Applied Biological Research 27(3): 340-346; (2025)
DOI: 10.48165/abr.2025.27.01.33

RESTORATION OF NATURAL COLOURATION IN INDIAN ROSY BARB (*Pethia conchonius*) USING ONION PEEL POWDER IN HOME AQUARIA

Sunita Pradhan and Joydev Maity*

Department of Fishery Sciences, Vidyasagar University, Midnapore 721 102, West Bengal (India)
*e-mail: *jmaity2003@gmail.com, sunita.pradhan1956@gmail.com*

(Received 15 January, 2025; accepted 12 August, 2025)

ABSTRACT

The use of natural carotenoids in fish-feed to intensify the colouration is a sustainable and cost-effective choice for ornamental fish farmers to meet the demand of radiant ornamental fish within budget. Onion (*Allium cepa*) peel powder in formulated feed was used to restore natural colour in Indian rosy barb (*Pethia conchonius*) fish. A basal diet was prepared by using fishmeal, soybean meal, groundnut oil cake, rice bran, wheat flour, starch powder, soya oil and vitamins & minerals. Experimental diets were prepared by replacing equivalent rice bran with onion peel powder at three concentrations *viz*., 5% (D_1), 10% (D_2), and 15% (D_3) of the basal diet (D_0). The feeding trial was conducted for 60 days. At the end of feeding trial, a significant increase in total carotenoids concentration (4.3 ± 0.2 µg g^{-1}; $p < 0.05$) and weight gain (35.17%) was observed in rosy barb of D_2 (10%) diet. All the tanks showed 100% fish survival. The result revealed that 10% inclusion of onion peel powder in diet of Indian rosy barb can give a fruitful impact on restoration and retention of rosy barb's natural colour in home aquaria.

Keywords: Carotenoids, colouration, ornamental fish, onion peel, pigment, rosy barb, weight gain

INTRODUCTION

Indian waters host a rich diversity of over 300 ornamental fish species. The visual appeal of these aquarium fishes can help to relax mind and reduce stress. A variety of aquarium fish are characterised by their wide diversity of colours and colour patterns. Intensity of fish colour, shape and size of fish fins are the main factors that reflect the market price of ornamental fishes (Paripatananont *et al.*, 1999). Colour is one of the essential prerequisites that confirms the value of ornamental fish within the global market (Saxena, 1994). However, fish that kept under captivity for long time often experienced fading of natural pigmentation due to the absence of dietary carotenoids (Chapman, 2000; Sinha and Asimi, 2007).

Fish body colour is mainly associated to the presence of chromatophores which contain pigments, such as melanins, pteridines, purines and carotenoids (Chatzifotis *et al.*, 2005). Fish like other vertebrate animals cannot synthesize colouring pigments on their own (Goodwin, 1951). Dietary carotenoids along with carotenoid-protein complexes are the major source of fish body pigmentation (Sales and Janssens, 2003; Chatzifotis *et al.,* 2005). Therefore, to maintain the fish body colour in captivity, they must obtain an optimum level of carotenoids through their diet (Sinha *et al.*, 2007). In Nature, the fish diet contains lots of carotenoids which are responsible for pigmentation in fish, but in a home aquarium, with time fish lose their bright colour due to a lack of natural diet availability. As the success in the aquarium fish trade is well dependent on the dazzling colour and alluring appearance of the fish (Pandey and Mandal *et al.*, 2017); aquarium keepers are exploring various methods to

retain the body pigmentation in fish. Perusal of literature suggests that various agro-industrial wastes, such as fruit and vegetable wastes (e.g., peels, pomace, etc.), are rich in carotenoids and can serve as cost-effective natural pigment sources (Sharma *et al.,* 2021). The Indian rosy barb (*Pethia conchonius*) is known for its red colour, which rapidly fades in captivity. The present study studied the potential of onion peel powder, a carotenoid-rich agro-industrial waste, as a natural dietary pigment source for restoring colouration in the fish.

MATERIALS AND METHODS

Collection and acclimatization of experimental fish

Indian rosy barb fish (*Pethia conchonius*) of Cyprinidae family were purchased from the ornamental fish market of Gallif Street, Kolkata (India) for experimental purpose. The collected fish were brought to the laboratory packed in oxygen sealed plastic bags. Fish were then left for 15 days under laboratory condition to acclimatize. During this period, fish were fed with formulated basal diet @ 4% body weight. Healthy disease-free fish specimens (250 in number with average weight: 1.92 ± 0.19 g; average length: 3.3 ± 0.43 cm) were selected for the experiment.

Preparation of onion peel powder

Onion peel was collected from local vegetable market. Washed thoroughly to remove dirt and soil; sun dried for 7-8 h followed by grinding in fine powder and stored in air tight zip lock pouches. Carotenoid content of peel was measured as 21.2 µg 100 g^{-1} following spectrophoto-metric method (Gavril *et al.*, 2024).

Experimental setup and aquarium management

The experiment was conducted in a laboratory of Fishery Sciences Department, Vidyasagar University, Paschim Medinipur, West Bengal (India). Fish were randomly allotted to four treatment groups: D_0 (control), D_1 (5% onion peel), D_2 (10%), and D_3 (15%). Each treatment was replicated three times with 20 fish specimens reared in each 60 L glass aquarium; along with air stone for extra oxygen diffusion. The initial carotenoids content along with weight and length were noted. Food particles and metabolic wastes were siphoned off from the bottom of aquarium on every three days.

Analysis of water quality parameters

Water quality parameters like temperature, pH, dissolved oxygen (DO) level, alkalinity, and hardness were analysed every fortnight following standard APHA methods (1998). Partial water exchange was carried out in every 15 days to maintain acceptable water quality parameters.

Preparation of experimental diet

For the preparation of 30% protein-basal feed, the selected basic ingredients such as fish meal, soybean meal, rice bran and wheat flour were ground separately and thoroughly mixed. The basal diet, which served as control, comprised of (per 100 g diet) fish meal 30 g, soybean meal 40 g, rice bran 18 g, wheat flour 8 g, and starch powder 3 g. A dough was prepared by adding the required amount of lukewarm water. This dough was steamed in a water bath at 70°C for 40 min and then left to cool. After cooling, soya oil (2 mL) and vitamins & mineral premixes (1 g) were added and mixed thoroughly. The dough was placed in a hand pelletizer to prepare 1-2 mm size feed pellets. The pelleted feed was then air-dried in hot air oven since carotenoids lose their efficacy at high temperatures and under light conditions. The feed was then kept in airtight containers until use. The experimental diets were prepared by adding onion peel powder as a carotenoid source at three concentrations (5, 10, and 15%) to the basal feed replacing the equivalent amount of rice bran.

Analysis of total carotenoid in fish tissue

The UV spectrophotometric analysis of total carotenoid was done to observe the change in carotenoid concentration in body tissue of Indian rosy barb fed with onion peel powder incorporated diet at

different concentration. Total carotenoid content of different fish tissues like skin, muscle and fins was analysed as per Olson **(**1979**)** method at 450 nm wavelength spectrophotometerically (Shimadzu UV-1800). During the trial period, carotenoid concentration of fish body was analysed fortnightly.

$$\text{Total carotenoid } (\mu g\ g^{-1}) = \frac{\text{Maximum absorption at wavelenght} \times \text{Dilution Factor}(10)}{\text{Extinction Factor } (0.25) \times \text{Sample weight in g}}$$

Data analysis

Body weight gain was evaluated to assess the growth performance of fish under study. FCR (feed conversion rate) was calculated by depending on biomass gain of the fish. At the end of experiment, all the data were statistically analysed using statistical package SPSS. A homogeneity of variance was tested using Levene's test to ensure the validity of statistical analysis. Comparison among all the treatments was done by one-way ANOVA. The level of significance employed was 0.05. Comparison between treatments at different time intervals was done by Tukey's HSD Test. Standard error mean (SEM) and critical differences (CD) between the treatments was calculated by statistical package SPSS.

RESULTS AND DISCUSSION

Water quality maintenance

During feeding trial, water quality parameters maintained were: temperature - 20-25°C, pH - 6.6-7.4; dissolved oxygen - 7.9-8.3 mg L^{-1}, water hardness - 210-250 mg L^{-1} and alkalinity - 50-54 mg L^{-1} (Table 1). The water quality values were quite similar in control and treatment tanks. Throughout experimental period, the water quality parameters were monitored and maintained within the optimal values fortnightly. During and after completion of 60 days experiment all fish were found healthy with no signs of stress or external infection. Indian rosy barb fish is highly adaptable to culture conditions and capable of living in a wide range of environments (Chapman, 2000). According to Pailan *et al.* (2012), optimum range of water temperature and pH for Indian rosy barb rearing varied between 18 to 22°C and 6.0 to 8.0, respectively. The water quality in present study revealed that water in experimental tanks is likewise optimal for Indian rosy barb rearing; however, no statistical analysis or comparison between treatments were done on water quality data.

Table 1: Water quality of water in experimental tanks rearing Indian rosy barb fish

	Temperature (°C)		pH		D.O. (mg L^{-1})		Hardness (mg L^{-1})		Alkalinity (mg L^{-1})	
	Max.	Min.	Max.	Min.	Max.	Min.	Max.	Min.	Max.	Min.
Initial	21	19	6.6	6.5	7.9	7.8	210	204	53	50
Day 15	22	20	7.2	7.0	8.0	7.9	250	245	50	49
Day 30	22	19	7.0	6.9	8.2	8.0	220	210	53	51
Day 45	24	22	7.4	7.2	8.3	8.2	233	229	54	52
Day 60	25	23	6.9	6.8	8.3	8.1	243	237	50	49

Proximate composition of the formulated diets

Table 2 reveals the proximate composition of all formulated diets. Similarities of ingredients was maintained, except for carotenoid sources in various diets. The proximate analysis of each test diets

Table 2: Proximate analysis (%) of formulated fish feeds for Indian rosy barb

Chemical components (%)	Control feed (D_0)	D_1 feed	D_2 feed	D_3 feed	SEM	$CD_{0.05}$
Crude protein	29.86	29.70	29.78	29.80	0.742	1.71
Crude fat	13.72	13.76	13.73	13.78	0.482	1.11
Carbohydrate	36.20	36.08	36.14	36.18	0.244	0.56
Ash content	08.67	08.78	08.70	08.45	0.466	1.07
Carotenoid content (μg g^{-1})	0^a	1.065^b	2.13^c	3.195^d	0.295	0.68

D_0, D_1, D_2, and D_3 were supplemented with 0, 5, 10, and 15 g onion peel powder 100 g^{-1}, respectively

demonstrated the adequate compositions of protein, fat and carbohydrates. The targeted protein in all diets was 30%, resulting in 29.70% to 29.86% for all feeds that were adequate for rearing and maintaining good health of ornamental fish.

Feed utilization and growth performance

All formulated diets were well accepted by fish throughout the experiment. At the beginning of study, the average weight of fish specimens was 1.92 ± 0.1 g. Table 3 presents the data regarding growth performance at the completion of study. Initial and final weight, and length of fish were estimated at the beginning and end of 60 days experimental period. Highest weight gain was observed in D_2 group *i.e.* 35.17%, followed by D_1 (32%), D_3 (26.57%) and D_0 (26.15%). The highest length gain was observed in control (D_0) group (9%), followed by D_2 (8.7%); while lowest length gain was observed in D_1 and D_3 tanks, both having 8.6% growth. The increased growth in D_2 group may be due to the synergistic effect of increased feed palatability, presence of bioactive antioxidant, and efficient carotenoid supplementation. All these seem to have resulted in increase in feed consumption and digestive efficacy, and simultaneously minimized oxidative stress, resulting in an improved feed conversion ratio (0.98) and increased weight gain (35.17%). Conversely, increased inclusion (15%) seemed to have brought about an overload of flavonoids such as quercetin that disrupts the functionality of enzymes as well as nutrient uptake, thereby inhibiting further growth enhancement (Nile *et al.*, 2018; Sharma *et al.,* 2021).

The survival rate of fish was 100% in all the tanks which is an indication of non-toxic nature of carotenoid source incorporation in the feed. The feed conversion ratio (FCR) was highest in control (1.28), followed by D_2 (0.98) and lowest in D_3 (0.96). Fish growth efficiency is high when FCR value is close to 1.0 (Fry *et al.,* 2018). In this study FCR of formulated diet-fed fish varied in between 0.96 and 1.28, which is an indication of good feed utilization. The FCR value showed reduction for fish fed with control diet, whereas fish fed with experimental diets showed optimum values. In comparison with other feed the FCR value of D_2 feed is good which is 0.98, close to the value of 1.

Total carotenoid concentration study in fish tissue

Ornamental fishes are particularly distinguished by their wide range of body colours and different colour patterns. Colour is an important criterion that controls the price of ornamental fish in global market (Torrisen, 1989; Saxena, 1994). As fish cannot produce their required carotenoids they get it from the diets in nature. But in captivity they are compelled to get carotenoids from the formulated diets incorporated with different carotenoid sources. Using carotenoid content of onion peel powder as the natural pigment source in Indian rosy barb's formulated diet resulting into its natural colour restoration in home aquarium. In this study, the fish group fed on D_2 diet *i.e.* 10% onion peel powder incorporated diet showed most carotenoid accumulation, followed by D_3 (15%), and D_2 (5%) onion peel powder incorporated diet. Initially the total carotenoid content in Indian rosy barb was 2.29 $\mu g\ g^{-1}$. After feeding onion peel incorporated diets for 60 days the total carotenoid concentration increased, highest (4.3 ± 0.2 $\mu g\ g^{-1}$) being at 10% level, beyond which no further increment in carotenoids content was observed in other treatment groups (Fig. 1).

Table 3: Growth performances, feed utilization and survival of fish fed with onion peel powder

Parameters	Control (D_0)	D_1	D_2	D_3	SEM	$CD_{0.05}$
Initial weight (g)	1.92 ± 0.19	1.80 ± 0.16	1.88 ± 0.19	2.10 ± 0.19	0.149	0.32
Final weight (g)	2.60 ± 0.49	2.65 ± 0.54	2.90 ± 0.35	2.86 ± 0.38	0.364	0.84
Initial Length (cm)	3.30 ± 0.44	3.40 ± 0.39	3.30 ± 0.44	3.40 ± 0.46	0.345	0.80
Final Length (cm)	3.50 ± 0.43	3.70 ± 0.39	3.60 ± 0.43	3.70 ± 0.45	0.367	0.85
FCR	1.28 ± 0.30	1.03 ± 0.49	0.98 ± 0.32	0.96 ± 0.40	0.314	0.72
Survival rate (%)	100	100	100	100	-	-

D_0, D_1, D_2, and D_3 were supplemented with 0, 5, 10, and 15 g onion peel powder 100 g^{-1}, respectively.
The data presented in mean ± SD
The initial and final data were measured at the start and end of 60 days experimental period.

Levene's test indicated that there was no significant variance difference ($p = 0.116$), affirming the assumptions of ANOVA. One-way ANOVA indicated a significant variation in carotenoid content between treatment groups ($F(4, 25) = 84.53$, $p < 0.0001$). Tukey's post-hoc test affirmed that D_2 contained significantly greater carotenoid content than all the other groups ($p < 0.05$).

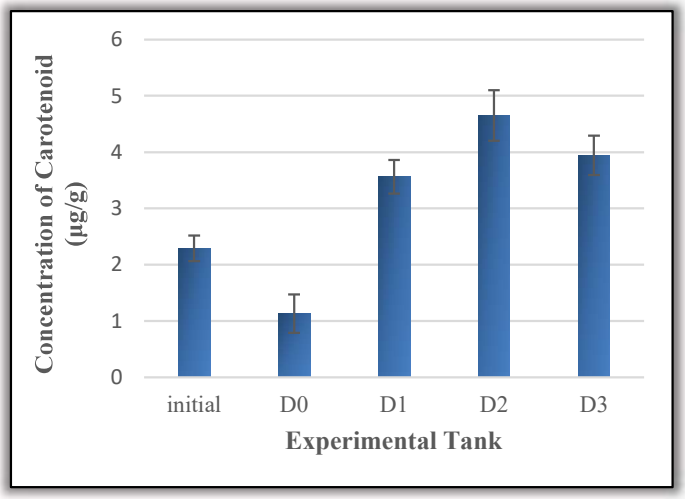

Fig. 1: Carotenoid concentration in rosy barb fish tissue; D_0, D_1, D_2, and D_3 were supplemented with 0, 5, 10, and 15 g onion peel powder 100 g^{-1}, respectively.

Various studies on the effect of using different carotenoids source in ornamental fish diets have shown significant impact on fish body colouration. A significant improvement in skin colouration combined with growth improvement was observed in goldfish (*Carassius auratus*) and orange swordtail (*Xiphophorus helleri*) when fed with a diet containing 5% of China rose petals (Sinha *et al.*, 2007; Joseph *et al.*, 2011). Significant growth and colour improvement was also seen in Koi carp (*Cyprinus carpio*), dwarf Gourami (*Colisa lalia*) and rosy barb (*Puntius conchonius*) when fed with diet containing 7-8% of rose petal meal incorporated as a carotenoid source (Pailan *et al.,* 2012; Jain and Kaur, 2016). Extracted carotenoids from algal powder was used by Sommer *et al.* (1992) for pigmentation trial in rainbow trout. The effect of red pepper, marigold flower and synthetic astaxanthin on pigmentation, growth and proximate composition was studied by Gocer *et al.* (2006). Several studies on colour enhancement of *Puntius sophore* with rose petal powder (Jain and Kaur, 2016) and marigold petal powder (Jagadeesh *et al.,* 2015) have revealed good colour enhancement in fishes but poor body growth. Marigold petal meal was used by Boonyaratpalin and Lovell (1977) for the tiger barb who found tiger barbs more brightly coloured than the fishes fed with control diet. Notable colour improvement has been reported in swordtail (*Xiphophorus hellerii*) when fed with 30 mg dried carrot powder 100 g^{-1} diet for 35days (Wagde *et al.,* 2018). However, the reduced carotenoid deposition noted in D_3 group (15% onion peel inclusion) in present study could be due to the high flavonoid content, especially quercetin, in onion peels. Although moderate inclusion rates (10%) were found to intensify pigmentation, higher doses could have anti-nutritional activities to restrict carotenoid absorption or inhibit digestive enzyme activity. Onion peel reportedly contains as much as 40 mg quercetin g^{-1} (Nile *et al.,* 2016), which at higher dietary intake may interfere with nutrient absorption. Also, carotenoid deposition could achieve a physiological saturation point, after which additional dietary input does not find its way into greater tissue accumulation. The same dose-dependent phenomena have also been reported in other research involving plant-based pigment sources (Mukherjee and Maity, 2024).

At the end of experiment, it may be concluded that the supplementation of onion peel powder in diet can increase the total carotenoid concentration in body tissue of Indian rosy barb fish; whereas absence of onion peel powder in diet (control) can cause a reduction of carotenoid concentration in body tissue. Natural carotenoids are pocket friendly and easily available source of pigments. Utilizing agro-industrial waste such as onion peel can revolutionize aquaculture in an eco-friendly manner. The incorporation of 10% onion peel powder in the diet of Indian rosy barb can greatly restore and improve the carotenoid deposits in tissues as well as growth performance. These findings justify the use of onion peels, as a cost-effective substitute to the synthetic pigments.

Acknowledgement: The authors express their gratitude to the Department of Fishery Sciences, Vidyasagar University, Midnapore, West Bengal (India) for allowing the smooth conduction of the experiments.

Ethical statement: All live fish experimental procedures were performed following the guidelines accepted by the Institutional Animal Ethics Committee, Department of Fishery Sciences, Vidyasagar University. Fish handling was done carefully to avoid any stress and discomfort during the study.

REFERENCES

AOAC. 1990. *Official Methods of Analysis (15th edn.). Methods 932.06, 925.09, 985.29, 923.03.* Association of Official Analytical Chemists. Arlington, USA.

APHA. 1998. *Standard Methods for the Examination of Water and Wastewater* (20th edn.). American Public Health Association, Washington, USA.

Boonyarapatin, M. and Lovell, R.T. 1977. Diet preparation for aquarium fish. *Aquaculture*, **12**: 53-62.

Boonyaratpalin, M., Thongrod, S., Supamattaya, K., Britton, G. and Schlipalius, L.E. 2001. Effects of β-carotene source, *Dunaliella salina*, and astaxanthin on pigmentation, growth, survival and health of *Penaeus monodon*. *Aquaculture Research*, **32**: 182-190.

Chapman, F.A. 2000. Ornamental fish culture, freshwater. pp. 602-610. **In:** *Encyclopedia of Aquaculture*, Vol. 3 (ed. R.E. Stickney). John Wiley & Sons, New York, USA.

Chatzifotis, S., Pavlidis, M., Jimeno, C.D., Vardanis, G., Sterioti, A. and Divanach, P. 2005. The effect of different carotenoid sources on skin coloration of cultured red porgy (*Pagrus pagrus*). *Aquaculture Research*, **36**(15): 1517-1525.

Fry, J.P., Mailloux, N.A., Love, D.C., Milli, M.C. and Cao, L. 2018. Feed conversion efficiency in aquaculture: Do we measure it correctly? *Environmental Research Letters*, **13**(2): 024017. [*DOI: 10.1088/1748-9326/aaa273*].

Gavril, R.N., Constantin, O.E., Enachi, E., Stoica, F., Lipa, F. D., Stănciuc, N., *et al.,* 2024. Optimization of the parameters influencing the antioxidant activity and concentration of carotenoids extracted from pumpkin peel using a central composite design. *Plants*, **13**: 1447. [*https://doi.org/10.3390/plants13111447*].

Göçer, M., Yanar, M., Kumlu, M. and Yanar, Y. 2006. The effects of red pepper, marigold flower, and synthetic astaxanthin on pigmentation, growth, and proximate composition of *Penaeus semisulcatus*. *Turkish Journal of Veterinary & Animal Sciences*, **30**(4): 359-365.

Goodwin, T.W. 1951. Carotenoids in fish. pp. 63-82. **In:** *The Biochemistry of Fish*. *Biochemical Society Symposia No. 6.* Cambridge University Press, Cambridge, UK.

Jagadeesh, T.D., Murthy, H.S., Surendranath, S.V., Panikkar, P R., Manjappa, N. and Mahesh, V. 2015. Effects of supplementation of marigold (*Tagetes erecta*) oleoresin on growth, survival and pigmentation of rosy barb, *Puntius conchonius* (Hamilton). *An Internal Quarterly Journal of Life Sciences*, **10**(3): 1431-1435.

Jain, A. and Kaur, V.I. 2016. Rose petal meal as a potential natural carotenoid source for pigmentation and growth of freshwater ornamental fish, koi carp, *Cyprinus carpio* (Linnaeus). *Indian Journal of Ecology*, **43**(1): 239-244.

Joseph, B., Sujath, S., Shalin, J.J. and Palavesam, A. 2011. Influence of four ornamental flowers on the growth and colouration of orange sword tail (*Xiphophorus hellerei*). *International Journal of Biological and Medical Research*, **2**(3): 621-626.

Lovell, R.T. 2000. Nutrition of ornamental fish. pp. 1191-1196. **In:** *Kirk's Current Veterinary Therapy XIII – Small Animal Practice* (ed. J. Bonagura). W.B. Saunders, Pennsylvania, USA.

Mukherjee, R. and Maity, J. 2024. Colour retention in ornamental fish, Indian rosy barb (*Pethia conchonius*), using petal meal of Asian pigeonwings (*Clitoria ternatea*). *Applied Biological Research,* **26**(1): 78-83.

Nile, A., Nile, S.H., Kim, D.H., Keum, Y.S., Seok, P.G. and Sharma, K. 2018. Valorization of onion solid waste and their flavonols for assessment of cytotoxicity, enzyme inhibitory and antioxidant activities. *Food and Chemical Toxicology,* **119**: 281-289.

Olson, J.A. 1979. A simple dual assay for vitamin A and carotenoids in human liver. *Nutrition Report International*, **19**: 807-819.

Pailan, G.H., Sinha, A. and Kumar, M. 2012. Rose petals meal as natural carotenoid source in pigmentation and growth of rosy barb (*Puntius conchonius*). *Indian Journal of Animal Nutrition*, **3**: 291-296.

Pandey, P.K. and Mandal, S.C. 2017. Present status, challenges and scope of ornamental fish trade in India. pp. 1-10. **In:** *Aqua Aquaria India Conference*, May 14-16, 2017, Mangalore, India.

Paripatananont, T., Tangtrongpairoj, J., Sailasuta, A. and Chansue, N. 1999. Effect of astaxanthin on the coloring of goldfish *Carassius auratus*. *Journal of the World Aquaculture Society*, **30**: 454-460.

Sales, J. and Janssens, G.P.J. 2003. Nutrient requirements of ornamental fish. *Aquatic Living Resources*, **16**(6): 533-540.

Saxena, A. 1994. Health and coloration of fish. pp. 94. **In:** *International Symposium on Aquatic Animal Health: Program and Abstracts*. University of California, Davis, California, USA.

Sharma, M., Usmani, Z., Gupta, V.K. and Bhat, R. 2021. Valorization of fruits and vegetable wastes and by-products to produce natural pigments. *Critical Reviews in Biotechnology*, **41**(4): 535-563.

Sinha, A. and Asimi, O.A. 2007. China rose (*Hibiscus rosasinensis*) petals: A potent natural carotenoid source for goldfish (*Carassius auratus* L.). *Aquaculture Research*, **38**: 1123-1128.

Sommer, T.R., D'Souza, F.M.L. and Morrissy, N.M. 1992. Pigmentation of adult rainbow trout, *Oncorhynchus mykiss*, using the green alga *Haematococcus pluvialis*. *Aquaculture*, **106**: 63-74.

Torrissen, O.J. 1989. Biological activities of carotenoids in fishes. pp. 387-399. **In:** *Proceedings of the Third International Symposium on Feeding and Nutrition in Fish* (eds. H. Takeda and T. Watanabe). August 28-September 1, Laboratory of Fish Nutrition, Faculty of Agriculture, Mie University, Toba, Japan.

Wagde, M.S., Sharma, S.K., Sharma, B.K., Shivani, A.P. and Keer, N.R. 2018. Effect of natural β-carotene from carrot (*Daucus carota*) and spinach (*Spinacia oleracea*) on colouration of an ornamental fish-swordtail (*Xiphophorus hellerii*). *Journal of Entomology and Zoology Studies*, **6**(6): 699-705.