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## EFFECT OF DIETARY ASTAXANTHIN ON GROWTH AND PIGMENTATION IN BALLOON MOLLY FISH (*POECILIASPHENOPS*)

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### ABSTRACT

*These studies were conducted to investigate the effect of astaxanthin on the growth performance and skin pigmentation of Poeciliasphe nops (Balloon Molly). Three astaxanthin supplemented diets were prepared to contain astaxanthin @ 40, 80 and 120mg kg<sup>-1</sup> of diet. The basal diet without astaxanthin supplementation were used as control. Color changes and growth parameters i.e. weight and length and hydrobiological parameters were observed during the study. In each treatment, 5 fishes were introduced in three replicates. The maximum colour changes in fishes fed on diet containing astaxanthin @12mg/100 gm of feed was shown whereas the maximum growth were also observed in the treatment where fishes were fed on diet containing astaxanthin@12mg/100 gm of feed (3.040 gm, 4.733 cm) followed by @8mg (2.913gm, 4.467 cm), 4mg (2.653 gm, 4.267 cm) and control (2.447 gm, 4.100 cm). The water parameters were found in optimum range during the experimental period. The enhancement of coloration can be achieved by pigment enriched feed and improve the quality and cost of the fishes i.e. attractive colorations determine the commercial value of ornamental fish.*

### History of Article

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### **Introduction**

Ornamental fishes fascinate people of all ages. Keeping aquarium at home, office and public places is one of the most popular hobbies around the world. Coloration is one of the most important factors to deciding the aesthetic and

market values of the ornamental fishes. Pigmentation in the skin is responsible for coloration in the fish. The body colors of the fish are predominantly dependent on the presence of special cells in the tissue, called chromatophores. The enhancement of coloration

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can be done by administrating pigment enriched feed. It will definitely improve the quality and cost of the fish. It has been recognized that changes of fish color can be divided into two categories; one is physiological color change, which is attributed to rapid motile responses of chromatophores and the other is morphological color change, which results from changes in the morphology and density of chromatophores. Chromatophores can concentrate the pigment granules to the center of the cell to make the skin become lighter, or the pigments be dispersed, hence darkening the skin.

Carotenoids are so heavily focused upon because they are most influenced by the diet. The various carotenoid resources have been suggested natural resources such as crustacean and insects and some scientist believed synthetic carotenoid (including  $\beta$ -carotene, canthaxanthin, zeaxanthin and astaxanthin) can be used in daily diet (1,2). Other coloring/shades are affected more indirectly by food, requiring a balanced diet in order to produce pigment in fish.

Carotenoids also improve survival rate, metabolism and reproduction enhance resistance to several stress conditions as well as pigmentation of fishes (3,4,5,6). The main carotenoid is astaxanthin which leads to red-pink coloured aquatic animals and has used in aquaculture, so far. There is an increasing demand for carotenoids in the international market due to the numerous applications of carotinoids in the pharmaceutical trade. The aqua feed trade seeks environment friendly sources of pigments to improve coloration and enhance commercial acceptability within fish feeds trades, and fat-soluble pigments such as carotenoids are promising for optimal nutritional requirements and coloration of fish (7).

### Material and Method

Balloon Molly, *P. sphenops* Cyprinidae of uniform size group (1.33 to 1.38gm and 3.53 to 3.63 cm) were purchased from a commercial fish farm and were acclimatized to laboratory conditions for one week before the start of experiment. The experiments were conducted for a period of 45 days and were carried out in aquarium containing 30litrecapacities. These aquariums were grouped into four sets and each set consists of three aquariums. The fishes were fortnightly weighed and length was recorded. For the present investigation, the feed was purchased from the market and mixed astaxanthin 40, 80 and 120 mg per Kg of diet in the form of dry pellets. The fishes were fed with 5% of their body weight daily. The basal diet without astaxanthin supplementation was used as a control. Water in the experimental troughs was changed everyday for the sufficient supply of oxygen to the fishes. At the end of the experiment, all fish were starved for one day to take the final wet weight, length and colour changes.

### Result

Present study was carried out in Aquaculture laboratory, CCSHAU, Hisar for a period of 45 days. After 45 days of study, the change in colour was observed which is shown in Plate 1 (A-D). The maximum colour changed was observed in fishes fed on diet containing astaxanthin @12mg/100 gm of feed followed by @ 8 mg/100gm and @ 4mg/100gm. In the present study, increased muscle carotenoid content was observed in all groups in line with increasing dietary astaxanthin inclusion. The growth of fishes in different treatment recorded in the table 1 and 2. The maximum growth was observed in the treatment where fishes were fed on diet containing astaxanthin@12mg/100 gm of

feed (3.040gm, 4.733cm) followed by @8mg (2.913gm, 4.467 cm), 4mg (2.653 gm, 4.267 cm) and control (2.447 gm, 4.10 cm). The water

parameters were found in optimum range during the experimental period (Table 3).



A.



B.



C.



D.

Plate1: Fish showing changes in colour after fed on control diet (A); fish fed on diet containing astaxanthin @4mg/100 g of feed (B); fish fed on diet containing astaxanthin @8mg/100 g of feed (C); fish fed on diet containing astaxanthin @12mg/100 g of feed (D)

**Table 1 Effect of astaxanthin on weight of Balloon Molly fish (*P.sphenops*)**

| Treatment         | Weight of Balloon Molly fish (g) after days |              |              |              | Mean         |
|-------------------|---|--------------|--------------|--------------|--------------|
|                   | 0   | 15           | 30           | 45           |              |
| Control           | 1.337                                       | 1.670        | 2.140        | 2.447        | <b>1.898</b> |
| 4 mg/100gm feed   | 1.377                                       | 1.957        | 2.380        | 2.653        | <b>2.092</b> |
| 8 mg/100 gm feed  | 1.383                                       | 1.953        | 2.623        | 2.913        | <b>2.218</b> |
| 12 mg/100 gm feed | 1.357                                       | 1.980        | 2.537        | 3.040        | <b>2.228</b> |
| Mean              | <b>1.363</b>                                | <b>1.890</b> | <b>2.420</b> | <b>2.763</b> |              |

CD (p=0.05) for Treatment (T) =0.051; SE(m)= 0.018

CD (p=0.05) for Observation period(OP) =0.050; SE(m)= 0.017

CD (p=0.05) for TxOP = 0.102; SE(m)= 0.035

**Table 2: Effect of astaxanthin on length of Molly fish (*P.sphenops*)**

| Treatment        | Length of Molly fish (cm) after days |              |              |              | Mean         |
|------------------|--------------------------------------|--------------|--------------|--------------|--------------|
|                  | 0                                    | 15           | 30           | 45           |              |
| Control          | 3.533                                | 3.733        | 3.967        | 4.100        | <b>3.833</b> |
| 4 mg/100g feed   | 3.667                                | 3.767        | 4.067        | 4.267        | <b>3.942</b> |
| 8 mg/100 g feed  | 3.567                                | 3.867        | 4.267        | 4.467        | <b>4.042</b> |
| 12 mg/100 g feed | 3.633                                | 3.900        | 4.267        | 4.733        | <b>4.133</b> |
| Mean             | <b>3.600</b>                         | <b>3.817</b> | <b>4.142</b> | <b>4.392</b> |              |

CD (p=0.05) for Treatment (T) =0.054; SE(m)= 0.019

CD (p=0.05) for Observation period(OP) =0.054; SE(m)= 0.019

CD (p=0.05) for TxOP = 0.108; SE(m)= 0.037

**Table 3: Hydro biological parameters of the glass aquarium water during the experiments**


| Parameters                         | Range     |
|------------------------------------|-----------|
| Temp.(°C)                          | 25-29     |
| Dissolved oxygen (DO)(mg/l)        | 6.0-6.8   |
| pH                                 | 7.4-7.8   |
| Conductivity ( $\mu$ S $cm^{-1}$ ) | 580 – 720 |
| Alkalinity (mg/l)                  | 160-210   |

### Discussion

Ornamental fishes are recognized for their bright and beautiful coloration and shapes. Coloration is due to the skin pigments but background may also be due to underlying tissue and body fluids. As the aquaculture feed trade seeks a pigment to improve coloration and to enhance commercial acceptability so there is a great potential for use of carotenoids for pigmentation in aquaculture. Red carotenoids, mainly associated with astaxanthin and canthaxanthin, are the predominant organic natural pigments of interest in aquaculture that should be provided through diet as fishes are unable to synthesis these important compounds *de novo* (8, 9). It is well documented that these pigments play vital roles in variety of different concepts of fish physiology including behavior, immunity,

growth, nutrition and reproduction (10, 11, 12, 13, 14, 15). These results are strongly supported by finding of other studies in which carotenoids were used for fish pigmentation (16, 17, 18, 19). The fishes in control and treatment were fed with astaxanthin @50 and 100 mg  $kg^{-1}$  diet demonstrated almost the same CV% of the muscle carotenoid content. The most familiar reaction of carotenoid metabolism in fish is primarily oxidative (20). Fish can bio-convert main carotenoids through reductive metabolic pathways (21, 22). The basal diet which is used in this study was made from different ingredients involving fish oil and fish meat.

It is reported that muscle carotenoid concentration of 11.8 mg  $kg^{-1}$  would be obtained in rainbow trout after four weeks of feeding with diet including synthetic astaxanthin (23). The

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similar improvements in the muscle carotenoid contents after feeding with supplemented carotenoid also reported in discus fish (24) and in olive flounder fish (25).

Several carotenoids are known to have a positive role in the intermediary metabolism of fish and crustaceans, improving growth and enhancing feed utilization (25) in present study, feeding European seabass has been observed positive effects on growth or feed utilization as well as on survival of natural carotenoids obtained from marigold petals meal (ML) or crab waste meal (CM), had positive effects on growth or feed utilization as well as on survival (26). Carotenoids like astaxanthin are categorized as micronutrients in fish feeding. So, the physiological effects of these compounds are not directly inserted into somatic growth as they act and put effects, such as antioxidants, indirectly in various physiological aspects and may ultimately result in improved growth.

The present study on the skin carotenoid content demonstrated that the dietary astaxanthin supplements has significant effects on the fish skin pigmentation with increasing along with the concentration of the astaxanthin inclusion. At the end of the experiment, the concentration of the skin pigmentation was significantly high in diet containing @ 120 mg/kg astaxanthin followed by @ 80 mg/kg astaxanthin then @ 40 mg/kg astaxanthin. With the increasing the muscle carotenoid content in the experimentally fed fishes with dietary astaxanthin is obviously resulted as ingestion and accumulation of supplemented astaxanthin in diet.

### Conclusion


The aim of the present study was to investigate the effect of dietary astaxanthin on growth and skin pigmentation in terms of carotenoid concentration in body tissues of Balloon Molly

fish (*P. sphenops*) during a 45-days period of administration of supplemented feed with astaxanthin carotenoid. This is best achieved by the use of quality manufactured feeds and supplements with a balanced mixture of colour enhancing ingredients. It paves the way to many aqua based industries to promote the product as natural synthetic ingredients and colorants.

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