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"Assessing The Efficacy Of *Spirulina* Algae-Based Feed For Goldfish (Carassius Auratus) And Black Molly (Poecilia Sphenops)''

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Article History	Abstract		
	Abstract: <i>Spirulina</i> sp a type of cyanobacteria characterized by its spiral shape. <i>Spirulina</i> is comprised of 60% vegetable protein, essential vitamins, and β -carotene. The dark green color of <i>Spirulina</i> is attributed to a combination of pigments, including carotenoids (orange), phycocyanin (blue), and chlorophyll (green). <i>Spirulina</i> sp. exclusively contains chlorophyll a, with levels ranging from 0.8% to 1.5% of its dry weight. Additionally, freeze-dried <i>Spirulina</i> sp. boasts a notable xanthophyll content, measuring at 6.9 g.kg-1. Other prominent carotenoids include β - carotene (28%), mycoxanthophyll (37%), and zeaxanthin (17%). Owing to its pigment composition, <i>Spirulina</i> sp. is employed for ornamental fish pigmentation, particularly in Goldfish and Black molly, as a feed additive. If Gold fish and Black Molly fails to achieve the desired coloration level, it can result in a substantial decrease in market demand and the value of the fish. Despite the availability of fish feeds enriched with carotenoids in the market, they tend to be costly. Therefore, our study aimed to explore the impact of <i>Spirulina</i> on both the pigmentation and growth of Goldfish and Black Molly.		
CC License	Keywords: Spirulina, Goldfish, Black Molly, Average body weight gain, Daily and Relative weight gain, Feed conversion ratio,		
CC-BY-NC-SA 4.0	Pigmentation and Survival rate.		

1. INTRODUCTION:

Spirulina, a type of blue-green algae, is characterized by its spiral-shaped structure and microscopic cells. Specifically, *Spirulina* is a filamentous and multicellular organism known for its anti-inflammatory properties and antioxidant effects. *Spirulina* is abundant in vitamins, particularly vitamin B12, minerals, protein, and carotenoids. *Spirulina*, a multicellular filamentous cyanobacterium thriving in alkaline conditions, is globally cultivated for its nutritional value as a health food. Additionally, it serves as a source of cyanophycin, a blue pigment utilized in cosmetics and food products. The skin pigmentation of ornamental fish is a critical quality

criterion that significantly influences market value and consumer acceptance. When kept in captivity, this material can complete a full diet, multi-ingredients were employed to manufacture fish feed. Feed components for ornamental fishes were *Spirulina*, GOC (Ground nut oil cake), rice bran, and wheat flour and jaggery.

2. METHODOLOGY:

2.1 Fish collection and maintenance

Ornamental fishes, goldfish and molly were brought from West Bengal. Fingerlings were brought to the lab in polythene bags filled with aerated water. All fingerlings were placed in aquarium tanks that had been thoroughly cleaned. The tank was filled with chlorine-free water and aerators were installed throughout the study period. The fishes were acclimatized in the laboratory for 15 days. Fishes were fed with the commercial feed. Healthy fishes were selected to investigate the effects of *Spirulina*. ornamental fish were tested for three months.

2.2 Arrangement of the Aquarium, Fish stocking Density, and Experimental Specimen:

To evaluate the effect of feed on ornamental fish, aquarium tanks of equal capacity were filled with water. The aquarium was covered with netting to prevent fish from escaping and insects from entering.

Each aquarium housed four fish. The experimental fish chosen for the study were the Common Goldfish and Black Molly. Before introducing the experimental feed, the fish were acclimatized by feeding them regular feed for one week.

2.3 Feed Application and Experimental Setup Maintenance:

Fish were fed prepared pellets at 5% of their body weight twice daily, at 10 a.m. and 5 p.m., over a period of three months. Regular netting was performed in each aquarium to prevent plankton growth. Weekly water changes were conducted, maintaining a temperature of 25-30°C and pH levels between 7-8. Excreta removal was performed daily using siphoning.

2.4 Feed preparation:

Fish were fed prepared pellets at 5% of their body weight twice daily, at 10 a.m. and 5 p.m., over a period of three months. Regular netting was performed in each aquarium to prevent plankton growth. Weekly water changes were conducted, maintaining a temperature of 25-30°C and pH levels between 7-8. Excreta removal was performed daily using siphoning.

2.5 Proximate Analysis of Feed and Ingredient Composition:

Proximate composition analysis of feed ingredients and prepared feed was conducted using standardized methods: crude protein by the micro-Kjeldahl method (IS: 7219, 2015), lipid by the Folch *et al.*, (1957) method, sugar by the sampling and analysis method for sugar confectionery (IS: 6287, 2015), and ash and moisture by the gravimetric method (AOAC, 2000). Table 2 presents the list of ingredients along with their proximate composition values for both ingredients and the experimental feed.

Table No:2 Proximate analysis of fish feed					
Sample	Protein	Lipid	Sugar	Ash	Moisture
<i>Spirulina</i> pellets ornamental	35.64%	0.007%	6.85%	0.003%	-

2.6 Impact of Feed on Ornamental Fish: Observational Study

- Measurement of growth performance and Utilization Parameters.
- i. **Body weight gain (g/fish)** = Mean of weight (g) at the end of the experimental period weight (g) at the beginning of the experimental period (Schmalhusen, 1926)
- ii. Daily weight gain (DWG) = Gain/ experimental period (Schmalhusen, 1926)
- iii. **Relative weight gain (RWG %)** = Gain / initial weight x 100 (Brown, 1957)
- iv. Feed conversion ratio (FCR) = Feed intake / Body weight gain (Uten, 1978)

Table No:1 Proportion of ingredients in the experimental diets (Ornamental fishes)			
Spirulina 45 gm			
GOC	15 gm		
Rice bran	15 gm		
Wheat flour	15 gm		
Jaggery	10 gm		
Total	100 gm		

v. Survival rate (%) = Surviving fish number / Initial number of fish x 100

vi. Determination of carotenoid content: (Harpaz and Padowicz, 2007)

A sample of fish was taken and it was placed into a container, frozen at -20°C, and it was freeze-dried using a Rockwell jumbo freezer. After two days, the dried fish was cut down. Three ml of acetone was added to the fish sample. The sample was covered with dark paper, and it was kept overnight in a dark room, and then centrifuged in a Remi Centrifuge R-8C model centrifuge for 5 min at 6000 rpm. The overall variation in skin carotenoid levels was observed at different spectrophotometric wave lengths (WL), namely 450 nm, 475 nm, and 500 nm. The amount of carotenoids in the sample was calculated based on their dry weight. **Equation:**

4 \times Optical density value \times total volume of sample taken/ weight of sample (mg)

2.7 Statistical analysis:

Statistical analysis was conducted using Microsoft Excel. (Mean±S.E)

3. RESULTS AND DISCUSSION:

- **i.** According to the results, adding *Spirulina* to fish feed (Gold fish) significantly improved growth in terms of average body weight gain, daily weight gain, relative weight gain, survival rate and color.
- **ii.** According to the results, addition of *Spirulina* to fish feed (Black Molly) showed survival rate of 100% but all other parameters were found to be negative. The experimental fish remained free from disease throughout the duration of the experiment.

Table No:3 Gold fish					
No	Average Body Weight gain (g)	Daily Weight gain (g/fish/day)	Relative weight gain (g/day)	Feed conversion ratio	Survival rate (%)
GOLD FISH	1.412 ± 0.268	0.0938±0.018	11.797 ± 1.861	3.812 ± 1.035	100

Table No:4 Results of carotenoid				
Name	Carotenoid	Carotenoid	Carotenoid	
Tame	(450 nm)	(475 nm)	(500 nm)	
Gold fish	36.269 ± 0.105	41.040 ± 0.124	32.171 ± 0.094	

Figures:

Spirulina powder	Ground nut oil cake	Rice bran	Wheat flour	Jaggery

Figure 1. Feed components



Figure 2. Feed preparation



Figure 3. Before (left side) and after (right side) experiment of gold fish and black molly



Figure 4. Carotenoids extract of gold fish and black molly

4. CONCLUSION:

To know the effect of this *Spirulina* on fish, the study was conducted by making fish feed using *Spirulina*, which showed positive results of gold fish. Apart from the survival rate in molly fish, the rest of the results were found to be negative.

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