fishes

Review

Role of Dietary Microalgae on Fish Health and Fillet Quality: Recent Insights and Future Prospects

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Abstract: An increase in the consumption of food fish, combined with a decrease in the harvest of fish, is driving the aquaculture industry at a fast pace. In parallel with the growth in the aquaculture sector and resulting stresses, the prevalence of diseases in farmed fish can increase. Although effective administration and prophylaxis are the main factors safeguarding fish species against diseases, recent approaches to mitigate the response caused by typical stressors include the uses of dietary additives. Microalgae are one of the main sources of nutrients, namely protein, lipids, vitamins, minerals, and pigments in aquatic animal diets. Numerous studies have proved the beneficial effects of microalgae on fish growth performance, feed utilization, disease resistance, and immunological and antioxidant activities. On the other hand, the administration of different microalgae to fish feed can enhance the fillet quality from several aspects, leading to an overall improvement in fillet shelf-life. This review focuses on the evidence supporting the beneficial effects of various microalgae on biochemical and organoleptic aspects as well as the proximate composition of carcasses in fish species.

Keywords: aquaculture; microalgae; feed; fish; fillet quality

Key Contribution: The microalgae in fish feed can enhance the fillet quality from several aspects, leading to an overall improvement in fillet shelf life.

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1. Introduction

The fish farming industry has turned into one of the most sustainable tactics to provide food for humans [1]. However, fish farming is altered by various interconnected factors, including the fish environment, diet, and farmed stock [2]. Boosting fish farming conditions via appropriate and healthy diets is considered as an important approach, causing improvement in fish growth, feed utilization, health, and welfare as well as carcass quality [3–6]. Among different feed additives, natural feed additives, such as microalgae, are preferable as they contain high nutritional value, are inexpensive, readily available, and environmentally safe [7–10]. The incorporation of these additives into fish feed can provide vitamins, polyunsaturated fatty acids, some amino acids, polysaccharides, minerals, and pigments [11]. The most important microalgae in fish farming are *Spirulina*, *Chlorella*, *Dunaliella*, and *Haematocussus*, which are available commercially [12]. In fish feed, microalgae are viable as a partial replacement for fish meal and positively affect the fish growth, disease resistance, skin coloration, and physiological activity [13,14]. Meanwhile, the influence of dietary microalgae on fish meat quality traits is well documented in different studies. Accordingly, this review addresses the potential benefits of microalgae application on meat quality in different fish species. In particular, the effects of different microalgae on various aspects of fillet quality, including biochemical, microbiological, and organoleptic traits as well as proximate composition are discussed.

Although there are numerous studies on the use of microalgae on fish health, growth, and fillet quality, some studies have reported on the safety aspects and constrains of some microalgae in aquaculture, including the high levels of toxins and trace elements and high production costs [10,15,16]. Therefore, in order to enter the market, it is important to ensure that these feed additives do not pose any risk to aquatic animals [15].

2. Commonly Used Microalgae in Aquaculture

Microalgae offer a sustainable alternative to terrestrial plants. This is due to their higher growth rate, ease of farming, and lack of competition for arable land. There are approximately 50,000 classified species of these unicellular aquatic microorganisms, including notable ones like *Arthrospira platensis*, *Chlorella vulgaris*, *Isochrysis*, and *Tetraselmis* [17]. Microalgae have a well-balanced protein, vitamins, lipids, carotenoids, carbohydrates, and minerals (Table 1), which are often lacking in plant-based ingredients [18].

Table 1. Chemical compositions in various microalgae.

Microalgae Nutrient (per 100 g)	*Tetraselmis suecica*	*Isochrysis galbana*	*Chlorella*	*Tetraselmis chuii*	*Phaeodactylum tricornutum*	*Nannochloropsis granulate*	*Botryococcus braunii*	*Porphyridium aerugineum*	*Schizochytrium* sp.
Sodium (mg)	3223.1	2735	-	890	1430	310	940	810	100
Calcium (mg)	1790.7	1145.5	331,00	2990	1910	980	100	640	340
Potassium (mg)	1349.4	1314.4	714	1860	1720	600	750	670	550
Phosphor (mg)	964.6	625.3	29820	1460	269	0.073	1450	1390	470
Sulfur (mg)	-	-	-	1380	1050	0.058	410	640	740
Magnesium (mg)	601.4	400.4	335	430	555	700	360	550	-
Iron (mg)	30.9	31	53.9	477.5	477.27	0.0102	620.3	1110.07	13
Zinc (mg)	6.6	13.4	217	5	37.3	0.0025	2.78	4.1	36
Manganese (mg)	3.5	3.6	-	4.51	31.4	0.0019	45.37	25.85	-
Copper (mg)	1.1	2.8	-	5.48	8.4	0.0064	3.52	4.53	2
Selenium (mg)	-	-	2.2	0.05	0.05	0.05	-	-	0.13
Lipid (g)	14.68	31.9	12.18	01.07	12.73	18.4	14.3	7.55	50.00
Protein(g)	26.05	28.98	51.45	19.57	26.95	28.8	19	40.8	19.22
Carbohydrate (g)	24.01	17.67	11.86	79.36	16.91	37.6	18.74	39.2	24.88
Ash (g)	17.99	15.16	9.50	14	27.95	8.6	39.73	2.1	3.67
References	[19]	[19]	[20,21]	[22]	[19,23]	[24,25]	[26]	[19]	[27]

Approximately 50% of the global protein and peptide market, which is prepared from terrestrial plant sources, could be potentially replaced with insects, unicellular bacteria, yeast, and microalgae [28]. Meanwhile, microalgae can be convenient alternatives to fish meal on account of their high protein level and richness of essential amino acids. *Spirulina*, for instance, contains proteins that have a higher quantity of essential amino acids compared to legumes, which are typically considered as the standard plant protein sources [29]. Additionally, *Spirulina maxima* contains up to 71% protein [30]. The protein apparent digestibility coefficient was also high for diets supplemented with *Nannochloropsis granulata* for rainbow trout (*Oncorhynchus mykiss*) [31]. A previous study conducted in this respect also showed that administration of 20% *Arthrospira* sp. in golden barb (*Puntius gelius*) meal increased growth rates in fish significantly [32]. Similarly, 0.5–2% *Arthrospira* *platensis* in Nile tilapia (*Oreochromis niloticus*) feed could enhance fish health status through tissue protection and antioxidant effects [33].

The lipid content of microalgae can range from 1–70% of their dry weight [34]. For example, *Phaeodactylum* sp. contains up to 57% of oil [30]; *Isochrysis galbana* also contains fatty acids and ascorbic acid as bioactive compounds [35]. Some PUFA-rich microalgae are *Schizochytrium*, *Crypthecodinium*, *Nannochloropsis*, *Isochrysis*, *Nitzschia*, *Diacronema*, *Porphyridium*, and *Desmodesmus* that produce eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) [36,37]. In the previous study, *Nannochloropsis* sp. and *Isochrysis* sp. were substituted with fish meal and fish oil in a rainbow trout (*Oncorhynchus mykiss*) diet [27]. Oral use of 15% *Nannochloropsis* sp. and *Isochrysis* sp. in juvenile Atlantic cod (*Gadus morhua*) also improved fish growth, feed intake, and survival, revealing no difference in muscle ω-3 and ω-6 fatty acid contents among treatment groups [38].

Microalgae are sources of various pigments, including astaxanthin, lutein, zeaxanthin, canthaxanthin, β-carotene, chlorophyll and phycobiliproteins. These bioactive compounds have antibacterial, anticancer, anti-inflammatory, antioxidative, neuroprotective, and antimicrobial effects, as well as skin and fillet pigmentation [39,40]. For example, *Spirulina* contains additional carotenoids such as ß-carotene and astaxanthin that can be converted to astaxanthin and other brightly colored pigments after being used by ornamental koi and other fish. Microalgae can also improve antioxidant effects, disease resistance, and growth rate [40]. *Phaeodactylum tricornutum* produces massive quantities of fucoxanthin, which causes the golden yellow coloration in gilthead sea bream. It has an anti-inflammatory effect on fish [41]. In addition, carotenoid-source *Dunaliella salina* is a popular microalga for mass production (up to 14% dry weight) [42].

Microalgae contain about 10–85% carbohydrates, with varying amounts depending on their culture age and growth conditions. Examples of microalgae with a high carbohydrate content include *Porphyridium cruentum* (40–57% of dry weight) and *Spirogyra* sp. (33–64% of dry weight) [43]. *Porphyridium cruentum* (40–57%) and *Spirogyra* sp. (33–64%) have a high innate carbohydrate content [44]. These unicellular organisms are the main sources of modified polysaccharides, such as xylooligosaccharides, galacto-oligosaccharides, alginate oligosaccharide, neoagaro-oligosaccharides, galactans, arabinoxylans, and β-glucans [45], which are categorized as prebiotics. These substances assist in enhancing the growth of probiotics in animal gastrointestinal tracts [46]. *Chlorella* contains up to 26% of carbohydrates that contains many potential prebiotic bioactive compounds [30]. In general, the carbohydrate digestibility of microalgae species varies between 22% and 83%, depending on the presence of some non-starch polysaccharides (cellulose, gums, pectins, and hemicellulose) and fibers, which make them difficult for digestion. In general, differences in fish species, physiology, and the pretreatment of microalgae could cause different digestibility for each microalgae [18]. For example, a previous study indicated the high carbohydrate digestibility of *S. maxima* and *C. vulgaris* (nearly 70%) in Nile tilapia [47].

Microalgae are an excellent potential source of vitamins, namely vitamins A, B, C, D, E, nicotinate, biotin, folic acid, pantothenic acid, and niacin [39]. Although microalgae do not naturally produce vitamin A, they can store vitamin some precursor substances like carotenes (i.e., α- and β-carotenes) and retinol [48]. The findings of a study indicate that *Nannochloropsis oceanica* has the highest vitamin D3 content at 1 μ/g dry weight. Additionally, the formulated powders of *Chlorella* sp. and *Nannochloropsis* sp. exhibited vitamin B9 contents of 25.9 and 20.8 μg/g, respectively. These results suggest that these microalgae species are promising sources of vitamin D3 and vitamin B9, which are essential micronutrients [49]. The crucial vitamin imperative for good health is vitamin B_{12} which is limited in plant foods. *Hapalosiphon fontinalis*, *Tolypothrixtenuis*, *Cylindrospermum* sp., *Nostoc* sp. *Chollera* sp. (2.4 μg/g), and *Spirulina* sp., have more vitamin B_{12} than plants or animal-based foods [35,50]. *Euglena gracilis* and *Eisenia arborea* brown microalgae contain a high amount of vitamin E (3.7 mg/g) and vitamin C content (3.44 mg/g), respectively [51,52].

Microalgae include plenty of organic minerals ranging from about 2.2% to 4.8% of total dry weight [51–53]. The mineral composition of microalgae demonstrates extensive variations between marine and freshwater microalgae on the strain, species, generic basis, and environmental conditions [52]. For example, Tibbetts et al. [53] reported the calcium, phosphorus, magnesium, potassium, sodium, and sulfur levels of *Phaeodactylum tricornutum*, *Nannochloropsis granulate*, *Botryococcus braunii*, *Porphyridium aerugineum*, and *Tetraselmis chuii* range in 0.26–2.99%, 0.73–1.46%, 0.26–0.71%, 0.67–2.39%, 0.81–2.66%, and 0.41–1.38%, respectively. The highest calcium and phosphorus levels were shown by the chlorophyte, *Tetraselmis chuii* (2.99% and 1.46%, respectively). The highest magnesium, potassium, sodium, and sulfur contents were measured in the bacillariophyte *Phaeodactylum tricornutum*. In another study, Fithriani and Melanie [19] also showed that *Chlorella* contained K minerals (714 mg/100g), Ca (331 g/100g), Mg (335 mg/100g), Fe (539 mg/kg), Zn (21.7 mg/kg), Se (0.22 mg/kg), and P (2982 mg/kg). *Isochrysis galbana* showed levels of sodium (2735 mg/100 g), calcium (1145.5 mg/100 g), potassium (1314.4 mg/100 g), phosphor (625.3 mg/100 g), magnesium (400.4 mg/100 g), and also levels of trace minerals such as iron (31 mg/100 g), zinc (13.4 mg/100 g), manganese (3.6 mg/100 g) and copper (2.8 mg/100 g); additionally, *Tetraselmis suecica* showed contents of sodium (3223.1 mg/100 g), calcium (1790.7 mg/100 g), potassium (1349.4 mg/100 g), phosphor (964.6 mg/100 g), magnesium (601.4 mg/100 g), and contents of trace minerals such as iron (30.9 mg/100 g), zinc (6.6 mg/100 g), manganese (3.5 mg/100 g), and copper (1.1 mg/100 g) [19]. According to these data, the levels of some mineral elements such as sodium, magnesium, manganese, and iron are much higher in microalgae than in most common food items when expressed on a dry matter basis. The addition of mineral-rich microalgae to salmon diets improved the texture and flavor of the fish [54]. The mineral leaching before fish ingestion could be avoided using mineral-rich microalgal biomass [55]. Also, the minerals can protect against lipid oxidation in fish products [56]. One of the essential minerals for fish growth and survival is phosphorus. The form of phosphorus found in plants is organic phytic acid, which is anti-nutritional and indigestible for fish species [57–59]. However, some studies suggest that phosphorus in microalgae is found in the form of polyphosphate granules, and the digestibility of microalgal phosphorus in fish ranges from 38% to 100% [47,59,60]. The phosphorus digestibility of *Nannochloropsis gaditana*, *Spirulina maxima*, *Chlorella vulgaris*, and *Scenedesmus dimorphus* for juvenile Nile tilapia (*Oreochromis niloticus*) was reported to be 92%, 93%, 86%, and 39%, respectively [39,60]. The phosphorus digestibility of *Nannochloropsis gaditana*, *Spirulina maxima*, *Chlorella vulgaris*, and *Scenedesmus dimorphus* for juvenile African catfish (*Clarias gariepinus*) was 77%, 92%, 84%, and 45%, respectively [47,60].

3. Effects of Microalgae on Fish Fillet Proximate Composition

In light of the fact that lipids are critical to meeting energy needs and offering essential fatty acids, they are important nutrients included in aquafeeds for marine fishes. These fatty acids are necessary for sustaining health, reproduction, and growth [61]. Fish oil is an excellent digestible source of energy that also contributes to a significant amount of essential fatty acids, particularly omega-3 (n-3) highly unsaturated fatty acids, such as EPA and DHA that are widely acknowledged as the primary lipid sources used in the production of aquafeeds. These fatty acids are widely valued for their health advantages in the development of the nervous system and the prevention of inflammatory illnesses, as well as neurological and cardiovascular problems, and thus are recommended as an integral part of natural medications for improving human health. Due to issues with sustainability, economics, and the environment, the production of aquafeeds that rely on fish oil is constrained [61]. The capture fisheries are a major source of essential fatty acids and protein for the aquaculture of finfish. In 2018, 19% of capture fishery production was converted to fish oil and fish meal for aquafeeds. Finding compositions of alternative and sustainable sources of oil and protein, especially DHA and EPA, has become a top challenge for the aquaculture industry due to the limited supply of these compositions and the expansion of fish farming [62]. For the aquaculture business to continue to expand and be sustainable, fewer fish meals must be used in aquaculture diets in favor of other affordable and environmentally-friendly foods [38]. Fish meal and fish oil have been lowered, but not entirely eliminated, by aquafeed makers, who are now looking for alternatives that are less expensive [27]. In recent years, various studies have been carried out to replace fish meal with diverse sources, like plant-derived protein sources and microalgae [13,63–66]. Microalgae contain many biological compounds such as n-3 LC-PUFA, polysaccharides, and carotenoids [67]. DHA and EPA have significant effects on human health by reducing cardiovascular diseases and mitigating inflammatory diseases [67]. A fish fillet (a dark muscle), rib meat (a light muscle), mesenteric fat, and liver are a few of the tissues where lipids are stored. These tissues have several purposes for storing and digesting lipids. Light and dark muscles serve as greater short-term storage for localized energy requirements, the liver processes lipids, and mesenteric fat normally offers long-term lipid storage [68]. However, the nutritional value of fish fillets as a health-promoting food may be harmed since the fatty acid profile is greatly impacted by the fatty acid profile of diets [61,69]. So far, various microalgae/extracts of microalgae have been used to increase the fillet quality of fish in aquaculture (Table 2).

Table 2. Effects of dietary microalgae on the proximate composition of fish fillets.

Microalgal Culture	Fish Species	Microalage Replacement (%) with Fish Meal/Fish Oil/Dietary Inclusion Level	Influence	References
Combination of *Nannochloropsis* sp. and *Isochrysis* sp.	Juvenile Atlantic cod (*Gadus morhua*)	15%, 30%	No changes in n-3 and n-6 fatty acids content ↑ level of 20:1(n-9) fatty acid ↑ feed intake ↑ fish growth, ↑ survival, ↓ feed conversion ratios	[38]
Pavlova viridis and *Nannochloropsis* sp.	European sea bass (*Dicentrarchus labrax* L.).	50%, 100%	↑ DHA ↑ Total PUFAs	[70]
Nannochloropsis oculata *Schizochytrium* sp.	Nile tilapia (*Oreochromis niloticus*)	*N. oculata*: 3%, 5% and 8% *Schizochytrum* sp.: 3.2%	↑ DHA ↑ Total PUFAs ↑ crude lipid content ↓ arsenic content crude protein and ash content, total n-3 PUFA, n-6 PUFA, n-3 LC PUFA, and n-6 LC PUFA, SFA, MUFA not significantly different	[71]
Desmodesmus sp.	*Atlantic salmon*	10, 20% diet 10% diet 20% diet	No significant changes in proximate composition ↓ Lipid content ↓ protein content ↓ ash content	[72]
Schizochytrium sp.	*Juvenile tilapia*	1.75%, 5.26%, 8.77%	↑ Omega 3 ↑ total n-3 ↑ LC-PUFAs ↑ DHA ↑ Total SFA ↓ Total MUFA ↓ Omega 6 ↓ Total ash ↓ Crude protein	[68]

Table 2. *Cont.*

Microalgal Culture	Fish Species	Microalage Replacement (%) with Fish Meal/Fish Oil/Dietary Inclusion Level	Influence	References
Isochrysis galbana	*Trachinotus ovatus*	4.5–5.0 wt %	↑ DHA contents ↑ EPA contents ↑ PUFAs ↑ Docosapentaenoic acid ↑ LC-n-3 PUFAs ↑ total lipid contents ↑ total PLs ↑ α-linolenic acid content ↑ SDA content SFAs, MFAs, n-6 FAs, neutral lipids and TAG, crude protein contents and moisture not significantly influenced	[73]
Spotted wolffish (*Anarhichas minor*)	*Nannochloropsis oceanica*	7.5% and 15%	↑ omega-3 fatty acid EPA ↑ PUFA ↓ Total saturated fatty acids ↓ Saturated fatty acid C14:0, C16:0 ↓ monounsaturated fatty acid C22:1n-9 No changes in EPA and DHA level	[74]
Combination of *Nannochloropsis oculata* and *Schizochytrium* sp.	Nile tilapia (*Oreochromis niloticus*)		↑ growth performance ↑ DHA, protein, lipid	[75]
Schizochytrium limacinum	*Totoaba macdonaldi*	33%, 66% and 100%	↑ DHA ↑ TGC ↑ total n-3 FA, 22:6n-3 ↑ n-3/n-6 ratios, ↑ Crude fat	[61]
Mixture of: *Schizochytrium limacinum* and *Nannochloropsis oceanica*	Rainbow trout (*Oncorhynchs mykiss*)	9% and 17%	↑ n-3/n-6 ratio ↓ MUFA No changes in total SAFA, total PUFA, n−3 PUFA, EPA, DHA, and 22:6n−3 fatty acid content	[75]
1. *Chlorella vulgaris* 2. blend of *Schizochytrium* sp. and *Microchloropsis gaditana*	Gilthead seabream (*Sparus aurata*)	1. 10%, 20%, 30% 2. 50% and 100%	1. ↑ level of 18:3n-3 and 18:2n-6 No significantly changes in DHA and EPA 2. ↑ level of n-6 PUFA No changes in DHA and EPA	[76]
Oedocladium carolinianum	*Trachinotus ovatus*	1% and 5%	In the whole body: ↑ The total MUFA ↑ oleic acid (C18:1) and Eicosenoic acid (C20:1) content ↓ Total PUFAs andtotal n-6 PUFA, linoleic acid (C18:2n6) content in 5% of the diet No changes in total SFAs, total n-3 PUFA profiles, EPAand DHA concentrations	[77]

Table 2. *Cont.*

Microalgal Culture	Fish Species	Microalage Replacement (%) with Fish Meal/Fish Oil/Dietary Inclusion Level	Influence	References
Spirulina platensis	Nile tilapia (*Oreochromis niloticus*)	0.25, 0.5%	↑ protein content ↑ EPA ↓ lipid content ↓ MUFA ↓ SFAs ↓ PUFA ↓ linoleic acid concentrations ↓ n-6/n-3 ratio	[78]
filamentous microalga Tribonema ultriculosum	Rainbow trout (*Oncorhynchs mykiss*)	50 g/kg 100 g/kg	↓ polyunsaturated fatty acids (linoleic acid and linolenic acid) ↑ DHA contents ↑ n-3/n-6 polyunsaturated fatty acids ↑ Saturated fatty acids ↑ palmitoleic acid ↑ EPA	[67]
Oedocladium sp. or *Tribonema ultriculosum*	Yellow catfish (*Pelteobagrus fulvidraco*)	4% and 5%	↑ MUFA ↑ TFA content ↑ palmitoleic acid ↑ EPA ↑ C14:0 profile or the ↑ C14:0 and C16:0 profiles ↑ EPA ↓ PUFAs, n-6 PUFA No changes in n-3/n-6 ratio and EPA	[79]

Docosahexaenoic acid (DHA), eicosapentaenoic acid (EPA), saturated fatty acid (SFA), mono-unsaturated fatty acids (MUFA), polyunsaturated fatty acid (PUFAs), N-3 long-chain polyunsaturated fatty acids (LC-n-3 PUFAs), Triglyceride (TG). (↑): indicates a rank increase and; (↓): indicates a rank decrease.

Atlantic cod (*Gadus morhua*) growth, body composition, and feed intake were studied after fish were fed with diets containing fish meal protein partially replaced with microalgae. For instance, the fish fed isocaloric diets containing protein substituted by a mixture of dried *Isochrysis* sp. and *Nannochloropsis* sp. (15 and 30%) for 84 days exhibited no changes, in terms of feed conversion ratios, survival, viscero-somatic indices, or n-3 and n-6 fatty acids in muscles compared to the treatment groups. No change was seen in fatty acid profiles of fish muscle in the control and fish fed with 15% replacement; however, the eicosenoic acid 20:1(n-9) level was increased in the treated fish. The growth and feed intake, which were related to algal incorporation level, considerably decreased in algae-fed fish, most likely due to palatability issues. The authors assumed that the amount of specific fatty acids could not guarantee the level in the cod fillet since differential metabolism of fatty acids took place in response to energy requirement. Therefore, a higher amount of EPA in the cod diet could not increase the quantity in fish muscle [38].

Haas et al. [70] conducted a research study that compared the effectiveness of using microalgae *Pavlova viridis* and *Nannochloropsis* sp. as n-3 PUFA sources in the diets of juvenile European sea bass (*Dicentrarchus labrax* L.). The study involved administering different isonitrogenous and isoenergetic diets, which were prepared using a fish oil diet, plant oil (a combination of linseed, rapeseed, and sunflower oil), and a basal diet consisting of fish oil and *Pavlova viridis* and *Nannochloropsis* sp. lipids. The total PUFA was the highest in the *Pavlova* 100% group, followed by *Pavlova* 50%, *Nannochloropsis* 100%, and *Nannochloropsis* 50%, due to the high content of linolenic and linoleic acids coming from microalgal products and plant oils, whereas the lowest levels were observed in the fish oil group. The fish oil diet had the highest amounts of total fatty acids and DHA, but not significantly higher than that in the *Pavlova* 50% and *Pavlova* 100% groups. Also, the highest content of EPA was in the *Pavlova* 100% diet. The results demonstrated that replacing 50% of the fish oil in the diet with *Nannochloropsis* sp. meal and 100% of it with *Pavlova viridis* meal could be implemented without posing a detrimental impact on the nutrient utilization and growth performance of juvenile sea bass [70].

Sarker et al. [71] showed that the complete substitution of fish oil with microalga *Schizochytrium* sp. in juvenile Nile tilapia feeds improves growth and the deposition of n3 -LC PUFA content in tilapia fillets. Higher contents of DHA were observed in fillets which led to high DHA: EPA ratios in fillets compared to controls. Saturated fatty acid (SFA) was higher in *Schizochytrium*-fed fish and in the SFA profile, the composition of 15:0, 18:0, 20:0, and 22:0 did not significantly differ in dietary treatments; despite this, palmitic acid (16:0) content increased, and the highest concentration was observed in the *Schizochytrium*-fed fish. Fish fed a control diet showed the highest content of mono-unsaturated fatty acids (MUFA), which was related to MUFA amount in the tested diets. Meanwhile, the 100% substitution of fish oil with *Schizochytrium* resulted in n3: n6 and DHA: EPA ratios of 1.4:1 and 34.2:1, respectively, in fish fillets, suggesting the possible inclusion of this microalga in tilapia feed with tailoring the n3 PUFA composition of tilapia muscles. The researchers assumed that fish feed with fish oil makes the fish fillet susceptible to lipid oxidation, producing the adverse effects of fillet quality. In return, the advantage of administrating DHA-rich whole cell *Scizochytrium* sp. biomass in fish feeds is the natural encapsulation provided by the cell wall that can protect valuable fatty acids from oxidation [71].

The efficacy of the defatted biomass of marine microalga, *Desmodesmus* sp., in *Atlantic salmon* (*Salmo salar*), was assessed for 70 days. In the treatment diets, the algal mass replaced a portion of the fish meal, with the results indicating that the growth indices, such as specific growth rate, condition factor, and survival of the *Desmodesmus*-fed (10 and 20%) fish were not different from the values of the control-fed fish. The whole-body proximate composition of fish, and the lipid and protein digestibility from all diets did not significantly differ. Fish fed 10% *Desmodesmus* in their diet demonstrated lower lipid amounts in their fillets and less digestibility of energy than the control fish. Authors concluded that the inclusion of *Desmodesmus* did not significantly change the Atlantic salmon growth, health, and fillet quality in comparison with the fish meal [72].

Several experimental diets containing n-3 fatty acids from fish oil (1%, 3%, and 5%), algal meal (1.75%, 5.26%, and 8.77%), or a control diet (with 6.3% corn oil) were fed to tilapia. With an increase in fish oil and algal meal levels in the diets, all tissues showed better fatty acid profiles, higher n-3 content, lower n-6, and n-6: n-3 rates. Diet with algal meal 8.77% was the most effective for enhancing the lipid profile in fish fillets. Total n-3 level in the fillet increased from 151.2 mg to 438.7 mg, while the n-6: n-3 ratio decreased from 5.19 mg to 1.29 mg, suggesting such formulated diets as a workable solution for increasing the n-3 content of tilapia fillets. The treated fish also showed the capacity to lengthen and desaturate shorter-chain PUFAs into longer-chain PUFAs. Authors indicated that *Schizochytrium* sp. and fish oil can boost n-3 fatty acids in tilapia fillets over a period of four weeks, which is of high nutritional benefit to consumers [67]. Five isonitrogenous and isolipidic feed diets were prepared by replacing fish oil with *Isochrysis galbana* in pompano (*Trachinotus ovatus*). Results demonstrated that the inclusion of microalga *Isochrysis galbana* (4.5–5.0 wt %) in the fish diet enhanced growth performance, lipid deposition, DHA, EPA, and total n-3 fatty acid contents in lipids of fish muscles [73]. Feeding spotted wolfish (*Anarhichas minor*) with isocaloric and isonitrogenous diets containing 7.5% or 15% microalgae *Nannochloropsis oceanica* and replaced with wheat and fish meal in the diets for 12 weeks exhibited an increase in omega-3 fatty acid EPA value in fish fed microalgae, which could be explained by the high content of EPA in the microalga. On the other hand, muscle protein level decreased in the algal-fed groups, indicating a lower ability of fish to absorb and then utilize protein due to the complex carbohydrates in the cell walls of this microalgae [74].

A combination of two microalgae, a protein-rich defatted biomass of *Nannochloropsis oculata* replaced with fish meal and a whole cell of DHA-rich *Schizochytrium* sp. as a substitute for fish oil, was used to produce a high-performing fish-free feed for Nile tilapia (*Oreochromis niloticus*) [75]. Fish-free feeds revealed a higher growth, specific growth rate, weight gain, and a better feed conversion ratio than the control diets. Higher levels of fillet lipid, protein, and DHA were also observed in fish fed fish-free meals than control diets although the data were insignificant. Furthermore, in vitro protein digestibility and protein hydrolysis were higher in fish-free meals than the control diets. The median feed cost for experimental diets was marginally higher than the control diet. However, the median economic conversion ratio of the fish-free feed was lower than the reference diet. This research provides evidence of a cost-competitive microalgae-based tilapia diet that enhances growth performance and reduces dependency on fish oil and fish meal [75].

A 6-week feeding *Totoaba macdonaldi* juvenile diets containing 33%, 66%, and 100% lipid of soybean oil and microalgae (*Schizochytrium limacinum*) sources replaced with fish oil exhibited higher levels of weight gain, with the thermal growth coefficient being higher in fish fed *S. limacinum* meals than in fish fed soybean oil. The proximate composition of liver was influenced by lipid source; apart from that, crude fat was higher in fish-fed soybean oil than in fish-fed *S. limacinum*. Also, soybean oil increased the concentrations of total n-6 FA and 18:2n-6 FA in fish fillet, whereas it decreased growth, n-3/n-6 ratios, total n-3, 20:5n-3, and 22:6n-3 FA. In contrast, *S. limacinum* meals elevated n-3/n-6 ratios, total n-3 FA, and 22:6n-3 FA, which is beneficial in foods destined for human consumption [61]. Considering the daily need of 200 mg DHA+EPA for the prevention of coronary heart disease, the authors assumed that consuming 200 g totoaba fillet administrated with *S. limacinum* meals, with the intake of nearly 380 mg DHA+EPA, can satisfy the daily consumption for providing the health benefit to consumers [61].

When rainbow trout was fed with a mixture of two microalgae meals, including *Nannochloropsis oceanic* and *Schizochytrium limacinum* in a 1:1 ratio (9% and 17%) replacing fish oil, the feed conversion ratio and growth performance were reduced at a higher level (17%) as compared to the fish-oil fed group [75]. The authors suggested that the current result might be related to the indigestible carbohydrates in the microalga cell wall, which decrease the digestibility of proteins and lipids. A reduction of 23% was also seen in EPA value in the muscle of fish fed 17% microalgae meal, but DHA+EPA content was similar among the dietary treatments. Authors indicated that the need for EPA might be covered by the available fish oil or fish meal or the trout capacity to metabolically retro-convert DHA into EPA as the dietary inclusion of *S. limacinum* increased [75]. Karapanagiotidis et al. [76] studied the effects of replacing fish meal with *Chlorella vulgaris* and fish oil using a mixture of *Microchloropsis gaditana* and *Schizochytrium* sp. on growth performance and the fatty acid composition of muscle in gilthead seabream (*Sparus aurata*). The fish meal protein of the control diet was replaced with *C. vulgaris* meal at 10%, 20%, and 30%; and the fish oil of the control diet was replaced with a mixture of *Microchloropsis* sp. and *Schizochytrium* sp. at 50% and 100%. The results exhibited that C. *vulgaris* meal at 30% and the mixture of *Schizochytrium* sp. and *M. gaditana* at 100% had no harmful effect on fish feed intake. Diets containing *C. vulgaris* revealed an increase in lipid deposition, especially in fish liver, while no other diet-related changes were seen in fish muscle and total body composition. *C. vulgaris* diets increased values of 18:3n-3 and 18:2n-6 in muscle, while an insignificant decrease was seen in DHA and EPA contents, probably due to their deposition by the dietary fish oil. The diet containing a mixture of *Microchloropsis* sp. and *Schizochytrium* sp. enhanced n-6 PUFA, especially 22:5n-6 and 20:4n-6, in fish muscle while maintaining similar DHA and EPA levels compared to the control group, suggesting that a biomass of *M. gaditana* and *Schizochytrium* sp. could replace dietary fish oil in a gilthead seabream diet [76]. Zhao et al. [77] assessed the growth performance and flesh quality of golden pompano (*Trachinotus ovatus*) fed with astaxanthin-rich *Oedocladium carolinianum* at 1% and 5% powder for 6 weeks which exhibited a significant increase in total MUFA, eicosenoic acid (C20:1), and oleic acid (C18:1) contents in fish fed *O. carolinianum* at 5%. Despite a decline in palmitoleic acid content, fish fed the *O. carolinianum* 5% diet exhibited lower levels of n-6 PUFA, PUFA, and linoleic acid (C18:2n6) compared to those fed the control and *O. carolinianum* 1% diets. No significant change was noted in total SFA profiles, total n-3 PUFA, n-3/n-6 ratio, and values of whole-body EPA and DHA, suggesting O. *carolinianum* at 5% could promote the synthesis of endogenous DHA and EPA in golden pompano and produce astaxanthin, which is attractive to consumers and contributes to the increase in sales price [77].

Only in a handful of studies were the effects of replacing some microalgae with other sources except fish oil and fish meal evaluated. For example, flesh quality and growth of Nile tilapias (*Oreochromis niloticus*) fed with *Saccharomyces cerevisiae*, *Spirulina platensis*, and *Rubrivivax gelatinosus* at 0.25% and 0.5% for 72 days demonstrated no effect on the pH and textural characteristics of fish fillet. However, compared to the control group, use of this microbial biomass enhanced protein content, due to the probiotic effects of *Saccharomyces cerevisiae*, *Rubrivivax gelatinosus,* and lowered the n-6/n-3 ratio of fish fillets. Such data show that consuming small amounts of biomass of these microbials has no negative effect on fish growth, but rather may improve fish nutritional quality, and preserve the textural characteristics of fish fillet [78]. According to Chen et al. [66], the supplementation of 50 and 100 g/kg *Tribonema ultriculosum* meal replaced with portions of wheat flour and soybean oil increased rainbow trout growth and enhanced contents of DHA, EPA, and palmitoleic acid in fish fillet. In addition, the benefits of DHA, EPA, and palmitoleic acid enhance the cell membrane fluidity, and decrease the inflammation related to diabetes, heart disease, and various health problems in human. Therefore, *Tribonema ultriculosum*, as a source of palmitoleic acid in fish fillet, could be beneficial to consumers. Chen et al. [79] also assessed the effect of *Oedocladium* sp. (4% meal) and *Tribonema ultriculosum* (5% meal) on growth performance, skin pigmentation, immune response, and the fillet fatty acid content of yellow catfish (*Pelteobagrus fulvidraco*) after a 40-day feeding trail. The outcomes demonstrated that fish performance was unaffected by microalgal supplementation. However, EPA and palmitoleic acid values in fillets increased by 12.51% and 100.44%, respectively. In fish fed *T. utriculosum* and *Oedocladium* meal supplementation, fish skin ventral lutein content (61.6%) was considerably enhanced. These findings showed that the nutritional quality of fillets may be improved using *T. utriculosum* and *Oedocladium* sp. [79].

4. Effects of Microalgae on Organoleptic Parameters of Fish Fillet

Moreover, chemical and microbial properties, sensorial characteristics, including appropriate color, are important determinants in consumer's choice of food fish [80]. The impression of natural and bright colors often reflects high-quality fish food while pale color unconsciously is associated with inferior quality [54]. To acquire an appropriate color and a suitable appearance, the dietary administration of pigments in the optimized levels for fish species is necessary. Previous studies proved that microalgal biomasses have significant impacts on fish pigmentation [41,81] since they are rich in substances such as carotenoids, chlorophylls, and xanthophylls. Besides color, textural indices are also crucial for fillet quality [82]. Few studies have addressed the effects of dietary microalgae on the texture of food fish, e.g., enhanced firmness, gaping decrease, and overall improved fillet quality attributes (Table 3). For example, studies conducted on the potentiality of microalgae biomass from the diatom *Phaeodactylum tricornutum*, as an effective component for *Sparus aurata* diets, exhibited that the use of diet containing 2.5% of *Phaeodactylum tricornutum* biomass did not affect the impression of flavor, odor, fatness perception, and whiteness in cooked fillets. Macroscopically, the *Phaeodactylum tricornutum* biomass diet caused a significantly lighter and more intense yellow coloration in the operculum of seabream. Also, the lightness of ventral skin coloration was affected by the *Phaeodactylum tricornutum* diet although this variation was not perceptually potent [41]. A three-week addition of a diet containing *Chlorella pyrenoidosa* at 2.5, 5, and 7.5 g/kg added into a common carp (*Cyprinus carpio*) diet significantly affected meat juiciness, color, and complete acceptability better than the control one [83]. In another study, Güroy et al. [84] showed that a four percent inclusion of *Spirulina* sp. in a trout diet increased the red/green tonality (*a**) fillet compared with the control diet. The luminosity (*L**) value of all fish fillets increased during the storage period [84], suggesting that *Spirulina* could induce pigmentation in trout fillet that is more acceptable by fish markets since this microalga contains pigments such as xanthophyll (yellow), carotene (orange), except chlorophyll (green) phycoerythrin (red) and phycocyanin (blue). In parallel, Sáez et al. [81], in their study showed that the quality characteristics of gilthead seabream (*Sparus aurata*) fillets fed *Nannochloropsis gaditana* (2.5 and 5%) in a 42-day feeding trial manifested a positive dose-dependent effect on fillet harness and skin color, suggesting an extended shelf life of gilthead seabream fillet using this dietary strategy.

Table 3. Effects of dietary microalgae on the sensory qualities of fish fillets.

Microalgal Culture	Fish Species	% Replacement of Fish Meal/Fish Oil/Dietary Inclusion Levels	Effects	References
Phaeodactylum tricornutum	Gilthead seabream	2.5%	No significant effect on flavor, odor, flavor, whiteness, and fatness perception in cooked fillets Skin color: lighter and more vivid yellow	[41]
Chlorella pyrenoidosa	Common carp (*Cyprinus carpio* L.)	2.5 g/kg, 5 g/kg 7.5 g/kg	↑ color ↑ j uiciness ↑ complete acceptability than the ↑ freshness ↑ Flavor (except 2.5 g/kg)	[83]
Spirulina (*Arthrospira platensis*)	rainbow trout (*Oncorhynchs mykiss*)	4%	↑ red/green tonality (*a**) No significant differences in the sensory evaluation including smell, texture, and general acceptability among diet groups	[84]
Nannochloropsis gaditana	*Sparus aurata*	2.5% and 5%	Skin color: ↑ *L** (more lightness) ↑ *b** (more yellowish) ↓ *a** (more greennish)	[81]

(↑): indicates a rank increase and; (↓): indicates a rank decrease.

5. Effects of Microalgae on Fish Fillet Biochemical and Microbiological Parameters

Intensive fish culture can expose fish species to various factors, such as pollutants and drugs that can heighten the production of free radicals inside their body. The oxidation of lipid and protein by the produced free radicals will reduce fillet quality, induce an unpalatable flavor and odor, reduce the fillet shelf life, cause loss of nutritional values, and lead to unhealthy molecules. Using some feed additives such as microalgae for reducing the negative effects of oxidative stress on fish fillets is a novel approach, as indicated in Table 4. For example, substitution of fish meal and fish oil at 0, 30, 50, 70, and 100% with both *Arthrospira platensis* and linseed oil in a mullet (*Mugil liza*) diet exhibited an enhancement in antioxidant activity, especially at 50% inclusion; whereas 100% inclusion showed a diminution in the antioxidant capacity [85]. For ameliorating the deteriorative effects of arsenic in rainbow trout fillet, Sheykhkanlu Milan et al. [86] investigated the effects of dietary *Haematococcus pluvialis* at 0.28, 0.56, and 1.12% of their diets on fish fillet quality. The researchers discovered that when fish were fed with *H. pluvialis*, it resulted in reduced levels of pH, peroxide value, and the oxidation of proteins and lipids. Additionally, the expression of antioxidant genes (CAT, GPX, SOD, and GST) was significantly higher in the treated fish compared to the control group. This indicates that *H. pluvialis* can enhance the fish's ability to defend against reactive oxygen species by improving their antioxidant defense system. Therefore, dietary *H. pluvialis*, as a potent antioxidant, could protect fish fillets during the storage period. Protective effects of *Spirulina platensis* against the toxic impact of sodium sulfate in Nile tilapia muscle revealed a higher GPX level in fish muscle fed the microalga and challenged with sodium sulfate compared with challenged tilapia [87]. It was concluded that dietary *S. platensis* can be recommended to counteract the oxidation induced by sodium sulphate toxicity in fish species. A 12-week feeding Nile tilapia with microalgae mixture (*Nannochloropsis oculate*, *Schizochytrium* and *Spirulina* species) at 0.75%, 1.5%, and 3% exhibited a higher antioxidant capacity and reduced ROS, H2O2, and MDA levels especially at 3% inclusion [88], Meanwhile, contents of EPA and DHA in tilapia fillet were significantly enhanced with an increase in NSS (microalgae mix (NSS) containing *Nannochloropsis oculate* and *Schizochytrium* and *Spirulina* species) levels. Such antioxidant activity could be in part due to the availability of tocopherols, phenolic compounds, and carotenoids that can reduce the oxidative stress responses in different fish species [88].

Table 4. Effects of dietary microalgae on biochemical qualities of fish fillets.

Microalgal Species	Fish Species	Microalgae (%) Replacement with Fish Meal/Fish Oil/Dietary Inclusion	Effects	References
Spirulina platensis	Mullet (*Mugil liza*)	30, 50, 70, 100% substitution with fish meal	↑ antioxidant capacity No significant changes in MDA level	[85]
Spirulina platensis	Nile tilapia	1% diet	↑ GPX level	[87]
Haematococcus pluvialis	Rainbow trout (*Oncorhynchus mykiss*)	0.28, 0.56, 1.12% diet	↓ levels of pH ↓ peroxide value ↓ protein and lipid oxidation levels ↑ CAT, GPX, SOD and GST gene expressions	[86]
filamentous microalga Tribonema ultriculosum	Rainbow trout (*Oncorhynchus mykiss*)	50 g/kg 100 g/kg	↓ MDA level	[67]
Oedocladium carolinianum	*Trachinotus ovatus*	5% diet	In the liver: ↑ T-AOC and CAT, GSH-PX, andSOD activities ↓ the MDA level	[77]
Nannochloropsis oculate, *Schizochytrium*, and *Spirulina*	Nile tilapia	0.75%, 1.5%, 3% diet	↓ ROS, H_2O_2, and MDA levels ↑ Total antioxidant capacity	[88]

(↑): indicates a rank increase and; (↓): indicates a rank decrease.

Usually, fresh fish fillet is contaminated with different microorganisms available on skin, gill, and intestine tissues. However, specific spoilage organisms can penetrate the fish's muscle tissue, resulting in an unpleasant flavor and odor. Using natural additives in fish diets for improving the microbial characteristics of fillet quality is a novel approach [89]. In a study by Gulhan et al. [90] and Selamoglu et al. [91], rainbow trout and common carp administered with dietary propolis showed lower mesophilic and psychrophilic bacteria counts following a challenge with cypermethrin and arsenic. Similarly, Öz [92] showed that garlic-supplemented feed could decrease the number of total bacterial count as well as *Enterobacteriaceae* and psychotropic bacterial populations in rainbow trout fillet during storage at −18 °C. In contrast, no positive effects of dietary zeolite/chitosan and zeolite/nanochitosan composites on the rainbow fillet microbiological characteristics were observed [93]. The authors concluded that lower doses of these additives in fish diets might be the main reason for unchanged bacterial counts in fish fillets. Meanwhile, using alginate and carrageenan in the formulation of edible films showed antimicrobial properties against *L. monocytogenes*, e.g., in cold smoked salmon [94]. Despite the presence of few studies on the effects of some additives on fish fillet microbiological indices, in just one study, the effects of *Spirulina platensis* on rainbow trout microbiological aspects were assessed [84]. At the conclusion of the storage period, the total coliform measurements in fish that were given the PM/S diet were lower compared to all other diets.

6. Conclusions and Future Prospective

The inclusion of various microalgae in the diet of food fish has been found to positively impact fillet quality. The omega-3 fatty acid content, for example, has been found to increase significantly with the addition of certain microalgae. In addition, the presence of specific antioxidants and pigments in microalgae has been shown to improve the sensory properties of fish fillets, such as color, flavor, and aroma. Furthermore, microalgae have been found to enhance the preservation of fillets during storage. This is due to the presence of bioactive compounds that possess antimicrobial and antioxidant properties, which help to maintain the quality and safety of the fish product. However, regulatory and safety issues of microalgae and current challenges on the microalge productions should be considered.

In conclusion, the administration of microalgae to food fish can yield numerous benefits, including improvements in nutritional value, sensory properties, and shelf-life. The present review provides compelling evidence for the inclusion of microalgae in fish feed formulations which could have significant implications for the aquaculture industry as well as the seafood market. The findings suggest that microalgae could serve as a sustainable and cost-effective alternative to traditional fish feed ingredients, while also improving the nutritional quality of the final product. The potential benefits of microalgae in fish feed extend beyond economic considerations, as they could help to reduce the environmental impact of aquaculture and contribute to the development of a more sustainable food system. Overall, this review underscores the importance of further research and development efforts aimed at exploring the full potential of microalgae in fish feed formulations.

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