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EFFECTS OF SPIRULINA POWDER IN COLOURATION AND GROWTH ENHANCEMENT OF AN INDIGENOUS ORNAMENTAL FISH *TRICHOGASTER LALIUS*

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ABSTRACT

Body colouration of fin fishes is one of the most important factors that increase their ornamental value in the market. In the present study juveniles of *Trichogaster lalius* were exposed to different concentration of spirulina powder incorporated diets to observe the impact in pigmentation of the respective fish in the captive condition. Three different concentration feed were prepared by incorporating spirulina powder at the rate of 2 g/kg (A-2), 4 g/kg (A-4) and 6 g/kg (A-6) feed and one kept as control. Eight set of glass aquarium of 50 l capacity (30 × 30 × 30 cm) were used for 4 set of experiment with their respective replicates. The impact of spirulina treated feed on pigmentation of fishes was analyzed for a period of 60 days. After the 60 days of experiment growth rate was found increasing significantly ($p<0.05$) when they were fed with different dose of spirulina powder incorporated diet and best result were obtained at 2 g/kg treated feed in aspects of growth rate and enhanced colouration. Findings of the result concluded that spirulina powder incorporated feed has very good potential to enhance the pigmentation of fishes.

KEYWORDS: Spirulina powder, growth parameters, carotenoid, pigmentation, *Trichogaster lalius*

INTRODUCTION

Trichogaster lalius (dwarf gourami) locally known as "lalia or kholse" an indigenous ornamental fish under Perciformes belongs to the family Osphronemidae was selected for the present study. In West Bengal the species faced good demand in ornamental as well as commercial fish market and sold with Rs. 10-20 per pair while sold as ornamental purposes. It also has a high foreign demand as an aquarium fish. It is naturally collected and exported to foreign countries such as USA, Japan, Sri Lanka, Germany, Taiwan, Thailand, Bangladesh, Malaysia and China. 'Lalia' widely distributed in South Asia from Pakistan, India and Bangladesh. Male 'dwarf gourami' usually larger then female in size and possess a good market demand due to having orange colour with an iridescent blue stripes, where females are dull in colour. Male dwarf gourami can reach up to a length of 7.5 cm and female of 6 cm. 'Dwarf gourami' is a peaceful fish and can live in a community tank with other ornamental fishes. For this reason, neon blue dwarf gouramies (*Trichogaster lalius*) prefer for the colour enhancement treatment due to increase its demand in local as well as global market. Use of cheap and easily available natural additives to enhance colouration of fishes to minimize the expenditure and will help ornamental fish entrepreneurs to maximize their earnings. Spirulina powder, easily available in local market and can be smoothly incorporate as feed ingredients. Spirulina powder has been chosen for this reason as a colour enhancer and increasing fish growth. The present study was undertaken in laboratory condition to observe the impact of spirulina powder on colouration and growth enhancement of an indigenous ornamental fish *Trichogaster lalius*.

MATERIALS & METHODS

Adult males of Dwarf gourami (*Trichogaster lalius*) of uniform size group was purchased from local ornamental fish markets. Fishes were transported to the laboratory by plastic bag with oxygen packing. The experiment was carried out in the Department of Fisheries Resource Management, Faculty of Fishery Sciences, West Bengal University of Animal and Fishery Sciences, Kolkata, West Bengal. In the laboratory the fishes were given a short bath treatment with 3 ppm $KMnO_4$ solution for 3 to 5 minutes for disinfection and subsequently, they were transferred carefully to the rectangular tanks containing chlorine free tap water for acclimatization. Fishes were fed with prepared pellet feed (without spirulina) in two equal rations *i.e.* at 09.00 and 18.00 hours at the rate of 2% of their body weight. Healthy and uninjured juveniles with weight and length ranged from 0.9 ±0.1 g and 4 ±0.2 cm were selected for further experiment. After proper acclimatization healthy specimen were stocked in to the experimental tanks for feeding experiment. A total 8 set of glass aquarium of 50 l capacity (30 × 30 × 30 cm) were used for 3 set of experiment with replicate and one set kept as control. In each tank 10 numbers of juveniles were reared and the total experiment was conducted for 60 days. In the experimental tank fishes were first kept for a week with normal feed to make them adjusted with the

environment. Suitable aeration and floating aquatic weed Hydrilla (*Hydrilla verticillata*) were provided to the tank to give the fishes a natural habitat. Spirulina powder was mixed with the prepared pelleted feed at the rate of 2 g/kg (A-2), 4 g/kg (A-4), 6 g/kg (A-6) feed and one remained as control. The feed ingredients used to prepare the experimental diet provided in the Table 1.

TABLE1: Proportion of feed ingredients used in formulated diet

Name of ingredients	Percentage content in 100 gm feed
Fish meal	25
Soyabean meal	22
Groundnut oil cake	15
Rice bran	20
Wheat flour	12
Starch	3
Soya powder oil	2
Vitamin & mineral mix	1

Fishes were fed with experimental and control feed twice daily (09.00 and 18.00 hours) at 2% of body weight. Water was exchanged in each alternative day and left over feed and excreta were siphoned out every day. In every fortnight, sampling was done randomly from the stocked specimen for their growth and carotenoid estimation. In each sampling, 20 numbers of fishes were taken for growth estimation and 2 healthy fishes were selected for carotenoid estimation. For estimation of total carotenoid content in skin, fin and muscle of test fish procedure was followed as described by Harpaz and Padowicz, 2007 with following equation: 4 × Optical density value × total volume of sample taken / weight of sample (mg). Water quality parameters in each tank maintained with optimum ranged. The persistence rate of carotenoids in skin, fin and muscle as well as growth of dwarf gourami were statistically analysed through one Way ANOVA and Paired t-Test by using the software packages (SPSS Statistics v16 and Microsoft Excel 2010).

RESULTS & DISCUSSION

Effects of spirulina power in growth enhancement of *Trichogaster lalius*

The fortnightly mean length of the fish fed with spiruline incorporated fed and control feed were provided in the spirulina Table 2. The mean feed length increment was found to be highest when spirulina powder mixed with 2 g/kg feed. Student't-test' showed that length of fish was increasing significantly ($p<0.05$) when they were fed with different dosed of spirulina powder incorporated diet. The fortnightly mean weight increment was also found to be highest when the feed mixed with 2g/kg spirulina incorporated diet and weight increment was significantly different ($p<0.05$) when they were fed with different dosed of spirulina powder incorporated diet (Table 3).

TABLE 2: Fortnightly mean length increment of *T. lalius* fed with spirulina incorporated feed and control feed

Days	Control	A-2	A-4	A-6
Initial (cm)	4.0±0.0	4.0±0.0	4.0±0.0	4.0±0.0
15 days (cm)	4.0±0.01	4.23±0.1	4.11±0.01	4.13±0.02
30 days (cm)	4.1±.07	4.31±0.07	4.26±0.07	4.27±0.02
45 days (cm)	4.1±.07	4.52±0.03	4.38±0.04	4.4±0.2
60 days (cm)	4.1±.07	4.66±0.01	4.5±0.07	4.59±0.03

TABLE 3: Fortnightly mean weight increment of *T. lalius* fed with spirulina incorporated feed and control feed

Concentration	Days					Weight gain
	Initial (g)	15^{th} day (g)	30^{th} day (g)	45^{th} day (g)	60^{th} day (g)	(mg/g/day)
A-2	0.9±0.02	0.91±0.0	0.91±0.0	0.93±0.0	0.93±0.0	0.06
A-4	0.9±0.02	1.38±0.2	1.47±0.1	1.65±0.6	1.72±0.08	3.18
A-6	0.9±0.02	0.97±0.1	1.13±0.1	1.13±0.0	1.35±0.1	0.42
C	0.9±0.02	1.28±.05	1.34±0.05	1.35±0.1	1.56±0	2.89

Nandeesha *et al*. (2001) found that growth rate was improved in carp when fed with spirulina. Domenio and Edo (2005) observed that high feed intake; body weight gain and specific growth rate was found in fish silver sea bream (*Rhabdosargus sarba*) when consumed spirulina containing diet. The growth of fish is species specific in respect of weight gain and certain concentration of additives. In case of dwarf gourami 2 g/kg of spirulina powder while incorporated with feed was found to be most suitable for weight gain. Nakazoe *et al*. (1986) reported that, 5% supplementation of spirulina resulted the higher body weight gain in nibbler, *Girella punctatus* which may be due to the higher size of fish.

To estimate total carotenoid concentration in skin, fin and muscle of *T. lalius* four different spectrophotometric wave lengths (WL) such as 380, 450, 475 and 500 nm were measured for the respective samples. In the current study total amount of carotenoids were determined by following the path way of Alagappan (2004) used for Blue gourami, *Trichogaster trichopterus*. In case of *Etroplus maculatus*, black tiger shrimp, 200-600 nm wave lengths were used to measure carotenoid present in tissue extract by Manimegalai *et al*. (2010). Therefore, it was agreed that for accurate estimation of carotenoid concentration in skin, fin and muscle of fish the ideal wave length should be ranged from 200 to 600 nm.

Effects of Spirulina Power in Colouration

The initial carotenoid value in skin was 0.54µg/g. A maximum carotenoid concentration of 6.52 µg/g (at 380 nm) was obtained in skin of those fishes which fed on 2 g/kg spirulina powder incorporated diet followed by 4 g/kg and 6g/kg spirulina powder incorporated diet fed fish. The overall variation in carotenoid level of fin was observed at 380 nm as shown in Figure 1. Significant level ($p \leq 0.05$) of skin colouration was observed when the fishes were fed with different level of spirulina powder treated feed. The initial carotenoid value in fin was of 0.2µg/g and at the end of experiment the carotenoid value went up to 6.36µg/g (at 380 nm). A maximum amount of carotenoids were recorded in diet incorporated with spirulina powder at concentration of 2 g/kg followed by 6g/kg, 4g/kg spirulina and control fish (Figure 2). The overall variations in carotenoid level of muscle were presented in Figure 3. It was ranged from 0.04 to 2.2µg/g. A maximum amount of carotenoids were recorded in 6 g/kg spirulina powder incorporating diet fed fish followed by 4 g/kg, 2 g/kg and control fish.

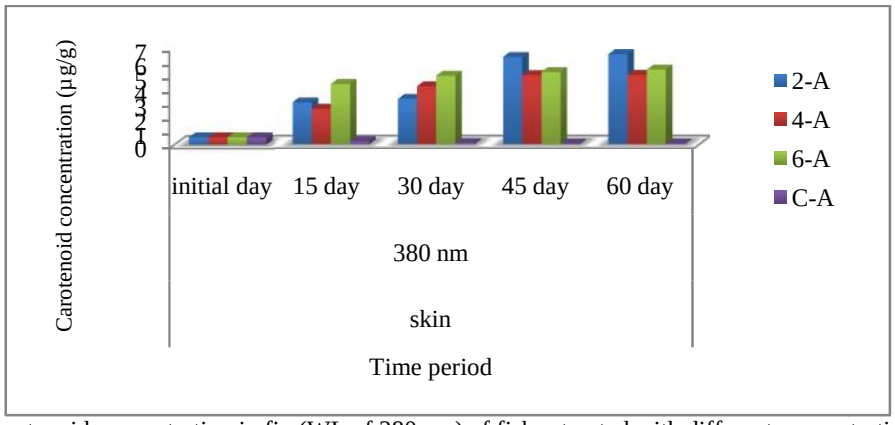

FIGURE 1: Carotenoid concentration in fin (WL of 380 nm) of fishes treated with different concentrations of spirulina treated feed and control feed

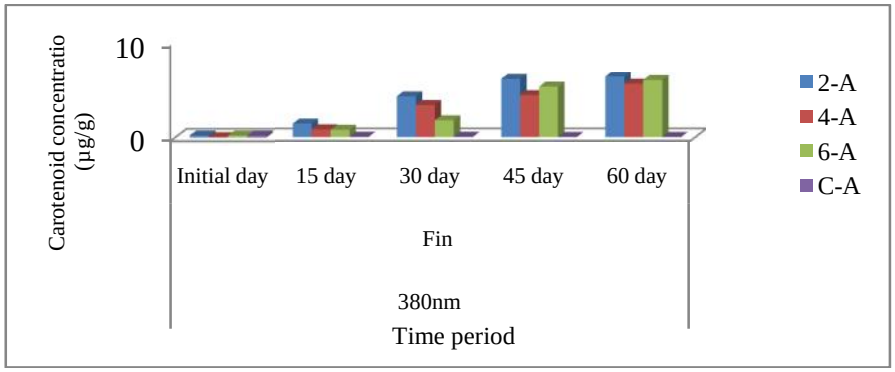

FIGURE 2: Carotenoid concentration in fin (WL of 380 nm) of fishes treated with different concentrations of spirulina treated feed and control feed

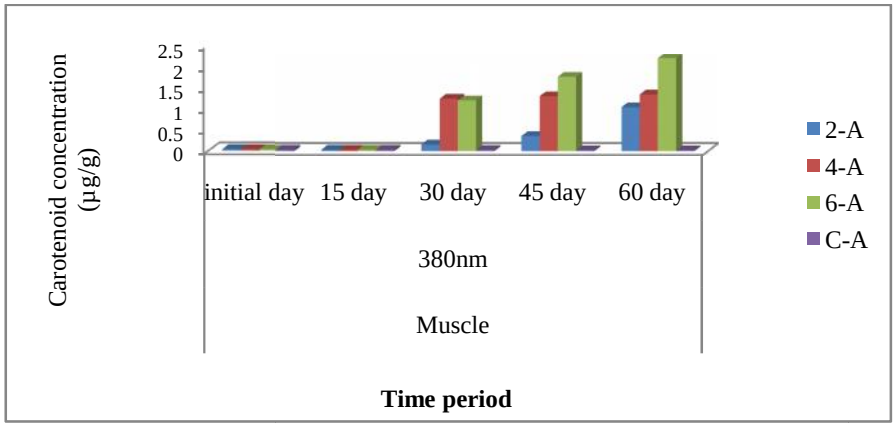

FIGURE 3: Carotenoid concentration in muscle (WL of 380 nm) of fishes treated with different concentrations of spirulina treated feed and control feed

Colour plays a major role in the overall preference of any pet fish. As fish cannot synthesize pigments into their body, they rely upon dietary supplementation of carotenoids to maintain their natural colouration. Synthetic carotenoid sources mostly used to enhance colouration in fishes but it is expensive and sometime cause's negative impact on fish metabolism (Teimouri *et al*., 2013). Natural carotenoids provide better colouration to fishes. Several authors (Ako *et al*., 2000; Barbosa *et al*., 1999; Torrissen and Naevdal, 1988 and Storebakken *et al*., 2004) used natural carotenoid source to increase colouration of swordtail, *Xiphophorus hellerii*; rainbow fish, *Pseudomugil furcatus* and topaz cichlids, *Cichlasoma myrnae*. Ako *et al.* (2000) found that topaz cichlids, *Cichlasoma myrnae* became significantly coloured when they fed a diet containing 1.5-2.0 g of a carotenoid-rich strain of *Spirulina platensis*. Alagappan *et al.* (2004) mentioned that carotenoid value in skin of blue gourami was 13.64 ±0.03 µg/g. According to Vasudhevan *et al*. (2013) each fish has a specific capacity to utilize carotenoids from diet. Total skin carotenoid value of *T. lalius* differed from blue morph and blue gourami in spite of using same carotenoid source.

In case of Rainbow trout higher dose that was 10% of spirulina in diet resulted highest carotenoid deposition reported by Teimouri *et al*. (2013). On the other hand Allaf *et al*. (2014) reported that 2% beet root juice caused adequate colouration in both skin and fin of Rainbow trout. Allaf *et al.* (2014) reported that the addition of beet-root juice powder in diet had a positive effect on fillet of rainbow trout. According to Ranjan (2016) only about 5 to 15 g of the dietary carotenoids was utilized for muscle pigmentation of fish. May be due to this reason 6 g spirulina powder fed fish showed more carotenoid deposition in muscle as compare to other doses of spirulina powder. Vasudhevan *et al*. (2013) who had conducted similar experiment on gold fish and found that 50 g *Azolla* per kg provide more carotenoid deposition in muscle as compare to 0, 25, 100 and 200 g/ kg *Azolla* incorporated diet. A proportionate increased in carotenoid content of muscle was observed with dietary supplementation of spirulina in *Xiphophorus helleri* (James *et al*., 2006) but on the other hand in case of rainbow trout lowest dose of astaxanthin caused more pigment deposition in muscle rather than higher doses of astaxanthin (Choi *et al*., 2016). As reported by Xiangjun *et al*. (2012) more amount of carotenoids derived from some plant sources were not improved the growth and feed utilization of Japanese ornamental carp (Koi) due to high levels of carbohydrate and cellulose in plant sources. May be due to this reason 4 g/kg and 6 g/kg spirulina in diet fed fish showed a weight gain which was less as compare to 2 g/kg spirulina in diet fed fish. *Spirulina* was used by many scientists (Ezhiland Narayanan, 2013; Mori *et al*., 1987, Okada *et al*., 1991) to enhance colouration of Blue morph, *Pseudotropheus lombardoi*, Swordtail, *Xiphoporus helleri*. The result of the present experiment suggested that dietary inclusion of 2 g/kg spirulina powder incorporated diet was able to improve body colour of dwarf gourami more effectively as compare to other doses of spirulina powder in diet.

CONCLUSION

Carotenoids are absorbed in animal diets, sometimes transformed into other carotenoids, and incorporated into various tissues. Fishes in the wild obtain the food of the quality required for proper growth, pigmentation, and nutrient profile. Spirulina powder, easily available in local market and can be smoothly incorporate as feed ingredients. Spirulina powder not only useful in pigmentation in fishes but also enhanced growth efficiency in fishes and 2-4 g/kg spirulina powder incorporated diets may be recommended in commercial culture to enhance the growth and pigmentation in dwarf gourami.

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