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Review Article

Effects of *Spirulina platensis* and *Chlorella vulgaris* on the Immune System and Reproduction of Fish

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ABSTRACT

This review briefly highlights previous studies on the effects of *Spirulina platensis* and *Chlorella vulgaris* on the health and reproduction of fish. These microalgae have diverse potentials. This study can be used as a stepping stone in advancing the aquafeed industry by formulating microalgae-based feeds. It can be made to specifically enhanced the health status of fish and its reproductive system through the supplementation and/or replacement of fishmeal or other plant proteins such as soybean meal. Hence, it could be more sustainable than depending on natural fish stocks. The usage of antibiotics and vaccines to solve the issue of disease outbreak in aquaculture, as well as the usage of hormones for the growth and reproduction of fish, can also be replaced by the usage of *S. platensis* and *C. vulgaris*. The inclusion of these microalgae in fish feed has affected hemathological parameters and survival in fish as it boosts the numbers of white and red blood cells and thus affecting the immunity-stimulating capacity in fish. Besides, these microalgae also affect the fecundity and survival of fish eggs and thus directly affecting the reproduction performance of fish. *Spirulina platensis* affects eggs production and survival in fish whereas *C. vulgaris* enhances oxidative stress that affects the reproduction of White rabbits. This review aimed to deliver the results on the research of *S. platensis* and *C. vulgaris* on the immunity and reproduction of various fish species.

Keywords: *Chlorella vulgaris*, immunity, microalgae, reproduction, *Spirulina platensis*

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INTRODUCTION

The increasing human population worldwide has pressured the natural stocking of fish. Hence, aquaculture industries are blooming all over the world to support the growing demand for protein sources. This industry also provides food security and directly responsible in developing the livelihood of less-privileged communities. Aquaculture industry is being enforced and promoted as an important tool that drives economic growth in Malaysia and has been enlisted in the National Key Economic Area (NKEA), which highlights the 16 agro-food's entry point projects (EPP) (Yusoff, 2015). Statistics from the Department of Fisheries Malaysia (DOF) (2018) reported that the production of freshwater aquaculture was 102,500 tonnes in 2017 and increased to 105,700 tonnes in 2018, while the production of brackish water aquaculture was 324,300 tonnes in 2017 and 290,900 tonnes in 2018. The rapid growth of the aquaculture industry, particularly fish farming, has raised a number of issues in the health management of fish in terms of fish immunity against disease outbreak and the continuous supply of good quality fish seeds in terms of fish reproduction and developments.

This scenario is often related to the use of chemicals in aquaculture for a successful production (Subasinghe, 2004). However, such application is not widely encouraged as it introduces several risks to the production system, environment, and human health (Melba & Rohana, 2008). Thus, chemotherapeutics has been replaced with other alternative sources that are more acceptable in aquaculture practises, such as adding microalgae in fish feed as supplements. Various forms of algal meal applications have been studied. Generally, microalgae are used as larval feed by some fish farmers, and different levels of inclusion are added in fish feeds to boost their beneficial effects on fish conditions (Brown, 2002). Several microalgae possess high protein, lipid, and carbohydrate contents. Besides, the biomass of microalgae is rich in proteins, and it can strive and compete fairly for its quality and quantity compared with regular food proteins, such as fish, soybeans, and eggs (Ejike et al., 2017). A handful of researches and studies had been carried out in the past to analyse the potential effects of microalgae in fish, and such attempts are still in progress with various developments on specific parameters in fish, such as the immune and health system as well as the reproductive system. In this review, the effects of *S. platensis* and *C. vulgaris* on the immunity and reproduction of fish are briefly elaborated.

POTENTIALS OF MICROALGAE IN AQUACULTURE

The growth of the aquaculture industry has gone hand in hand with the growth of the population worldwide to ensure that ample fish supply as a part of protein sources can be fulfilled. The aquaculture industry is expected to solve food security and nutritional well-being, reduce poverty, and develop the economy (Melba & Rohana, 2008). However, the focus on developing the aquaculture industry to fulfil such expectation has raised several issues in aquaculture. Among the most recognisable problem is the use of chemotherapeutants in aquaculture. Chemotherapeutics has benefits and is a key to a successful aquaculture production in activities, such as pond and tank constructions, feed formulation, growth promotion, health management, and booster of natural production (Melba & Rohana, 2008; Subasinghe, 2004). Nevertheless, the use of chemicals in aquaculture is not widely encouraged. Various researches and tests have been carried out to develop products that are friendlier to replace the use of chemicals in aquaculture. The organism that is known to be a more sustainable choice in replacing the use of chemotherapeutants in aquaculture is microalga. The growing interest in the multipurpose properties of microalgae has developed various microalgae applications in daily life. Microalgae are an alternative to sustainable aquaculture practices by playing roles in wastewater treatment and ingredient replacement in fish feed. The development of fish feed with the inclusion of microalgae is widely studied because of its encouraging effects on farmed fish. Microalgae can be used in aquaculture as feed, growth enhancers, and immunostimulants (Ahmad et al., 2018). However, microalgae must be easily cultured and nontoxic to be used in aquaculture (Spolaore et al., 2006). The criteria that are taken into consideration before incorporating microalgae as feed ingredients for fish are that they need to have the correct size, are easily ingested and digested by fish, and have high nutritional quality profiles (Brown et al., 1999; Renaud et al., 2002). Research on microalgae, such as *S. platensis* and *C. vulgaris*, their effects in enhancing the immunity and reproduction performance of fish and other organisms is continuous and evolving.

GENERAL INTRODUCTION

Spirulina platensis

Spirulina or *Arthrospira platensis* is a blue-green filamentous alga that inhabits freshwater bodies. Its name is acquired from its cylindrical shape with multicellular trichomes in an open left-handed helix (Figure 1) (Jung et al., 2019). Morphologically, *S. platensis* is a helicoid alga with a radial that is distinct from those of other species (Promya et al., 2008). This microalga has a protein content of up to 70% and is rich in vitamins, minerals, and essential fatty acids, such as linolenic and linoleic acids, and palmitic acid (Abdel-Tawwab & Ahmad, 2009). *Spirulina* has been used as a dietary supplement for a long time by the community that resides near the alkaline lakes where it is habitually found (Jung et al., 2019). Today, *Spirulina* is produced in numerous countries in Africa and America, such as Benin, Burkina Faso, Chad, Brazil, Chile, and Costa Rica, as well as Asian countries, such as Thailand, India, China, Vietnam, and Taiwan (Habib et al., 2008). However, the production of *Spirulina* is carried out in control conditions to avoid contaminations from other sources, such as blue-green algae, pesticides or heavy metal; thus, its general composition is affected by the location and type of production (Table 1) (Jung et al., 2019). The rich contents of this microalgae species made it an interesting ingredient for testing as feed in fish. Various studies have analysed the immunology, disease resistance, oxidative stress, and growth performance of a number of aquatic animals based on *S. platensis*-supplemented feeds (Abdel-Latif & Khalil, 2014; El-Sheekh et al., 2014; Kim et al., 2013; Macias-Sancho et al., 2014; Promya & Chitmanat, 2011; Teimouri et al., 2013; Yeganeh et al., 2015).

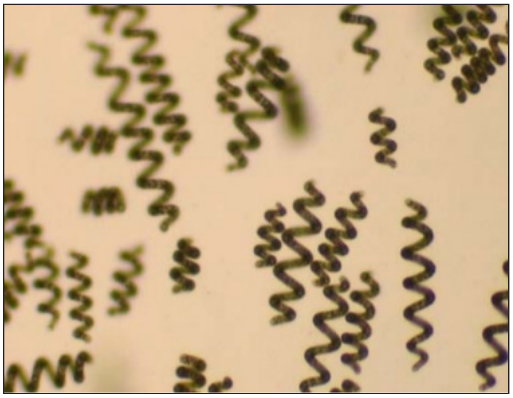

Figure 1. The microscopic view of *Spirulina*
Note. Adapted from "Earth food *Spirulina* (*Arthrospira*): Production and quality standarts", Retrieved June 01, 2020, from https://www.intechopen.com/books/food-additive/earth-food-spirulina-arthrospira-production-and-quality-standarts. Copyright 2012 by Koru. Adapted with permission

Table 1
The general composition of Spirulina*. Adapted from "*Spirulina platensis*, a super food?" (Jung et al., 2019)*

Components	Percentage (%)
Proteins	55-70
Carbohydrates	15-25
Lipids	6-8
Minerals	7-13
Humidity (dried algae)	3-7
Dietary fibers	8-10

Chlorella vulgaris

Chlorella vulgaris is a freshwater species and unicellular alga that contains a nutrient-dense super food, including various vitamins and minerals, 18 amino acids, and 60% protein (Khani et al., 2017). A report by Nick (2003) also stated that *Chlorella* possessed excess minerals, such as iron, calcium, potassium, magnesium, phosphorous, and 20 vitamins, such as pro-vitamin A, vitamins C, B1, B2, B2, B5, B6, B12, E, K; biotin, inositol, and folic acid. Morphologically, Beijerinck (as cited in Safi et al., 2014, p. 266) discovered *C. vulgaris* as the first microalga with a well-defined nucleus. *Chlorella* has a rigid cell wall that varies according to each growth phase and provides protection against invaders and harsh environment (Safi et al., 2014) (Figure 2). In addition,

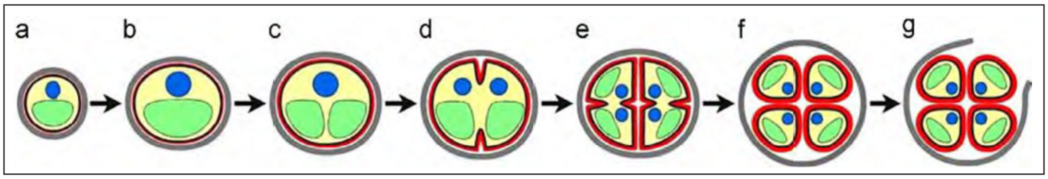

Figure 2. The growing phases of *Chlorella vulgaris*
Note. Adapted from "Morphology, composition, production, processing and applications of *Chlorella vulgaris:* A review" by C. Safi, B. Zebib, O. Merah, P. Y. Pontalier, and C. Vaca-Garcia, 2014, *Renewable and Sustainable Energy Reviews*, *35*, pp. 265–278. Copyright by Elsevier

this microalga has many fundamental components that are identical to actual plants, including cytoplasm, mitochondrion, and chloroplast. Besides, it is one of the microalgae that contain the highest quantity of chlorophyll compared with other plants (Raji et al., 2018). *Chlorella* has a unique phytonutrient property that is made up of vitamins, nucleic acid-related substances, amino acids, proteins, peptides, and sugars known as the *Chlorella* growth factor (CGF), which is found in abundance in the nuclei of the alga (Nick, 2003).

ENHANCEMENT OF FISH IMMUNE SYSTEM

Spirulina platensis

The properties of *S. platensis* that are rich in proteins and vitamins have a positive impact on the immunity of the animals that consumed it (Promya & Chitmanat, 2011). However, the effects of *S. platensis* on the immune system of fish may vary according to the levels of supplementation or inclusion. Promya and Chitmanat (2011) stated that a 5% supplementation of *Spirulina* in the feed resulted in higher red blood cell (RBC) and white blood cell (WBC) counts in African sharptooth catfish as well as advanced its immunity-stimulating capacity. The result is remarkedly different compared to a 3% supplementation of *Spirulina*, which gave a lower immunity in African sharptooth catfish. Besides, a feeding experiment on Nile tilapia with five levels of *Spirulina* inclusion (1.25, 2.5, 5.0, 7.5, and 10.0 g kg^{-1} diet) added to the basal diet showed that the RBC and WBC counts increased from 1.92×10^6 μL^{-1} to 2.54×10^6 μL^{-1} and 3.21×10^6 μL^{-1} to 4.02×10^6 μL^{-1}, respectively, as the *Spirulina* levels increased, although 5.0 g kg^{-1} *Spirulina* is the optimum amount that gave the highest specific growth rate, weight gain, and food conversion ratio with no substantial difference in the survival rate from one treatment to another (Abdel-Tawwab & Ahmad, 2009). These results showed that different levels of *Spirulina* inclusion in fish diet affect blood counts. Blood biochemical indexes can be an indicator that reflects the health status, physiological condition, metabolism, and immunity of fish (Zhou et al., 2001). In addition, alkaline phosphate (ALP) activity in fish is another parameter that is commonly used to study the immunity of fish. ALP is a non-specific phosphate hydrolase that is involved in fish metabolic regulation and plays a vital role in non-specific immune response in organisms (Huang et al., 2005). Lin et al. (2016) showed an increasing trend in the ALP activity of golden pomfret (*Trachinotus ovatus*) based on the 0%–6% supplementation of *S. platensis* in the fish diet and suggested that further experiment should be carried out for the ALP activity in fish. C-phycocyanin, a property of *Spirulina*, helps build immune capacity (Vonshak, 1997) and could be the reason for the enhancement of the immunity of fish based on the feeding trials with the inclusion or supplementation of *Spirulina*. Furthermore, a different approach of administering *Spirulina* through direct or oral method was also investigated. A test on carp (*Cyprinus caprio*) intubated with 0 (control), 1, 10, and 25 mg doses of *Spirulina* suspended in sterilised physiological saline (0.85% NaCl) and subjected to *Aeromonas hydrophilia* (strain MU9901) infection showed that the number of bacterial cells in the liver and kidney of carp treated with *Spirulina* were lower compared with those of the control groups at the treatment intervals of 4, 8, and 12 hours and at 1 and 4 hours post-bacterial challenge (Watanuki et al., 2006). Another challenge test using sturgeon (*Huso huso*) that was intraperitoneally injected with *Streptococcus iniae* (strain ATCC29178) after 8 weeks of feeding period with 0%, 2.5%, 5%, and 10% of *Spirulina* added to the basal diet showed that the cumulative mortality of sturgeon decreased with the increasing supplementation of *Spirulina* (Adel et al., 2016). These challenge tests suggested that *Spirulina* could activate leucocyte activities in fish and thus increased the resistance against bacterial infections. Leucocytes are involved in superoxide production, cytokine release, and phagocytosis (Watanuki et al., 2006). The effects of *Spirulina* in enhancing the immunity of fish from disease infections and increasing the blood biochemical indexes in fish could create a specific market for *Spirulina*-based feed or supplements for aquaculture use. *Spirulina* could also be an alternative to replace the usage of antibiotics and vaccines, which are less environmentally friendly and have higher cost.

Chlorella vulgaris

Chlorella vulgaris is a green microalgae species with an engaging immunostimulant property that enhances the health and increases the life expectancy of fish (Gouveia et al., 2002). Formulated diet supplemented with 2%, 5%, 7%, and 10% of *C. vulgaris* dry powder gave higher values of C4, total immunoglobulin and lysozyme in koi carp than the control group (0% *Chlorella vulgaris* inclusion); thus, *C. vulgaris* could be involved in the modulation of the innate immunity of fish during the experimental period of 8 weeks (Khani et al., 2017). Khani et al. (2017) also found that the koi group fed with *C. vulgaris*-supplemented diets also had higher haemoglobin and haematocrit levels, and the highest values were obtained by 5% *Chlorella vulgaris* supplementation. These components are important for the survival of fish and linked to the oxygen-binding capacity of blood (Bielek & Strauss, 1993). The addition of *C. vulgaris* powder supplement to the formulated diet increased the levels of IgM, IgD, interleukin-22 (IL)-22, and chemokine (C–C motif) ligand 5 in Gibel carp (*Carassius auratus gibelio*) (Zhang et al., 2014). The expression of various cytokines, such as IL-8 and IL-1, in fish fed with microalga-supplemented diet is greatly affected (Díaz-Rosales et al., 2008). This report supports the immunostimulant potential of *C. vulgaris* in boosting the immunity of fish. Besides, *C. vulgaris* contains abundant carotenoid and thus subjected to various studies in yielding different types of carotenoids (Markou & Nerantzis, 2013). Fish with high carotenoid content are more immune to fungal and bacterial infections (Gupta et al., 2007). Besides, carotenoids are soluble lipid pigments that play a role in the formation of skin colouring in ornamental fish [Kestemont et al. (as cited in Liang et al., 2012, p. 2); Paripatananont et al., 1999]. The biomass of *C. vulgaris* is more efficient in enhancing skin colouring and produces the highest deposition of carotenoid and red hue in three varieties of chromatic koi carps, namely, Kawari (red), Showa (black and red), and Bekko (black and white) (Gouveia et al., 2003). Colouring and pattern formation are important in ornamental fish as those traits will determine their quality (Li et al., 2008). The high bioavailability of carotenoids in *C. vulgaris* and the thin cellular membrane of this microalga are the possible characteristics that gave better efficiency for the skin colouring of fish, even for species that have relatively short digestive tracts (Gouveia et al., 1998). *Chlorella vulgaris* also remarkably enhances the growth performance and feed intake of koi carp at 5% inclusion in feed and other parameters, such as specific growth rate and protein and lipid efficiency rate, in contrast to the group without any *C. vulgaris* inclusion; thus, the growth enhancement is possibly caused by the high digestibility of this microalga (Khani et al., 2017). This enhancement may also be due to growth promoter properties, such as the adequate amounts of macronutrient and CGF in *C. vulgaris* (Badwy et al., 2008; Yamaguchi, 1996) and its high-quality protein content (Kang et al., 2013). However, further research on the growth effects of *Chlorella* should be carried out in various fish species and might not give the same positive effects. Its effect can be affected by the levels of supplementation, species-specific reactions to *Chlorella*, and the experimental period (Rahimnejad et al., 2016). Hence, the addition of *C. vulgaris* in feed for its carotenoid contents and property in stimulating the growth and immunity of fish is a promising idea as *C. vulgaris* is also known as a fast-growing microalga and can be a sustainable aquafeed ingredient in the future.

The addition or inclusion of *S. platensis* and *C. vulgaris* in fish diet and their effects on fish immune system are summarised in Table 2. Further trials on these microalga species on different fish stages, sizes, age, and environment should be investigated to improve the knowledge of suitable fish diet formulation with the addition of microalgae as natural supplements that could boost the immune system of fish. However, such approach requires a broad understanding on the abilities of different fish species to digest or utilise the target properties in microalgae, particularly the immunostimulant properties.

IMPROVEMENT OF FISH REPRODUCTION SYSTEM

Spirulina platensis

The effects of *S. platensis* on the reproduction and survival rates of fish were studied by several researchers throughout the years. The quality of fish brooders is the important key to a successful fish propagation. The features that define the reproductive capability and

Table 2
Summary of addition or inclusion of Spirulina platensis *and* Chlorella vulgaris *in fish diet and their effects on fish immunity*

Microalgae	Fish species	Diet / Test type(s)	Effects on immunity	Reference(s)
Spirulina platensis	*Clarias gariepinus* (African sharptooth catfish)	D1 (Control): Instant fish feed (obtained from animal feed supply) D2: Instant fish feed + 3% *Spirulina* algae D3: Instant fish feed + 5% *Spirulina* algae	Higher counts of WBC, RBC, and lysozyme activity assay in 3% and 5% *Spirulina* supplementation diets as compared to control	Promya and Chitmanat (2011)
	Oreochromis niloticus, L. (Nile tilapia)	D1 (Control): Basal diet (30.6% crude protein, 9.1% lipids, 4.72 kcal GE g^{-1}) D2: Basal diet + 1.25 g *Spirulina*/kg diet D3: Basal diet + 2.5 g *Spirulina*/kg diet D4: Basal diet + 5.0 g *Spirulina*/kg diet D5: Basal diet + 7.5 g *Spirulina*/kg diet D6: Basal diet + 10.0 g *Spirulina*/kg diet	Increasing number of WBC, RBC, and lymphocytes as the amount of *Spirulina* added in diets increased	Abdel-Tawwab and Ahmad (2019)
	Trachinotus ovatus (Golden pomfret)	D1 (Control): Basal diet (50.27% crude protein, 8.35 crude lipid) D2: Basal diet + 1.0% *Spirulina* D3: Basal diet + 2.0% *Spirulina* D4: Basal diet + 3.0% *Spirulina* D5: Basal diet + 4.0% *Spirulina* D6: Basal diet + 5.0% *Spirulina*	Increasing trend in the alkaline phosphate (ALP) activity as the amount of *Spirulina* in the diet increased	Lin et al. (2016)
	Cyprinus caprio (Carp)	Direct intubation (0.1 ml suspension) Dose 1: 0 dose (0.85% NaCl) Dose 2: 0.85% NaCl + 1 mg *Spirulina* Dose 3: 0.85% NaCl + 10 mg *Spirulina* Dose 4: 0.85% NaCl + 25 mg *Spirulina*	Lower number of bacterial cell (challenged with *Aeromonas hydrophilia* - strain MU9901 infection) in liver and kidney as compared to control	Watanuki et al. (2006)
	Huso huso (Sturgeon)	D1: Basal diet (Control) D2: Basal diet + 2.5% pure dried *Spirulina* D3: Basal diet + 5.0% pure dried *Spirulina* D4: Basal diet + 10.0% pure dried *Spirulina*	Cumulative mortality of the sturgeon decreased with the increasing supplementation of the *Spirulina* in basal diet (challenged with *Streptococcus iniae* - strain ATCC29178) through intraperitoneal injection	Adel et al. (2016)

Table 2 (*continue*)

Microalgae	Fish species	Diet / Test type(s)	Effects on immunity	Reference(s)
Chlorella vulgaris	*Cyprinus caprio* (Carp)	D1: Formulated feed (Control) D2: Formulated feed + 2% dry powder *C. vulgaris* D3: Formulated feed + 5% dry powder *C. vulgaris* D4: Formulated feed + 7% dry powder *C. vulgaris* D5: Formulated feed + 10% dry powder *C. vulgaris*	Higher values of immune parameters: C4, total immunoglobulin, and lysozyme than the control Higher values of haemoglobin (Hb) and haematocrit (Ht) than control – 5% of *Chlorella vulgaris* supplementation gave highest values	Khani et al. (2017)
	Carassius auratus gibelio (Gibel carp)	D1: Formulated diet (Control) D2: Formulated diet + 0.4% *Chlorella* powder D3: Formulated diet + 0.8% *Chlorella* powder D4: Formulated diet + 1.2% *Chlorella* powder D5: Formulated diet + 1.6% *Chlorella* powder D6: Formulated diet + 2.0% *Chlorella* powder	Increasing immunoglobulin (Ig) M and D, interleukin-22 (IL)-22, and chemokine (C-C motif) ligand 5 (CCL-5) as the *Chlorella* supplementation in diet increased	Zhang et al. (2014)

performance of brooders rely on the eggs' fecundity, diameter, and hatchability rates (Chong et al., 2004; Kumaraguruvasagam et al., 2007; Izquierdo et al., 2001). *Spirulina platensis* provides a remarkably higher egg survival rate of 73%, shorter hatching and faster larval development compared with the commercial flake diet-fed group when given as sole food for zebrafish broodstock with the hypothesis that commercial flake does not contain as much Omega-6 fatty acid as *S. platensis* (Geffroy & Simon, 2013). In addition, *S. platensis* contains omega-6 fatty acid (which made up 41.2% of fatty acids), specifically gamma linolenic acid and linoleic acid (Qiang et al., 1997). These properties are the precursors of arachidonic acid, which is a remarkable constituent in the formation of prostaglandin that interferes in oocyte maturation, ovulation, and steroidogenesis (Pati & Habibi, 2002; Patiño & Sullivan, 2002). Hence, *Spirulina* is a promising diet for adult fish. *Spirulina platensis* also considerably increases the hatching percentage and total egg reproduced by yellow tail cichlid (*Pseudotropheus acei*) during a 12-week observation period (Güroy et al., 2012). This microalga species could possibly replace the administration of artificial hormones in enhancing reproduction performance by incorporating it in the diet of brooders. Besides, raw *Spirulina* was also tested on Nile tilapia as the primary feed and has increased the egg production, hatching percentage, and survival rates of fish compared with conventional fish feed (Promya & Chitmanat, 2011). *Spirulina* as a replacement fish meal for the feeding of three-spot gourami (*Trichopodus trichopterus*) provides greater gonadosomatic indices (19.4%–21.85%) and affects the absolute fecundity of fish between 7,300 and 12,700 eggs per female at 2.5%–10% *Spirulina platensis* replacement levels than the group fed with fish meal only (Khanzadeh et al., 2016). The addition of this microalga can result in the enhancement of gonad maturation. Apart from that, *S. platensis* possesses fat soluble pigments (carotenoids), such as xanthophylls, B-carotene, echinenone, cryptoxanthin, and zeaxanthin (Nakagawa & Montgomery, 2007). Reproductive performance can be enhanced by dietary carotenoids (Watanabe & Vassalo-Agius, 2003). Increasing dietary carotenoid supplementation enhances the reproduction capability of different fish species, such as rainbow trout (*Oncorhynchus mykiss*) (Dabrowski et al., 1987), gilthead seabream (*Sparus aurata*) (Scabini et al., 2010), and yellow tail (*Seriola quinqueradiata*) (Vassallo-Agius et al., 2001, 2002). These discoveries show that the carotenoids in *S. platensis* play multiple roles, that is, it affects the colouration of fish and improves their reproductive performance. Thus, *S. platensis* can be one of the primary sources to be developed in ornamental fish industry by focusing on its capability to enhance the colouration and reproductive performance of the fish.

Chlorella vulgaris

Based on the knowledge obtained in this review, very scarce information is available on the effects of *C. vulgaris* on the reproduction of fish. Some of the studies found were only focused on mammals. For example, Sikiru et al. (2019) reported that *C. vulgaris* enhances oxidative stress, which was an exclusive biochemical complication that affected the reproduction in New Zealand White rabbits. Besides, the extract of *C. vulgaris* improves the histological adjustment of monosodium glutamate in ovarian tissue, the level of sex hormones, and increases the level of ovarian enzymatic antioxidants in adult female albino mice (Abdel-Aziem et al., 2018). Hence, its effects on fish should be further explored for potential use in aquaculture.

FUTURE PROSPECT AND CONCLUSION

The inclusion of *Spirulina platensis* and *Chlorella vulgaris* as feed supplement has substantial impacts on fish health and immunity. Planning better tests on a bigger scale of fish culture system is important to see the pattern of immunity and hematological parameters in natural culture conditions than in controlled laboratory environments. Besides, *S. platensis* has a potential in affecting the performance of fish brooder, which directly influences the quality and survival of eggs. Well-modified feed products based on *S. platensis* that focus more on the reproductive effects in fish could have a great future in aquaculture. Apart from that, more research should be done to analyse the effects of *C. vulgaris* on fish to highlight the capability of carotenoids in evaluating the growth performance and immunity of fish. In conclusion, this review briefly presented the effects of *S. platensis* and *C. vulgaris* on the immunity and reproduction of fish. It is recommended that more studies should be conducted on the possible contributions of these microalgae in the aquafeed industry to create more environmentally effective antibiotics and vaccines to combat fish diseases in a culture system and enhance the reproduction of fish. Using renewable natural products, such as microalgae, will also benefit the aquaculture industry by having better practices that conserve the environment.

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