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Effect of Dietary Supplementation of Carrot Meal on Growth, Survival, Whole-Body Composition and Total Carotenoid Content of Zebrafish, *Danio rerio*

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Abstract: A six-months feeding trial was carried out to evaluate the effects of diets supplemented with carrot meal [0%(CM0), 1%(CM1), 3%(CM3), 5%(CM5), 7%(CM7) and 10%(CM10)] on growth, survival, proximate body composition and total carotenoid content in zebrafish, *Danio rerio*. Complete randomized design (CRD) was implemented where 18 aquaria were used, each with 40 juveniles (0.033 g). After feeding trial, significantly highest final weight (0.70 ± 0.03 g), body weight gain (0.66 ± 0.03 g) and specific growth rate (SGR) ($1.71 \pm 0.03\%$) was observed in the fishes fed with diet (CM5) containing 5% carrot meal, followed by CM3 set and then in CM7 set and the lowest values were found in CM10 set. Carotenoid content in fish skin and muscle was noted maximum (6.67 ± 0.09 $\mu\text{g/g}$) in CM5, followed by CM3 set (5.72 ± 0.07 $\mu\text{g/g}$), while the lowest (2.59 ± 0.04 $\mu\text{g/g}$) in CM0 (control) set. Survival was 100% in CM5 and CM7 sets, while CM10 set had the lowest value which was insignificantly varied with control set. Fishes fed with CM1 diet containing 1% carrot meal, showed significantly highest values for growth and feed utilization parameters, survival and carotenoid content, compared to control (CM0). No significant effect of dietary treatment was found in final length and whole-body composition of experimental fishes. The results demonstrate that carrot meal could be suitably supplemented in the diet up to 5% level to ensure improved growth and feed utilization, survival and enhanced pigmentation in zebrafish. Hence, carrot meal can be successfully incorporated as an alternative, easily available, cost-effective, natural carotenoid source in zebrafish diets.

Keywords: Carotenoid content, Carrot meal, Feed utilization, Growth, Survival, Zebrafish

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Introduction

One of the nature's wonderful creations is the ornamental fishes as they possess brilliant colourations with different patterns which have impact on human's psychology, emotions, behaviour and on the cognitive performance (Kaushik, 2011; Hulshof, 2013). Ornamental fish

keeping as home décor is the second most preferred hobby throughout the world (Jain *et al.*, 2019) due to their aesthetic value. Hence, ornamental fish farming is an emerging field of aquaculture industry and has the potential to contribute to the economic development in

underdeveloped and developing countries (Yanar *et al.*, 2008) as this field plays a crucial role in the international fish trade with high export value (Jain, 2016). The astounding growth in ornamental fish production has impelled the researchers to enhance the colour quality, its main characteristic, and to improve the health status of the fishes in the last three decades.

Among the most cultured ornamental fishes, the zebrafish (*Danio rerio*), a freshwater omnivorous small-sized fish, belong to the family Cyprinidae, and order Cypriniformes, stands out as choicest one because of its brilliant red, yellow and orange colouration and remarkable characteristics, including simple husbandry, gregarious behaviour, faster growth, development and maturation, small adult size, high fecundity and productivity and disease resilience (ADB, 2005; Aketch *et al.*, 2014). These features make them a suitable model species for pilot feed studies with novel and alternative ingredients (NC3R's, 2014). However, limited researches were conducted using them as a model for their nutrition and skin pigmentation mainly due to lack of standardized or defined diets and husbandry conditions.

Attractive colouration and the colour intensity of ornamental fishes is one of the major factors that determine their commercial value in the world market (Tiewsoh *et al.*, 2019). Fish skin colouration depends on their skin pigmentation that primarily depend on the presence of integumentary chromatophores, containing lipid soluble pigments, carotenoids (Chatzifotis *et al.*, 2005). Fishes are not able to synthesize carotenoids endogenously (Gupta *et al.*, 2007; Das and Biswas, 2016), but their dietary carotenoids, synthesized by photosynthetic organisms, are responsible for their skin colouration (Sefc *et al.*, 2014; Jorjani *et al.*, 2019). The fishes in their natural habitat, obtain colouration from their wild food sources. But aquarium fishes and farmed fishes, cultured under high density in captive condition without supplementation of dietary carotenoids leads to a faded coloration which

decreases their commercial acceptability and thereby their market value (Harpaz and Podowicz, 2007; Awasthi *et al.*, 2014). Faded colouration due to inadequate carotenoid in fish diets, can be improved by inclusion of carotenoid in their diet. Therefore, fish diets must contain natural carotenoid-rich ingredient supplementation that efficiently provide the carotenoid, necessary for their intensified target colour achievement (Gupta *et al.*, 2007).

Natural and synthetic carotenoids are the two common sources of the fish diet carotenoid. The synthetic sources are expensive, not species specific and also harmful (Bano *et al.*, 2020). High cost of synthetic carotenoids pushes the researchers to explore easily available cost-effective natural carotenoid sources from plant and animal origin, and to reveal their species-specific potential for fish skin pigmentation. Prior studies addressed this issue and investigated several carotenoid-rich potential natural pigment sources as feed supplementation for ornamental fishes to determine whether they could be an alternative to synthetic carotenoids. Spirulina was used to enhance colour of rainbow trout, fancy carp and yellow tail cichlids (Choubert, 1979). Marigold petal meal was reported as colour enhancer in tiger barb, red sword tail and koi carp (Ezhil *et al.*, 2008; Swain *et al.*, 2014). Dietary beet and tomato powder had enhanced the skin pigmentation of sword tail and guppy fish, respectively (Mirzaee *et al.*, 2012; Singh and Kumar, 2016). A number of researches had supplemented the carrot meal, one of the cheapest natural carotenoid sources, to enhance the skin colouration of ornamental fishes and notable among them are *Amphiprion ocellaris* (Ramamoorthy *et al.*, 2010), guppy fish (Mirzaee *et al.*, 2012), botia fish (Andriani *et al.*, 2020) and goldfish (Karo-karo *et al.*, 2015). Carrot has an excellent nutritional profile with high β -carotene which could be used as potential functional fish feed ingredient and also as an alternative of synthetic carotenoid without significantly increasing the diet price (Andriani *et al.*, 2020).

As no records are available on zebrafish showing the effect of dietary natural carotenoid supplementation as colour enhancer, the present experiment was carried out to investigate the effects of carrot meal (*Daucus carota*) as dietary carotenoid supplementation on growth, survival, whole-body composition and total carotenoid content of skin and muscle tissue of zebrafish, *Danio rerio* in order to increase their cost-effective production with enhanced colouration.

Materials and Methods

Experimental fish:

The juveniles of zebrafish were purchased from the Galiff Street ornamental fish market, Kolkata, India. Apparently, healthy ones were acclimatized under indoor conditions for two weeks in a glass aquarium (36"×12"×12") with control diet containing 40% crude protein. For the six-month feeding trial, a total number of 720 juveniles (0.033 g) were chosen and distributed in 18 glass aquaria (18"×12"×12") having six dietary treatment sets with three replications, each aquarium with 40 juveniles. Diets were given twice daily at 9 am and 6 pm, to the fishes, at the amount of satiation level and daily food consumption was noted. Water exchange was done twice a week at one-third level in each aquarium. Continuous aeration throughout the feeding trial was provided to the aquaria by the mini air blower pumps.

Experimental diet:

Carrots were purchased from nearby local vegetable market, washed, grated and dried under shade and then ground by electric grinder into powder form to prepare the carrot meal. All the feeding ingredients, used for the preparation of the experimental diets were also powdered and kept in airtight glass container at room temperature until used for diets preparation.

Six different fishmeal-based iso-proteic experimental diets were formulated with 40% of crude protein to meet the nutritional requirement of zebrafish. Practical diets were prepared by incorporating carrot as carotenoid source at the

level of 0%(CM0), 1%(CM1), 3%(CM3), 5%(CM5), 7%(CM7) and 10%(CM10) by adjusting the amounts of feed ingredients, rice bran and wheat.

For the preparation of each experimental diets, all the dried dietary ingredients were well mixed and thoroughly blended with double-distilled water into dough, and pelleted by pelletizer through 1.5 mm die. The obtained moist noodles were dried under shade for 48 h and then crushed into desirable particle size and kept in airtight glass container at room temperature until used. Dietary ingredients proportion and proximate composition of all the experimental diets are listed in Table 1.

Growth efficiency:

At the termination of the feeding trial, final weight and length of the experimental fishes from the six dietary sets were recorded in order to estimate their growth performance parameters like weight gain, specific growth rate (SGR%) and feed utilization parameters in terms of food conversion ratio (FCR).

Proximate Composition Analysis:

Fish from experimental sets were collected at the end of feeding trial to estimate whole-body composition. The proximate composition such as crude protein, crude lipid, ash and moisture contents of whole-body of the fishes from the six experimental sets, were analyzed by following the methods of AOAC (2006).

Estimation of carotenoid:

Carotenoid content of experimental diets were estimated by following the method of Cyanotech (2002). At the end of the feeding trial, coloured region of skin and muscle tissue of fishes from the six experimental sets were collected to measure the total carotenoid content by Tiewsoh *et al.* (2019) method.

Survival:

Survival percentage of experimental fish from the six different dietary sets, was calculated by subtracting the number of fish collected at the end

Table 1: Ingredients, their proportion and proximate composition of experimental diets

Diet ingredients (g/100 g)	CM0	CM1	CM3	CM5	CM7	CM10
Fishmeal	42	42	42	42	42	42
Carrot meal	0	1	3	5	7	10
Soybean	25.20	25.20	25.20	25.20	25.20	25.20
Corn	5.00	5.00	5.00	5.00	5.00	5.00
MOC*	12.50	12.50	12.50	12.50	12.50	12.50
Rice bran	5.29	4.83	3.86	2.78	0.97	2.02
Wheat	8.01	7.47	6.44	5.52	5.33	1.28
Vit-Mineral Premix**	2	2	2	2	2	2
Total	100	100	100	100	100	100
Crude Protein	40.33±0.64ns	40.25±1.18ns	40.20±1.21ns	40.14±1.59ns	40.04±1.69ns	39.92±1.11ns
Crude lipid	6.26±0.56ns	6.40±0.60 ns	6.23±0.43ns	6.36±0.33ns	6.37±0.54ns	6.57±0.38ns
Ash	2.78±0.21ns	2.80±0.45ns	2.81±0.47ns	2.84±0.25ns	2.82±0.61ns	2.78±0.65ns
Crude fibre	8.63±0.86 ns	9.11±0.91ns	9.38±0.54ns	9.63±0.68ns	9.92±0.68ns	10.26±0.77ns

*MOC- mustard oil cake; **Vit-Mineral Premix (mg/kg diet): retinol-18,000 IU, Choleclaciferol-2000 IU, thiamine-15, menadione sodium bisulphate-10, riboflavin-25, pyridoxine-5, α -tocopherol-35, nicotinic acid-200, Ca-pantothenate-50, biotin-1.5, folic acid-10, cyanocobalamin-0.03, ascorbyl monophosphate-50, inositol-400, copper sulphate-20.2, dibasic calcium phosphate-5.9, sodium fluoride-2.21, potassium iodide-0.78, zinc oxide-37.5, ironsulphate-200, magnesium oxide-840, manganese oxide-26, cobalt sulphate-1.85, sodium selenite-0.65, potassium chloride-1.17, sodium chloride-0.45.

Values of proximate composition are presented as mean $\pm$ SE. Values with ns letters are not significantly different ($P>0.05$) in one way ANOVA

Table 2: Water quality parameters of experimental aquarium sets during the experiment

Parameter	Minimum	Maximum
Temperature ($^{\circ}$ C)	25	36
Dissolved Oxygen (ppm)	8.1	8.4
pH	7.2	7.5
Free Carbon dioxide (ppm)	1.32	2.21
Hardness (ppm)	205	236
TDS (ppm)	167	195

of the feeding trial from the fishes, stocked at the initiation of the feeding trial.

Water Quality Analysis:

Water quality parameters in terms of water temperature, dissolved oxygen, pH, free carbon dioxide, total hardness and total dissolved solids (TDS) of experimental aquaria were analysed and recorded at fortnightly intervals, throughout the experimental period using the standard methods of APHA (2012).

Statistical Analysis:

The experimental data were presented as mean $\pm$

standard error (SE) of the three replications. One-way analysis of variance (ANOVA), followed by Duncan's multiple range tests (DMRT) for multiple comparisons at the significance level of 0.05 was used to compare the differences among the six dietary treatments.

Results

Water quality:

During the experimental period, water quality parameters (Table 2) such as water temperature, pH, dissolved oxygen, free carbon dioxide, TDS and total hardness did not vary significantly

Table 3: Growth and food utilization parameters of zebrafish, *D. rerio* fed with six experimental diets

Growth parameters	Diets					
	CM0	CM1	CM3	CM5	CM7	CM10
Initial weight (g)	0.033±0.00a	0.033±0.00a	0.032±0.00a	0.032±0.00a	0.032±0.00a	0.033±0.00a
Initial length (cm)	1.70±0.00a	1.67±0.03a	1.70±0.00a	1.63±0.03a	1.70±0.00a	1.67±0.03a
Final weight (g)	0.39±0.01b	0.52±0.02c	0.61±0.02d	0.70±0.03e	0.55±0.01c	0.29±0.02a
Final length (cm)	3.97±0.07a	3.93±0.09a	3.93±0.09a	4.10±0.12a	4.13±0.07a	3.97±0.07a
Weight gain (g)	0.35±0.01b	0.49±0.02c	0.58±0.02d	0.66±0.03e	0.52±0.01c	0.26±0.02a
FCR	3.21±0.02e	3.06±0.01d	3.00±0.01b	2.96±0.01a	3.03±0.01c	3.38±0.05f
SGR (%)	1.37±0.02b	1.54±0.02c	1.63±0.02e	1.71±0.03f	1.58±0.01d	1.22±0.03a
Food consumption (g)	1.13±0.03b	1.49±0.05c	1.74±0.05e	1.97±0.07f	1.58±0.02d	0.88±0.05a

Values are mean ± SE. Values with different letters are significantly different ($P < 0.05$) using DMRT after one way ANOVA

among aquarium water of the dietary treatments and were within the recommended range for the ornamental fish culture (Rinna *et al.*, 2013).

Experimental diet:

Dietary ingredients proportions and nutrient composition of all the experimental diets are displayed in Table 1. Fishmeal was incorporated at the level of 42 g per 100 g in all the experimental diets and 0%, 1%, 3%, 5%, 7% and 10% of carrot meal was added in the diet by adjusting the amount of rice bran and wheat, and named the diets as CM0, CM1, CM3, CM5, CM7 and CM10, respectively. Relatively, a consistent percentage of crude protein content among the experimental diets (39.92% to 40.33%) were observed.

Growth performance:

The effect of different dietary treatments on the growth performance and feed utilization of zebrafish are shown in the Table 3. Initial length and weight of the juveniles of all the dietary sets did not vary significantly ($P > 0.05$). After the six-months of feeding trial, significant variations were found in the growth efficiency parameters such as final body weight, weight gain, specific growth rate in per cent (SGR%) and also in feed utilization parameters like total food consumption and food conversion ratio (FCR). Significantly highest body

weight was observed in the fishes fed with the CM5 diet, followed by the CM3 set and a significantly lower value was noted in the CM10 set. Whereas the body weight of the CM1 and the CM7 sets did not show any significant variation, but were found higher than the control set (CM0). The results of weight gain of the experimental fishes from the different dietary sets, followed a similar trend as noted in the results of final body weight, which was significantly higher in the CM5 set and lower in the CM10 set. No significant effect of dietary treatments was found in final body length of the experimental fish from the six treatments. Juveniles, fed with the CM5 practical diets containing 5% carrot meal, reached to a significantly higher ($P < 0.05$) SGR, followed by the CM3 set and then in the CM7 set, and a lower SGR was noticed in the fishes fed with the diet having 10% carrot meal (CM10). The value of SGR was significantly higher in the CM1 set, when compared with the control set (CM0). In case of total food consumption, the maximum amount of food was taken by the fishes of the CM5 set, followed by the CM3 set, while a minimum amount was consumed by the fishes of set, CM10. Feed utilization in terms of FCR also improved in the CM5 with the best value, followed by the CM3 set and then in the CM7, whereas the poorest FCR was noticed in the CM10 set among the dietary

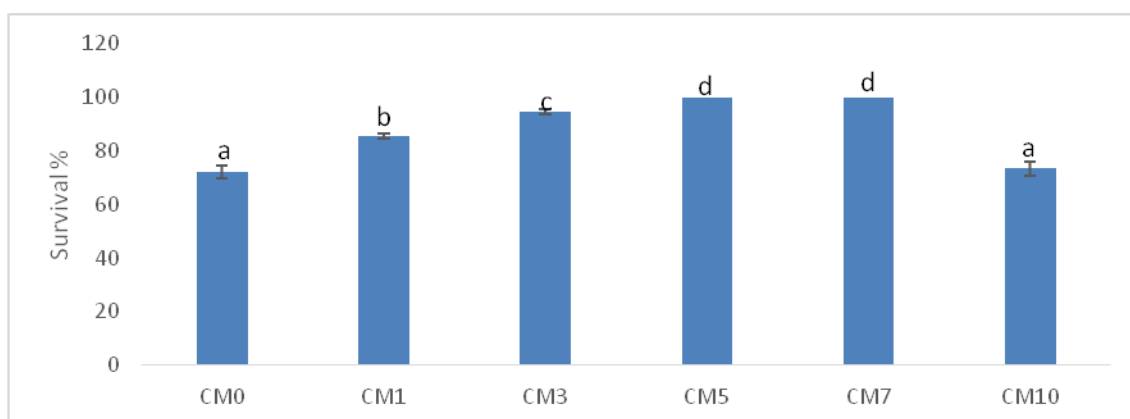


Fig. 1: Survival percent of *D. rerio* fed with six experimental diets. Values are mean $\pm$ SE. Bars with different letters are significantly different ($P < 0.05$) using DMRT after one way ANOVA.

Table 4: Proximate composition (%) of the whole-body of zebrafish, *D. rerio* fed with six experimental diets in six-months feeding trial

Diets	Crude Protein	Crude lipid	Moisture	Ash
CM0	15.02 $\pm$ 0.64ns	4.40 $\pm$ 0.25ns	74.63 $\pm$ 0.81ns	2.32 $\pm$ 0.82ns
CM1	15.04 $\pm$ 0.58ns	4.47 $\pm$ 0.32ns	74.40 $\pm$ 0.80ns	2.14 $\pm$ 0.68ns
CM3	15.70 $\pm$ 0.89ns	4.53 $\pm$ 0.38ns	74.71 $\pm$ 0.60ns	2.11 $\pm$ 0.64ns
CM5	15.60 $\pm$ 0.59ns	4.54 $\pm$ 0.47ns	74.40 $\pm$ 1.19ns	2.18 $\pm$ 0.71ns
CM7	15.47 $\pm$ 0.80ns	4.13 $\pm$ 0.40ns	74.93 $\pm$ 1.14ns	2.79 $\pm$ 0.60ns
CM10	15.23 $\pm$ 0.47ns	4.37 $\pm$ 0.45ns	74.03 $\pm$ 1.32ns	2.75 $\pm$ 0.80ns

Values are mean $\pm$ SE. ns= not significantly different ($P > 0.05$) one way ANOVA

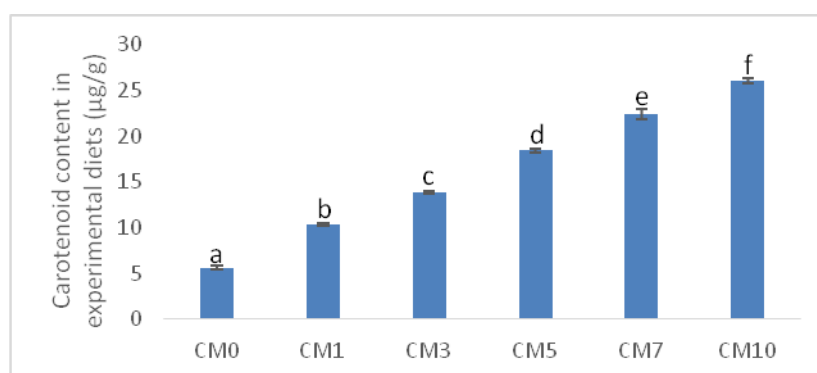


Fig. 2: Carotenoid content ($\mu\text{g/g}$) in six experimental diets. Values are mean $\pm$ SE. Bars with different letters are significantly different ($P < 0.05$) using DMRT after one way ANOVA.

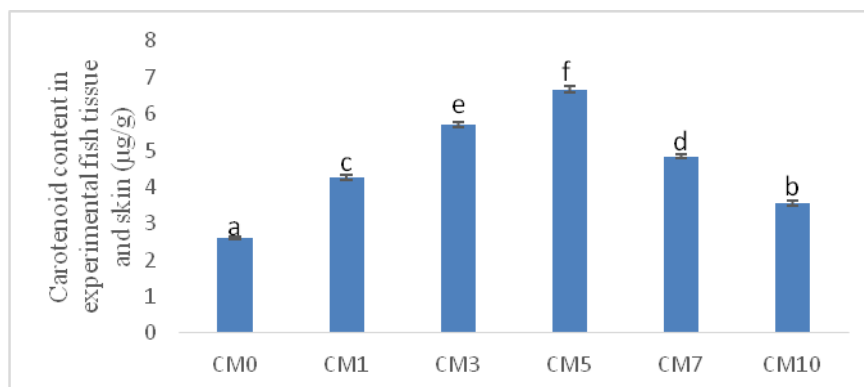


Fig. 3: Carotenoid content ($\mu\text{g/g}$) in six experimental diets fed fish tissue and skin. Values are mean $\pm$ SE. Bars with different letters are significantly different ($P < 0.05$) using DMRT after one way ANOVA.

treatments.

Survival percentage:

The effect of different dietary treatments on the survival percentage of zebrafish after six months of feeding trial is presented in Figure 1. Survival of experimental fishes was noted 100%, fed with diets supplemented with carrot meal at the level of 5% and 7% (CM5 and CM7), while it was 94.44% in the CM3 set. A higher survival was recorded in the CM1 set, when compared with the control (CM0) which showed insignificant variation with the value of survival of the CM10 set.

Whole-body composition:

The effect of dietary supplementation of carrot meal on the whole-body proximate composition for crude protein, crude lipid, moisture and ash content of zebrafish from the dietary sets are presented in Table 4. Whole-body composition of the fishes was relatively consistent across all the dietary treatment with no significant differences ($P > 0.05$).

Carotenoid concentration of experimental meal and of zebrafish skin and muscle tissue:

The result of total carotenoid content in the experimental diets showed an increasing trend with the increased amount of carrot meal supplementation in the experimental diets where significantly higher value of carotenoid content was observed in the CM10 diet and lower in the control diet (Fig. 2).

After six-months of feeding trial, carotenoid

content in the skin and muscle tissue of fishes, fed with experimental diets with increasing carrot meal levels, showed significant variations among the fishes of the dietary sets and also varied significantly with that of the control one (Fig. 3). Colour improvement of the experimental fishes in terms of the carotenoid deposition in fish skin and muscle tissue showed an escalating trend of their values up to 5% carrot meal inclusion in their diet. Carotenoid content of fishes from CM7 and CM10 was found maximum than the control set (CM0).

Discussion

In the current study, carrot meal was used as a natural carotenoid supplementation in the experimental diets, which had significant effects on growth, feed utilization, fish skin pigmentation and survival of zebrafish, *D. rerio*.

Growth is an important criterion that indicates the health and physiology of a fish (Huntingford *et al.*, 2006). In the earlier studies, it was depicted that carotenoid had a positive role in the intermediary metabolism of fish that enhance feed utilization and ultimately resulted in an improved growth (Amar *et al.*, 2001). In this regard, Maiti *et al.* (2017) stated that natural carotenoid had a growth promoting role. Carotenoids not only function as a precursor of vitamin A synthesis but also produce other essential nutrients through carotenoid bioconversion in fishes, which justify its role as growth promoter (Maiti *et al.*, 2017). This finding was also in agreement with the investigation of Jha *et al.* (2012), where spirulina

and marigold supplemented diets improved growth and feed utilization of *Barilius bendelisis* and Ezhil *et al.* (2008) and Jagadeesh *et al.* (2015), where marigold added diet enhanced the growth and nutrient utilization of red sword tail and rosy barb, respectively. Pan *et al.* (2010), Pailan *et al.* (2012), Maiti *et al.* (2017), Jain *et al.* (2019), Bano *et al.* (2019), and Tiewsoh *et al.* (2019) reported the correlation between the supplemented carotenoids and growth improvement in rosy barb, koi carp, gourami, goldfish and characins respectively. In contrary, Seyedi *et al.* (2013), Wang *et al.* (2006) and Kop *et al.* (2010) observed no significant growth on carotenoid supplemented diet in clownfish, characins and cichlid, respectively. Whereas, the present study supported the view of positive impact of carotenoid supplemented diets on growth with improved feed utilization in zebrafish.

In the current study, the zebrafish received significantly higher carotenoid deposition in their skin and tissue in all the dietary sets compared to the control set, which confirmed that dietary carotenoid leads to the deposition of carotenoid in the fish skin and muscle, which consequently resulted an enhanced colouration of the fish. A similar result was found for the goldfish fed with spirulina (Kiriratnikom *et al.*, 2005), jewel cichlid fed with carrot meal (Mirzaee *et al.*, 2013), cichlid fed with carrot meal (Kop *et al.*, 2010) and for the marine ornamental fish fed with carrot meal (Ramamoorthy *et al.*, 2010).

In this study, it was noted that carrot meal supplemented diets had no significant effect on proximate composition of whole-body of zebrafish. Pailan *et al.* (2012) observed a similar finding when rose petal supplemented diets were fed to the rosy barb. But Christiansen *et al.* (1995) and Christiansen and Torrisen (1996) found higher protein and moisture content and lower lipid content in *Salmo salar* fry and juveniles when fed with the diets, low in astaxanthin. This conflicting result of proximate composition might be due to species difference or due to negative effect of artificial carotenoid, which enhance the

colouration temporarily but hamper the health status of fish.

Prior studies revealed that survival of ornamental fishes influenced positively with the carrot meal supplemented diets. Jain *et al.* (2019) reported that 100% survival of Koi carp was found in the fishes, fed with diets containing 5% and 7% carrot meal. A similar result was obtained in the current study with zebrafish where also 5% and 7% carrot meal supplementation showed the 100% survival. In this regard, Krinsky (1993) stated that absorption of carotenoid, provided through diet, enhances fish survival. This finding supports the result of survival recorded in the present study.

In past few investigations with the natural carotenoid sources from plant origin proved their effectiveness for enhancing skin colouration in the ornamental fishes (Pan *et al.*, 2010; Pailan *et al.*, 2012; Maiti *et al.*, 2017; Jain *et al.*, 2019; Bano *et al.*, 2019; Tiewsoh *et al.*, 2019). Results of feeding trial showed that in the experimental fish sets, the carotenoid content in fish skin and muscle tissue was increased with the increasing amount of carrot meal inclusion in the diet, which was maximum up to 5% incorporation level. This indicates that up to a definite amount of carotenoid in the diet could be utilized maximally by the fish which might contribute to the maximum carotenoid deposition and pigmentation in the fish skin and muscle tissue. Therefore, the amount of carotenoid in fish diet for a maximum carotenoid deposition in fish skin, is species specific (Ha *et al.*, 1993). In the present study, a maximum carotenoid deposition in fishes fed with experimental diet containing 5% carrot meal, might be due to a higher transforming ability of alimentary carotenoid and subsequently depositing in the fish skin and muscle tissue.

Earlier studies also revealed that carrot meal supplementation in ornamental fish diet showed superior result of colour enhancement, when compared with other natural carotenoid sources such as red pepper (on jewel cichlid by Mirzaee *et al.*, 2013; on cichlid by Kop *et al.*, 2010) and,

marigold petal, China rose petal and rose petal (*Amphiprion ocellaris* by Ramamoorthy *et al.*, 2010). In the present study, carrot meal as a natural carotenoid source of plant origin, is examined on zebrafish and considered to be the suitable one for optimum growth and improved skin colouration in *D. rerio*.

Conclusion

The results of present study revealed that the zebrafish could efficiently utilize the natural carotenoid from the carrot meal supplanted diets containing up to 5% carrot meal and could be able to store the carotenoid in their skin and muscle tissue and also able to enhance their skin pigmentation. Hence, it can be concluded that the carrot meal supplanted diets have a positive impact on growth, feed utilization, survival and skin pigmentation of zebrafish, *D. rerio* in the indoor rearing conditions. Carotenoid supplemented diet with 5% carrot meal is recommended for the zebrafish culture as 5% inclusion level of carrot meal was more effective as colour enhancer and growth promoter.

From the present investigation, it could be inferred that for enhancing and maintaining the proper skin colour and health in the ornamental fishes, their diet must be supplemented with suitable skin pigment enhancing natural carotenoid.

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