EFFECT OF DIFFERENT SPIRULINA SUPPLEMENT FORMULATED DIETS ON GROWTH AND SURVIVAL OF FRIES OF BLACK MOLLY (*Poecilia sphenops*)

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Abstract

In this study growth performance and survival rate of molly fish fries subjected to different feeding diets were evaluated. Different types of formulated diets comprised of some low cost and easily available ingredients; like, Spirulina Powder (SP), Fresh Water Fish (FWF), Marine Water Fish (MWF), Wheat flour (WF), corn flour (CF), Soyabean Meal (SbM) etc ingredients were used in the diets. The result was compared with applied commercial diet as control. After 40 days experimental period, diet 2 (5 % spirulina) showed the best result in case of length gain (0.73 cm) in molly fries whereas lowest (0.24 cm) was reported from control. The body weight gain in fries was also found higher (0.48 g) in diet 2 (5 % spirulina) than other diets. Specific growth rate (SGR) of all formulated diets was higher than the commercial diet. Maximum survival rate (95%) was recorded in formulated diet 2 (5% spirulina). Growth of black molly fries from all the supplied formulated diets resulted better rather than the commercial diets. So, Commercial diets might be replaced by experimental diets for the better production of black mollies.

***Keywords*:** Spirulina, black molly fries, formulated diet; growth; survival rate; SGR

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).

Molly fish is one of the most popular ornamental fish due to varieties of colour, species varieties, its high growth rate, birth size and brood number. Mollies appear to be a hardy and highly adaptable species. The name Molly was come from the genus *Mollienisia*, as it was originally classified in 1846, but now it is comes under family poecilidae, and thus it renamed as genus *Poecilia*. Black mollies (*Poecilia sphenops*) are one of the popular ornamental fishes produced in many Asian countries as live bearing species (Chong*et al.*, 2004). 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 fish fries to quality feed for better growth and survival. Black mollies can easily survive in small water tanks also and if they have good quality and balanced feed, they can definitely grow fast and survive longer. Therefore the present study was focused on production of quality feed at low cost to note the growth and survival of molly fish fries.

2. Materials and Methods

2.1. Fry collection, stocking and experimental unit design

Total 360 black molly (*P. sphenops*) fries were purchased from aquarium fish shops with special order, Navsari district Gujarat and acclimatized in a large glass aquarium in laboratory condition for three days. Then the fries were divided into six experimental groups including a control, each with three replicates. The divisions were designed on the basis of formulated feeding trial. The study was conducted with 20 black molly fries (almost uniform size) in each 20 L 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.

2.2. Feed preparation, composition and fish feeding

Six types of diets were provided to black molly fries in the present study. Diet 1, diet 2, diet 3, diet 4 and diet 5 were prepared using ingredients according to Table 1 whereas control was a commercially available pellet feed. No preservatives, synthetic colour and flavour materials were added. Feeds were made pellet through standard process and kept in fully air tight aquariam after sun drying. Feeding was carried out twice a day at 9:00am and 5:00pm throughout the experimental period. Fry were initially fed at a rate of 100% of their body weight however the remaining food was removed every day after half an hour of feeding time using cotton cloth net. Fish health was monitored regularly though eye observation and dead fish were taken away immediately without any replacement. Feed amount was reduced as per the dead fries count.

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	

2.3. Growth

Molly fries 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* fry 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 fish fry at the end of the experiment (g); Wo = Average fish fry 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 fish fry at the end of the experiment (g); Wo = Average fish fry 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 fish fry at the end of the experiment (cm); Lo = Average fish fry 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 fish fry at the end of the experiment (cm); Lo = Average fish fry 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 fry at the end of the experiment (g); ln Wo = Natural Logrithum value of average weight of fish fry 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 fish fry at the end of the experiment (g); Wo = Average fish fry weight at the beginning of the experiment (g) and Nt = final numbers of fry 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 fish fry at the end of the experiment (g); Wo = Average fish fry weight at the beginning of the experiment (g) and t = Rearing period (days)

❖ **Molly fry survival**:

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

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

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

2.4. 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.

2.5. 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 fry. 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

3.1 Growth of molly fry

The growth was analyzed and compared with control diet. Growth pattern, survival and weight gain (%) had significantly varied among all experimental diets (Table 2). The maximum body length gain (0.73 cm) was measured from fries fed with diet 2 (5% Spirulina); whereas the lowest (0.20 and 0.24 cm) was found in diet 1 (0% spirulina) and in control respectively (Figure 1). The highest body weight increase (0.0085 g) was found in diet 2 (5% Spirulina); whereas the lowest (0.0005 g) was found in control. As the stocking density remained same, the variations resulted for the ingredient composition of the diets. Highest (0.48 g) body weight gain was observed in diet 2, on the other hand, lowest weight gain (0.02 g) was investigated from the control and others were also lowest (figure 2). It was resulted the highest (20%) percent body weight increase (PBWI) while lowest (1.35 %) was reported from commercial diets. Specific growth rate (SGR) was recorded highest (5.30 %/day) in diet 2 followed by 5.29 %/day in diet 3, 5.13 %/day in diet 5 and 4.98 %/day in control (Figure 3).

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	1.13 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 %/day	5.07 %/day	5.30 %/day	5.29 %/day	5.08 %/day	5.13 %/day
Survival fish fries	50%	60%	95%	70%	80%	65%

In the present experiment, mollies fed with commercial feed had the survival rate of 50% which is similar with survival of both diet 1(60%) and diet 5 (65%). By contrast, the highest survival rate (95%) was reported from diet 2 than other diets 3 (70%) and diet 4 (80%). Mollies were recorded remain live from majority of Spirulina containing diet at the end of the 40 days experiment (Figure 6). The results indicated that mortality rates were higher in fishes feed with commercial feed rather than formulated diets.

Growth parameters data from different diets were positively and significantly correlated with each other throughout the experiment ($p<0.05$). It is resulted from the study that diet 2 evidenced the better growth rate in molly fries compared with the other prepared diets and control. Moreover, formulated diets reported the better growth pattern than that in commercial feed. No toxicity, abnormal behavior and disease prevalence was recorded and mortality rate was also lower during the study.

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

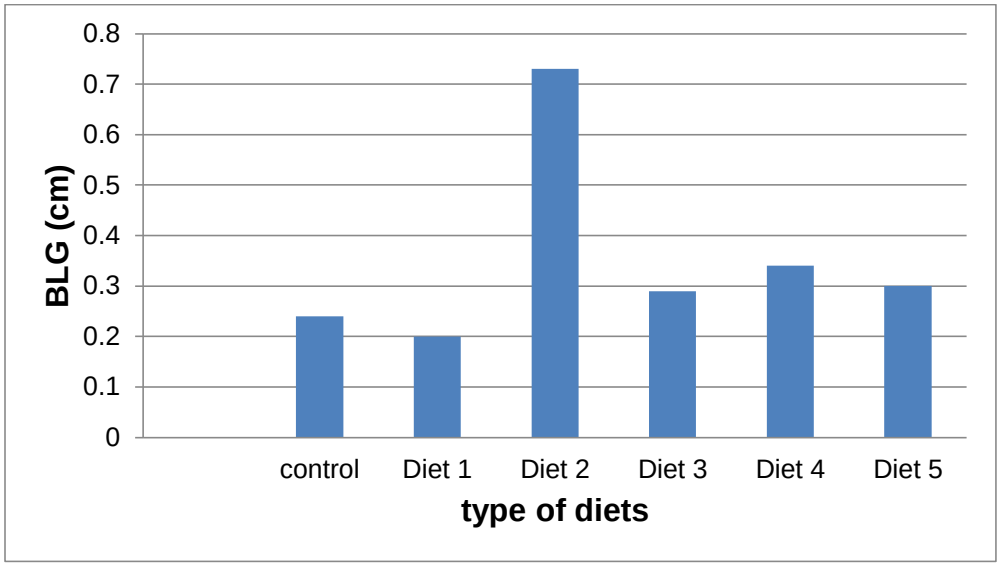
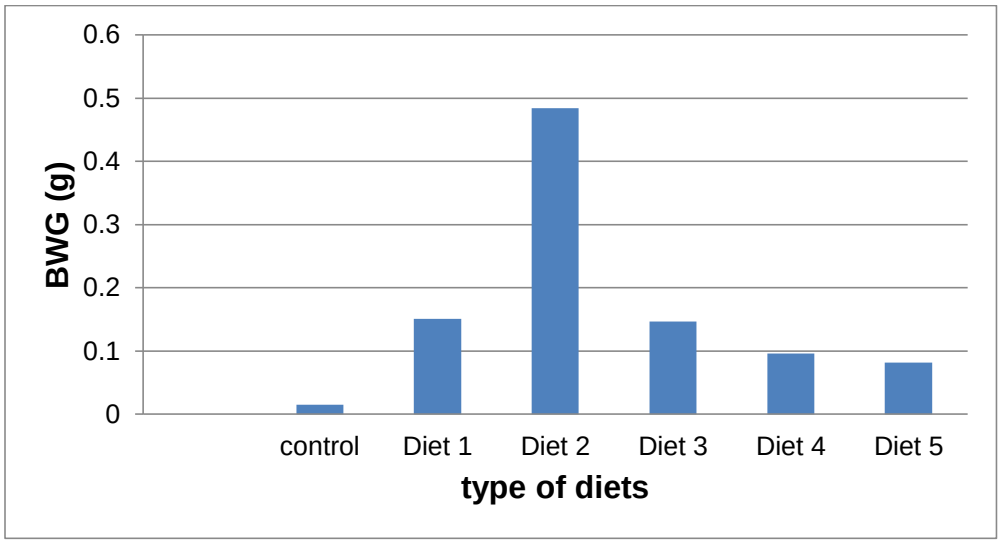


Figure 2. Body Weight gain (BWG) of *P. sphenops* fries with different types of diets



Note : Diet 1=0% Spirulina, Diet 2=5% spirulina, Diet 3=10% spirulina, Diet 4=15% spirulina and Diet 5 =20% spirulina in the figures

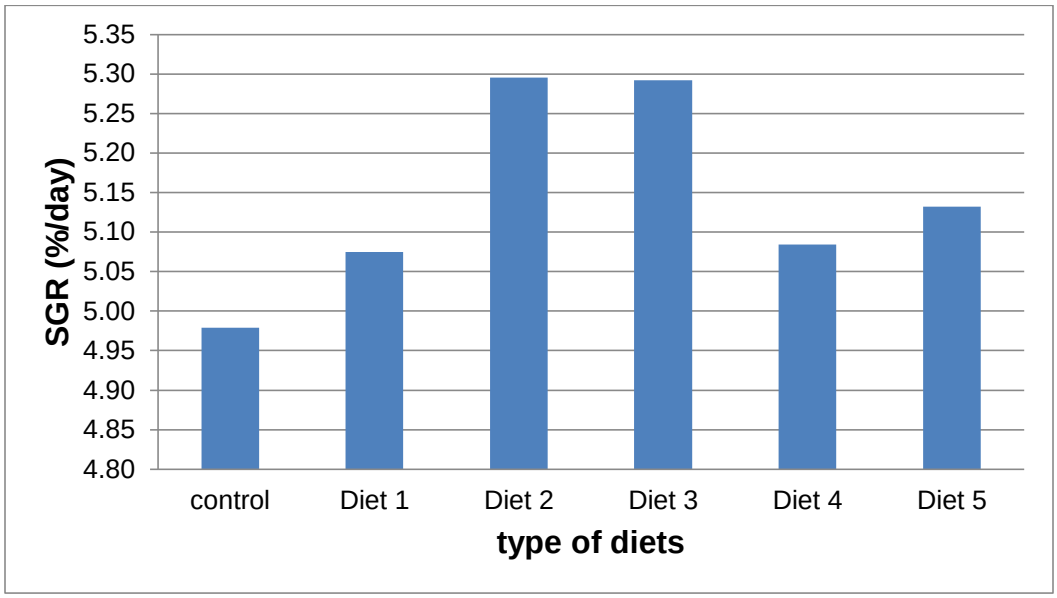


Figure 3. Specific growth rate (SGR) of *P. sphenops* fries with different types of diets

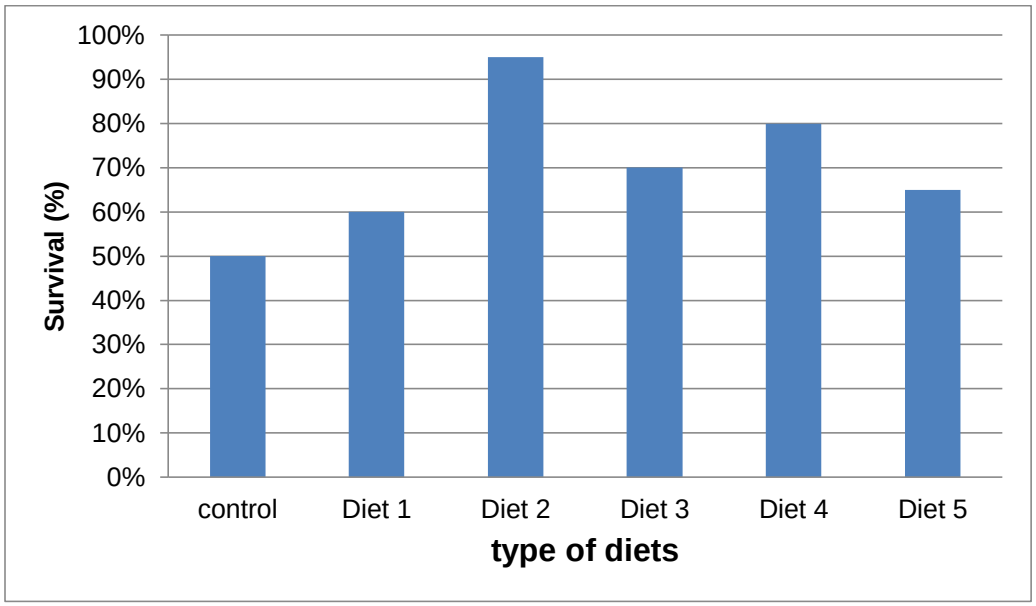


Figure 4. Survival rate of *P. sphenops* fries with different types of diets

Note : Diet 1=0% Spirulina, Diet 2=5% spirulina, Diet 3=10% spirulina, Diet 4=15% spirulina and Diet 5 =20% spirulina in the figures

Table 3. One Way ANOVA test result of weight gain of *P. sphenops* fries with different types of diets

Source of Variation	SS	df	MS	F	F crit
Between Groups	0.00197	5	0.000394	11.59596	2.250725
Within Groups	0.008357	246	3.4E-05	-	-
Total	0.010327	251	-	-	-

Growth parameters data from different diets were revealed that calculated F value was found greater than the F critical value (Table 3). Therefore, null hypothesis will be rejected and the experimental diets made significant effects on the growth of fish fry. Hence, It was resulted from the study that diet 2 (5% Spirulina) evidenced the better growth rate in molly fries compared with the other prepared diets and control. The poor growth from commercial feeds might be resulted because commercial feeds are not always prepared following recommended requirement of major nutrient components for specific species of fish and also not for specific life stage of fish. Among those, protein is considered as one of the crucial nutrients for the early stages of fish. Diets that used in the experiment, all diets with different quantity of ingredients but protein level maintained same for the study concern. So that it can be helpful for the investigator to find out the percentage of Spirulina as a protein source used as supplementary to molly fries. Moreover, formulated diets reported the better growth pattern than that in commercial feed. No toxicity, abnormal behavior and disease prevalence was recorded and mortality rate was also lower during the study.

4. Conclusion

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. Also the fish fries fed the 5 % diet had a significantly ($p<0.05$) higher growth. The higher cost involvement, poor growth, less survival and higher mortality rate of imported commercial feeds brought less success in molly fish farming as well as in ornamental aquaculture industry. 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.

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