The role of carotenoids in enhancing the health of aquatic organisms

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Abstract
Carotenoids constitute a class of pigments derived from photosynthetic plants, algae, bacteria and some fungi. Carotenoids act as potent antioxidants scavenging reactive oxygen species. They hold a vital role in the antioxidant defense system of humans. Several studies have shown carotenoid can increase the growth overall well being of aquatic animals. This review examined the effect of carotenoid in aquatic organisms on growth, survival, immunity, pigmentation and reproductive performance. The review recommends the use of carotenoid as feed additive for enhance the overall improvement of the aquatic organisms.

Keywords: carotenoids, growth, survival, pigmentation, immunity, reproductive performance

1. Introduction
Carotenoids are group of natural pigments contributes many of the hue in nature. The basic structure of carotenoid is a symmetrical tetraterpene skeleton formed by the conjugation of two C20 units, which could be deemed the backbone of the molecule. Based on their composition, carotenoids are subdivided into two groups. Carotenoid which comprises solely carbon and hydrogen atoms are collectively assigned as carotenes. The majority of natural carotenoids contain at least one oxygen functional group, referred to as xanthophylls. Only plants, bacteria, fungi and algae can synthesize carotenoids; animals cannot biosynthesize them thus, they must be obtained from the diet [1]. Carotenoids play a critical role in the photosynthetic process and they carry out a protective function against damage by light and oxygen. Antioxidants, immunoregulators, pro-vitamin A is the distinct roles of carotenoids. Furthermore, the mobilization of the pigment from muscle to ovaries implies a purpose in reproduction [2]. It has similarly mentioned that fishes with a large content of carotenoids are further resistant to microbial diseases [1]. The long conjugated double-bond system is the major feature of carotenoid which make them able to absorb light of wavelength 400-500nm from electromagnetic spectrum [3]. The chemical structure of the carotenoids plays an important role in their oxygen scavenge properties [4].

2. Application of carotenoids in aquaculture
Aquaculture is the farming of aquatic organisms, including fish, molluscs, crustaceans and aquatic plants. Farming intends some sort of intrusion in the rearing method to improve production. The feed choice and feed management practices have a significant impact on the economic performance of a production system [5]. Several feed additives have been incorporated in shrimp feed to generate resistance against various stressors and thereby increase aquaculture production. Carotenoid is one such compound that plays a significant function in industrial aquaculture. Carotenoids are chiefly employed in diets of crustaceans, salmonids and other farmed and ornamental fishes being pigment sources for desirable coloration. Besides, carotenoids will serve as antioxidant that helps to mitigate the oxidative stress.

2.1 Growth performance and survival
Various studies are reported the influence of carotenoid pigment on the development and
survival of the aquatic organisms (Table: 1). The carotenoid source of these studies varies from synthetic carotenoids [6, 7, 8, 9, 10] to natural carotenoids [11, 12]. Chien [13] reported a 77% increase in survival rate for shrimp fed 100mg/kg astaxanthin supplemented diet in contradiction to shrimp enriched with β-carotene which equated to 40%. An 88.2% increased survival was observed in L. vannamei fed 350 ppm carotenoids enriched diet (Tagetes erecta) for 5 weeks rearing contrasted to 76.5% in the control [14]. Petit [15] noticed that feeding astaxanthin-based diet at 60 mg kg\(^{-1}\) over 8 weeks showed notable decrease in mortality of adult shrimp (Penaeus japonicas) than those individuals receiving carotenoid-free diets. Yamada [9] proclaimed an increased survival rate of 91% for P. japonicas supplemented with 100 ppm carotenoid contrasted to 57% in the control group. The authors further elucidated that astaxanthin is more effective than β-carotene or canthaxanthin as a pigment source in P. japonicus. P. indicus larvae exhibited markedly greater survival rate (88%) from PZ1 stage until metamorphosis when fed the astaxanthin-enriched nematodes Panagrellus redivivus (1.43 μg astaxanthin g\(^{-1}\) dry weight of nematode), while neither larval growth nor development was affected [18]. Survival (100%) was greater in shrimp (L. vannamei) fed paprika (Capsicum annuum) than in those fed basal diets (80.5%) [17]. Hyphessobrycon callistus was supplied with nine pigmented diets containing AX-astaxanthin, BC- β-carotene and MX-1:1 mixture of AX and BC at different concentrations (10, 20, and 40 mg/kg). No differences in growth and survival of the fish among treatments were found after 8 weeks rearing [18].

**Table 1: Effect of carotenoids on various parameters to aquatic organisms**

| Carotenoid                        | Source                        | Organism studied                  | Effect                                                                 | Reference |
|----------------------------------|-------------------------------|-----------------------------------|----------------------------------------------------------------------|-----------|
| Cantaxanthin/ astaxanthin         | Synthetic                     | *Salmo salar*                     | Astaxanthin and cantaxanthin supplemented diet promoted growth rate during the early start-feeding periods. No significant effect on survival and pigmentation to the eyed, hatching, and alevin stage. | [6, 5]    |
| Astaxanthin β-carotene            | Synthetic                     | *Penaeus japonicas*              | No notable variations in daily feed intake, percent gain or feed efficiency on feeding various pigments diets. | [7]       |
| Astaxanthin β-carotene algal meal | Synthetic                     | *Dunaliella salina*              | The average weight gain was higher in algal meal fed group. Prawns fed the astaxanthin diet had a higher rate of survival than those supplemented with β-carotene or algal meal diets. | [19]      |
| Cantaxanthin astaxanthin          | Synthetic                     | *Penaeus japonicus*              | The shrimp fed canthaxanthin had a growth rate higher that of individuals receiving the other three diets and least growth rate for standard diet. | [8]       |
| Astaxanthin                       | Synthetic                     | *Salmo salar L.* (First-feeding fry) | The conclusions recommend a minimum of 3.1 mg astaxanthin/kg diet to achieve maximum growth and supreme endurance throughout the start-feeding period. | [20]      |
| Astaxanthin                       | Synthetic                     | *Salmo salar L.* (Juveniles)      | The mean weight of the organism supplemented with astaxanthin was significantly higher than those fed the unsupplemented diet. The highest survival was achieved by 125-300 mg Dunaliella extract/kg fed group related to the control. | [21]      |
| Astaxanthin                       | Synthetic                     | *Penaeus monodon*                | Increased survival rates, higher total antioxidant status and improved hepatopancreatic function were shown by dietary astaxanthin fed group under ammonia stress. | [22]      |
| β-carotene                       | Synthetic                     | *Penaeus monodon*                | Shrimp fed 125 - 300 mg of the Dunaliella extract/kg diet for 8 weeks showed higher weight gain and survival related to the control. Survival of all groups fed β-carotene supplemented diet were significantly higher than control groups during 9 days of low dissolved oxygen stress. | [23]      |
| Astaxanthin                       | Synthetic                     | *Litopenaeus vannamei*            | Supplemented astaxanthin at 90mg kg\(^{-1}\) improved growth, survival and moult frequency in shrimp. Under salinity stress, shrimp fed astaxanthin supplemented diet (80 mg/kg) had significantly greater concentration of glucose, haemocyanin, lactate in haemolymph and total haemocyte count. | [9]       |
| Natural astaxanthin               | Synthetic                     | *Litopenaeus vannamei*            | Postlarvae given Natural astaxanthin diet had significantly higher growth performance and astaxanthin content. The mRNA expression levels for the antioxidant enzymes (cMnSOD and GPx) also increased for Natural astaxanthin (90 ppm) fed shrimp under the stress of Vibrio parahaemolyticus infection. | [24]      |
| Synthetic astaxanthin             | Synthetic                     | *Litopenaeus vannamei*            | Astaxanthin diet fed group shows better growth performance, with the best performance exhibited by in the 400 mg kg\(^{-1}\) diet astaxanthin supplemented group. | [10]      |
| Synthetic astaxanthin             | Synthetic                     | *Marsupenaeus japonicus*          | No notable variations among treatments concerning the total growth parameters (p > 0.05). However, the Pollen extract with carotenoid (50mg kg\(^{-1}\)) diet rendered a positive effect on growth parameters. | [11]      |
| Astaxanthin                       | Synthetic                     | *Litopenaeus vannamei*            | Specific growth rate (SGR) and weight gain were significantly greater in treatment groups compared to control group. Optimal dose of dietary astaxanthin activated metabolic pathway to enhance growth by favoring the expression of many vital genes. | [12]      |
2.2 Immunity
An early research revealed that dietary intake between 230 and 810 mg astaxanthin kg⁻¹ diet for 4 weeks improved the immunity of postlarvae giant tiger prawn (*P. monodon*) against salinity shock [20]. Another study pointed out that astaxanthin (200 mg kg⁻¹ diet) was effective in increasing the endurance of *P. monodon* postlarvae to low salinity stress [23]. Additionally, Chien [21] noticed that dietary inclusion of astaxanthin (360 mg kg⁻¹ feed) for 1 week appeared to induce optimal tolerance in the larval stages of *P. monodon* upon exposure to 4 h of low dissolved oxygen level (<1 mg L⁻¹). The observations made when different stress factors were tested on *P. monodon* juveniles that received astaxanthin (80 mg kg⁻¹ diet) over 8 weeks also exhibited enhanced antioxidant defense capability, better hepatopancreatic function and subsequent improvement recovery against osmotic and thermal stresses [28]. Similarly, *P. monodon* juveniles fed diet supplemented with 71.5 mg astaxanthin kg⁻¹ feed displayed a sounding antioxidant status and elevated resistance to ammonia stress [24]. Supamattaya [30] found that *P. monodon* supplemented 200–300 mg Dunaliella extract kg⁻¹ diet were more endurable to hypoxic conditions (0.8–1 mg L⁻¹) along with significantly greater resistance to white spot syndrome virus (WSSV), while measures of phenoloxidase assay and total haemocyte count were negatively correlated.

Wang [18] studied antioxidant activities of *H. callistus* modified with dietary carotenoid type viz; astaxanthin, β-carotene and combination of both (1:1) at 10, 20, and 40 mg/kg concentrations. Dietary astaxanthin had more numbers of negative correlations with antioxidant parameters in fish than β-carotene. Pham [24] authenticated lesser liver and plasma SOD activities in *Paralichthys olivaceus* supplemented with carotenoid than the control group.

2.3 Pigmentation
Menasveta [26] reported a 318% increase in carotenoids from the tissue of carotenoid fed group than those fed the commercial diet without carotenoid had a carotenoid increase of only 14%. A noticeable increment of carotenoid content in the exoskeleton was reported when animals were provided with *Spirulina*-supplemented diets [33, 34, 35]. Arredondo-Figueroa [14] unveiled that pigmentation of *L. vannamei* was influenced by carotenoid supplemented diet. Abdomen coloration produced by 200 ppm carophyll and unesterified marigold diet is insignificant. Red porgy (*P. grapsus*) were fed with 100 ppm astaxanthin obtained 27.7 µg g⁻¹ carotenoid from skin, while fish fed non-carotenoid supplemented diet (control) had 4.33 µg g⁻¹ skin [16]. Nine pigmented diets includes carotenoid diet (CD) and its combination (AX-astaxanthin, BC- β-carotene, MX-1:1 combination of AX and BC) at three concentrations (10, 20, and 40 mg/kg) were used for feeding *H. callistus*. Body AX and BC content increased with increasing dietary CD concentration [18]. Pham [24] reported that skin coloration and total carotenoid content of olive flounder (juvenile) is increased by dietary supplementation of carotenoid (paprika, *H. pluvialis* extract and raw *H. pluvialis*). Ruangsomboon [37] discovered that 30% inclusion of *Arthrospira platensis* in feed as carotenoid supplement improved fish color in Red tilapia (*Oreochromis sp.*).

2.4 Reproductive performance
Pangantihon- Kuhlmann [38] provided valuable insight on the improved fecundity, ovarian development and spawning of *P. monodon* broodstock when fed with astaxanthin (100 mg kg⁻¹ diet) for 61 days. In another related study, *P. monodon* broodstock performance assessed in terms of number of spermatooza in male shrimp and amount of eggs in gravid female was greatly enhanced when fed with 500 mg astaxanthin kg⁻¹ diet [39]. Dietary intake of 150 mg astaxanthin kg⁻¹ feed (compared to 50 and 100 mg levels) for 150 days significantly promoted the spermocrit value, sperm concentration, motility, osmolality and fertilization rate of goldfish *Carassius auratus* [42]. In rainbow trout *O. mykiss*, supplementation of astaxanthin are deemed necessary for optimum reproduction [41], Scabini [42] reported the influence of supplemented carotenoids from paprika oleoresin on gilthead seabream broodstock performance and seems a significant improvement in broodstock performance via egg viability, hatching rates and fecundity.

3. Conclusion
Investigations on supplementation of carotenoids to the diet of aquatic organisms came out with promising results that the dietary inclusion of carotenoids would enhance the growth and general performance of the animal simultaneously with marked reduction in their mortality. In addition, a large amount of empirical data suggests that sufficient carotenoids supply is essential for the wellbeing of the animal.

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