance, energy metabolism, and reproduction. The phosphorus requirements of fish depend on several factors, such as species, life stage, growth rate and water temperature (Kraft, 1992; Sambraus et al., 2020; Lall & Kaushik 2021). Higher phosphorus requirements are associated with growth and skeletal development (Jahan et al., 2002; Ye et al., 2006; Sugiura, 2015), and this is particularly interesting for the aquacultured species. Salmonids and other carnivorous fish species are widely cultivated in open flow aquaculture systems and require high levels of protein in their diet. The protein requirement varies depending on the size and life stage of the fish, but generally ranges from 32-45% of the diet. The phosphorus content of salmonids may vary depending on diets composition but in Europe, it typically ranges from 0.7-1.4% (Chatvijitkul et al., 2018; Mavraganis et al., 2017).

1.2. The Environmental Impact of Aquaculture, with Emphasis on Open Flow Fish Farms

The demand for farmed fish can lead to increased fish farm production which can result in higher loads of phosphorus influenced by biomass and water flow rate (Mavraganis et al., 2017; 2021). Typically, in temperate climate zones, land-based rainbow trout aquaculture occurs near rivers and at high altitude where it's more likely than agriculture or industry to pollute the aquatic ecosystems. Aquaculture management, flow rates, and other spatial and seasonal factors affect phosphorus release in trout aquaculture. Before discharging, some farms use effluent treatments of varying efficacy, while others use different feeding regimes or discharge diluted waste (Bergheim and Brinker, 2003; Moraes et al. 2016).

Intensive aquaculture is frequently based on open flow fish farms (Rico et al 2019) which must quickly release the water outflow on neighbouring rivers, allowing for limited time for water treatment and resulting in phosphorus release downstream (Tahar et al., 2018; Mavraganis et al., 2021). In most freshwater ecosystems, phosphorus (P) is the limiting nutrient, whereas nitrogen (N) is the limiting nutrient in marine ecosystems. As a result, monitoring anthropogenic sources of phosphorus in freshwater ecosystems is a useful tool for determining the causes of eutrophication and their environmental impact. One possible source for phosphorus in rivers and lakes is aquaculture feed. However, farmed fish require phosphorus in their diet. Diets lacking in phosphorus can lead to severe pathological problems of farmed fish (Sugiura et al. 2018; Fjellidal et al. 2012). Phosphorus-containing fish feeds can contribute to aquatic pollution by releasing uneaten feed, faeces, and metabolic wastes of farmed fish (Beveridge & Brummett, 2015).

There is uncertainty regarding the range of potential ecological parameters affected by phosphorus pollution in aquatic ecosystems; however, fish farm effluents can account for most of downstream river eutrophication (Webb, 2012) with ecological effects such as loss of biodiversity according to nutrient load released and mainly via increased levels of phosphorus (Camargo, 2019). The ecological effect of fish farms is evaluated by monitoring the levels of phosphorus produced by them expressed in kilograms of phosphorus produced by fish farms per metric ton of fish produced. The phosphorus load generated can be estimated with direct measurements of water samples or with fish production and feed records or feed conservation rates (FCR) combined with chemical analyses of feed and fish (OSPAR, 2000).

2. Morphological, Physiological, and Dietary Factors Affecting Phosphorous Absorption in Fish

2.1. Intestinal Morphology and Physiological Mechanisms

Fish intestine morphometric characteristics, including the surface area, as well as the presence of specialized cells, can reveal an intriguing relationship between anatomy and nutrient absorption efficiency (Bakke et al., 2010 et al., 2020; Wu et al., 1998; Debnah et al., 2021). The intestinal phosphorus absorption process in fish can be divided into two main phases: luminal and brush-border absorption. Luminal absorption involves the uptake of phosphorus from the intestinal lumen into enterocytes,

while brush-border absorption involves the transport of phosphorus across the brush-border membrane of enterocytes and into the bloodstream (Ndiaye et al., 2020; Debnath et al., 2021). Several nutritional experiments have demonstrated the length of villi and the number of goblet cells to be indicators of feed utilization efficiency (Rakovi et al., 2011; Caspary, 1992). Rainbow trout (*Oncorhynchus mykiss*) with higher phosphorus retention efficiencies exhibit longer intestinal villi and higher goblet cell densities compared to those with lower phosphorus retention efficiency (Vidakovic et al., 2020). Similarly, studies on Nile tilapia (*Oreochromis niloticus*) have explored the effects of dietary phosphorus levels on villi morphology and nutrient retention. Digestibility of protein and phosphorous was associated with intestinal villus length (Adesina et al., 2023). In fact, a complex interplay of transporters, enzymes, and hormones facilitates the process of phosphorous absorption. So, the anterior part of the intestine is considered as the primary site of dietary phosphorus absorption since it exhibits higher concentration of alkaline phosphatase, an enzyme responsible for phosphate ester hydrolysis, and other phosphorus transporters (Denstadli et al., 2006). Several transporters and channels have been identified as playing key roles in intestinal phosphorus absorption in fish. These include sodium-dependent phosphate transporters (NaPi-IIb), which are responsible for the luminal uptake of phosphorus, and type III sodium-dependent phosphate transporters (Pit-1), which mediate brush-border uptake (Sugiura, 2009; Sugiura et al., 2003). Additionally, calcium-sensing receptor (CaSR), transient receptor potential vanilloid 6 (TRPV6), and plasma membrane calcium ATPase (PMCA) are other transporters involved in regulating phosphorus absorption in fish (Denstadli et al., 2006).

Regarding vitamins, cholecalciferol, also known as vitamin D3, is a type of fat-soluble vitamin that plays a crucial role in calcium and phosphate metabolism. Cholecalciferol converted into its active form, calcitriol, regulates the absorption of calcium and phosphate in the intestine and influences their levels in the blood. In the context of trout and other farmed fish, cholecalciferol supplementation has been studied for its effects on plasma phosphate levels and phosphorus utilization and the results indicate that cholecalciferol supplementation can modulate plasma phosphate concentrations affecting the overall phosphorus balance in fish (Vielma et al. 1999).

Intestinal phosphorus absorption in fish can also be affected by hormones such as calcitriol, parathyroid hormone, and fibroblast growth factor 23. Calcitriol stimulates the expression of NaPi-IIb and Pit-1 transporters, while parathyroid hormone inhibits NaPi-IIb expression and stimulates PMCA expression. Fibroblast growth factor 23 inhibits calcitriol production and promotes phosphorus excretion in urine.

2.2. Relationship between Fish gut Structure and Feed Conversion Efficiency

The relationship between fish gut structure and feed conversion efficiency (FCE) is crucial in understanding the potential for reducing phosphorus release in aquaculture. FCE plays a vital role in both economic viability and environmental sustainability. It refers to the ability of fish to convert feed into body mass, and improving FCE means that less feed is required to produce the desired amount of fish biomass. This reduction in feed input has the potential to decrease the release of phosphorus-rich waste into the surrounding aquatic environment.

The gut structure of fish, including factors such as the length and surface area of the intestine, the thickness of the intestinal wall, and the presence of intestinal villi and microvilli, can significantly impact the digestion and absorption of nutrients from the feed, ultimately affecting Feed Conversion Efficiency (FCE) and phosphorus retention efficiency (Thaib et al., 2021). Studies have shown that fish with a higher gut surface area or longer intestine tend to exhibit higher FCE, indicating their enhanced ability to digest and absorb nutrients from the feed (Ringo et al., 2010; Stevens & Devlin, 2005).

Moreover, the relationship between FCE and phosphorus pollution in aquaculture is crucial. Low FCE is associated to decreased phosphorus retention efficiency in farmed fish, resulting in increased phosphorus excretion into the water (Bureau & Cho, 1999; Morales et al., 2018; Li et al., 2017; Miao et al., 2019). Understanding this relationship enables the creation of effective management strategies to reduce phosphorus pollution in aquaculture systems.

2.3. *The Role of Fish Feeds in Nutrient Uptake with Emphasis on Phosphorous*

This intricate interplay of transporters and dietary factors reflects the complex nature of nutrient absorption in farmed fish. Phosphorus retention can be influenced by several dietary parameters, such as the raw materials used as protein and phosphorus sources, and the ratio of phosphorus to other minerals, such as calcium (Milian-Sorribes et al., 2021; Chanpaisaeng et al., 2021; Avila et al., 1999&2000; Ye et al, 2006; Morales et al., 2018; Jin et al., 2022). The development of new fish feed formulations allows aquaculture to tailor the nutritional composition of the feed to meet the specific requirements of different fish species and use alternative protein sources to reduce the reliance of aquaculture on fish meal and fish oil. However, efforts to replace fish meal with plant proteins in fish diets face challenges related to the impact on the functional integrity of farmed fish intestines (Aslaksen et al., 2007; Santigosa et al., 2011). Plant natural defense mechanisms, such as protease inhibitors, phytates, glucosinolates, saponins, tannins, lectins, oligosaccharides, and non-starch polysaccharides, can induce intestinal inflammation (Behera, 2021). This inflammation is linked to changes in gene expression within the intestine, including the absorption of phosphorus. Furthermore, anti-nutritional factors, such as phytate, can affect how efficiently aquacultured species absorb phosphorus. Phytate is a type of phosphorus present in plant-based feed ingredients commonly used in aquaculture diets. However, many aquatic species have limited ability to digest phytate because they lack the necessary enzyme, called phytase, which is responsible for breaking it down (Lall & Kaushik, 2021). To mitigate the negative effects of these anti-nutritional factors, fish feed processing methods are employed to neutralize harmful compounds present in plant ingredients and prevent adverse effects on fish. This includes techniques to destroy or reduce the presence of plant natural defense mechanisms. Phytase, for example, an enzyme that can break down phytic acid, is a useful tool that can help fish make the best use of phosphorus available in plant-protein-based feeds. Studies have shown that its addition, can increase the availability of phosphorus in fish diets, leading to improved growth and health (Liebert & Portz, 2005; Cao et al., 2008; Yigit et al., 2018; Dias & Santigosa, 2023; Zheng et al., 2020). Interestingly, the commonly used soybean meal (SBM) in fish diets has been found to impact nutrient absorption (Hernandez et al., 2016), down-regulating the expression of fabp2, a fatty-acid binding protein responsible for lipid absorption in the gut. This disruption in fabp2 expression can interfere with the transport and absorption of lipids, leading to reduced lipid intestinal uptake (Venold et al 2012). Decreased fabp2 expression has been particularly observed in fish experiencing SBM-induced inflammation in the distal portion of the intestine (Perera & Yufera, 2016). Overall, when fed plant proteins, farmed fish often exhibit intestinal inflammation, which can be addressed by incorporating functional feed additives or employing processing techniques for plant proteins (Yang et al., 2011; Vélez-Calabria et al., 2021). This becomes especially important given the growing trend of substituting fish meal with plant protein in aquafeed.

The physiological mechanisms involved in intestinal phosphorus absorption in farmed fish are complex and multifaceted, and are influenced by various nutritional and physiological factors such as pH, calcium and anti-nutritional factors (Figure 2). Understanding these mechanisms is important for optimizing the formulation of fish diets and improving the efficiency and sustainability of aquaculture production.

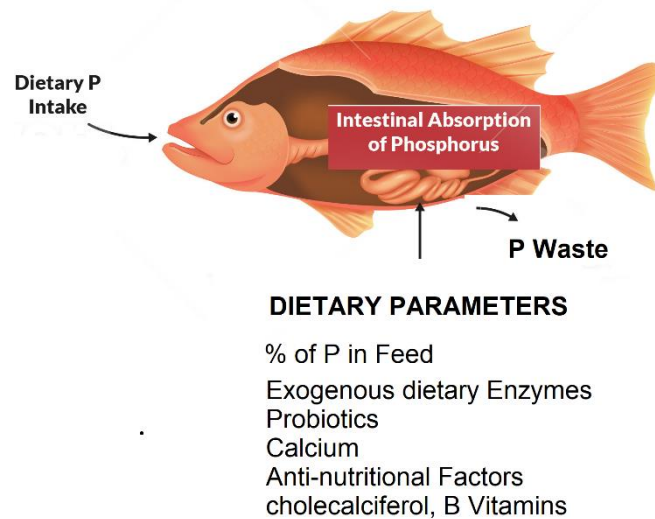


Figure 2. Dietary Factors affecting phosphorus waste of farmed fish.

Dietary phosphorus levels can also affect phosphorus absorption in farmed fish, influencing the expression of phosphate transporters like NaPi-IIb and Pit-1 with the highest expression observed in the anterior intestine, followed by the posterior and middle intestines (Chen et al., 2016; Dai et al., 2021). Increasing the dietary levels of calcium and vitamin D has been shown to enhance phosphorus absorption by upregulating the expression of sodium-phosphate co-transporters in the fish intestine, particularly in the anterior segment (Sugiura et al., 2003; 2018).

2.4. Effect of Probiotics on the Environmental Impact of Freshwater Fish Farms:

The utilization of probiotics in aquaculture offers a promising approach for reducing phosphorus pollution and implementing effective nutrient management strategies in fish farm effluents. Probiotics, particularly *Bacillus* strains, have shown the ability to modulate various water quality parameters, including phosphates (Hlondzi et al., 2020). This is because probiotics utilize phosphates for their own metabolic processes, effectively decreasing the concentration of this nutrient in aquaculture waters (Rao, 2002). The positive impact of probiotics on reducing orthophosphate concentrations in treated ponds, thereby removing phosphorus, nitrogen, and organic matter from aquaculture systems illustrates the potential of this method for reducing phosphorus pollution by aquaculture. (Sunitha & Padmavathi, 2013; Choi et al., 2002). Wang & Xu (2006) found that the application of commercial probiotics in *Penaeus vannamei* ponds resulted in reduced nitrogen and phosphorus concentrations and increased shrimp yields. Similar findings were reported by Kumar et al. (2016), who observed a reduction in total $\text{NH}_4\text{-N}$, nitrogen, $\text{NO}_2\text{-N}$, $\text{NO}_3\text{-N}$, and phosphorus concentrations with the use of probiotics.

Probiotic supplementation in fish feed offers several physiological and anatomical benefits that positively impact nutrient absorption including phosphorus retention. The elongation of villi, stimulated by probiotics, results in an increased absorptive surface area, thereby enhancing nutrient bioavailability. Specific strains of probiotics can stimulate the elongation of villi in the midgut and hindgut of fish, as evidenced by a comparison of probiotic-treated groups with control groups (Amoah & Dong, 2020; de Moraes et al., 2018; Yang et al., 2020; Standen et al. 2016; Yeganeh Rastekenari et al., 2021). The production of short chain fatty acids (SCFAs) by probiotic bacteria further stimulates the production of gastrointestinal peptides, enhancing nutrient absorption capacity and ultimately contributing to improved growth performance (Reda & Selim, 2015; Zhou et al., 2021; Nimalan et al., 2023).

In addition to the above, there is emerging evidence that probiotics can have a positive effect on gut health (Camara-Ruiz, 2020) and positively influence enzyme levels through various physiological pathways, ultimately improving fish digestion and nutrient utilization (Encarnaç o, 2016). For example, probiotics may directly influence the cells lining the intestinal tract, such as enterocytes or

goblet cells, which are responsible for intestinal enzyme synthesis and secretion (Islam et al., 2021; Standen et al., 2013; Elsabagh et al., 2018). It can be assumed that probiotics can promote the expression and release of digestive enzymes by these specialized cells, leading to higher enzyme levels in the gut consequently affecting the utilisation of nutrients including phosphorus (Mass et al., 2021).

2.5. The Modulatory Effect of Temperature and Metabolism on Intestinal Absorption of Phosphorus in Farmed Fish

Apart of the nutritional parameters, phosphorus absorption in farmed fish is influenced by several factors, including water temperature and fish size (Zhang et al., 2022). Temperature in open flow fish farms can vary seasonally but also other factors such as fish size, feeding regimes, water flow may change, contributing to a range of interactions affecting phosphorus release from freshwater Open flow fish farms. Glencross et al. (2005) found that increasing water temperature from 15°C to 25°C resulted in a significant increase in phosphorus absorption in rainbow trout. Growth rate is significantly affected by thermal condition and by parameters of growth (Jobling, 2010). The changes in phosphorus requirements with temperature, fish size and life stage highlight the importance of considering the growth trajectory and physiological status of fish when formulating diets and managing phosphorus levels in their aquaculture systems. For example, body size affects the physiological processes and energy metabolism, triploid fish may exhibit higher phosphorus requirements (Martinez-Llorens, 2020); larvae and juvenile fish exhibit significant phosphorus requirements due to their rapid growth and increased tissue turnover (Wang et al., 2016; Fraser et al., 2019); larger fish have greater skeletal mass and overall body size, which necessitates dietary absorption of phosphorus (Lall & Kaushik, 2021), whereas at older stages, gonadal development can also result in increased phosphorus requirement (Bruce, 1924). The above parameters can also interact with temperature, low temperature resulting in poor nutrient absorption and lower retention of dietary phosphorus, while high temperatures increase metabolic rate and nutrient requirements, potentially leading to increased feed intake and pollution (Preston & Lamb, 2021; Jaworski et al., 2020).

Water temperature also affects the expression of genes related to nutrient absorption in the rainbow trout intestine (Volkoff & Rønnestad, 2020). The relationship between temperature and gene expression in fish intestine is complex and influenced also by diet, fish size, and water quality. High-protein diets and higher water temperatures influence specific gene expression related to amino acid and glucose transport (Hassan et al., 2019; Jaworski et al., 2020).

3. Current and Potential strategies for reducing phosphorus pollution of FW fish farms

3.1. Phosphorus Waste Reduction Initiatives

Technological developments are playing a significant role in reducing the ecological impact of open flow aquaculture, driving changes in water treatment methods for aquaculture systems, including addressing phosphorus pollution. Several advancements have emerged to improve water treatment efficiency and minimize the environmental impact of aquaculture effluents (Martins et al., 2021). Open flow aquaculture systems may face greater challenges in managing and controlling phosphorus pollution compared to closed Recirculating Aquaculture Systems (RAS), nevertheless, there are some similar principles of mechanical and biological filtration which can be applied to treat aquaculture effluents and minimize downstream phosphorus pollution. These include sedimentation and settling ponds which can reduce the organic load and particulates from the effluents before they are discharged downstream in the aquatic ecosystem. The construction of wetlands or the cultivation of algae downstream of open flow fish farms can offer natural filtration mechanisms to utilize vegetation and soil to filter and absorb nutrients, including phosphorus, from the effluents. By promoting the growth of specific algae or plants, phosphorus can be effectively removed from the aquaculture effluents (Khairunisa et al., 2022).

Nutrient management and feed optimization is also a highly effective method for reducing phosphorus pollution in fish farms (Cho & Bureau, 2001) and the industry has made significant strides in this regard; the level of phosphorus in fish feeds has been significantly reduced and feeding regimes have been optimized to reduce the phosphorus content of aquaculture wastes (Sugiura et al., 2006; Huang et al., 2019). For example, supplementary feeding with cereals, such as wheat and other grains, is commonly practiced in semi-intensive aquacultural ponds for species like common carp. Cereals provide a low cost and readily available source of energy in fish feeds, but they contain antinutritional substances, including enzyme inhibitors, phytoestrogens, and oligosaccharides, which can reduce feed intake and nutrient bioavailability. These factors hinder phosphorus digestion and utilization, leading to slower growth and increased excreta in the water. Heat treatment, grinding, and removal of hulls can mitigate the impact of antinutritional factors and improve feed digestibility. Furthermore, the use of pelleted or extruded feeds enhances digestibility, minimizes water pollution, and promotes better fish growth. Applying thermal and mechanical treatments to supplementary feeds prior to their use in aquaculture ponds can help reduce undigested or poorly digested feed, further improving efficiency and decreasing environmental impacts (Hlaváč et al., 2014;2016)

As a result, significant progress has been made in the past through the implementation of phosphorus waste reduction initiatives that have been developed and refined over the years (MacMillan et al., 2003; Muhammetoglu et al., 2022). These initiatives have relied on the application of best management practices, optimization of feeding regimes, and the utilization of low-phosphorus feed ingredients. Moreover, promising results have been reported by incorporating feed supplements such as α -ketoglutarate (Ali et al., 2019) phytase enzymes (Lee et al., 2020; Priya et al., 2023), organic acids (Pandey & Satoh, 2014), and low-phosphorus plant protein combinations (Sarker et al., 2011). These strategies offer promising avenues to not only reduce phosphorus waste but also optimize fish growth and foster sustainable freshwater aquaculture practices. Likewise, exogenous enzymes can serve as a safe and efficient bio-additive to regulate various aspects of fish performance and reduce phosphorus pollution into the environment (Zheng et al. 2020) . It can be concluded that incorporating exogenous enzymes into fish feed has the potential to improve growth performance, digestibility, feed utilization, whole-body composition, immune performance, and subsequently reduce phosphorus pollution in open flow freshwater fish farms.

3.2. Management Strategies for Reducing Phosphorous Pollution from Aquacultures

In fact, aquaculture – environment interaction is an interesting paradox. On one hand, aquaculture effluents containing excess nutrients into surrounding waters contributes to phosphorus release with detrimental effects on ecosystems' ecological state. Aquaculture, on the other hand, is susceptible to the effects of eutrophication, as excessive phosphorus levels can disrupt the ecological balance and the water quality, fish health and growth. Recognizing this paradox, legislative initiatives are being implemented to address phosphorus pollution from various sources, including aquaculture and agriculture, to mitigate environmental impacts and promote sustainable aquaculture practices (Brownlie et al, 2021).

As a result, there are a variety of emerging or refined management strategies that can be used to reduce phosphorus pollution from aquaculture, including developing existing and new approaches such as substituting fish meal with plant proteins and also reducing the amount of phosphorus in the feed (Milián-Sorribes ety al., 2021), optimizing feeding regimes to reduce feed conversion ratio (FCR), using water treatment technologies to remove phosphorus from wastewater, and developing sustainable aquaculture practices that reduce the environmental impact of fish farming (Sindilariu et al., 2007; Stojanović et al., 2019; Luo, 2023; True et al 2004). However, the feasibility and effectiveness of these strategies depend on various factors, such as the type of aquaculture system, the type of fish being farmed, and the local environmental conditions. Phosphorus removal in open flow aquaculture systems within rivers is a critical concern for maintaining water quality and mitigating environmental impacts. Emerging solutions such as phytoremediation and adsorbents/filtration offer promising approaches to address phosphorus pollution. These methods are based on aquatic plants, mechanical

and biological filters to remove excess phosphorus from the water (Alfeus & Gabriel, 2023). Phytoremediation in river aquaculture can also be based on the use of floating aquatic plants, such as water hyacinth (*Eichhornia crassipes*) and duckweed (*Lemna* spp.), which have demonstrated effective nutrient removal capabilities (Snow et al., 2008). These plants can be strategically placed in the aquaculture system or in constructed wetlands along the flow path of the river to help mitigate phosphorus pollution. Another example involves the utilization of effluent collected after wastewater treatment with *Rhodopseudomonas sphaeroides*. This effluent can be reutilized for microbial feed, medicament, and aquaculture water, specifically for the culture of Common Carp *Cyprinus carpio* (Xu et al., 2019). The integrated system of wastewater treatment and the use of effluent containing *R. sphaeroides* offers several benefits for Common Carp culture. Studies have shown that Common Carp raised in effluent containing *R. sphaeroides* exhibit improved survival rates, increased yield, and enhanced whole-body composition compared to control groups and this effect is attributed to the presence of B vitamins in the effluent with *R. sphaeroides*, which enhance the activity of various enzymes and genes related to digestion, immunity, and antioxidant defense mechanisms (Xu et al., 2019). Furthermore, the presence of *R. sphaeroides* in the effluent contributes to the improvement of aquaculture water quality, leading to reduced water pollution and wastewater discharge.

Adsorbents and filtration systems are effective approaches for mitigating phosphorus in river-based aquaculture and reducing eutrophication impacts. Modified clays or activated carbon, acting as adsorbents, can bind to phosphorus particles in water, facilitating their removal. Similarly, filtration systems equipped with specific media or membranes can capture phosphorus particles. Zeolites, for instance, have demonstrated potential in removing phosphorus from aquaculture effluents (van Rijn et al., 2006). Additionally, biomaterials derived from lodgepole pine have been utilized to reduce aquaculture waste and mitigate micronutrient-induced eutrophication. Treating rainbow trout effluents with these biomaterials for up to 60 minutes resulted in the removal of 150 to 180 grams of phosphorus per metric ton, providing a method for eutrophication reduction in aquaculture (Bare et al., 2023). The economic costs associated with these strategies can be a determinant of their potential applications in aquaculture. It is important to conduct thorough economic feasibility studies and cost-benefit analyses specific to each aquaculture operation to determine the financial viability and return on investment of these solutions. Factors such as potential cost savings from reduced water pollution, improved fish health, and regulatory compliance should also be considered.

3.3. The role of Probiotics

Efforts to reduce the organic load of fish farms can utilize probiotics which can affect phosphorus dynamics released by fish farms through their interaction with the intestinal microbiota of farmed fish. By incorporating probiotics into the fish diet or introducing them into the water column, it is possible to modulate the composition and activity of the gut microbiota, thereby enhancing the digestive capacity of fish in relation to phosphorus assimilation and utilization. Probiotics, when added to the fish diet or introduced into the water column, can alter gut microbiota and enhance the digestive capacity of fish. As a result of the improved functionality of the intestinal epithelium and enhanced nutrient transport mechanisms facilitated by probiotics, nutrients, including phosphorus, are assimilated more efficiently. This enhanced assimilation leads to a reduction in phosphorus wastes, which is a critical issue in freshwater fish farms due to its environmental impact (Tahar et al., 2018; Mavraganis et al., 2021; Nathananailides et al., 2021). By increasing the efficiency of nutrient utilization, the amount of phosphorus excreted into the environment can be minimized. Following probiotic administration, the gut microbiota can contribute to enhance nutrient utilisation of feed components and synthesize vitamins and amino acids, which can improve the nutritional value of the feed and enhance the digestion and absorption efficiency of nutrients. Several studies have shown that probiotics and prebiotics, which can promote the growth of beneficial gut bacteria, can improve the feed conversion efficiency and growth performance of fish. Certain strains of probiotic bacteria, when administered to aquaculture systems, have shown promise in improving phosphorus

utilization and assimilation and reducing its release into the surrounding water (Jahangiri & Esteban, 2018; Nathanailides et al., 2021).

Additionally, probiotics can also promote the growth of beneficial bacteria in the gut of fish, leading to enhanced nutrient absorption and utilization. This can result in improved feed conversion efficiency and reduced waste production, including phosphorus excretion. However, the effectiveness of probiotics in reducing phosphorus pollution can vary depending on several factors, such as the specific probiotic strains used, the aquaculture system's characteristics, and the feed composition.

Apart from the traditional method of administering probiotics through diet, they can also be introduced into the aquatic environment, either by adding them to the water column or incorporating them into filtration systems (Jahangiri & Esteban, 2018; Nathanailides et al., 2021). This alternative approach allows probiotics to exert their effects on gut function and directly interact with the aquaculture water and sediment, potentially enhancing their remediation effects. For example, a study by Yi et al. (2021) investigated the use of commercial probiotics immobilized on different carriers for aquaculture water and sediment remediation. Probiotics immobilized within oyster shells, vesuvianite, and walnut shells reduced nutrient content of aquaculture water and sediment. Likewise, through competitive exclusion, the application of a mixture of probiotics, such as lactic acid bacteria, phototrophic bacteria, and yeast, can inhibit the growth of pathogenic and harmful bacteria in fish farms, as well as reduce phosphorus wastes. Jówiakowski et al. (2009) reported a significant decrease (77.6%) of phosphorus concentrations in the water from the aquaculture pond following the application of a mixture of probiotics. These findings suggest that probiotics can not only function as dietary components but also contribute to bioremediation efforts, ultimately improving water quality parameters and reducing nutrient loads in aquaculture effluents. However, further research is still needed to optimize the use of probiotics for phosphorus management in freshwater aquaculture, as their effectiveness can vary depending on factors such as probiotic strains, aquaculture system characteristics, and feed composition.

4. Conclusion and Perspectives

Aquaculture, particularly open flow fish farming, poses a significant environmental challenge due to the release of nitrogen and phosphorus into waterways, leading to eutrophication. Global legislation is in place to protect and restore freshwater and marine ecosystems affected by aquaculture. In the case of open flow fish farms, phosphorus-rich feeds contribute to phosphorus pollution through the release of uneaten feed, feces, and metabolic waste. To address this issue and enhance aquaculture sustainability, a comprehensive and integrated approach to aquaculture management is essential. For example, the development of new fish feed formulations can have an impact on phosphorus absorption in fish (Rosenthal, 1994 Wilfart et al., 2023). Increased efficiency of phosphorus absorption can lead to a decrease in the amount of phosphorus wasted in both soluble and solid forms (Igwegbe et al., 2023). In other words, if the fish can absorb most of the dietary phosphorus, there will be less phosphorus excreted as waste.

Phosphorus is an essential nutrient required for various physiological processes, including bone formation, energy metabolism, and cellular functions. However, phosphorus availability and absorption in fish can be influenced by the composition of the feed. New feed formulations can be designed to optimize phosphorus utilization and absorption in fish. Another important aspect is managing gut health in farmed fish. A healthy gut microbiome is essential for efficient nutrient absorption and digestion. Probiotics, for example, can enhance the gut health of fish and improve their ability to retain and utilize nutrients effectively. By maintaining a balanced and diverse gut microbiota, fish can optimize nutrient absorption, reducing the amount of undigested phosphorus that is excreted and released into the surrounding water. Likewise, gut inflammation commonly observed in farmed fish, can disrupt the efficient absorption of phosphorus and increase the potential for phosphorus pollution. Inflammation of the fish gut, often associated with poor water quality, dietary imbalances, or infectious agents, can compromise the structure and function of the intestinal lining. Inflammatory processes can lead to changes in the morphology and functionality of the fish

intestine, including the villi and microvilli. Disruption of these structures reduces the surface area available for nutrient absorption, including phosphorus. Additionally, gut inflammation can alter the expression and activity of nutrient transporters in the intestinal epithelium, including those responsible for phosphorus absorption. The downregulation or impaired function of sodium-dependent phosphate transporters, such as NaPi-2b, can hinder the active transport of phosphorus across the intestinal cells. As a result, the absorption efficiency of dietary phosphorus decreases, leading to a higher excretion of unabsorbed phosphorus in fish waste. Overall, intestinal phosphorus absorption in fish is a complex process influenced by enzymes, transporters, and factors such as pH, calcium, and vitamin D. Anti-nutritional factors, such as phytic acid, can also affect phosphorus availability. Techniques like phytase supplementation and appropriate processing methods can mitigate these factors and improve phosphorus utilization. In conclusion, reducing phosphorus pollution from open-flow fish farms requires a holistic approach that encompasses various aspects of aquaculture. By optimizing feed composition, incorporating functional feed ingredients, and monitoring gut health parameters of farmed fish, the issue of phosphorus pollution of aquaculture can be reduced.

The data reviewed in the previous sections indicate that phosphorus retention efficiency of different farmed fish species can vary due to factors such as their physiological characteristics, feeding habits, and digestive physiology. The phosphorus requirements and absorption mechanisms can also differ among fish species, with some species having specialized adaptations in their intestines. Environmental factors like water temperature can affect phosphorus retention efficiency, as can the composition and formulation of the feed. Different fish species may respond differently to specific feed formulations, affecting their phosphorus retention efficiency. While several methods and strategies can contribute to reducing phosphorus pollution from fish farms, a holistic approach encompassing various factors should be considered. Cost analysis, proper feed management, water quality monitoring, and nutrient cycling play pivotal roles in effectively addressing phosphorus-related environmental concerns. By understanding the mechanisms of phosphorus absorption, dietary factors, anti-nutritional substances, and intestinal morphology, we can optimize aquaculture practices to reduce phosphorus release.

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