ALGAE IN NUTRITION AND COLOURATION OF ORNAMENTAL FISH : A REVIEW

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ABSTRACT

Algae grow in simple aquatic environments and about a fifth of the annually harvested 5 million kg of the biomass nourishes the cultured fish and shellfish, mainly for colouring and inducing biological activities, besides nutrition. These are potential fish feed, with varying nutritional values and are used as a source of carotenoids, astaxanthin, lutein and zeaxanthin for various fishes especially ornamental fishes. The deteriorating effects of synthetic pigments on environment emphasises the need for natural agents as ecofriendly alternatives. Their dietary application for better growth and attractive colouration needs further research. This review discusses the potential of algae for nutrition and colouration in ornamental fish in particular keeping the objective of their growth and profitability.

Keywords : Algae, aquaculture, colouration, fish growth, nutrition, ornamental fish

INTRODUCTION

Algae are a diverse group of largely aquatic, photosynthetic organisms. They are the most important primary producers in any aquatic ecosystem contributing to the first ring of the food chain in nature. Algae represent a valuable source of nearly all essential nutrients including PUFA (Polyunsaturated Fatty Acid), vitamins and minerals. As aquatic relatives of plants, microalgae thrive in aerated, liquid cultures with sufficient access to light, carbon dioxide and other nutrients (Rosenberg *et al.*, 2008). Approximately one fifth of 5 million kg/year of the algal biomass is used to nourish the fish and shellfish that are cultivated in aquaculture hatcheries (Muller-Feuga, 2004). The main applications of microalgae for aquaculture are associated with nutrition for colouring and for inducing other biological activities. Navarro and Sarasquete, 1998 and Biedenbach *et al.*, 1990 have found that algae have tremendous potential as fish feed, with varying nutritional values particularly for crustaceans and fish larvae. Researchers (Avron and Ben-Amotz, 1992; Yamaguchi, 1997) have concluded that algal genera like *Spirulina, Chlorella, Scenedesmus, Dunaliella, Nannochloropsis* are popularly used as aquaculture feed for their high nutritious value. Algal genera are used as a source of pigments like carotenoids, astaxanthin, lutein and zeaxanthin in fish farming especially for the coloured fishes (Hanaa *et al.*, 2003). Ornamental fish is one of the important items among the various types of commercially important fish marketed nationally and internationally. The modern ornamental fish culture and breeding operations are getting intensified with an increasing continuous supply of nutritionally balanced cost effective diets that provide all the essential nutrients to the fish (Sarkar, 1997). Pant *et al.*, (2002) observed that fish growth is influenced by the presence of natural food. In fish culture, the application of artificial feed is the most expensive input and therefore the supply of quality feed under various environmental conditions is necessary. Aquarium fish is significant not only for aesthetics but also for their immense commercial value in the global trade. Ideally, pigmentation is an accepted global parameter for ornamental fish 'quality'. Various scientists have found that colour is one of the major factors, for determining the price of aquarium fish in the world market (Lovell, 2000; Gouveia *et al.*, 2003). Pigments-fortified special diets are fed to trigger and enhance colouration it is an economical and practical way to achieve colouration, it is found that fish gain its colour through natural diets like algae as they synthesise and provide carotenoids, the pigmentation precursors. Bicyclic carotenoids like â-carotene and xanthophylls are converted to the most commonly occurring carotenoids in aquatic animals, astaxanthins - 3,3'–dihydroxy–*â,â*–carotene-4,4'-dione (Goodwin, 1986; Torrissen, 1989). Other essential nutrients besides pigments microalgal carriers also contain other essential nutrients, which determine aquarium species quality, survival, growth and resistance to disease. Pigmentation is affected by several factors, which include fish size, age, duration of supplementation, diet composition, source and concentration of carotenoids, sexual maturation and genetic factors. Environmental factors also influence pigmentation such as water temperature, salinity and light intensity (Torrissen *et al*., 1989; Storebakken and No, 1992).Successful commercial utilisation of microalgae has been established in production of nutritional supplements, antioxidants, natural dyes, and PUFA (Spolaore *et al*., 2006).

ALGAL TYPES

Algal species are morphologically varied and are economically important for various reasons. They also exhibit differential growth and anatomical characteristics. Table 1 summarises many such economically significant varieties, many of which overlap and are often one and the same thing with minute characteristic differences.

CHEMICAL COMPOSITION OF ALGAE

The main reason for considering the various sources of microalgae as a source of protein is their high protein content (Soletto *et al*., 2005). The amino acid pattern of almost all algae compares favorably with that of other food proteins. As the cells are capable of synthesising all amino acids, they can provide the essential ones to humans and animals (Guil-Guerrero *et al*., 2004). Carbohydrates in microlagae can be found in the form of starch, glucose, sugars and other polysaccharides. According to Becker (2004), their overall digestibility is high, which is why there is no limitation to using dried whole microalgae in foods or feeds. The average lipid content of algal cells varies between 1 and 70% but can reach up to 90% of dry weight under specified conditions (Metting, 1996). Microalgae also represent a valuable source of nearly all essential vitamins, *e.g.*, A, B1, B2, B6, B12, C, E, nicotinate, biotin, folic acid and pantothenic acid (Becker, 2004). In order to give a general overview on the major constituents, selected data of various microalgal species are compiled in Table 2. Further, the nutritional relevance of algae is not just restricted to aquatic animals. They have been useful supplements and/or compliments to conventional human foods, particularly as nutraceuticals or single cell proteins for the convalescing or the dilapidated. Table 3 describes the nutritional levels of single cell protein vis-à-vis various conventional foods for human consumption.

ALGAE FOR NUTRITION

Many fish including the ornamental fish varieties particularly of the tropical waters are good detriti- and plankti-vores. In fact, more than half of the total food requirements of even the culturable fish varieties are met from the primary productivity of a pond/aquatic body ecosystem. As per existing research reports, it is proven that particularly the ornamental varieties exhibit brilliant colours when fed with more natural food instead of artificial formulated feed. Table 4 summarises some such facts. Proper nutrition has long been recognised as a critical factor in promoting normal growth and sustained fish health. Prepared diets not only provide the essential nutrients that are required for normal physiological functioning, but also may serve as the medium by which fish receive other components that may affect their health (Gatlin, 2002). The value of dried microalgae as feed ingredients for crustaceans and fish larvae has been well demonstrated (Navarro and Sarasquete, 1998). Successful maintenance of 'difficult' species is often influenced by the aquarist's success in obtaining or rearing specialised food items (Floyd, 2002).

Unlike terrestrial plants that require irrigated fertile land, microalgae can grow in a wide range of habitats (Raja, 2009). Several hundred algal genera have been tested over the last four decades, and less than forty of them could pass the test in aquaculture as microalgae have tremendous impact on growth and vitality of fish. Khatoon *et al.* (2000) experimented with fresh *Nostoc ellipsosporum* and *Navicula minima* mixed with *Daphnia* to formulate the value-added feed and found that the experimental feed showed significantly higher protein, lipid and carotenoid, and increased level of amino acids and carbohydrate content. Goldfish fed with the feed showed a two-fold protein, glycogen and carotenoid level increase. They also found significant gain in body weight and specific growth rate together with high protein productive value, protein efficiency ratio and low feed conversion ratio in experimental fishes which signifies better utilisation of feed by fish. Khatoon *et al*. (2009) obtained similar results in the carotenoid content of prawn when fed with algal feed. The algal feed enhances the value of feed and promotes health conditions of aquaculture animals.

Spirulina platensis has been largely studied due to its commercial importance as a source of proteins, vitamins, essential amino acids, and fatty acids (Cifferi and Tibboni, 1985), and more recently, as a potential source of nutraceuticals (Borowitzka, 1995). Ahamad (1966) and Nell O'Connor (1991) found an overall improvement in the general condition, increase in weight, volume and size of fishes fed with the algae. Mustafa and Nakagawa (1995) reviewed the supplementation of macro- and microalgae meal enhanced growth, feed utilisation, lipid metabolism, body composition, disease resistance and carcass quality of a variety of fishes. Supplementing fish feeds with algae has certain physiological merits such as vitamin precursors, growth promoters and essential fatty acids. Research on the use of algae as a protein source in formulated feed is scant such as the reported enhanced growth in certain food-fish such as silver carp (Nakagawa *et al*., 1987; Chow and Woo, 1990).

Green alga *Ulva lactuca* (Chlorophyta) and red alga *Pterocladia capillacea* (Rhodophyta) are among the most abundant macroalgae available in excessive amounts round the year. This study showed that dietary supplementation significantly improved feed intake or palatability and protein intake. Fish fed this diet also produced significantly higher weight gain percent and daily weight gain (Wassef *et al*., 2005**)**. The importance of these algae, as possible alternative protein sources or supplement for cultured fish has been recently recognised (Mustafa and Nakagawa, 1995).

According to Chakraborty *et al*., (2007), microalgae feeds are currently used mainly for the culture of larvae and juvenile shell- and fin-fish, and raising the zooplankton to feed juvenile animals. Algal food is provided to newly-hatched rotifers (zooplankton) until it reaches the desired size, then a boost of algae just prior to harvesting to increase the nutritional value of the target culture species feeding on it. Nile tilapia juveniles obtain major nutrition (more than 48%) for growth from feeding only on algae (Elnady *et al*., 2010). Sommer *et al*. (1992) reported enhanced growth in trouts by adding carotenoid-rich microalgae *Haematococcus pluvialis*. Boonyratpalin and Unprasert (1989) observed a positive effect of dietary carotenoid on the growth of red tilapia.

Spirulina positively affected the growth performance and feed efficiency of Nile tilapia as well as its resistance to *A. hydrophila* infection (Mohsen Abdel-Tawwab *et al*., 2008). They reported a 05-10 g *Spirulina* in a kg of formulated diet as optimum, and found significant increase in weight and specific growth rate. *Spirulina* had high quality protein content (59-65%), more than many conventional sources, *viz*., 35% in dry soybean, 25% in peanuts or 8-10% in grains (Sasson, 1997). Thus, *Spirulina* can be a cheaper part supplement or complete replacement for protein in aqua-feeds (Habib *et al*., 2008).

Spirulina is regarded as a super food for ornamental fish as it increases and promotes uniform growth rates when fed at 0.5-2.0% inclusion rate. It improves the intestinal flora and feed conversion ratio in fish by breaking down the otherwise indigestible feed components. The beneficial internal flora produce vitamins and displace harmful microbes which is why fish fed *Spirulina* have less intestinal compaction, slimmer abdomen, and better infection resistance (Anon, 2010). It stimulates the production of enzymes that transport fats within the fish body. The fish utilises the fat to power growth instead of just storing it and becoming flabby. The chlorophyll and phycocyanin also facilitate skin colouration which is especially important for koi and goldfish for commanding a higher market price. Studies on marine yellowtail showed that the fingerlings fed a ration of 0.5% *Spirulina* resulted in a significant gain in survival over the non-*Spirulina* fed group similar was the experience of professional Discus fish breeders (Anon, 2010).

ALGAE FOR COLOURATION

Fish are naturally coloured, and the colouration fades under intensive culture conditions. It is found that fish can't synthesise carotenoid and depend on dietary carotenoid content for colouration. Hence, a direct relationship between dietary carotenoids and pigmentation exists (Halten *et al*., 1997). Fish skin colour is mainly attributed to the presence of chromatophores that contain pigments including melanins, pteridines, purines and carotenoids (Chatzifotis *et al.*, 2005). Carotenoids are a group of over 600 natural lipid soluble pigments primarily produced in phytoplankton, algae and plants. These are absorbed through the diets in animals, sometimes transform into other carotenoids, and incorporate into various tissues. Fish skin colours depends primarily on the presence of chromatophores (xanthophores and erythrophores) containing carotenoids (*e.g.*, astaxanthin, canthaxanthin, lutein, and zeaxanthin). Fish, like in many other animals, can't perform *de novo* synthesis of carotenoids (Goodwin, 1984); they modify alimentary carotenoids and store them in the integument and other tissues. Different species have different carotenoids metabolism and storage mechanisms. Carotenoid-protein complexes and dietary carotenoids are the main source of fish skin and muscle pigmentation (Sales and Janssens 2003, Lovell 2000, and Chatzifotis *et al*., 2005). Hence, to increase skin and flesh colour in captivity, fish must obtain optimal level of carotenoids from their diet (Sinha and Asimi, 2007).

According to Anderson (2000), carotenoids also have excellent antioxidative characteristics. Various scientists have observed that if colour-enhancement can be done by administering pigment enriched feed, it will definitely improve the quality and cost of the fish. *Chlorella vulgaris* was found to be as efficient pigments synthesiser in rainbow trout *Oncorhynchus mykiss* (Gouveia *et al*., 1996), gilthead seabream *Sparus aurata* (Gouveia *et al*., 2002), koi carp *Cyprinus carpio* and goldfish *Carassius auratus* (Gouveia *et al*., 2003; Gouveia and Rema, 2005). Gouveia *et al*. (2003) reported increased total skin carotenoid content in Kawari (red), Showa (black-n-red), and Bekko (black-n-white) varieties of koi carp (*Cyprinus carpio*) and goldfish (*Carassius auratus*) by feeding *Chlorella vulgaris*, *Haematococcus pluvialis* and *Arthrospira maxima* dietary carotenoid supplements. *C. vulgaris* was found to be most effective in koi carp and goldfish colouration as ascertained by total carotenoid content. A red hue in goldfish was maximal with *H. pluvialis* supplementation. Algal genera are being used in fish farming especially for the coloured fishes as a source of pigments like carotenoid, astaxanthin, lutein and zeaxanthin (Hanaa *et al*., 2003). The economic importance of microalgae has been explored since long. A general overview on some chemical characteristics of *Spirulina* is given in Table 5. *Spirulina* is very suitable as a dietary supplement. The table also describes the composition of another alga, *Haematococcus pluvialis*, found worldwide that produces astaxanthin having a potential for use in aquaculture feeds. Chemically synthesised astaxanthin is a different stereoisomer from natural astaxanthin (Ako *et al*., 2000) and is a costly proposition for the small grower to incorporate into fish diets. (Ako *et al*., 2000) incorporated carotenoids from *Spirulina platensis* and *Haematococcus pluvialis* in coloured swordtails, rainbowfish and topaz cichlids and also similarly in case of rosy barbs, 24K mollies, kissing gouramis.

Dunaliella salina is a microalgae occurring naturally in a number of locations worldwide. The microalga has been introduced in two forms, liquid paste and dry powder to aquaculture. The paste is a *D. salina* concentrate with levels of high-carotenoids, minerals, vitamins, and fatty acids. The paste is spray dried to create the dry form (www.nutracol.com). Gouveia and Rema (2005) investigated the effect of concentrations of carotenoid sources on goldfish skin pigmentationin relation to temperature and found best results in *C. vulgaris* at 26-30°C.

Zatková *et al*. (2010) examined the effect of carotenoid-rich microalgal (*Scenedesmus, Chlorella* and *Haematococcus*) biomass as feed supplement (12-60 mg/kg feed) in an albino form of wels catfish (*Silurus glanis*) and reported an increase in specific growth rate and physical condition index. Lorenz (2000) reported that Koi fed with *Haematococcus* (NatuRose) assimilated xanthophylls exhibiting dark red colouration. He also reported significant progress in colour and pigmentation in freshwater and marine ornamental fish tetras, cichlids, gouramis, goldfish, danios, swordtails, rosy barbs, rainbow fish, discus and clown anemone fish. *Spirulina* added in the diet of goldfish and fancy red carp enhanced pigmentation (Miki *et al*., 1986; Borowitzka, 1994). Comparative analyses of various microalgae (*Chlorella vulgaris, Haematococcus pluvialis* and *Spirulina maxima*) with synthetic astaxanthin (Carophyll Pink) showed that microalgal biomass, especially of *C. vulgaris*, contributed to enhanced skin colour in koi and goldfish (Gouveia *et al*., 2003). Skin colour affects market price and play a major role in the overall appraisal (Gouveia and Rema, 2005). Feeding *Chlorella vulgaris* produced positive pigmentation in certain fish (Gouveia *et al*., 1996; 2002). Microalga *Chlorococcum* sp. is a promising source of astaxanthin, canthaxanthin and adonixanthin (Higuera-Ciapara *et al*., 2006). Although some authors claim that the biological functions of carotenoids in fish are still speculative (Choubert *et al*., 2005), other consider these compounds as important micronutrients that fish are unable to synthesise and, therefore, must be included in the diet (Baker *et al*., 2001).

Studies with microalgal biomass supplementation have shown that *Chlorella vulgaris* is efficient as synthetic pigments in the pigmentation of rainbow trout, *Onchorhynchus mykiss* (Gouveia *et al*., 1996), gilthead seabream, *Sparus aurata* (Gouveia *et al*., 2002), koi carp, *Cyprinus carpio* and gold fish, *Carassius auratus* (Gouveia *et al*., 2003).

Table 1 : Various economically significant algal forms in aquaculture practices

Algal form	General characteristics
Slime/smear algae (Blue-green algae)	Grow rapidly as blue-green, slimy sheets, spreading rapidly over almost everything. Usually, indicates poor water quality. These can fix nitrogen and may be seen in aquaria with extremely low nitrates; can also be seen between the substrates and aquarium sides.
Brown algae	Found in soft brown clumpy patches in freshwater aquaria, these are usually diatoms. Presence of these usually indicates a low light or an excess of silicates.
Green algae	Green unicellular algae sometimes reproducing so rapidly that the water turns green, commonly referred as an ‘algal bloom’. It is usually caused by too much light, including direct sunlight. Green water is very useful in raising *Daphnia* and brine shrimp. Algal film grows on aquarium glass and forms a thin haze.
Hair algae	Grows in bright green clumps in the gravel, around the base of plants like *Echinodorus* and around mechanical objects. It has a coarser texture than beard algae. Hair algae tend to form matted clumps. Individual strands can get to 5cm or more. This is a popular food supplement for fish among European aquarist.
Thread algae	These algae grow as long and thin strands up to 30 cm or more. They tend towards a dull green colour. They usually indicate an excess of iron (> 0.15 ppm).
Staghorn algae or Horse-hair algae	These algae look like individual strands of hair algae but tend to grow in single branching strands like a deer antler and it is grey green in colour. They seem to grow mostly on tank equipment near the surface.
Spot algae	Normally these algae grows in thin, hard, circular, bright green spots, usually on the aquarium glass but also it found on plants under high light conditions.
Fuzz algae	These grow mostly on plant leaves as separate, short (2-3mm) strands. These might be a less virulent form of beard algae. They can be controlled by algae eaters such as black mollies, *Otocinclus*, *Peckolita* and Siamese algae eaters.
Beard algae	They grow on plant leaves and are bright green in colour. Individual strands have a very fine texture but grows in thick patches and looks just like a green beard, grows upto 4 cm.
Black brush algae	They grow in feathery black tuts 2-3 mm long and tend to collect on slower growing leaves like *Anubias*, some *Echinodorus* and other wide leaf plants.

Source : George Booth, http://www.aquaticscape.com/articles/algae.htm

Table 2 : General composition of some common economically significant algae (% dry matter basis)

Alga	Protein	Carbohydrates	Lipids
Anabaena cylindrical	43–56	25–30	4–7
Aphanizomenon flosaquae	62	23	3
Chlamydomonas rheinhardii	48	17	21
Chlorella pyrenoidosa	57	26	2
Chlorella vulgaris	51–58	12–17	14–22
Dunaliella salina	57	32	6
Euglena gracilis	39–61	14–18	14–20
Porphyridium cruentum	28–39	40–57	9–14
Scenedesmus obliquus	50–56	10–17	12–14
Spirogyra sp.	6–20	33–64	11–21
Arthrospira maxima	60–71	13–16	6–7
Spirulina platensis	46–63	8–14	4–9
Synechococcus sp.	63	15	11

Source : Becker, 2007

Table 3 : General composition of different human food sources and algae (% of dry matter basis)

Commodity	Protein	Carbohydrate	Lipid
Bakers' yeast	39	38	1
Meat	43	1	34
Milk	26	38	28
Rice	8	77	2
Soybean	37	30	20
Anabaena cylindrica	43–56	25–30	4–7
Chlamydomonas rheinhardii	48	17	21
Chlorella vulgaris	51–58	12–17	14–22
Porphyridium cruentum	28–39	40–57	9–14
Dunaliella salina	57	32	6
Scenedesmus obliquus	50–56	10–17	12–14
Spirulina maxima	60–71	13–16	6–7
Synechococcus sp.	63	15	11

Source: Becker, 2004

CONCLUSION

Some microalgae that have been exploited for the millennia are *Nostoc* in china and *Arthospira* in Chad and Mexico. However, closed system cultivation of these has begun in Japan and Israel, and *Chlorella* in Germany. In view of deteriorating effects on the environment due to use of synthetic pigments, researchers are emphasising on the need for natural pigmenting agents as alternatives to synthetic chemicals. Further, algal production system needs improvement to become more competitive and more economically feasible through integrated farming approaches. This review emphasises on the application of different types of potential algae for nutrition and colouration in ornamental fish for their optimal growth and higher commercial profitability. Their application through formulated feed for better growth and attractive colouration needs further emphasis.

Table 4 : Various algae-eating ornamental fish along with their feeding bahaviour

Fish form	Characteristic behavior
Siamese algae eaters	Otherwise known as SAEs, siamese algae eaters (*Crossocheilus siamensis*) eat black brush hair (red) algae but have not been known to eat forms of beard algae. They are good algae eaters and very mild mannered.
Otocinclus cats (dwarf sucking catfish)	Otocinclus are diligent algae eaters, but are kept in schools due to their small size. Most are good algae eaters but some seems to prefer the slime coats on fish to algae. Otocinclus cats, or dwarf sucking catfish *Otocinclus affinis* are good algae eaters though they mainly deal with algae in its beginning stages when it is short. They eat mainly diatoms.
Plecos	Plecos, *Hypostomus punctatusare*, widely known as the algae eater fish store. The bristle nose is a nice addition to any planted tank eating many forms of algae.
Butterfly goodied	Butterfly goodied (*Ameca speldens*) simply devours hair/ thread/beard algae.
Black mollies	Black sailfin mollies (*Poecillia latipinna*) are excellent candidates for the break in periods of a planted tank since they are cheap and easy to find. Usually considered expendable and are removed after a month or so. If they are fed, they will not be quite so eager to consume algae. When they are hungry, they are eager algae consumers; seen during the break period.
Plecostomus sp.	A generic name for a wide range of sucker mouth fish. Only the smaller types are useful in a planted tank, since the larger varieties tend to eat the plant along with the algae. Two common useful types are the bristle-nose *Plecostomus* and the clown *Plecostomus* or pekolita. Both stay under for long and do not seem to cause too much plant damage.

Source : George Booth, http://www.aquariumsecrets.com

Table 5 : Mean proximate dry weight composition of *Spirulina* and *Haematococcus*

Parameter	*S. platensis*	*H. pluvialis*
Protein (%)	≤ 70	20-30
Carbohydrate (%)	14-19	30-40
Fat (%)	5-6 (mainly ã-linolenic acid –w-6 fatty acids)	7-25
Ash (%)	3-9	5-15
Fibre (%)	3-7 (especially mucilage)	-
Moisture (%)	4-9	4-9
Vitamins (%)	10 (Vit. B, C, B_{12}, Folic acid, niacin, riboflavin)	-
Nitrogen-free extract (%)	15-20	-
Colour	Dark blue to green	Red to dark red
Pigments (%)	*â*-carotene, xanthophylls, zeaxanthin, *â*-cryptoxanthin etc.	Total catotenoids > 1.75 Astaxanthin > 1.5
Particle size	64 mesh through	5-25 ì
Density (tapped; g/ml)	0.35-0.60	0.37-0.44 g/ml

Sources: http://www.botanical-online.com/english/spirulina.htm for *Spirulina*; Dore and Cysewski, http://www.ruscom.com/cyan/web02/pdfs/naturose/nrtl09.pdf for *Haematococcus*

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