EFFECTS OF DIFFERENT FORMULATED SPIRULINA FEEDS ON THE GROWTH OF BLACK MOLLY FISHES

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Abstract

For captive breeding and intensive culture of ornamental fish requires a good quality of diet and surrounding environment. Nutritionally low quality diet inhibits their growth and productivity. Therefore the study was conducted to produce a diet having optimal nutritionally ingredients and the aim of the study was to investigate the effects of dietary Spirulina supplementation on growth of the ornamental Black Molly Fish (*Poecilia sphenops*). The present experiments were performed to investigate the influence of different levels of Spirulina (0, 5, 10 15 and 20 %) on growth and fertility on the ornamental Black Molly (*Poecilia sphenops*). After 40 days experimental period, the body weight gain in mollies was found higher (0.13 g) in diet 3, whereas lowest (0.01g) was reported from control. Diet 3 (10% Spirulina) showed the best result in case of length gain (0.29 cm) than other diets in mollies. Specific growth rate (SGR) of diet 3 was also found higher (1.03 %) than all other formulated diets and commercial diet. Survival rate (SR) of adult fish in diet 3 was also found higher (93.75 %) than all other diets. Fish fed the diet containing spirulina were significantly ($p<0.05$) enhanced the growth compared to commercial diets. Hence, Commercial diets might be replaced by experimental diets for the better production of molly fish.

Keywords: Black molly fish, spirulina , formulated diet, SGR, Survival rate.

1. Introduction

Spirulina is one of the most concentrated natural sources of nutrition for all animals. Early interest in Spirulina focused mainly on its potential as a source of protein and vitamins. Dried spirulina contains 5% water, 24% carbohydrates, 8% fat, and about 60% (50–70%) protein (phang et al., 2000). Spirulina is the richest source of vitamins B-12 and beta carotene, and is loaded with essential fatty acids and minerals. Essential amino acids such as isoleucine, leucine, lysine, methionine, phenylalanine, threonine, tryptophane, and valine are present in Spirulina. Spirulina improves the intestinal flora in fish by breaking down indigestible feed components to extract more nutrients. Spirulina stimulates the production of enzymes that transport fats within the fish for growth instead of storage (Amer, 2016).

The cell wall of Spirulina is rich in muco-proteins that enhance the natural mucus layer of the skin, resulting in the shiny appearance of fins and skin and improving resistance to skin infections (James, et. al. 2009). Mosha (2019) review suggests that " 1-20% inclusion level of Spirulina can be used to replace fish meal in a diet for effective low feeding costs in both omnivorous and herbivorous fish species." Many factors influence the behaviour, growth and fertility in female fish. Among them size, age, endocrine control, physical condition, physiological factors, previous Pregnancy, environmental factors, stress and nutritional state of female are important. Various studies on spirulina as an alternative feed for animal and aquaculture have been done (Shivakiran, et. al., 2015). Several studies have been conducted in both finfish and shellfish using dried Spirulina powder as a supplement in diets with promising results. In finfish, Broun (1980) and Zeinhom, et. al. (2004) have been reported positive fish growth performance when fed diets containing algae cells. Dernekbasi (2010) also reported positive results in ornamental fish such as Guppy fish. Previous authors have studied the effect of a

Spirulina rich diet or its extract on growth and immune responses in various animals (Hayashi, et. al., 1994; Qureshi, et. al., 1995; Scaria, et. al., 2000). However, no such study has been especially carried out on the effects of Spirulina on growth and fertility in ornamental fishes like molly. The present study was carried out to analyze the impact of different levels of dietary Spirulina on growth of the ornamental Black Molly (*Poecilia sphenops*).

2. Materials and Methods

2.1 Fish and maintenance:

Two hundred (200) adult *Poecilia sphenops* were collected from fish aquarium shops. They were divided into six groups. They were named as control, diet I, diet II, diet III, diet IV and diet V group. Total thirty two fishes of each groups were reared in separate glass aquarium tanks (30 cm x 30 cm x 45 cm) containing 40 l of water capacity. The water was clean, unchlorinated well water. 20 % of water from each tank was changed everyday throughout the experiment period (Marshall, 2024)). Settled waste particular matters were removed from tank everyday two times using Siphon method. Tape water was used for the experiment. The tanks were completely drained and dried and also properly cleaned before started the experiment to maintain hygienic condition of the tanks throughout whole experiment.

2.2 Feed and feeding:

Six diets (including control) were used contain different Spirulina levels - 0%, 5%, 10%, 15%and 20%, and were provided to molly adult fish in the present study. Different diets, like Diet-1 to Diet-5 (0% Spirulina to 20% Spirulina) were prepared using ingredients according to Table 1 whereas control (Diet-C) was a commercially available pallet feed. Spirulina and fresh water fish meal were used as key ingredients in diet-1(0% Spirulina), diet-2 (5% Spirulina) and diet-3 (10% Spirulina) respectively. The quantity of marine water fish, wheat flour, corn flour and soya bean meal were kept constant to prepare 100 g diet. No preservative was added. Feeds were made pellet through standard process and kept in fully air tight container after sun drying for three days. Fishes were fed at 9:00am and 5:30pm, twice a day during the 40-days experiment. Fish health was monitored regularly though eye observation and dead fish were taken away immediately without any replacement. Ingredients quantity of prepared diets of experimental diets is given in Table 1. The main ingredients of fish feed contain protein, fat and mineral substance, and raw materials of these ingredients mainly include waste or cheap fish meal, soybean meal, etc and easy available flours like, wheat, rice, corn etc. and other ingredients. Fishes need a mix of different nutrients in a balanced way which makes sure their minimum nutritional needs are met.

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 Fishes 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 fish at the end of the experiment (g); Wo = Average fish 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 at the end of the experiment (g); Wo = Average fish 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 at the end of the experiment (cm); Lo = Average fish 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 at the end of the experiment (cm); Lo = Average fish 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 fish at the end of the experiment (g); Wo = Average fish weight at the beginning of the experiment (g) and Nt = final numbers of fish 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 at the end of the experiment (g); Wo = Average fish weight at the beginning of the experiment (g) and t = Rearing period (days)

- **Molly fish survival**:

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

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

Where; Nt and N0 were final and initial numbers of fish 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 fish. 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 fish

The growth was analyzed and compared with different diets. Growth pattern, survival and weight gain (%) had significantly varied among all experimental diets. The highest body length increase was found in diet 3 (10% spirulina) whereas the lowest was found in control (figure 1). As the stocking density remained same, the variations resulted for the ingredient composition of the diets. Diet 3 resulted the highest body weight gain while lowest was reported from commercial diets (Figure 2). The maximum percent length gain was measured from fish fed with diet 3. The poor growth from commercial feeds might be resulted because commercial feeds are not always prepared following recommended requirement of major nutrient components. Among those, protein is considered as one of the crucial nutrients for the fish. Altaff et al., 2015 also observed the same thing that generally, the commercial feeds contain a minimum of fishmeal and additional animal by-products. Specific growth rate (SGR) was recorded highest in diet 3 than other diets (figure 3). In the present experiment, mollies fed with commercial feed had the survival rate of 56.25% which is very less than any other formulated diets. The highest survival rate (93.75%) was reported from diet 3 at the end of the 40 days experiment (Figure 4). The results indicated that mortality rates were higher in fishes feed with commercial feed rather than formulated diets.

Table 2. Growth performance of *P. sphenops* fish 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)	3.51	3.51	3.51	**3.51**	3.51	3.51
Final length (cm)	3.52	3.60	3.61	**3.79**	3.53	3.58
Initial weight (g)	0.59	0.58	0.59	**0.58**	0.58	0.59
Final weight (g)	0.60	0.65	0.68	**0.71**	0.65	0.65
Weight Gain (g)	0.01	0.06	0.09	**0.13**	0.07	0.06
Length gain (cm)	0.02	0.10	0.10	**0.29**	0.03	0.07
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 Length gain of *P. sphenops* with different types of diets

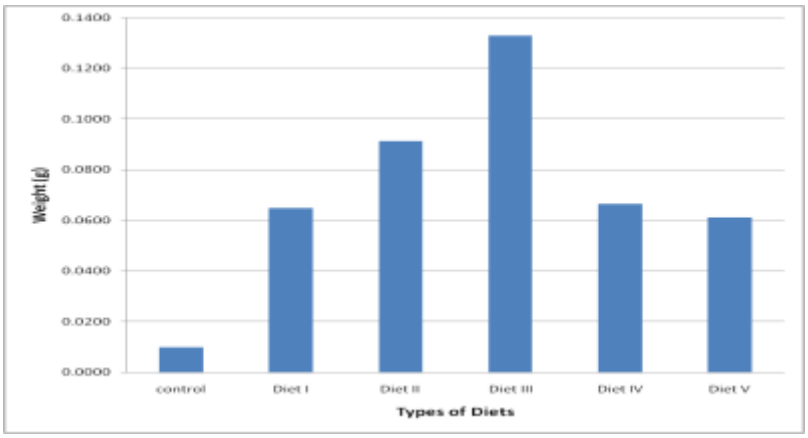


Figure 2 Weight gain of *P. sphenops* with different types of diets

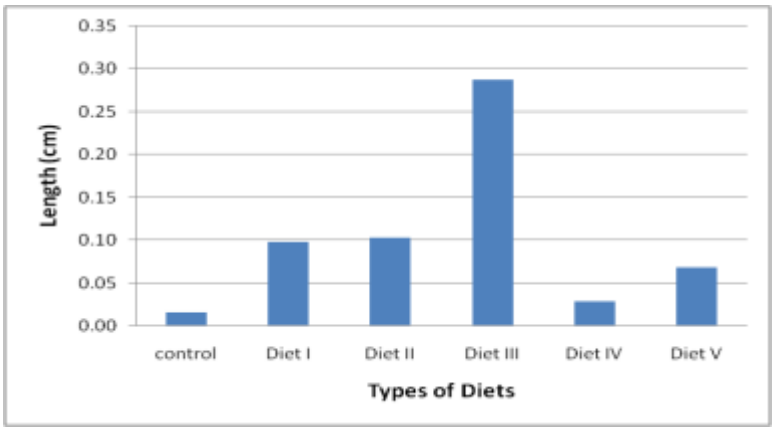


Figure 3. Specific growth rate of *P. sphenops* with different types of diets

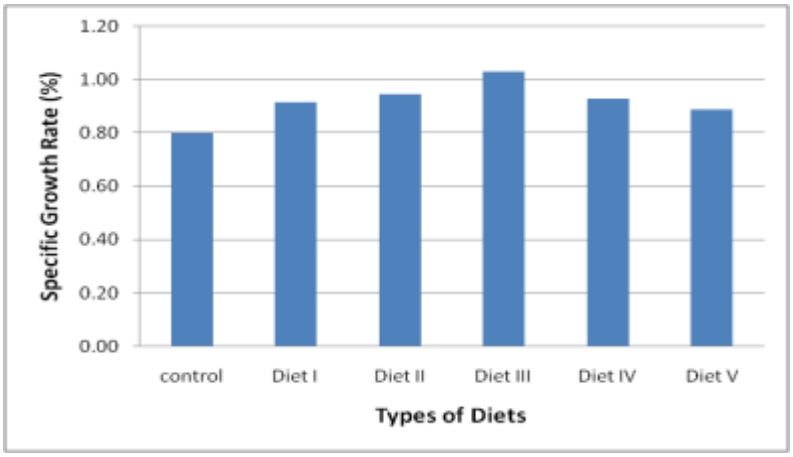
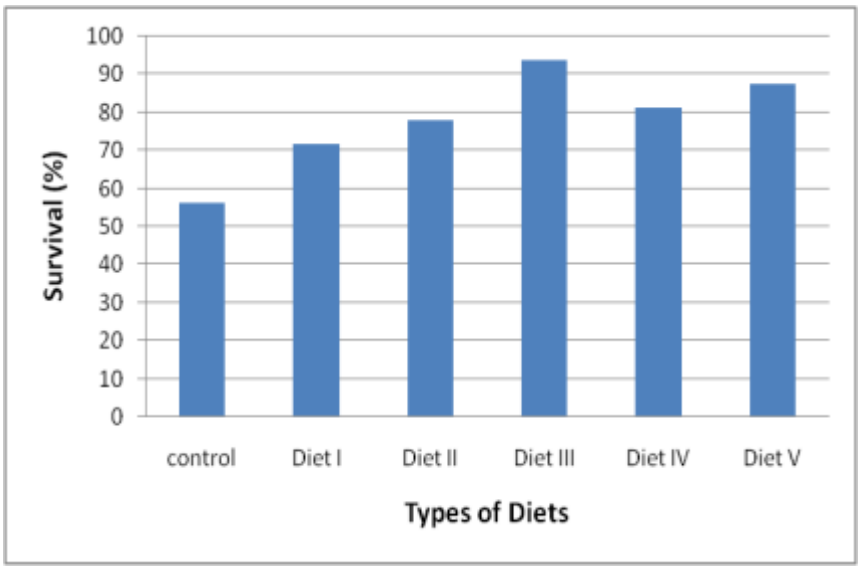


Figure 4. Survival rate of *P. sphenops* with different types of diets



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 1 to 4.

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

Source of Variation	SS	df	MS	F	P-value	F crit
Between Groups	1.324511	5	0.264902	4.277844	0.001173	2.277044
Within Groups	8.917089	144	0.061924	-	-	-
Total	10.2416	149	-	-	-	-

Table 4. One Way ANOVA test result of weight gain of *P. sphenops* with different types of diets

Source of Variation	SS	df	MS	F	P-value	F crit
Between Groups	0.154524	5	0.030905	3.041536	0.012186	2.277044
Within Groups	1.463169	144	0.010161	-	-	-
Total	1.617692	149	-	-	-	-

Growth parameters data from different diets were revealed that F critical value $<$ F value (Table 3 and 4). Also the p value, $p<0.05$. Therefore, null hypothesis will be rejected and the experimental diets made significant effects on the growth of fish. Hence, It resulted from the study that diet 3 (10% Spirulina) evidenced the better growth rate in molly 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.

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 fed the 10% 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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