Raja Rammohun Roy National Agency for ISBN
Department of Higher Education, Ministry of Education
Government of India

EFFECTS OF DIFFERENT CONCENTRATION OF *SPIRULINA* ALGAE AS FEED ADDITIVE ON GROWTH PERFORMANCE OF BLACK MOLLY FISH AT DIFFERENT STAGES

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Abstract

In the present study black molly fishes (at early fry stage and at adult stage) were exposed to different concentration of *Spirulina* as feed additive to observe the growth performance of black molly fish at different stages in the captive condition. Mollies are found in varieties of color and body shape; and are good considered as ornamental value. Black molly (*Poicilia sphenops*) fishes of almost similar length and weights were collected from local aquarium shops and reared in laboratory condition for forty days to study the effect of spirulina additive diets I to IV (0, 5, 10, 15 & 20 %) on their growth performance. Looking to the nutritional requirements at different stages, spirulina had significant effect on both adults and fries molly fish. The body weight gain , body length gain and specific growth rate in adult mollies was found higher (BWG 3.99 g , BLG 0.29 cm and SGR 1.03 %) in additional 10% spirulina diet (diet III), whereas lowest was reported from control (commercial diet). In case of molly fries, additional 5% spirulina diet (diet II) showed the best results in the body length gain (BLG 0.73 cm), body weight gain (BWG 0.48 g) and specific growth rate (SGR 5.30 %) than control and other diets. Survival rate of molly fries was found higher (SR 95%) in diet-II and survival rate of adult molly fishes was found higher (SR 93.75 %) in diet III than control (50 % and 56.25 % respectively). To increase the fish crop yield in the captive condition, the available food (commercial diet) may not be sufficient to fulfill the nutritional requirement at different stages of molly fish. In this situation, proper food can be managed by providing well formulated additive diets.

Keywords: black molly fish, commercial diets, feed additive, spirulina, SGR, survival rate.

1. Introduction

Ornamental fish culture is one of the most important fields of Aquaculture. There are various factors involved in ornamental fish culture and among these quality and quantity of food, density and water hardness are the most important (James, 1998) [1]. Fish or any animal needs an adequate balanced diet that contains all the required necessary essential nutrients to have optimal growth. These nutrient requirements vary from species to species, gender, the surrounding where it lives, different stages of life, and the health status of the species. The amount of energy required also differs with different life stages of fish. Bowen (1987) [2] reported that 'the energy required by fish is much low when compared with warm-blooded organisms and hence it needs a high level of protein to energy ratio in the diet. Compared to the life stages of fish, juvenile fishes require low energy high protein and mature adult fish requires high energy low protein diet'. Feed nutrition plays a vital role in the production of ornamental fishes. In recent years, feed production is updated with scientific technology in producing species-specific food formulation. Every feed prepared for aquaculture is a scientifically designed feed to obtain optimum growth within a short period. Inadequate nutrition can also

affect fish health. The feed should be prepared economically with limited resources at the same time it should enhance the productivity and health of the organisms. Looking to the available resources and nutritional requirements at different stages, spirulina algae have been chosen as main additive ingredient to study the its effects on both adults and fries molly fish. Black mollies are becoming more popular having high adptiaptive and reproductive behaviour, ease of availability and affordable price. They generally accept all kind of foods including dried and live. The feed requirements vary in quantity and quality according to their feeding habits. Therefore, it is necessary to focus on the feed supply according to their nutritional requirement. According to Elangovan and Shim (1997), the comparison of protein requirements between fish species is complex since this can vary according to the size and life stage (Lochmann and Phillips, 1994), diet formulation or farming condition (Fiogbe and Kestemont, 1995). The crude protein requirements in many fish species generally range between 25 and 55% (National Research Council, 1993). In ornamental fish, a correct formulation of the diet improves the nutrient digestibility; supply the metabolic needs and reducing the maintenance cost and at the same time the water pollution (Yohana and Wilson, 2011). It is also essential to provide the adults and fish fries to quality feed for better growth and survival.

2. Materials and Methods

2.1. Experimental fish stocking , experimental diets and experimental unit design

Experimental fish, Black molly, *Poecilia sphenops* L. were stocked in glass aquaria. All aquaria were filled by fresh tap water during the experiment of 40 days in the laboratory. Length and weight of individual black mollies were recorded before starting of the experiment using measuring scale and electric balance. Separate sets of aquaria were marked regarding six different diets. Among those five experimental diets were formulated using different commonly available ingredients; those types containing Spirulina powder with different level (0, 5, 10, 15, 20%) and first one type was commercially available pellet feed as a control. Continuous aeration was provided by aquarium air pump. Excreta and leftover feed were removed at alternate day from the bottom of each aquariam through siphoning and along with that one third volume of water was changed from each experimental unit.

Table 1. Ingredients quantity of prepared diets of experimental diets

Ingredients	**Diet1 (g/100g)**	**Diet2 (g/100g)**	**Diet3 ((g/100g)**	**Diet4 (g/100g)**	**Diet5 (g/100g)**	**Diet C. or Control**
Spirulina Powder (SP)	**0**	**5**	**10**	**15**	**20**	Commercial pellet feed
Fresh Water Fish (FWF)	40	35	30	25	20	
Marine Water Fish (MWF)	10	10	10	10	10	
Wheat flour (WF)	15	15	15	15	15	
Corn Flour (CF)	10	10	10	10	10	
Soyabean Meal (SbM)	20	20	20	20	20	

Sunflower Oil (SO)	4	4	4	4	4	
Vitamin tablet (Vt)	1	1	1	1	1	

2.2. Growth

Molly fries and adults were measured at the beginning and then at the end of the experiment (40 days). Growth was calculated as the difference between the length and weight at the beginning of the experiment and on the day of calculation. Specific growth rate (SGR) was calculated. Survival and growth rate of *Poecilia sphenops* was calculated for a period of 40 days using following equations:

❖ **Body Weight Increase (BWI) :**

According to Tacon (1990), the body weight increase was calculated based on the formula:

$$BWI\ (g) = Wt - Wo$$

Where; Wt = Average weight of molly at the end of the experiment (g); Wo = Average molly weight at the beginning of the experiment (g)

❖ **Percent Body Weight Increase (PBWI) :**

According to Bekcan et. al. (2006), the percent body weight increase was calculated based on the formula:

$$PBWI(\%) = [(Wt - W0)/ Wt] \times 100$$

Where; Wt = Average weight of molly at the end of the experiment (g); Wo = Average molly weight at the beginning of the experiment (g)

❖ **Body Length Gain (BLG):**

The body length gain was calculated based on the standard formula:

$$BLG\ (cm) = Lt - L0$$

Where; Lt = Average length of molly at the end of the experiment (cm); Lo = Average molly length at the beginning of the experiment (cm)

❖ **Percent length gain (PLG):**

The percent length gain was calculated based on the standard formula:

$$PLG\ (\%) = [(Lt - L0) / Lt] \times 100\%$$

Where; Lt = Average length of molly at the end of the experiment (cm); Lo = Average molly length at the beginning of the experiment (cm)

❖ **Specific Growth Rate (SGR) :**

According to Hevroy et al. (2005), the specific growth rate was calculated based on the formula:

$$SGR\ (\%/day) = [(\ln Wt - \ln W0) / t\ ] \times 100$$

Where; ln Wt = Natural Logrithum value of average weight of fish at the end of the experiment (g); ln Wo = Natural Logrithum value of average weight of fish at the beginning of the experiment (g) and t = Rearing period (days)

- **Body Weight Gain (BWG) :**

According to De Silva and Anderson (1995), the body weight gain was calculated based on the formula:

$$BWG\ (g) = (Wt - W0) \times Nt$$

Where; Wt = Average weight of molly at the end of the experiment (g); Wo = Average molly weight at the beginning of the experiment (g) and Nt = final numbers of in each treatment aquarium

- **Daily Growth Rate (DGR) :**

According to De Silva and Anderson (1995), the daily growth rate was calculated based on the formula:

$$DGR\ (\%) = [(Wt - W0) / t] \times 100$$

Where; Wt = Average weight of molly at the end of the experiment (g); Wo = Average weight of molly at the beginning of the experiment (g) and t = Rearing period (days)

- **Survival of mollies**:

According to Ai et al. (2006), the survival of the molly was calculated by subtracting the number of mollies harvested at the end of the experiment from the mollies stocked at the initiation of the experiment.

$$Survival\ (\%) = [Nt / N0] \times 100$$

Where; Nt and N0 were final and initial numbers of mollies in each treatment aquarium.

2.3. Water quality parameters

Physical and chemical water quality parameters were measured by following the methods of APHA (1991). From each aquarium, everyday water temperature and pH were measured. Dissolved oxygen, total alkalinity, total hardness and ammonical nitrogen were measured at alternate day of the experiment period.

2.4. Data processing and statistical analysis

The collected data from the feeding trials were incorporated in Microsoft office excel sheets. Processed data then were analyzed to compare the effect of different experimental diets on the growth and survival of molly. Growth parameters and water quality parameters according to six different diets were compared by using one-way ANOVA. Statistical data analysis was accomplished with SPSS software to evaluate the significant differences among treatments.

3. Results and Discussion

The growth was analyzed and compared with control diet. Growth pattern, survival and weight gain had significantly varied among all experimental diets provide to fry and adults mollies (Table 2 and 3). The maximum body weight gain (BWG 0.48 g) , body length gain (0.73 cm) and specific growth rate (SGR 5.30 %) were measured from fries fed with diet II (5% Spirulina); whereas the lowest (0.20 and 0.24 cm) was found in diet 1 (0% spirulina) and in control respectively (table 2). In case of adult molly; the body weight gain , body length gain and specific growth rate in adult mollies was found higher (BWG 3.99 g , BLG 0.29 cm and SGR 1.03 %) in additional 10% spirulina diet (diet III) than control and other diets. Survival rate of molly fries was found higher (95%) in diet-II and survival rate of adult molly fishes was found higher (93.75 %) in diet III than control (50 % and 56.25 % respective experiments). Comparative study of experiment result showed that the higher growth performance and survival were observed in fries of molly in the diet II (5% Spirulina) where as the higher growth performance and survival were observed in adult molly fishes in the diet III (10% Spirulina). Therefore, to increase the fish crop yield in the captive condition, the any single type of food may not be sufficient to fulfill the nutritional requirement at different stages of molly fish. In this situation, proper food can be managed by providing well formulated additive diets.

Table 2. Growth performance of *P. sphenops* fry fed with formulated and commercial diets.

Growth parameter	Control	Diet 1 (0% Spirulina)	Diet 2 (5% Spirulina)	Diet 3 (10% Spirulina)	Diet 4 (15% Spirulina)	Diet 5 (20% Spirulina)
Initial length (cm)	1.17 cm	1.18 cm	**1.16 cm**	1.11 cm	11.13 cm	1.15 cm
Final length (cm)	1.41 cm	1.38 cm	**1.89 cm**	1.40 cm	1.47 cm	1.45 cm
Initial weight (g)	0.0365 g	0.0365 g	**0.0340 g**	0.0315 g	0.0350 g	0.0340 g
Final weight (g)	0.0370 g	0.0407 g	**0.0425 g**	0.0350 g	0.0370 g	0.0361 g
BWI (g)	0.0005 g	0.0042 g	**0.0085 g**	0.0035 g	0.0020 g	0.0021 g
Length gain (cm)	0.24 cm	0.20 cm	**0.73 cm**	0.29 cm	0.34 cm	0.30 cm
BWG (g)	0.02 g	0.15 g	**0.48 g**	0.15 g	0.10 g	0.008 g
DGR (%)	0.0013 %	0.0105 %	**0.0213 %**	0.0088 %	0.0050 %	0.0052 %
PBWI (%)	1.35 %	10.32 %	**20 %**	10 %	5.41 %	5.82 %
PLG (%)	17.02 %	14.49 %	**38.62 %**	20.71 %	23.13 %	20.69 %
(SGR) (%/day)	4.98 %	5.07 %	**5.30 %**	5.29 %	5.08 %	5.13 %
Survival fish fries	50%	60%	**95%**	70%	80%	65%

Table 3. Growth performance of *P. sphenops* fish fed with formulated and commercial diets.

Growth parameter	Control	Diet 1 0%	Diet 2 5%	Diet 3 10%	Diet 4 15%	Diet 5 20%

		Spirulina	Spirulina	Spirulina	Spirulina	Spirulina
Initial length (cm)	3.51 cm	3.51 cm	3.51 cm	**3.51** cm	3.51 cm	3.51 cm
Final length (cm)	3.52 cm	3.60 cm	3.61 cm	**3.79** cm	3.53 cm	3.58 cm
Initial weight (g)	0.59 g	0.58 g	0.59 g	**0.58** g	0.58 g	0.59 g
Final weight (g)	0.60 g	0.65 g	0.68 g	**0.71** g	0.65 g	0.65 g
Body weight increase BWI (g)	0.01 g	0.06 g	0.09 g	**0.13** g	0.07 g	0.06 g
Body weight gain (g)	0.17 g	1.49 g	2.28 g	**3.99 g**	1.72 g	1.71 g
Body length gain (cm)	0.02 cm	0.10 cm	0.10 cm	**0.29** cm	0.03 cm	0.07 cm
DGR (%)	0.02 %	0.16 %	0.23 %	**0.33** %	0.17 %	0.15 %
PBWI (%)	1.62 %	10.02 %	13.49 %	**18.73** %	10.31 %	9.39 %
PLG (%)	0.45 %	2.72 %	2.84 %	**7.57** %	0.80 %	1.92 %
(SGR) (%/day)	0.80 %	0.92 %	0.95 %	**1.03** %	0.93 %	0.89 %
Survival (%)	56.25 %	71.88 %	78.13 %	**93.75** %	81.25 %	87.50 %

Figure 1. Body Length Gain (BLG) of *P. sphenops* fries and adults with different types of diets

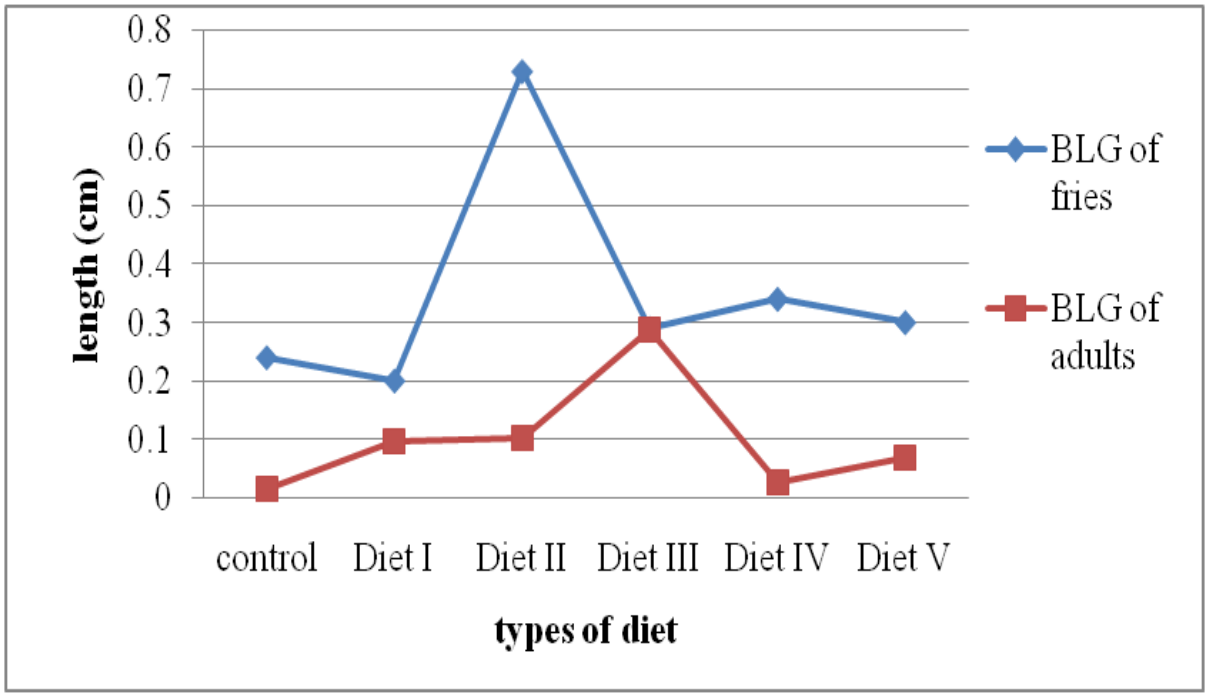
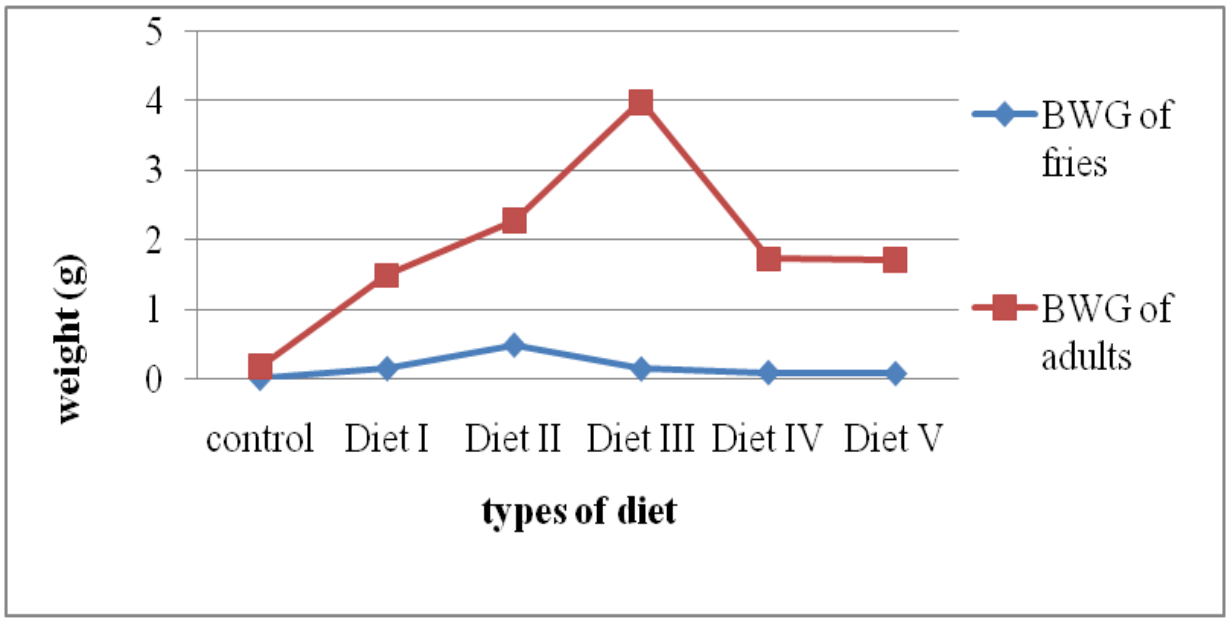


Figure 2. Body Weight gain (BWG) of *P. sphenops* fries and adults with different types of diets



Note : Diet I=0% Spirulina, Diet II=5% spirulina, Diet III=10% spirulina, Diet IV=15% spirulina and Diet V =20% spirulina in the figures

Figure 3. Specific growth rate (SGR) of *P. sphenops* fries and adults with different types of diets

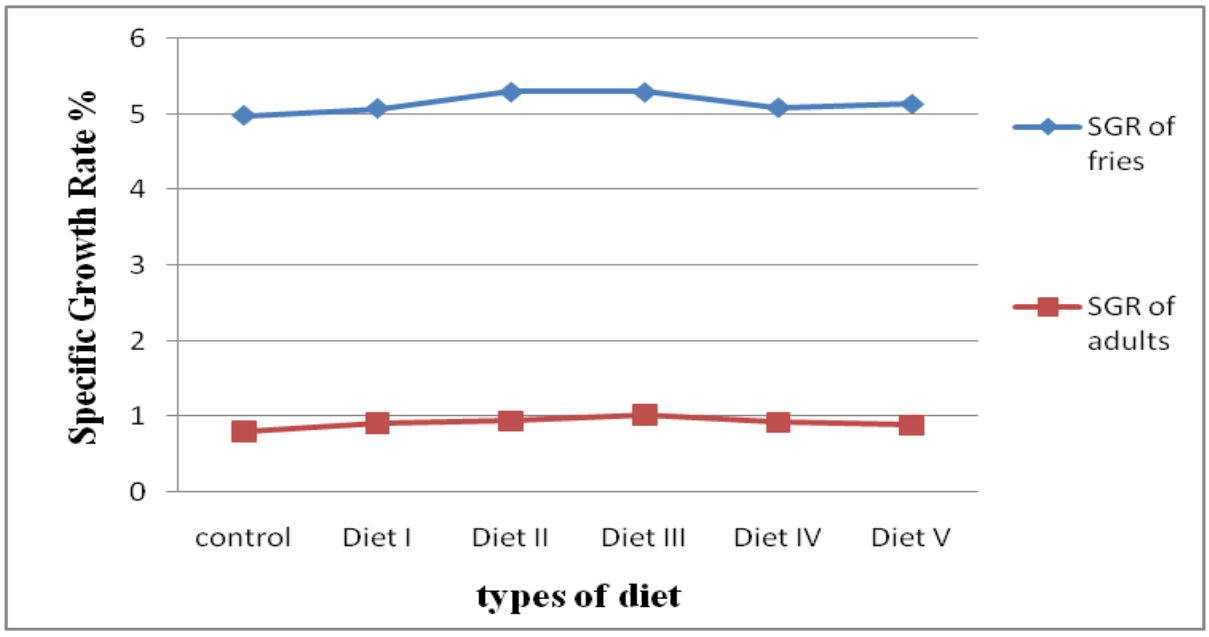
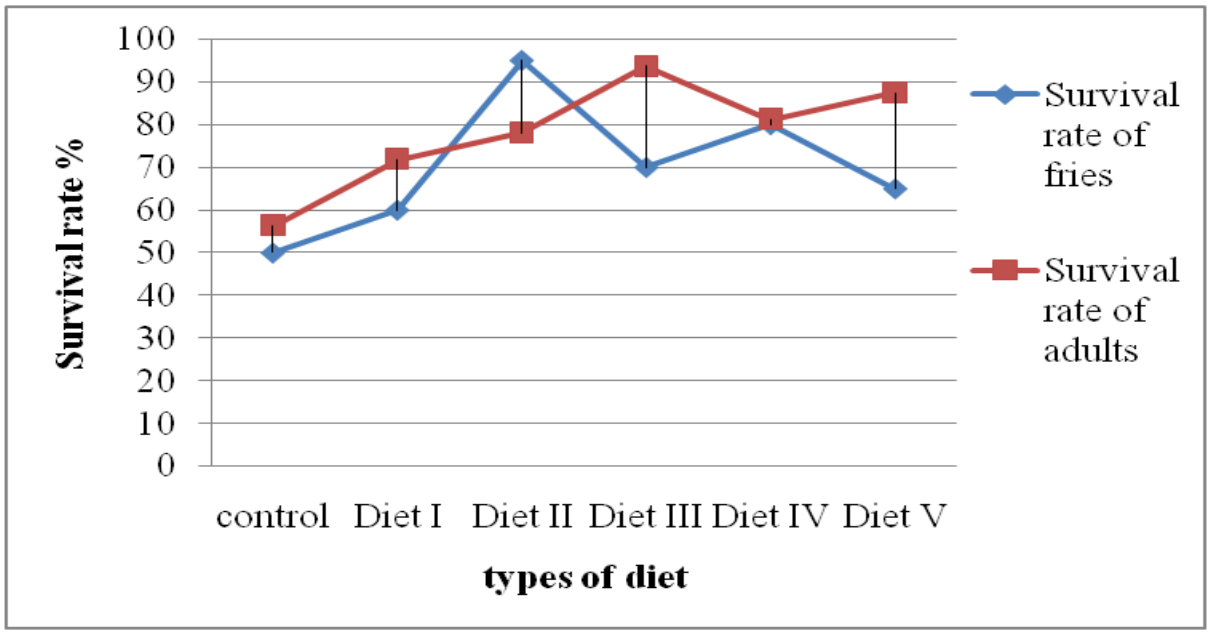


Figure 4. Survival rate of *P. sphenops* fries with different types of diets



Note : Diet I=0% Spirulina, Diet II=5% spirulina, Diet III=10% spirulina, Diet IV=15% spirulina and Diet V =20% spirulina in the figures.

4. Conclusions

In nature mollies are mainly plant and algae eater specie, the researcher tried to make diets according to fulfilling their nutritional requirements. The study showed that, among the formulated diets those which were prepared mainly by Spirulina, Fresh Water Fish (FWF), Marine Water Fish (MWF), corn flour (CF), Wheat flour (WF), Soyabean meal (SbM) etc; resulted better growth than the commercial pallet feed (table 1). Looking to the nutritional requirements at different stages, the 5% Spirulina additive diets had significantly higher growth in fries and 10% Spirulina additive diet had a significantly higher growth in adults. As Fish grow quickly during the early stages and during these stages they have different biological characteristics from adult. At early stages fish fries remained highly energetic, required high amount of protein and energy for their growth. Species at different stages of various species have different growth rates. Even the same species show different growth rates at different developmental stages. So, the diet with high protein from animal sources (table 1) made more preferable for molly fries (5% Spirulina) because of their high metabolic rate compared to adults (10% spirulina). In case of adults, the body weight and weight gain were found to be increased with increasing the inclusion level of Spirulina in the diets, although at much higher concentration of spirulina supplemented diets, there was a negative effects on growth observed. Spirulina has good source of protein and other nutrients. Thus, it was used as feed additive for ornamental fish molly. On the other side, the less cost involvement and quality assured formulated feeds can bring a lot of change in this sector. Thus, the experimental diets could be used in commercial molly fish farming by improving survival rate and its growth.

Acknowledgements

The authors are thankful to department of aquatic biology, faculty of science, Veer Narmad South Gujarat University Surat and also thankful to Mr. N. B. Patel, Principal, Adarsh nivasi kanya ST Shala Chikhli, Navsari for providing facilities to carry out this research work.

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