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Effects of Dietary Vitamin C on the Growth Performance, Antioxidant Activity and Disease Resistance of Fish: A Review

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Abstract:

Vitamin C is an essential micronutrient that cannot be synthesized by fish and must be present in fish diets for proper functioning of the physiological conditions. It is required for the biosynthesis of the collagen which is a pre-requisite for the formation of connective tissue and increases the absorption of iron in fish. It prevents various diseases; it is soluble in water and is easily oxidated by heat, light and metal. Most animals can generate vitamin C in sufficient quantities for normal growth and function, but many fish cannot because they lack the enzyme L-gulonolactone oxidase for its manufacture. Vitamin C facilitates the absorption of iron and is necessary for a maximum rate of immune responses and enables a good response to stressors. This updated review presents a general outline of the possible physiological function of vitamin C for fish, with an emphasis on the information on growth performance, antioxidant activity, immune response and disease prevention of fish as well as the synergistic effects of vitamin C with other micronutrients. The diets supplemented with vitamin C promote the growth performance, improve the structure of the intestinal mucosal epithelium, and have a positive impact on the hematological parameter. The addition of different dietary vitamin C to the basal diets significantly improved the growth performance, antioxidant activity, immune response and disease resistance of fish. vitamin C in the aquaculture, having a solid understanding of the positive functions and mechanisms that vitamin C possesses is of the utmost significance.

Keywords: *Vitamin C, Aquatic animals, Antioxidant, Physiological processes, Synergistic effects.*

Introduction

Vitamins are essential for the survival of fish due to their role as cofactors for enzymes. Vitamins facilitate the maintenance of optimal health and normal metabolic functions in organisms (Gasco et al., 2018; Gouda et al., 2020). Vitamin C, being a water-soluble vitamin, is of paramount importance in sustaining optimal physiological function and fish growth (Lim & Lovell, 1978; Ren et al., 2007; Shahkar et al., 2015; Dawood & Koshio, 2018; Ibrahim et al., 2020; Sandnes, 1992). Contrarily, VC has been shown to influence the formation of cartilage and bone, reproduction (Darias et al., 2011; Sarmento et al., 2020), normal growth and haematology, lipid metabolism, stress, the immune system, and interactions with other micronutrients (Kraus et al., 2004; Liu et al., 2019; Min et al., 2018). It has also been demonstrated that VC mitigates oxidative stress in fish, which is advantageous for their health (Caxico Vieira et al., 2018; Gao et al., 2013).

Teleost fish are incapable of synthesizing VC or L-ascorbic acid due to a mutation in the L-gulonolactone oxidase gene, which encodes the enzyme responsible for catalyzing the de novo synthesis process (Drouin et al., 2011). For this reason, farmed fish require VC-rich diets to promote optimal growth and the maintenance of other physiological functions. Inadequate VC supplementation would reduce enzyme activities, consequently leading to impaired growth performance, diminished survival rate, and increased vulnerability to fish diseases (Liang et al., 2017; Huang et al., 2011). Nonspecific symptoms may also manifest in the absence of dietary VC, including fluctuations in serum triglyceride and cholesterol concentrations (Deng et al., 2019). Extensive research has been conducted on the dietary VC needs of numerous farmed fish species. However, the optimal dietary VC levels vary from species to species and are influenced by growth rates, age, size, diverse environmental factors, and nutrient interrelationships (Zhou et al., 2012). Therefore, it is imperative to conduct an analysis of the dietary VC needs of a specific species of fish.Extensive documentation exists regarding the correlation between VC deficit and the pathogenesis of various disease processes (Clijsters et al., 2020; Xu et al., 2016). Nevertheless, it is critical to acknowledge that a significant proportion of commercially farmed aquatic organisms, including crustaceans and fish, have a limited capacity for endogenous synthesis of vitamin C. This is primarily attributed to the lack of the enzyme L-gulonolactone oxidase. Hence, it has been recommended that individuals incorporate vitamin C supplementation into their dietary plans as a viable approach to mitigate the risk of VC deficiencies or fulfil particular nutritional requirements (Liang et al., 2017; Daniel et al., 2021). The utilization of exogenous compounds possessing antioxidant properties, such as vitamins C and E, can aid in safeguarding cells and tissues from oxidative stress by scavenging free radicals (Winston and Digiulio, 1991). VC is a water-soluble antioxidant and a vital nutrient that is involved in numerous physiological processes in fish. Additionally, VE functions as a widely recognized antioxidant, providing protection against oxidizing agents (Tocher et al., 2002). Venous endothelial depletion occurs in animals due to the production of free radicals during lipid peroxidation (McDowell, 1989). A review of the literature reveals that the amount of VE required by fish varies in response to dietary lipid peroxidation (Zhong et al., 2008). Moreover, supplementing fish with higher dosages of VE can impede lipid peroxidation and enhance their antioxidant capacity. Multiple studies have documented an interactive relationship between VC and VE in preventing lipid peroxidation (Gao et al., 2013). Furthermore, Lee and Dabrowski (2003) have found that VC can preserve VE in a variety of fish species.

Ongoing investigations into the application of VC in aquaculture have been prompted by these inquiries into the potential advantages of VC for fish growth, antioxidant activity, and disease resistance (Liu et al., 2021; Kaur et al., 2022). As a result, a comprehensive evaluation is required to consolidate and analyze the numerous information sources.

Potential synergistic Effects of Vitamin C and Other Supplements

In the realm of research on feeding stimulation, inosine and inosine monophosphate (IMP) have garnered significant attention as particular nucleotides. However, their primary focus lies in their potential as functional nutrients for the growth and health benefits of aquatic species.

Both inosine and vitamin C are significant factors in the growth performance, feed consumption, and health condition of fish. Research has been conducted in the domain of aquaculture to explore the potential application of inosine as a feed attractant. According to Hossain et al. (2018), this particular practice has been observed to have the capacity to induce many physiological effects in a wide range of species. These effects include promoting development, enhancing the immune response, increasing tolerance to oxidative stress, and influencing the architecture of the intestines.

Inosine is a purine nucleoside that consists of the base hypoxanthine and the sugar ribose. It is naturally present in transfer RNAs. Inosine is generated through the process of deamination of adenosine, rendering it a recognized functional nutrient. The primary focus of study in aquaculture has been on the use of inosine as a particular nucleoside for stimulating feeding behavior in aquatic species, rather than exploring its potential as a functional nutrition for promoting growth and enhancing health benefits.

However, several studies have indicated that the inclusion of dietary inosine or inosine monophosphate (IMP), either alone or in combination with specific free amino acids, can have positive effects on the growth performance, survival, and feed intake of juvenile Eel (Takeda et al., 1984), Red sea bream (Hossain et al., 2016a). Additionally, it has been observed that the supplementation of inosine or IMP can enhance disease resistance and immune responses in aquatic species such as Japanese flounder (*P. olivaceus*), as evidenced by increased lysozyme activity and nitro-blue-tetrazolium activity (Song et al., 2014).

Furthermore, in Grouper (*E. malabaricus*), the inclusion of inosine or IMP has been found to enhance the production ratio of head kidney leucocyte superoxide anion (O_2^-), indicating improved immune function (Lin & Shiau, 2005). Moteki et al. (2012) have reported that a considerable proportion of fish species demonstrate inadequate proline synthesis, a chemical that plays a pivotal role in the cellular metabolism and physiological activities of aquatic animals.

Tyrosine amino acid oxidation and phenylalanine are processes in which vitamin C serves as a crucial coenzyme (Brander and Pugh, 1977). By facilitating protein synthesis, vitamin C can enhance growth rate and weight gain (Andrade et al., 2007; Faramarzi, 2012). Antioxidants such as ascorbic acid (Vitamin C) and α-tocopherol (Vitamin E) are critical for the survival of the majority of cultured fish species. Furthermore, it has been established that these vitamins are critical nutrients that influence the immune system, growth, and survival of fish, among other things (Blazer and Wolke, 1984; Li and Lovell, 1985; Waagbo, 1994; Chen et al., 2003; Lin and Shiau, 2004). Vitamin E plays a crucial role in fish immunity (Waagbo, 1994) as it safeguards macrophage membranes against peroxidative damage caused by free radicals (Beharka et al., 1997).

The antioxidative properties of vitamins C and E in the face of free radical oxidation have been extensively documented (Hunter and Willet, 1994). Several research studies have suggested that vitamin C promotes growth by inhibiting muscle atrophy and collagen tissue injury (Wang et al., 2003; Chen et al., 2004). Vitamin C, a potent antioxidant, safeguards numerous fish tissues, including red blood cells, from oxidative injury (Sahoo and Mukherjee, 2003). Dietary vitamin C is implicated in various functions of the immune system, such as facilitating macrophage activity, promoting cell proliferation, stimulating natural killer cell activity, and regulating complement and lysozyme activities (Chen et al. 2003; Lin and Shiau 2004).

Vitamins, which are organic compounds, are essential for the maintenance and proliferation of living organisms, including fish (Lin and Shiau, 2004). The natural cytotoxic activity of leucocytes was enhanced by high dietary levels of vitamin C (Cuesta et al., 2001). Previous research has examined the potential synergistic effects of vitamin C and E on certain fish species, including Nile Tilapia, *Oreochromis niloticus* (Kim et al., 2003). The growth performance, feed consumption, and variety of intestinal microbiota in aquatic animals were similarly enhanced by the inclusion of dietary β-glucan (Dawood et al., 2018; El-Murr et al., 2019). β-glucans (BGs) are a type of biological modifiers composed of D-glucose monomers that are connected by β-glycosidic linkages. These BGs have been widely recognized for their capacity to stimulate the immune system, as evidenced by studies conducted by Ringø et al. (2012) and Volman et al. (2008). Ascorbic acid, also known as vitamin C, is a crucial element that must be included in the dietary intake of finfish due to their inability to produce L-gulono-γ-lactone oxidase, an enzyme important for the manufacture of vitamin C (NRC, 2011).

The role of vitamin C in several parts of the immune system has been extensively studied. Key components of the immune system, such as leukocytes and lymphoid organs, have been found to store much higher quantities of vitamin C compared to circulating plasma (Volman et al., 2008). According to Carr and Maggini (2017), there exists a hypothesis suggesting that vitamin C possesses antioxidant qualities that have the potential to augment immunological cells. This is achieved by the scavenging of free radicals generated as a result of regular metabolic processes, exposure to stress, and the respiratory burst that occurs during immunostimulation. Ultimately, the antioxidant properties of vitamin C serve to safeguard these cells from oxidative harm.

The utilization of β-glucans in aquafeeds has been extensively studied and has shown significant positive effects on both humoral and cellular immune responses, as well as growth performance improvement. The linear β-1,3-glucan known as paramylon shows potential and can be obtained from microalgae (*Euglena gracilis*). These microalgae can be cultivated using waste-water derived from the agriculture industry (Barsanti et al., 2011). The potential additives for aquaculture have been shown to be crude *Euglena* sp. (Das et al., 2009) and paramylon (Skov et al., 2012; Yamamoto et al., 2020). These additives have been demonstrated to enhance the immunological responses of certain farmed fish species and increase the stress tolerance of *Artemia* sp. (Vismara et al., 2004). Therefore, they offer benefits for the aquaculture industry. These molecules are commonly present in the cell walls of bacteria, yeast, and fungi. Upon interaction with fish leukocytes, their pattern recognition receptors (PRRs) identify the molecular structures of these compounds as foreign antigens. This recognition triggers an immunostimulatory response through an inflammatory cascade, leading to the production and release of cytokines (Volman et al., 2008). In addition, it has been observed that vitamin C has the ability to facilitate the regeneration of oxidized vitamin E within the cytoplasm of cells, hence augmenting the antioxidant capacity at an intracellular level (Petit & Wiegertjes, 2016).

The utilization of β-glucan as a microelement in aquafeed has been documented for its ability to enhance the immune system, specifically in terms of humoral immunity and antioxidative properties (Dawood and Koshio, 2016; Pilarski et al., 2017). According to Narra et al. (2015), the introduction of β-glucan in the body enhances the generation of lymphocytes, hence enhancing the organism's capacity to combat infections by facilitating the synthesis of antibodies. In addition to its role as an antioxidant, β-glucan has the capacity to safeguard the body tissues of aquatic organisms against oxidative harm by the elimination of free radicals within the organism (Pietretti et al., 2013).

Moreover, the hepatoprotective effect of β-glucan is mediated by the modulation of alanine aminotransferase (ALT), aspartate aminotransferase (AST), alkaline phosphatase (ALP), triglyceride, total serum cholesterol, and total protein concentrations. According to Dawood et al. (2018), the inclusion of dietary β-glucan resulted in enhanced growth performance, feed consumption, and increased variety of intestinal microbiota in aquatic animals.

Advantages of vitamin C in Aquaculture

According to Blazer (1992), there is evidence to suggest that elevated dietary levels of vitamin C have a significant impact on the immune system functioning in fish populations in perspective of antioxidant efficiency. Several studies have documented the distinct impacts of vitamin C on non-specific resistance mechanisms and the particular immune response in fish (Hardie et al., 1991; Verlhac and Gabaudan, 1994; Ortuno et al., 1999). Vitamin C, also referred to as VC, is recognized as a potent nonenzymatic antioxidant that plays a crucial role in the removal of reactive oxygen species. These species, if left unattended, have the potential to combine with nitric oxide and give rise to a highly reactive free radical called peroxynitrite (Gordon et al., 2020). Moreover, vitamin C functions as a protective agent against oxidative damage to cells.

According to Dawood and Koshio (2018), the role of vitamin C as an antioxidant involves the transfer of electrons to free radicals, effectively neutralizing their detrimental impact and inhibiting potential injury. Presently, there is a significant body of evidence that substantiates the direct participation of vitamin C in the antioxidative mechanisms of aquatic species (Yin et al., 2020; Wang et al., 2016). For example, previous research conducted by Yin et al. (2020) and Liu et al. (2021) have shown that the application of vitamin C can effectively alleviate the detrimental effects of cadmium (Cd) and lead (Pb) toxicity in Grass Carp kidney (CIK) cells. Moreover, Wang et al. (2016) revealed that Sea cucumber has also exhibited comparable beneficial outcomes. Moreover, the inclusion of vitamin C as an antioxidant in the diet of *Rhamdia quelen* has promise for offering additional safeguard against pesticide exposure (Gomes et al., 2022).

Falahatkar et al. (2006) found that the administration of dietary vitamin C had a minimal impact on mortality rates in juvenile great Sturgeon (*Huso huso*). The study also examined the effects of vitamin C on growth, blood parameters, body chemical composition, and alkaline phosphatase (ALP) activity. The study conducted by Shalaby (2009) demonstrated that the presence of ascorbic acid (vitamin C) had an opposing effect on the toxicity of Ochratoxin in Nile Tilapia (*Oreochromis niloticus*). The results indicated a significant decrease in the total erythrocyte count, haemoglobin content, and haematocrit value in fish that were exposed to either low or high levels of ochratoxin. However, when vitamin C was administered, the blood parameters of the fish exposed to both levels of ochratoxin were enhanced and approached values similar to those of the control fish.

The study conducted by Alam et al. (2009) investigated the impact of vitamin C in formulated feed on the feed, growth, and body composition of *Heteropneustes fossilis*. The results revealed a notable favorable effect of vitamin C on the growth, feed utilization, and body composition of the species. In a study conducted by Moein (2012), the author examined the effects of vitamin C on the weight of Common carp (*Cyprinus carpio*).

The findings of the study indicated a strong relationship between vitamin C levels and the weight of the Common carp. Multiple studies have shown evidence that the combination of vitamin C and VC with chitosan offers protection to fish against the harmful effects of $CdCl_2$ on various biochemical markers in liver tissues. In their study, Sharifinasab et al. (2015) examined the potential of Vitamin C and Chitosan to mitigate the toxic effects of paraquat on several biochemical parameters in hepatocytes of Common carp. The researchers found that the administration of vitamin C was successful in lowering the liver toxicity induced by paraquat.

However, the administration of a combination of vitamin C and chitosan has been found to be more effective and exhibit a synergistic impact. They have the potential to serve as a hepato-protective agent in mitigating paraquat-induced hepatotoxicity in fish. The study conducted by Asaikkutti et al. (2016) aimed to assess the impact of varying levels of dietary vitamin C on *Macrobrachium malcolmsonii*. The researchers suggested that a supplementation of 100 mg/kg of vitamin C could be beneficial for enhancing the antioxidant defense system and promoting the production of *M. malcolmsonii*. The absence of gulonolactone oxidase, an enzyme responsible for the production of vitamin C in the liver and kidney of several fish species (Dabrowski, 1990; Fracalossi et al., 2001), necessitates the presence of vitamin C in their food in order to fulfill their nutritional needs and achieve optimal growth performance (Dabrowski, 1990).

In the study conducted by Gannam and Schrock (2001), it was shown that the efficacy of immunostimulants can be influenced by a singular dietary ingredient, such as vitamin C. Therefore, it is imperative to thoroughly examine the stimulatory properties of vitamin C, particularly in relation to its interaction with other components. Vitamin C is a hydrophilic vitamin primarily involved in the synthesis of pro-collagen, growth regulation, immune system modulation, embryonic development, vulnerability to bacterial infections, and reproductive processes, among other physiological roles (Kumari and Sahoo, 2005; Zhou et al., 2012). Vitamin C exhibits a robust antioxidant activity when combined with vitamin E (α-tocopherol), effectively neutralizing reactive oxygen species (ROS) within the tissue. At lower quantities, reactive oxygen species (ROS) can have beneficial effects or even play a crucial role in various biological processes, such as the immune response against microorganisms, by enhancing the activity of phagocytes in killing bacteria. According to previous studies (Paperna et al., 1980; Tixerant et al., 1984), a deficit of vitamin C in fish has been associated with an alteration in the metabolism of tyrosine. This variation leads to the precipitation of tyrosine in tissues, which is believed to be responsible for the development of granulomas. The hypothesis suggests that an imbalance of dietary minerals in Rainbow trout (*Oncorhynchus mykiss*) may lead to the formation of granulomas with an unknown cause. This, in turn, could result in the precipitation of calcium phosphate or calcium carbonate (Dunbar and Heramn, 1971). Recently, the presence of granulomas with an unclear cause has also been observed in Atlantic salmon (Salmo salar) (Good et al., 2015).

In recent decades, there has been a growing body of evidence supporting the effectiveness of vitamin C as a dietary supplement in many aquatic animal species. This evidence has demonstrated its capacity to enhance growth and regulate physiological balance. The incorporation of dietary vitamin C at suitable concentrations has demonstrated a noteworthy beneficial effect on diverse growth indicators in aquatic organisms, including Red Sea Bream (*Pagrus major*) (Dawood et al., 2017; Hossain et al., 2017a), Freshwater Prawn (*Macrobrachium malcolmsonii*), Nile Tilapia (*Oreochromis niloticus*) (Ibrahim et al., 2020), and Common Carp (*Cyprinus carpio*) (Ghafarifarsani et al., 2022). However, a significant discrepancy existed in the optimal dosage of vitamin C that was incorporated into the dietary regimen of aquatic creatures.

The considerable variation in requirement values can be explained by metabolic differences between species, variations in developmental stages, differences in cultivation conditions, the presence of interacting nutrients in the experimental diets used for evaluation, and the duration of the trials (Dawood and Koshio, 2018; Xu et al., 2022). Previous research has indicated that the improved growth performance of aquatic animals may be linked to greater feed efficiency as a result of dietary supplementation with vitamin C (Ibrahim et al., 2020). Moreover, the consumption of vitamin C might exert a noteworthy influence on nutrient intake due to its vital role in protein metabolism, namely in the process of collagen production. According to Yusuf et al. (2021), collagen has a crucial role as a structural foundation for cells, tissues, and organs. Previous research has suggested that vitamin C may play a role in the metabolic pathway of carbohydrates, namely aiding in the conversion of glucose into energy in aquatic organisms (Krasnov et al., 1999). Furthermore, it is widely hypothesized that this particular vitamin exerts an influence on the overall trajectory of growth performance. The potential influence of venture capital vitamin C on the enhancement of growth performance can be attributed to its capacity to augment the metabolic functionality of aquatic organisms.

In addition, previous studies have demonstrated the positive effects of dietary vitamin C intake on multiple physiological factors, such as body composition, digestive enzyme activities, somatic parameters, condition factor, viscerasomatic index, hepatosomatic index, spleen somatic index, and head kidney somatic index (Kong et al., 2021).

As immunostimulant vitamin C plays a role in enhancing immune protection by facilitating many cellular processes within the innate and adaptive immune system (Carr and Maggini, 2017). Several studies have shown evidence for the beneficial impact of vitamin C on immunological responses, including enhancements in phagocytic and lysosomal function, respiratory burst and complement activity, as well as serum protein levels (Liang et al., 2017; Daniel et al., 2021; Zehra & Khan, 2021). In a study conducted by Daniel et al. (2021), it was observed that the introduction of a stable variant of vitamin C, specifically L-ascorbyl-2-polyphosphate, into the diet of Striped Catfish (*Pangasianodon hypophthalmus*) at a dosage of 350 mg kg^{-1} led to notable enhancements in respiratory burst activity, phagocytic activity, and serum lysozyme activity.

The positive correlation between the dietary level of vitamin C and the levels of serum alkaline phosphatase, total serum protein, and lysozyme activities was observed in fingerling of *Channa punctatus* (Zehra and Khan, 2021). Vitamin C may also have an influence on the immunoglobulin M (IgM) levels, which play a crucial role in mediating humoral adaptive immunity in aquatic organisms. In addition, vitamin C has the potential to promote the differentiation and proliferation of T-cells and B-cells, leading to an increased production of immunoglobulin M (Carr and Maggini, 2017; Kim et al., 2019).

At the molecular level, the consumption of vitamin C in the diet has been observed to influence the expression of genes associated to the immune system in several species of aquatic animals (Zhang et al., 2022a, b; Sahoo et al., 2020; Cheng et al., 2018b, b; Dawood et al., 2020). Vitamin C has a significant function in regulating the immune defense system and enhancing the defense mechanisms of aquatic animals against infections.

As the enhancement of reproductive organs, numerous studies have reported that vitamin C possesses the ability to autonomously stimulate diverse cellular pathways involved in the regulation of cell cycle, resulting in enhanced cell division and proliferation across different species (Francis et al., 2012). The requirements for vitamins may be affected by intrinsic factors to the animal (growth rate, body size, species, reproductive status), diet composition and interaction with other nutrients and are also influenced by the production system (Sanches et al., 2010; Combs, 2012).

The gonad histological analysis revealed a notable rise in spermatogonia count in Nile Tilapia when dietary vitamin C was increased (Navarro et al., 2012). This observed effect may be attributed to the protective role of vitamin C in safeguarding germ cells against DNA damage and the oxidation of seminal plasma proteins caused by reactive oxygen radicals. Additionally, vitamin C is known to influence collagen production by regulating prolyl hydroxylation, as highlighted in studies conducted by Perera and Bhujel (2021). The positive impacts of vitamin C on the reproductive performance of aquatic animals are commonly ascribed to its facilitation of hormone release and mitigation of oxidative stress

The several studies conducted by DiTroia et al. (2019) indicated that a shortage in vitamin C has a negative impact on the reproductive health. Specifically, it results in a decrease in the quantity of germ cells, a delay in the process of meiosis, and a reduction in fecundity. The involvement of vitamin C is of utmost importance in the biosynthesis of norepinephrine and epinephrine in aquatic organisms. Norepinephrine exerts direct stimulatory effects on the release of hypothalamic gonadotropin-releasing hormone, a factor closely associated with the reproductive development of animals (Francis, 2012). Betsy et al. (2021) observed an elevation in the gonadosomatic index (GSI) of Koi Carp (*Cyprinus carpio* L.) following the administration of feed enriched with 600 mg kg^{-1} of vitamin C. Martins et al. (2016) reported a comparable promotional impact of VC in Nile Tilapia (*Oreochromis niloticus*).

A diet free from or deficient in ascorbic acid reduces the availability of vitamin C in the ovary, reducing the number of eggs and their hatchability and increases both the number of larvae with deformities and the mortality rate (Montalvo et al., 2022). Some authors have reported the influence of vitamin C in the reproduction and performance of fish (Zhou et al., 2012; Gao et al., 2014).

As disease inhibitors in the field of aquaculture, it has been observed that Vitamin C has a beneficial impact on the prevention and management of diseases caused by pathogen infections (Le et al., 2021; Han et al., 2019). As reported by Le et al. (2021), the inclusion of vitamin C supplement in the dietary regimen of *Psammoperca waigiensis*, resulted in elevated levels of white blood cells and higher activity of plasma lysozyme. The dietary supplementation of vitamin C demonstrated a notable enhancement in the survival rate of Coral Trout when exposed to *Vibrio harveyi*, Striped catfish when challenged with *Aeromonas hydrophila*, and Nile Tilapia when challenged with *S. agalactiae*. This improvement can be attributed to the potent antioxidant properties and immune-stimulating capabilities of vitamin C (Daniel et al.,2021; Zhu et al., 2022). The dietary supplementation of VC also shown an enhanced immune protective effect against infection caused by the White Spot Syndrome Virus (Zuo et al., 2022). The vitamin C enhances the resistance of aquatic animals to pathogen attack is predominantly attributed to its antioxidant and immune-stimulating properties.

The use of chemotherapeutants for controlling diseases has been widely criticized for their negative impacts like accumulation of tissue residues, development of drug resistance and immunosuppression (Rijkers et al.,1980; Ellis,1988). Hence, there is an urgent need to look for ecofriendly disease preventative measures to promote sustainable culture of Indian major carps.

In order to reduce the risk of disease, the level of resistance to infection in the cultured organisms should be increased by the use of better feeds, vaccines, immunostimulants or by selective breeding for higher disease resistance (Raa et al.,1992). Vitamin C has been demonstrated to play an important role in the functioning of the immune system when supplied at dietary levels higher than standard doses in several fish groups (Blazer,1992). Specific effects of vitamin C on a variety of non-specific resistance mechanisms and the specific immune response have been reported in fish (Hardie et al., 1991; Verlhac and Gabaudan,1994; Ortuno et al.,1999).

In regulation of the intestinal microbial, several studies have reported that the presence of *Lactobacillus* and *Bacillus* in the digestive tract of aquatic animals increased, but the abundance of *Escherichia coli* dropped, as the dietary levels of vitamin C increased (Liu et al., 2011). Vitamin C is water soluble and is important in hormone, collagen, and carnitine synthesis by promoting iron ion absorption. It also contributes to the functioning of the immune system. Various studies on animal models have shown that vitamin C improves immune health and digestion and inhibits the growth of bacteria. The presence of intestinal bacteria plays a vital role in maintaining the nutritional and overall well-being of the host. However, in various disorders, there is often an imbalance in the composition of these microbial communities (Diwan et al., 2022; Fei et al., 2022). The existing body of research suggests that the manipulation of nutritional factors can have a positive impact on the composition and function of gut microbiota, hence offering potential therapeutic benefits (Hossain et al., 2017a).

Multiple studies have provided evidence that the supplementation of vitamin C has effects on the modulation of microbiota in persons who are in good health, resulting in various advantageous alterations in bacterial populations (Hossain et al., 2017b; Liao et al., 2023).

Conclusion

It has been well documented that Vitamin C is essential for growth, development, as well as reproduction in fish species. The mechanism of Vitamin C to regulate the intestinal microbiota and further affect the nutritional and health status of aquatic animals. Furthermore, there are differences in the requirement and metabolic level of Vitamin C for different species aquatic animal. Vitamin C is a hydrophilic compound which has been investigated by many researchers due to its important health properties and great antioxidant functions. In aquaculture, Vitamin C offers many benefits, such as improving growth performance, protecting cells from oxidative damage, enhancing immunity and reproductive. However, the effects of Vitamin C depend on the aquatic animal species and size, additive dosage, presence or absence of other micronutrients, which should be extensively studied. With the sustainable development of aquaculture, the benefits and application of Vitamin C need to be further investigated.

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