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IDENTIFICATION OF AGE GROUPS ON FEEDING SPIRULINA TO ENHANCE GROWTH, COLORATION AND REPRODUCTION IN XIPHOPHORUS HELLERI

Nagarajan, R.¹, R. Niranjani, J. Shoba and R. James^{*2}

¹P.G and Research Department of Zoology, Kamaraj College Tuticorin- 628003.

² P.G and Research Department of Zoology, V.O. Chidambaram College, Tuticorin-628008, Tamilnadu, India.

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ABSTRACT

To avoid the wastage of *Spirulina* diet, an experiment was conducted to identify the time and duration of feeding *Spirulina* diet on growth, coloration and reproduction in *X.helleri* for a period of 175 days as a function of age groups. Three age groups such as one month, two months, and three months old *X.helleri* were selected from the laboratory bred brooders. Feeding, growth and gonad weight of *X.helleri* were increased with increase in *Spirulina* levels. The feeding and growth parameters were high at pre-breeding as compared to post-breeding. Fish fed on 15% *Spirulina* diet had heavier gonad weight in all tested age groups and attained maturity earlier than fish fed on other diets. Two months old female *X.helleri* fed with *Spirulina* diet released maximum number of fry (570) followed by three months (478) and one month (368) old age groups. Similar trend was obtained in coloration also. However, carotenoid content of fry was maximum in one month old age group followed by two and three months old age groups; but they did not show any significant (t = 0.92; p > 0.05) difference between them. It suggests that, ideal supplementation of *Spirulina* diet to *X.helleri* at the age of two months to enhance growth, coloration and reproductive performances. As protein is the costliest component used in artificial feeds, it is necessary to reduce the cost of feeds and to stimulate growth, coloration and reproduction in ornamental fish culture.

Keywords: Age groups, Growth, Coloration, Reproduction, Spirulina, Xiphophorus helleri.

INTRODUCTION

Proper nutrition is one of the most important factors influencing the ability of aquatic organisms to attain the genetic potential for growth, coloration, sexual maturity and longevity (Nickell and Bromage, 1998; Shearer and Swanson, 2000). The nutrient requirements vary between species and within species between the different stages of its life-cycle. The nutritional energy is partitioned by fish between the physiological processes involved various in maintenance, growth and reproduction. The relative of energy between and partitioning growth reproduction varies between different age groups. Fishes obtain their energy from protein and lipid sources, which enhance the growth and reproduction (James and Sampath, 2002). The demand for protein rich diet is ever increased and blue green algae like *Spirulina* which triggered the growth and other physiological activities of fishes (James *et al.*, 2006). Feeding the *Spirulina* in ornamental fishes throughout the lifecycle is costly in their culture practice. Hence, to avoid the wastage of *Spirulina* diet, an extensive study is necessary to identify the time and duration of feeding *Spirulina* on growth, coloration and reproduction in ornamental fish as a function of age groups. The present study is aimed to assess the optimal requirement of *Spirulina* to enhance the maximum growth, coloration and reproductive

*Corresponding Author: P.G. and Research Department of Zoology, V.O. Chidambaram College, Tuticorin-628008, Tamilnadu, India, Email: piojames2014@gmail.com, Mobile: +91 9443528557.

performance in red swordtail, *Xiphophorus helleri* in relation to different age groups.

MATERIALS AND METHODS

Fish and Maintenance

Healthy and active fishes of three different age groups, such as one month old $(0.13 \pm 0.01 \text{ g})$, two months old $(0.38 \pm 0.01 \text{ g})$ and three months old $(0.90 \pm 0.03 \text{ g})$ *X. helleri* were collected from the laboratory bred brooders. For convenience of presentation thereafter, the chosen three age groups would be referred to as 1 MG, 2 MG and 3 MG respectively. They were divided into four different groups corresponding to four levels of *Spirulina* (0, 5, 10 and 15%) diets. Each group was consisting of 25 individuals and reared in circular cement aquaria containing 100 litre water (width: 58.5 cm; height: 40 cm; 120 litre capacity). The tanks were drained twice a week and replenished with fresh water to remove the accumulated feces from the bottom.

Feed Formulation

Spirulina diets were prepared following the square method (Hardy, 1980). The compositions of the diets are given in (Table 1). The dried ingredients were blended to make a homogenous mixture. Then the diets were mixed with an aliquot of boiled water and cooked by steam for 15-20 minutes. Pellets (2 mm size) were prepared with a hand operated pelletizer and dried in sunlight. After drying, the diets were stored separately in a refrigerator until use. The protein and lipid contents of the experimental diets were determined in a spectrophotometer following Lowry et al. (1951) and Bragdon (1951) respectively. The moisture content was analyzed by drying in an electric hot air oven at 100°C. The mineral content was estimated following the method of Paine (1964). Nitrogen free extract (NFE) was calculated by subtracting the protein, lipid and mineral contents from the dry weights of the feed samples. Three replicates were maintained for each test diets.

Feeding

Weighed quantity of the chosen *Spirulina* diets were offered daily at 0700 and 1700 hr to the test fishes. Unconsumed feeds were collected after 1 hr by a pipette with minimum disturbance to the animals and dried in a hot air oven at 60° C. Feed consumption was estimated by subtracting the amount of unconsumed dry feed from the dry weight of feed offered. To simplify the data for clarity, the mean feeding and growth parameters were calculated as the respective

25 days sampling data relating to number of samples prior to commencement of breeding referred to as prebreeding and after the commencement of breeding referred to as post-spawning during the experiment. The experiment was conducted for 175 days.

The feeding rate was computed as,

Feeding rate (mg g^{-1} live fish day⁻¹) =

Amount of feed consumed (mg)

Initial wet weight of fish $(g) \times No.$ of days

Growth and gonad estimation

Fish were weighed at the beginning of the experiment and every 25 days thereafter. Weight gain was calculated as the difference between the wet weights at the beginning of experiment and on the day of sampling. SGR was calculated as: SGR = ($wt_1 - wt_0$) $t_1 \times 100$ where wt_0 and wt_1 are the wet weights of the fish at the beginning of experiment and end of each sampling period and t_1 is the period between sampling in days. Two females from each treatment were sacrificed at 25 days intervals from the time of gonad development until the commencement of breeding. Their ovaries were removed and weighed and the gonadosomatic index (%) was computed according to Dahlgren (1979) as follows :

GSI = Wet weight of gonad / Wet weight of fish \times 100.

After sampling the fish for gonad estimation, muscle, skin and fins were collected simultaneously for color estimation following Bjerking (1992). Fish, feed samples, unconsumed feed and ovaries were weighed in an electric monopan balance to an accuracy of 1 mg.

Breeding

After attaining sexual maturity, two females were randomly chosen from each replicate tank and reared with a male in a separate tank containing a sufficient quantity of macrophytes of the *Hydrilla* species until the completion of the experiment. The remaining test animals were removed from the experimental tanks. When the breeding females released their young ones, they were isolated from the parents and counted and thereafter subjected for color estimation.

Statistical analysis

Students 't' tests was used to determine the significance of differences in mean values between experimental groups. Duncan multiple comparison range test was applied to detect the significance of differences between the mean values between experimental data (Zar, 1984).

RESULTS

The results showed that, the feeding rate was high in 1 MG *X. helleri* followed by 2 MG and 3 MG fish. The mean feeding rate was high at pre-breeding as compared to post-breeding in 1 MG group. However, the trend was reversed in 2 MG fish and 3 MG groups. Weight gain and specific growth rate of tested age groups showed the higher values at pre-breeding than the post breeding. For instance, the mean SGR of 2 MG *X. helleri* was 1.26, 1.29, 1.41 and 2.17 % day⁻¹ at pre-breeding and it significantly (P<0.05) declined to 0.60, 0.55, 0.74 and 0.60 % day⁻¹ in fish fed on 0, 5, 10 and 15% *Spirulina* diets respectively (Table 2). Irrespective of age groups, fish fed on 15% *Spirulina* diet showed the maximum feeding and growth rates as compared to other diets.

Gonad weight and gonadosomatic index were increased with time and they were more pronounced in fish fed on 15% *Spirulina* diet than those fed on other diets. Fish fed on 15% *Spirulina* diet had 10 times more gonad weight as compared to control diet in 1 MG and 2 MG and 4.5 times in 3 MG on day 75 (Table 3). Similar trend was obtained in GSI also. Duncan multiple range test revealed that, 15% *Spirulina* diet significantly (P < 0.05) influenced the more gonad weight and GSI as compared to other *Spirulina* diets.

Fry production of *X. helleri* was increased with an increasing of *Spirulina* levels in the diets of all age

groups and progress of breeding number. 2 MG and 3 MG bred 4 times while 1 MG bred 3 times. Female

X. helleri belongs to 1 MG, 2 MG and 3 MG fed on 15% *Spirulina* diet released 368, 570 and 478 fry during the experimental period (Table 4).

Tissue (fins, skin and muscle) carotenoid content was increased with time and Spirulina levels in all the age groups of X. helleri. Maximum carotenoid content was observed in fish consumed 15% Spirulina diet in all tissues (except muscle) and age groups while control group elicited low carotenoid content. Duncan multiple range test showed that carotenoid content between Spirulina treatments differed significantly (P < 0.05) with better values in chosen age groups fed with 15% Spirulina diet. Fins exhibited the maximum coloration followed by skin and muscle (Figure 1). In fins, there were 15, 11 and 5 fold enhancement of coloration observed in 1 MG as compared to 2 MG and 3 MG X. helleri fed with 15% Spirulina diets. Similarly, an enhancement of coloration in skin or muscle was 26 or 28, 19 or 19 and 6 or 8 times in 1 MG, 2 MG and 3 MG fish respectively. It showed that, coloration was declined with increase in age groups.

Irrespective of age groups and dietary treatments, the total carotenoid contents of the newly born fry showed an increasing trend over the progressing of the breeding cycles (Table 5). It is interesting to observe that, 15% *Spirulina* diet fed 1 MG *X. helleri* fry contained maximum carotenoid content (0.044 mg /100 mg wet weight) in first breeding and the same level of carotenoid content was observed in third breeding in 2 MG and 3 MG (0.046 and 0.044 mg/ 100 wet weight) *X. helleri*. Duncan multiple range test elicited that 1 MG *X. helleri* fry had high carotenoid content as compared to 2 MG or 3 MG (Table 5). The mean carotenoid content in fry was increased with increasing of *Spirulina* levels; however, it declined with increasing of age groups (Figure 2).

Ingredients (g/100g diet)	Spirulina levels (%)									
higredients (g/100g diet)	0	5	10	15	20					
Fish meal	50	35	30	25	20					
Groundnut oil cake	36	36	36	36	36					
Maida	6	6	6	6	6					
Tapioca flour	6	6	6	6	6					
Spirulina	0	5	10	15	20					
Vitamin and mineral mixtures	1	1	1	1	1					
Cod liver oil	1	1	1	1	1					

Table 1. Formulation and percentage composition experimental diets.

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Proximate composition (%)										
Ductoin	45.02	±	46.99	±	48.35	±	50.08	±	51.38	±
Protein	0.14		0.11		0.16		0.27		0.12	
Linid	6.96	±	5.74	±	5.35	±	4.76	±	4.60	±
Lipid	0.07		0.02		0.03		0.03		0.02	
4 - L	15.83	±	17.67	±	18.50	±	19.95	±	22.17	±
Ash	0.76		0.29		0.50		0.84		0.29	
Niture your fried antine at	32.19	±	29.60	±	27.80	±	25.21	±	21.85	±
Nitrogen free extract	0.82		0.63		0.74		0.43		0.34	

Each value is the mean \pm SD of three observations.

Table 2. Effect of different levels of *Spirulina* (%) on feed consumption (g dry matter), feeding rate (mg g⁻¹ live fish day⁻¹), weight gain (g wet weight) and specific growth rate (% day⁻¹) in different age groups of red swordtail, *Xiphophorus helleri*. Each value is the mean \pm SD of three observations.

Levels of <i>Spirulina</i> (%)													
I MG			2	2MG			6	BMG					
)	5	10	15	0	5	10	15	0	5	10	15		
Feed consumption 7.12 9.60 7.64 9.00 11.02 9.22 9.54 12.05													
7.13	8.60	7.94	10.68	7.09	7.64	8.98	11.83	8.23	8.54	12.85	10.21		
4.26	4.98	8.73	11.35	6.42	7.04	8.87	11.75	6.70	8.21	10.20	10.57		
Feeding rate													
39.47	42.85	50.39	55.24	20.56	20.15	22.44	30.86	14.44	16.54	23.85	17.89		
22.02	24.54	34.08	35.33	29.42	30.50	34.47	31.98	25.56	25.35	27.16	27.03		
Weigh	eight gain												
2.38	3.68	4.82	6.06	5.09	5.70	6.66	8.63	6.27	6.41	8.41	8.80		
1.00	0.88	2.36	2.84	1.31	1.29	1.95	2.67	1.89	3.21	3.05	2.69		
Specifi	ic growt	h rate											
1.31	1.63	2.36	2.64	1.26	1.29	1.41	2.17	0.98	1.06	1.28	1.30		
).49	0.39	0.77	0.77	0.60	0.55	0.74	0.60	0.68	0.64	0.58	0.48		
	MG Feed c 7.13 5.26 Feedin 9.47 2.02 Weigh 2.38 .00 Specifi .31	MG 5 Seed consump 5 Seed consump 6 2.13 8.60 2.26 4.98 Seeding rate 9 9.47 42.85 2.02 24.54 Veight gain 3.68 .00 0.88 Specific growt .31	MG 5 10 Seed consumption 2.13 8.60 7.94 2.26 4.98 8.73 Feeding rate 9.47 42.85 50.39 22.02 24.54 34.08 Weight gain 9.38 3.68 4.82 .00 0.88 2.36 Specific growth rate .31 1.63 2.36	MG 2 0 5 10 15 Feed consumption 7.13 8.60 7.94 10.68 2.13 8.60 7.94 10.68 2.26 4.98 8.73 11.35 Feeding rate 9.47 42.85 50.39 55.24 22.02 24.54 34.08 35.33 Weight gain 9.47 42.85 50.39 55.24 2.02 24.54 34.08 35.33 35.33 Weight gain 9.38 3.68 4.82 6.06 .00 0.88 2.36 2.84 34 Specific growth rate 31 1.63 2.36 2.64	MG 2MG 5 10 15 0 Seed consumption 5 10 15 0 2.13 8.60 7.94 10.68 7.09 2.26 4.98 8.73 11.35 6.42 Seeding rate 9 9 47 42.85 50.39 55.24 20.56 22.02 24.54 34.08 35.33 29.42 Weight gain 9 9 9 9 9 10.68 1.31 3.38 3.68 4.82 6.06 5.09 5 1.31 Specific growth rate 33 2.36 2.84 1.31	MG $2MG$ 5 10 15 0 5 Seed consumption 7.94 10.68 7.09 7.64 2.13 8.60 7.94 10.68 7.09 7.64 2.26 4.98 8.73 11.35 6.42 7.04 Seeding rate 709 7.64 7.04 7.04 Seeding rate 7.02 24.54 34.08 35.33 29.42 30.50 Veight gain 7.04 34.08 35.33 29.42 30.50 Veight gain 7.04 34.08 35.33 29.42 30.50 .31 1.63 2.36 2.84 1.31 1.29	MG $2MG$ 0 5 10 15 0 5 10 Seed consumption 2.13 8.60 7.94 10.68 7.09 7.64 8.98 2.26 4.98 8.73 11.35 6.42 7.04 8.87 Seeding rate 9.47 42.85 50.39 55.24 20.56 20.15 22.44 20.2 24.54 34.08 35.33 29.42 30.50 34.47 Veight gain 4.38 3.68 4.82 6.06 5.09 5.70 6.66 .00 0.88 2.36 2.84 1.31 1.29 1.95 Specific growth rate .31 1.63 2.36 2.64 1.26 1.29 1.41	MG 2MG 3 0 5 10 15 0 5 10 15 Seed consumption 7.09 7.64 8.98 11.83 2.13 8.60 7.94 10.68 7.09 7.64 8.98 11.83 2.26 4.98 8.73 11.35 6.42 7.04 8.87 11.75 Seeding rate 9.47 42.85 50.39 55.24 20.56 20.15 22.44 30.86 2.02 24.54 34.08 35.33 29.42 30.50 34.47 31.98 Weight gain 3.68 4.82 6.06 5.09 5.70 6.66 8.63 .00 0.88 2.36 2.84 1.31 1.29 1.95 2.67 Specific growth rate 3.1 1.63 2.36 2.64 1.26 1.29 1.41 2.17	MG $2MG$ $3MG$ 0 5 10 15 0 5 10 15 0 Seed consumption 2.13 8.60 7.94 10.68 7.09 7.64 8.98 11.83 8.23 2.26 4.98 8.73 11.35 6.42 7.04 8.87 11.75 6.70 Geeding rate 9.47 42.85 50.39 55.24 20.56 20.15 22.44 30.86 14.44 2.02 24.54 34.08 35.33 29.42 30.50 34.47 31.98 25.56 Weight gain	MG 2MG 3MG 5 10 15 0 5 10 15 0 5 Seed consumption	MG 2MG 3MG 5 10 15 0 5 10 15 0 5 10 Seed consumption -		

FC** : Mean feed consumption

Fr** : Mean feeding rate

Wg** : Mean weight gain

SGR**: Mean specific growth rate

Rearing					Le	vels of Sp	pirulina	(%)				
period		11	MG			2 N	ЛG			3 N	ЛG	
(days)	0	5	10	15	0	5	10	15	0	5	10	15
						Gonad	weight					
25	ND	ND	ND	ND	ND	12.50± 0.54 ^a	35.00 ± 1.00 ^b	68.33 ± 2.64^{d}	30.00 ± 1.07 ^b	$\pm 3.54^{\circ}$	70.00 ± 2.00^{d}	
50	ND	22.50 ± 1.54^{a}	43.75 ± 1.79 ^b	87.50 ± 4.61 ^c	30.00 ± 1.07^{a}	52.50 ± 3.61^{b}	90.00 ± 4.14 ^c	316.50 ± 3.54^{e_*}	72.50 ± 3.54 ^c	±	322.50 ± 3.54^{e_*}	±
75	25.00 ± 0.07^{a}	52.50 ± 3.54 ^b	±	267.50 ± 10.61 ^d *	± 7.07	257.50± 10.60 ^d			$272.50 \pm 10.6^{d_{*}}$			
100	67.5 ± 3.54 ^a	182.50 ± 10.61^{b*}										
125	130.21 ± 4.20*											
					G	onadosor	natic ind	ex				
25	ND	ND	ND	ND	ND	$\begin{array}{ccc} 3.64 & \pm \\ 0.07^{a} \end{array}$	6.66 ± 0.41^{b}	7.68 ± 0.54^{b}	$\begin{array}{c} 6.07 \ \pm \\ 0.86^{b} \end{array}$	8.30 ± 0.30^{b}	$9.49 \pm 0.65^{\circ}$	9.12 ± 0.57 ^c
50	ND	14.47 ± 0.68 ^b		21.86 ± 1.61°		7.45 ± 1.05 ^a	13.13 ± 0.73 ^b	29.21 ± 0.04 ^c *	9.12 ± 0.26^{a}	10.57 ± $0.17^{a_{*}}$	28.73 ± 0.16 ^c *	$18.98 \pm 0.40^{b_{*}}$
75	15.83 ± 2.93 ^a	22.65 ± 1.03 ^b	$25.38 \pm 0.02^{b}*$	$37.57 \pm 0.90^{\circ}*$	$22.62 \pm 0.60^{b}*$	36.86 ± 0.10 ^c *	$48.50 \pm 0.21^{d_{*}}$		$26.39 \pm 0.28^{b_{*}}$			
100	18.70 ± 0.31 ^a	27.26 ± 0.31 ^b *										
125	34.60± 0.22*											

Table 3. Effect of different levels of *Spirulina* (%) on gonad weight (mg wet weight) and gonadosomatic index (%) in different age groups of red swordtail, *Xiphophorus helleri*.

* Breeding commenced; ND – Gonad not developed.

Values (mean \pm SD) with different superscripts in the same row are significantly different (P < 0.05).

											Leve	ls of S	piruli	na (%)										
No. of				1	MG							2 N	MG							3 N	мG			
bree	0		5		10		15		0		5		10		15		0		5		10		15	
ding	a	b	a	В	а	b	a	b	a	b	a	b	а	b	a	b	a	b	a	b	a	b	a	b
Ι	$172 \\ .00 \\ \pm \\ 1.6 \\ 3$	26. 332 ± 1.1 5	121 .00 ± 0.8 2	33 .3 3 ± 1. 25	117 .00 ± 0.8 2	41. 67 ± 1.2 4	111 .00 ± 0.8 2	60. 67 ± 1.7 0	92. 00 ± 0.8 2	32. 67 ± 2.0 5	87. 33 ± 1.2 5	42. 35 ± 1.2 5	82. 00 ± 0.8 2	57. 00 ± 1.6 3	77. 00 \pm 0.8 1	$68. \\ 00 \\ \pm \\ 0.8 \\ 2$	93. 00 ± 0.8 2	35. 33 ± 1.7 0	87. 33 ± 1.7 0	45. 33 ± 1.2 4	77. 00 \pm 0.8 2	57. 66 ± 1.2 5	$82. \\ 00 \\ \pm \\ 0.8 \\ 2$	60. 33 ± 2.4 9
Π	NB	-	152 .67 ± 1.2 5	58 .0 0 ± 3. 27	145 .67 ± 1.8 9	86. 00 ± 1.6 3	136 .33 ± 0.9 4	131 .33 ± 1.7 0	123 .00 ± 0.8 2	45. 33 ± 1.2 5	119 .67 ± 0.4 7	53. 00 ± 1.8 2	111 .00 ± 0.8 2	111 .67 ± 2.8 7	102 .67 ± 1.7 0	145 .33 ± 1.2 5	123 .67 ± 0.9 4	44. 00 ± 0.8 2	115 .33 ± 0.9 4	53. 00 ± 0.8 2	$105 \\ .00 \\ \pm \\ 2.8 \\ 3$	119 .00 ± 2.4 9	109 .00 ± 0.8 2	130 .00 ± 2.9 4
III IV	NB NB	-	NB NB	-	172 .67 ± 2.0 5 NB	123 .00 ± 2.4 5	162 .00 ± 1.4 1 NB	176 .00 ± 1.6 3	151 .00 ± 0.8 2 NB	66. 00 ± 1.6 3	$147 \\ .00 \\ \pm \\ 0.9 \\ 4 \\ 173 \\ .67 \\ \pm \\ 0.4$	$120 \\ .67 \\ \pm \\ 1.2 \\ 5 \\ 147 \\ .67 \\ \pm \\ 2.0 \\ $	$137 \\ .00 \\ \pm \\ 0.8 \\ 2 \\ 163 \\ .33 \\ \pm \\ 0.4$	$143 \\ .33 \\ \pm \\ 2.0 \\ 5 \\ 164 \\ .33 \\ \pm \\ 2.2$	$128 \\ .00 \\ \pm \\ 1.4 \\ 1 \\ 153 \\ .33 \\ \pm \\ 1.2 \\ $	$167 \\ .67 \\ \pm \\ 1.2 \\ 5 \\ 188 \\ .67 \\ \pm \\ 2.8 \\$	152 .00 ± 1.6 3 NB	65. 00 ± 0.8 2	$144 \\ .00 \\ \pm \\ 0.8 \\ 2 \\ 170 \\ .33 \\ \pm \\ 0.4$	$120 \\ .00 \\ \pm \\ 3.2 \\ 7 \\ 149 \\ .67 \\ \pm \\ 2.8 \\ $	$132 \\ .67 \\ \pm \\ 0.4 \\ 7 \\ 158 \\ .33 \\ \pm \\ 0.0 \\ $	140 .33 \pm 2.0 5 149 .00 \pm 2.7	$136 \\ .00 \\ \pm \\ 1.4 \\ 1 \\ 162 \\ .00 \\ \pm \\ 2.1$	$140 \\ .67 \\ \pm \\ 1.2 \\ 5 \\ 146 \\ .67 \\ \pm \\ 2.8 \\ $
Total fry prod uced		26. 32		91 .3 3		250 .67		368 .00		144 .00	0.4 7	2.0 5 363 .33	0.4 7	3.3 0 476 .33	1.2 5	2.8 7 569 .67		144 .33	0.4 7	2.8 7 368 .00	0.9 4	3.7 4 475 .99	2.1 6	2.8 7 477 .67

 Table 4.
 Effect of different levels of Spirulina (%) on fertility in different age groups of red swordtail, Xiphophorus helleri.

NB - Not breed; a - No. of days; b - No. of young ones

Each value is the mean \pm SD of three observations.

Table 5. Effect of different levels of Spirulina (%) on carotenoid contents (mg-'100 mg wet weight) in young ones of Xiphophoru	S
<i>helleri</i> as a function of age groups and breeding. Each value is the mean ± SD of three observations.	

1

Number of	Levels of Spirulina (%)														
breeding	1 MG				2 MG				3 MG						
	0	5	10	15	0	5	10	15	0	5	10	15			
Ι	0.011 ± 0.002^{a}	0.029 ±0.001 ^{ab}	$\begin{array}{c} 0.036 \pm \\ 0.002^{b} \end{array}$	0.044 ± 0.002 ^b	$\pm 0.011 \pm 0.003^{a}$	0.017 ± 0.002^{a}	0.022 ± 0.002^{a}	0.027 0.004 ^{ab}	$\pm 0.012\pm 0.002^{a}$	0.014 ± 0.005^{a}	0.026 ± 0.004^{ab}	$\begin{array}{c} 0.020 \ \pm \\ 0.003^{a} \end{array}$			
II	NB	0.029 ± 0.004 ^{ab}	$\pm 0.036 \pm 0.001^{b}$	0.046 ± 0.008°	$\pm 0.017 \pm 0.003^{a}$	0.023 ± 0.004 ^a	$\pm 0.031 \pm 0.002^{ab}$	0.036 0.001 ^b	$\pm 0.014 \pm 0.002^{a}$	$\pm 0.019 \pm 0.002^{a}$	$\pm 0.033 \pm 0.003^{ab} \pm$	$\begin{array}{c} 0.026 \\ 0.004^{ab} \end{array} \pm$			
III	NB	NB	$\begin{array}{c} 0.038 \pm 0.004^{b} \end{array}$	0.049 ± 0.007 ^c	$\pm 0.017 \pm 0.002^{a}$	0.029 ± 0.001 ^{ab}	$\pm 0.035 \pm 0.004^{b}$	0.046 0.004 ^c	$\begin{array}{c} \pm \ 0.017 \ \pm \\ 0.001^a \end{array}$	$\pm 0.026 \pm 0.005^{a}$	$\pm 0.045 \pm 0.003^{\circ}$	$\begin{array}{c} 0.044 \\ 0.006^{b} \end{array} \pm$			
IV	NB	NB	NB	NB	NB	0.031 ± 0.004 ^{ab}	$\pm 0.036 \pm 0.006^{b}$	0.049 0.004 ^c	$^{\pm}$ NB	0.030 ± 0.008 ^{ab}	$\pm 0.047 \pm 0.008^{\circ}$	$\begin{array}{c} 0.045 \\ 0.002^{c} \end{array} \pm$			
Mean carotenoid contents	0.011 ± 0.000	± 0.029 ± 0.000	= 0.036 ± 0.001	0.046 ± 0.002	± 0.015 ± 0.002	0.025 ± 0.005	= 0.031 ± 0.005	0.040 0.008	$\pm 0.014 \pm 0.002$	0.022 ± 0.006	= 0.038 ± 0.008	$\begin{array}{c} 0.034 \ \pm \ 0.011 \end{array}$			

NB – Not breed.

Values (mean \pm SD) with different superscripts in the same row are significantly different (P < 0.05).

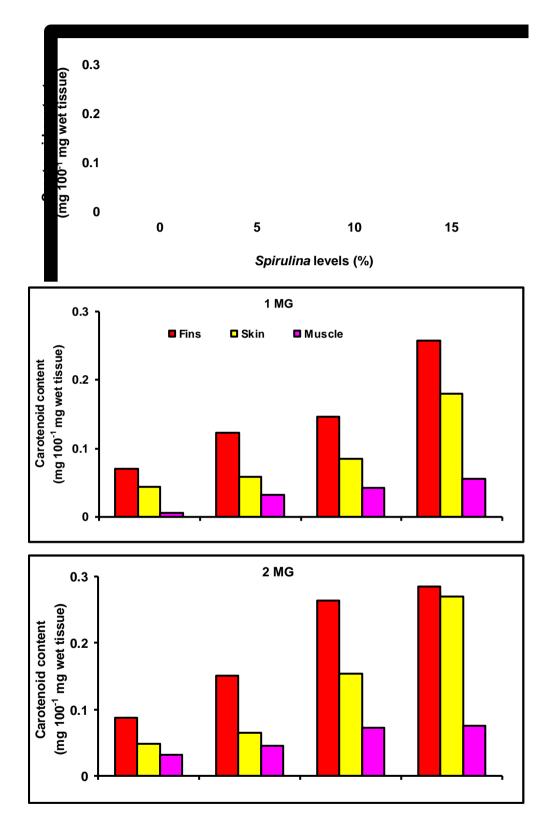


Figure 1. Effect of different levels of *Spirulina* on carotenoid contents in different age groups of *Xiphophorus helleri* on day 50.

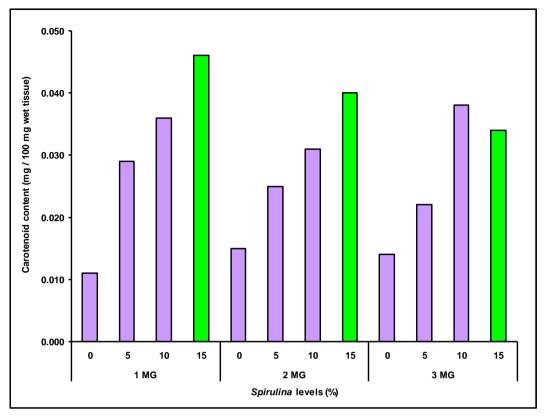


Figure 2. Effect of different levels of *Spirulina* on mean carotenoid contents in young ones of *Xiphophorus helleri* as a function of different age groups.

DISCUSSION

The present study revealed that high feeding rate in 1 MG might be due to the increased metabolic rate. Faster gastric evacuation can help the fish to digest more food per unit time and greater stomach volume can help ingest relatively large quantities of food (Elliott, 1975). However, the high mean feeding rate at post-breeding in 2 MG and 3 MG *X. helleri* was evidently due to the more quantum of food energy required for the gonad development and fry production. Townshend and Wooton (1984) found that females of the convict cichlid, *Cichasoma nigrofasciatum*, channel a higher proportion of ingested food energy to gonad development and fecundity, supports the present observation.

The slower weight gain and SGR in *X. helleri* at post-breeding could be due to diversion of more assimilated food energy to ovary development and fry production. James and Sampath (2003a and b) reported that, growth rate was higher in ornamental fishes before breeding and drastically declined afterwards; the significant reduction might have been due to the allocation of food energy to gonad development and fry/egg production. Rathinam (1993) reported that a loss of body weight during parturition in live-bearers. *X. helleri* is ovoviviparous. It bears young ones developed from eggs and retained in the mother's body, but without any close tissue connections between the two for the supply of extra nourishment to the embryo. The embryo is nourished from energy stored in the eggs. During development, the uptake of organic substances and intake of water molecules from the maternal body (Balinsky, 1970) could result in considerable loss of energy in *X. helleri*.

Many factors influence the reproduction in female fish. Among them size, age, physical condition, reproductive history and nutrition are important (Woodhead, 1960; James and Sampath, 2003a and b). The number of ova was positively correlated to the ovarian/gonad weight and body weight of female guppy, *Poecilia reticulata* (Dahlgren, 1980). Age of fish is directly related to breeding performance of the fish and as well as the growth of the fish (James and Sampath, 2004). The result showed that 2 MG *X. helleri* fed 15% *Spirulina* diet released more fry and bred more frequencies than other groups and it was probably due to the middle age of the fish and optimum *Spirulina* diet. In many egg laying fish species, size and number of egg productions were directly related to the age of fishes (Kamler, 2005). Usually, the largest and more number of eggs are produced by fish that are of middle age for their species (Bartel, 1971; Wilkonska *et al.*, 1994). However, Milton and Arthington (1983) reported that, the fecundity of fish species is related to body size.

Even though the red sword tail fish is brightly colored, dietary supplementation of Spirulina have significantly enhanced coloration in the fins and skin. James et al. (2006) reported that, increase in carotenoid content in the fins, skin and muscle of X. helleri was proportionate to the increase in dietary carotenoid, demonstrating that the fish has the capacity to efficiently utilize carotenoids. A dosedependent carotenoid content was reported in the muscle of Arctic char and Salmon (Bijerking et al., 1990; Ando et al., 1994; Halten et al., 1997; Wathne et al., 1998) supports the present observation. The low carotenoid content in the muscle indicates that the assimilated carotene is directly transported to the skin fins to provide necessary pigmentation. and According to Schiedt et al. (1985) this is achieved by establishing reductive metabolic pathways from the muscle to the skins and fins. In Salmon, Arctic char, and trout, the pigmentation of the integument and fins occurs only during sexual maturation and a reduction of muscle carotenoid indicates that carotenoids are mobilized directly to the integument and fins from the muscle during that season.

The results also showed that 1 MG younger fish and their fry have a higher turnover of carotenoid content than 3 MG older fish. This may explain the early feeding of Spirulina diets in 1 MG over long periods of time would appear to be an effective pigmentation in 1 MG X. helleri and their fry. Besides. the greater retention efficiency of carotenoids in parents tissues and transformation of coloration from parent X. helleri to their offspring. The findings of Smith et al. (1992) on Coho salmon suggested that feeding low astaxanthin concentrations throughout the grow-out period from fry to market size resulted the most efficient use of pigment and produced uniform pigmentation. Schiedt et al. (1989) observed that accumulation of idoxanthin was size or age dependent, since smaller fish had a higher percentage of idoxanthin in Salmo gairdnerii and in Arctic char, Salvelinus alpinus (Aas et al., 1997).

CONCLUSION

Based on the results, it is concluded that, feeding 15% *Spirulina* diet from 2 months old red swordtail is ideal to enhance the growth, coloration and reproduction and also reduce the wastage of *Spirulina* utilization

and production cost . To enhance the coloration in ornamental fishes, feeding the *Spirulina* at early stages is beneficial.

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