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Chudasma Niharika
 Department of Aquaculture,
 College of Fisheries Science,
 Kamdhenu University,
 Veraval, Gujarat, India

Joshi Nilesh
 Department of Aquaculture,
 College of Fisheries Science,
 Kamdhenu University,
 Veraval, Gujarat, India

Dabhi Rajkumar
 Department of Aquaculture,
 College of Fisheries Science,
 Kamdhenu University,
 Veraval, Gujarat, India

Tandel Dharmgna
 Department of Aquaculture,
 College of Fisheries Science,
 Kamdhenu University,
 Veraval, Gujarat, India

Effect of partial supplementation of *Chlorella* sp. on growth performance and survival of *Labeo catla* (Hamilton, 1822) fry

Chudasma Niharika, Joshi Nilesh, Dabhi Rajkumar and Tandel Dharmgna

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Abstract

This study was carried out to determine the effect of partial supplementation *Chlorella* sp. on growth and survival in *Labeo catla* fry as protein source in 60 days of feeding trial. A completely randomized experimental design was developed with 5 treatments and four replicates. *Chlorella* powder was added to the basal diet at a 0% (control) (T₀), 5% (T₁), 10% (T₂), 15% (T₃) and 20% (T₄) and fed to the *Labeo catla* fry. Sampling was carried out at every 15 days to monitor growth performance and to adjust rations. The water quality parameters *viz.* DO; alkalinity, pH and temperature were found in the permissible range during entire experimental period. At the end of feeding trial weight gain, specific growth rate, feed conversion ratio, protein efficiency ratio and proximate analysis of fish carcass were evaluated. Growth performance, FCR, SGR, and PER of fish affected by *Chlorella* sp. supplementation. Survival rate was not affected by different amount of *chlorella*. Treatment T₄, including 20% *Chlorella* in diet showed highest mean body weight, SGR, PER than those fed with other diets. FCR was lowest in treatment of *Chlorella* diet than control diets. In the proximate analyses of the fish showed highest protein in T₄ (16.92%), fat in T₄ (3.44%), ash in T₄ (4.39%) and moisture in T₀ (80.98%). results of the present study clearly showed that *Chlorella* sp. could be considered as a protein source for incorporation in *Labeo catla* fry diets to gain more weight.

Keywords: Aquaculture, *Labeo catla*, *Chlorella* sp., fish meal, partial feed supplementation

Introduction

Recently, aquaculture is one of the fastest growing in our world as it has a high level of protein, which is considered the lowest price in comparing with other sources of protein (Abd-El Alim *et al.*, 2018) ^[2]. The value of aquaculture goes beyond replacing capture fisheries as the main source of seafood; it is one of the most efficient ways of producing protein for human consumption (Lucas *et al.*, 2019) ^[17]. Fish is an essential dietary animal protein source in human nutrition. Production of sea-going species through freshwater fisheries and aquaculture for protein supply is being energized all through the world. As indicated by nutritionists, fish is an astounding substitute of protein for red meat (Akhil and Akhilesh, 2018) ^[4]. In India, 80% aquaculture production is composed of carps (Sivagami and Ronald, 2018) ^[31]. Freshwater aquaculture contributes over 89% of the total aquaculture production in the country and three Indian major carps (Arya *et al.*, 2019) ^[6]. The Indian major carps *Labeo catla*, *Labeo rohita* and *Cirrhinus mrigala* are the most important commercial fishes in India with a maximum market demand and acceptability as food by the consumers due to their taste and flesh (Kadhar *et al.*, 2012) ^[16]. Catla can be easily cultured in low input technology system due to its herbivorous feeding habit (Abarna, 2012) ^[1]. Its higher growth rate and compatibility with other major carps, specific surface feeding habit and consumer preference have increased its popularity in carp polyculture systems and it contributes a major share to the freshwater aquaculture production for the fish farmers in India (Mohanty *et al.*, 2019) ^[20].

Fish meal has been the protein source of choice in aqua feeds for many reasons, including its high protein content, excellent amino acid profile, high nutrient digestibility, generally lack in anti-nutrients, relative low price and its wide availability (Gatlin *et al.*, 2007) ^[13]. Sustainability of fish meal production from wild fish is questionable.

Corresponding Author:
Chudasma Niharika
 Department of Aquaculture,
 College of Fisheries Science,
 Kamdhenu University,
 Veraval, Gujarat, India

To reach a sustainable aquaculture, new alternative protein sources including cheaper plant or animal origin proteins are needed to be introduced for stable aqua feed production (Islam *et al.*, 2018) [14]. Among the potential substitutes, plant ingredients appear to be the best candidates. Many plant protein sources can be used to partially or almost totally replace dietary fish meal (Santigosa *et al.*, 2008) [27]. Plant ingredients have ANFs, are deficient in certain EAA, have less nutrient digestibility, have lesser nutrient bioavailability, and less palatability due to high levels of non-soluble carbohydrates such as fiber and starch (Daniel, 2018) [12].

In recent years, microalgae have emerged as a natural alternative nutritional source for novel bioactive compounds (Christaki *et al.*, 2011) [11]. *Chlorella* is commonly used as a dietary supplement (Abid *et al.*, 2018) [3]. *Chlorella* species are single-cell, spherical shaped (~2 to 10 μm in diameter) and photoautotrophic green microalga with no flagella (Mobin and Alam, 2017) [19]. *Chlorella* consists of green photosynthetic pigments chlorophyll-a and chlorophyll-b courtesy to which it is the richest source of chlorophyll availability. *Chlorella* consists of 55-60% protein, 1-4% chlorophyll, 9-18% dietary fiber, vitamins and minerals (Rani *et al.*, 2018) [26]. Owing to the high nutritional value, *Chlorella* has been used as dietary protein sources for marine and freshwater fish to improve weight gain and carcass quality (Shi *et al.*, 2017) [30]. Previous work showed inclusion of *Chlorella* in fish feed resulted in improved growth, survival and health status. Positive effect of *Chlorella* on feed utilization, stress and disease resistance of cultured fish has also been documented in earlier works (Rahimnejad *et al.*, 2017) [25]. Looking to the above aspects, the present study was undertaken to evaluate the effect of *Chlorella* as feed supplement on growth performance, survival, feed utilization and body composition of Catla (*Labeo catla*) fry.

Materials and Methods

Experimental setup

The experiment was conducted at the Wet Laboratory of Department of Aquaculture, College of Fisheries Science, JAU, Veraval, over a period of 60 days.

Collection of feed ingredient

The required ingredients for the preparation of the experimental diet such as fish meal (FM) and groundnut oil cake (GNOC) as a protein source, wheat flour (WF), fish oil, sunflower oil, tapioca powder (TP) and vitamin & mineral (V&M) for feed formulation were brought from the local market. *Chlorella* powder (CP) was brought from Oceanic Seaweed Private Limited, Rajkot.

Proximate analysis of Ingredients

Proximate composition of ingredients was determined by the standard methods (AOAC, 1995) [5]. Moisture content was obtained by heating the samples to 105°C for 30 minutes and then drying at 65 °C till a constant weight was obtained (Boyd, 1981) [9]. Crude protein values were obtained by the semi-automatic Micro-Kjeldahl digestion and distillation apparatus (Gerhardt, Germany). Crude fat was estimated with the Soxhlet apparatus. The total ash content was determined with the help of a muffle furnace (Table 1.)

Formulation and preparation of feed

The experimental diet was formulated with 30% protein level using locally available ingredients. The feed was formulated by Pearson's square method. The required quantity of ingredients was weighed accurately as per feed formula as shown in Table 1. In five experimental diets, the microalgae *Chlorella* sp. was added at the rate of 5%, 10%, 15% and 20% ingredients of control diet. Then prepared dough and then mixture of feed was thermally processed at 121 °C and 15 lbs pressure for 10-15 minutes. After steam cooking of feed mixture, vitamin and mineral mixture was mixed. The feed mixture was pelletized by using hand pelletizer. The pellets were spread on a plastic sheet, exposed to sunlight. Then feed pellets were packed in marked plastic jars

Treatments details

The experimental treatments are selected as follows:

T_0 = Diet prepared with 0% inclusion of *Chlorella* sp. (Control)

T_1 = Diet prepared with 5% inclusion of *Chlorella* sp.

T_2 = Diet prepared with 10% inclusion of *Chlorella* sp.

T_3 = Diet prepared with 15% inclusion of *Chlorella* sp.

T_4 = Diet prepared with 20% inclusion of *Chlorella* sp.

Experimental Design

Fry of *Labeo catla* (catla) were brought from Inland Fisheries Research Station, Junagadh Agricultural University, Junagadh (Gujarat) and transported in polythene bags by road to Veraval. The fish were brought to Aquaculture Wet Laboratory at College of Fisheries Science, Veraval and were allowed to remain in the plastic tank (500 L) with continuous aeration and feeding for 10 days. Fry with a total weight of 6.50 ± 0.108 to 6.80 ± 0.041 g was selected for the experiment. The experiment utilized 20 disinfected rectangular plastic aquaria (size 2x1x1 feet) filled with 30 liters of chlorine-free filtered freshwater. Each experimental treatment had three replicates which are arranged using completely randomized design (CRD), design. Each tank was stocked with 10 fishes with continuous aeration. Fry were fed twice daily at 10% of their body weight, and tanks were cleaned daily to remove excess feed pellets and fecal matter. Sampling occurred fortnightly to assess fish body weight and adjust feed quantities. Physiochemical parameters (temperature, pH, alkalinity and dissolved oxygen) were monitored weekly throughout the experimental period.

Growth Indices

At 15 days' interval sampling was performed to assess the growth performance of the fish. weight was measured using electric weighing balance respectively. Growth indices were evaluated using the below described formulae.

Mean Weight gain = Final weight-Initial Weight

Specific growth rate (SGR)

$\text{SGR} (\%) = \frac{\ln \text{Final Weight} - \ln \text{Initial Weight}}{100/\text{Number of Days Feed}}$

Survival

Survival (%) = total no. of live animal x 100/total no. of initial animal

Feed conversion ratio (FCR)

FCR = Feed given (dry weight)/Body weight gain (wet weight)

Protein efficiency ratio (PER)

PER (%) = Weight gain/protein intake

The calculated value indicates the average increase (%) of body weight per day

Biochemical Composition of Fish Carcass

Proximate composition of fish was estimated just after completion of the experiment. Whole meat of the fish was taken for proximate analysis. The fish meat was minced properly. The samples were analyzed for crude protein, crude fat, ash and moisture employing standard methods as explained earlier.

Water Physico-Chemical Parameters

Water quality parameters such as temperature, pH, dissolved oxygen, and total alkalinity were measured on weekly basis throughout the experimental period. The Temperature was measured by using thermometer. The pH was measured by using pH meter. Dissolved oxygen was measured using wrinkle's method. Total alkalinity was measured using HiMedia alkalinity testing kit.

Statistical Analysis

The results of partial replacement of fish meal with *Chlorella* sp. on the growth performance and survival parameters were statistically analyzed using completely randomized design (CRD) and one-way analysis (ANOVA) using IBM® SPSS® version 26.00 software and significant differences ($p<0.05$) among treatment means were compared using Duncan multiple range test (DMRT) (Snedecor and Cochran, 2014) [33].

Results**Physico-Chemical Water Parameters**

The water quality parameter recorded during experimental period shown in table 3. Water temperature ranged from 21.4 °C to 24.9 °C during the experimental period. The pH ranged from 7.5 to 7.7 during the experimental period. The data for dissolved oxygen ranged from 6.46 to 7.01 ppm during the experimental period. The alkalinity of water was ranged from 156 to 185 ppm during the experiment. All the physico-chemical parameters observed were within the optimum range required for the culture of the fish.

Growth Performance

This experiment was carried out to find the effect of *Chlorella* sp. supplemented diet on growth and survival of *L. catla* fry for 60 days of experimental period. The results of growth parameter include mean weight gain, and specific growth rate (SGR) shown in the in table 4. The initial weight of *L. catla* fry across treatments (T₀, T₁, T₂, T₃, and T₄) remained similar, ranging from 6.50±0.108 gm to 6.80±0.041 gm. The final average wet weight recorded were 20.52±0.75g, 20.58±0.085g, 22.90±0.238g, 23.88±0.165g and 33.50±1.132g in treatment T₀ (Control), T₁, T₂, T₃ and T₄ respectively. There was significant difference in growth observed in treatments and time. At the end of the experimental period (60 days), significant differences ($p<0.05$) were found between the treatment for wet weight

of fish. Final body weight was recorded in T₄ (33.50±1.132) treatment which was significantly higher ($p<0.05$) than the other treatments, followed by T₃ (23.88±0.165 gm), T₂ (22.90±0.238 gm), and T₁ (20.58±0.085 gm), while T₀ (20.53±0.075 gm) had the lowest final body weight. There was no significance difference between T₂ and T₃ treatments, and also in T₁ and T₀. But all treatment showed higher weight gain, except T₀ (control) treatment. Similarly, the total mean weight gain also different among treatments which T₄ showed highest (26.70±1.093) value, which was significantly ($p<0.05$) different from the control (T₀) and other treatments. No significance ($p>0.05$) difference among Treatment T₂ (16.30±0.158) and T₃ (17.38±0.131). While treatment T₀ (control) and T₁ (13.98±0.103) were at par with each other. Highest SGR was found in T₄ diet treatment (2.66±0.046) followed by T₃ (2.17±0.022), T₂ (2.07±0.010), T₀ (control) (1.90±0.018) and T₁ (1.87±0.016). Significant difference ($p<0.05$) was observed among the treatment diets; however, treatment T₀ and T₁ was at par with each other and T₂ and T₃ also at par with each other. However, T₄ was found to be higher as compared to the other treatments.

Feed Utilization

This experiment was carried out to find the effect of *Chlorella* sp. supplemented diet on feed utilization and body composition of *L. catla* fry for 60 days of experimental period. The results of feed conversion ratio (FCR) and protein efficiency ratio (PER) obtained in the present experiment is given in table 4. Feed conversion ratio was significantly higher ($p<0.05$) in T₀ diet (2.97±0.023) followed by T₁ (2.71±0.022), T₂ (2.60±0.020), T₃ (2.43±0.028) and lowest in T₄ (2.09±0.032) treatments respectively. There was significant difference observed between all the treatment ($p<0.05$). The results on protein efficiency ratio (PER) of *L. catla* fry of all the treatments were significantly differing with each other ($p<0.05$). The highest PER was found in T₄ (1.60±0.025) followed by T₃ (1.37±0.016), T₂ (1.28±0.010), T₁ (1.23±0.010) and the lowest in T₀ (control) (1.14±0.009). Statistical analysis showed significant differences ($p<0.05$) among all the treatments.

Survival Rate

Survival rate of *L. catla* fry in the respective treatments are presented in Table 4. survival rate was observed 100 % among all the treatment (T₀, T₁, T₂, T₃ and T₄) at 60 days of experimental period. There was no mortality indicate in this experiment throughout the culture period. This finding suggests that all dietary treatments supported optimal health conditions for the *L. catla* fry. Statistical analysis showed there was no significant differences ($p>0.05$) found between all the treatments.

Proximate Analysis of Fish Carcass

At the end of the feeding period, five fish from each tank were collected randomly for proximate analysis of fish carcass. The data for proximate analysis of fish whole body after the experiment (dry weight basis) are given in Table 5. The protein content was significantly higher (16.92 %) in T₄ compared to other treatments. The fat content was significantly highest (3.44 %) as compared to control with maximum in T₄ followed by T₁, T₀, T₃ and T₂. The ash content was significantly highest in T₄ (4.39 %) and the

moisture content was found highest (80.98 %) in T₀ as compared to other treatments and lowest found in T4 (75.25%).

Discussion

In the present study, the different percentages of *Chlorella* were added in the basal diet of *L. catla* and growth performance, survival and body composition were examined. The results indicated that *Chlorella* can be an alternate choice for fish diets. Based on the obtained data at the end of the 60th day experiment, it was observed that the final wet weight gain and SGR were significantly higher in T₄, while lower in T₀ and T₁ treatments. Significantly higher growth reported in fish fed with the diet containing 20% *Chlorella* (T₄), as compared to all the other experimental diets. The similar results were also observed by Xu *et al.* (2014) ^[34], they observed a significant increase in growth parameter when fed *Chlorella* incorporated diets at inclusion levels of up to 1.2% in Gibel carps *Carassius auratus gibelio* (Bloch). Sergejevova and Masojidek, (2013) ^[28] were also observed Positive effects on growth and skin coloration in freshwater sterlet, *Acipenser ruthenus L.* at 2.5% *Chlorella* inclusion. Similarly, Khani *et al.* (2017) ^[17] reported that dietary supplementation of 5% *Chlorella* powder significantly enhanced growth (98.06±0.73g), then control diets (57.8±0.48) in koi fish culture up to 60 days. The similar growth performance in *Catla catla* fish fed *Chlorella* diets may be credit to growth promoters, such as adequate amounts of macronutrients and naturally occurring bioactive ingredients (*Chlorella* growth factor (CGF)) found in *Chlorella vulgaris* (Badwy *et al.* 2008) ^[7]. The growth-promoting effect of dietary *Chlorella* in fish has also been reported in other aquatic species such as shrimp or prawn. According to Maliwat *et al.* (2017) ^[18] 4% *Chlorella vulgaris* diet allowed the highest growth performance when compared to the control diets and final body weight increased in *Macrobrachium rosenbergii* post larvae. Radhakrishnan *et al.* (2015) ^[24] reported that, the growth and level of digestive enzymatic activities of *M. rosenbergii* fed *C. vulgaris* meal up to 50% level increased significantly. In current experiment indicate that inclusion of microalgae can positively influence growth in *Catla catla* fry. The observed improvements in growth and feed utilization associated with *Chlorella* powder imply that microalgae contain a range of substances acting as dietary protein, minerals, vitamins, fiber, feeding stimulants, antioxidants, and other unidentified growth-promoting compounds. These bioactive constituents contribute to the widespread use of *Chlorella* as a feed supplement that promotes enhanced fish growth and feed intake (Mustafa and Nakagawa, 1995) ^[22]. It was also observed that initially the effect of *Chlorella* diet was not much significant in term of weight gain, but at as culture days' increase, the significant effect of *Chlorella* diets can be observed. However, the inclusion of *Chlorella* in this experiment is up to 20%, it can be assumed that if more percentage of *Chlorella* in diets were included, the growth may increase.

In the present study the lowest FCR was recorded in T₄ and highest in T₀. The highest PER was found in T₄ and lowest in T₀. With respect to FCR and PER, all the treatments were

significantly different with each other ($p<0.05$). Similarly, Zeinhom (2004) ^[35] found that, inclusion of algae in fish diets significantly improved the FCR, PER and PPV, whereas feed intake was significantly increased. In addition, Badwy *et al.* (2008) ^[7] reported that the inclusion of *Scenedesmus* sp. and *Chlorella* sp. up to 50% in diets could increase FI and PER, and reduced FCR in Nile tilapia. Similarly, Abid and Koyun (2018) ^[3], recorded that Food Conversion Ratio in treatment with 5 g (*Chlorella*/kg diet) and treatment with 7.5 g (*Chlorella*/kg diet) were significantly lower than control diet, protein Efficiency ratio was also significantly higher in the same treatment than control. Increased protein efficiency ratio in fish fed chlorella containing diets in this study indicated that inclusion of chlorella improved protein utilization of *labeo catla* mainly due to abundant amino acids and nucleic acid which have been reported to increase the growth of fish (Cai *et al.*, 2017) ^[10].

In this experiment, 100% survival was recorded throughout the experimental period. Similarly, Rahimnejad *et al.* (2017) ^[25] studied that inclusion of 15% *chlorella* meal fed to Olive Flounder, *Paralichthys olivaceus* gave 100% survival. Moosavi and Montajami (2013) ^[21] also reported 100% survival of angel fish (*Ptryphyllo scalare*) fed *Spirulina plantesis*. The higher survival rate may be attributed to the presence of carotenoids in chlorella algae, which enhance fish health and increase resistance to infections by reducing stress and strengthening immune function. In addition, stable water quality parameters, proper feeding management, and the absence of pathogenic challenges throughout the 60-day experimental period likely minimized stress and mortality, resulting in complete survival even without algal supplementation. These excellent survival rates indicated that the experimental diets supplied sufficient nutrition and had no harmful effects.

At the end of the 60th days of feeding period, whole body protein, fat and ash content were enhanced; however, moisture content decreased along with chlorella supplementation. The protein, fat and ash content was significantly highest in T₄ compared to other treatments. The moisture content was found highest in T0 as compared to other treatments. The moisture content of freshwater fish is reported to be between 72.1 and 83.6%, which supports the findings of the present study (Paul *et al.* 2018) ^[23]. The treatment T₄ exhibited higher protein (40 % more than control), which shows, *Chlorella* has higher acceptability in *catla* fry. Sivakumar *et al.* (2011) ^[32] reported that three live micro algal strains (*Chlorella* sp., *Tetraselmis* sp., and *Isochrysis* sp) and two cyanobacterial (*Synechococcus* sp. and *Phormidium* sp) strains fed *P. monodon* had significant improvement in body composition of protein, lipid and ash etc. In addition, similar results were also obtained by Bai *et al.* (2001) ^[8]; who recorded that increase in protein and fat content in Korean rockfish fed with diet inclusion of 2% and 4% *chlorella* powder as compared to control diets. Physico-chemical parameters of water were found inside the acceptable range in the present study. Temperature, pH and dissolved oxygen and alkalinity were found within the favorable range to enhance the growth of *L. catla*.

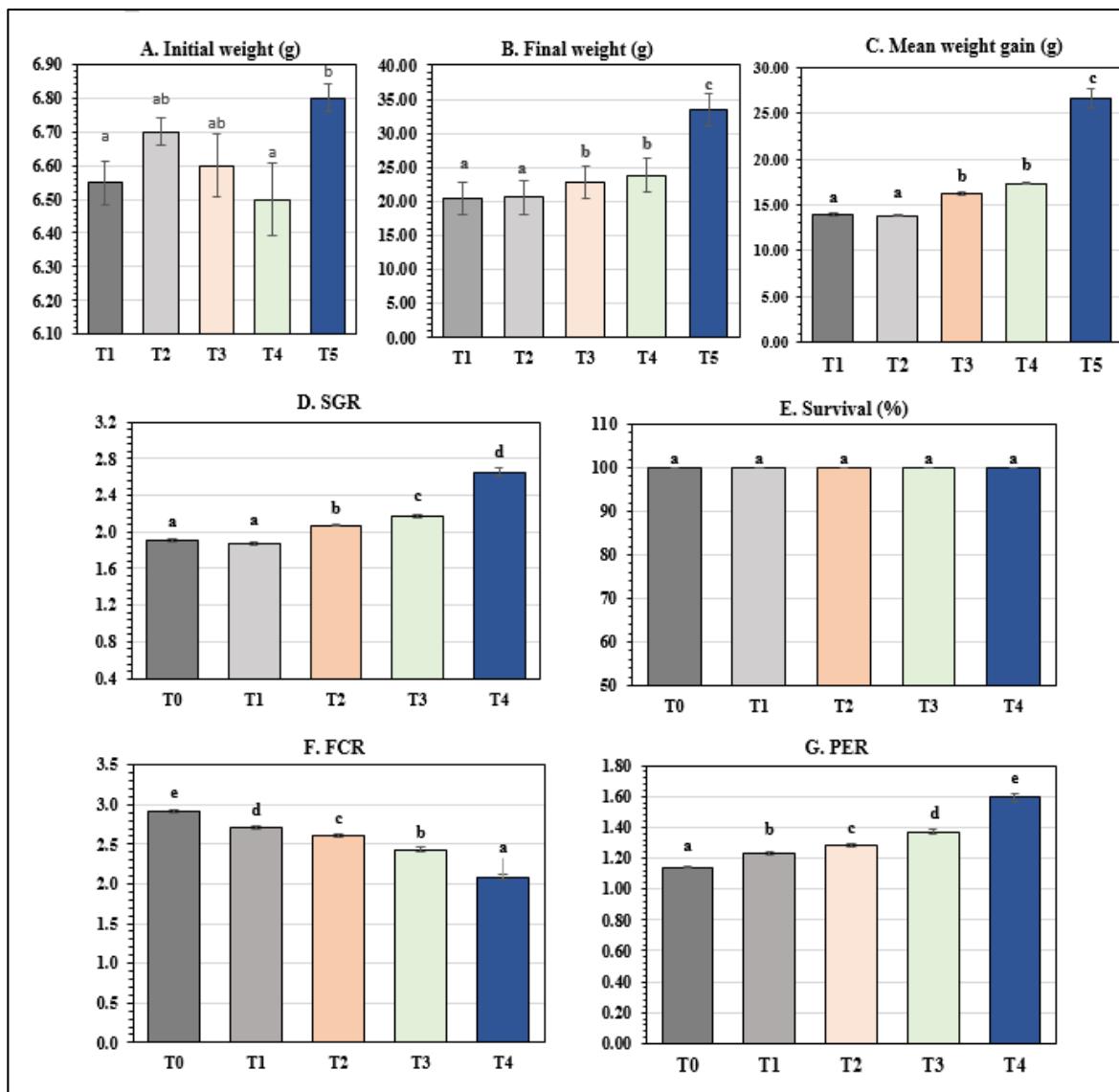


Fig 1: A-F. (A) initial weight (g) (B) Final weight (g) (C) Mean weight gain (g) (D) Specific growth rate (SGR), (E) Survival rate (%), (F) Feed conversion ratio (FCR) and (G) Protein efficiency ratio (PER) of *Labeo catla* recorded in different treatments during the culture period.

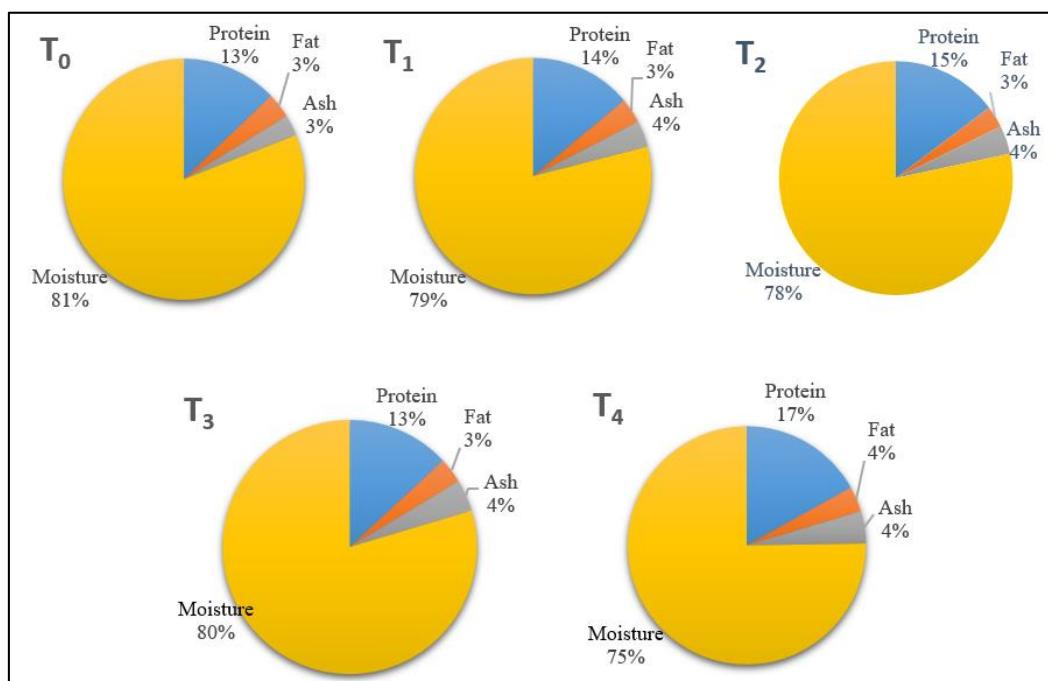


Fig 2: Proximate composition of fish body carcass of *Labeo catla* recorded in different treatments at the end of experiment.

Table 1: Proximate composition of feed ingredients (%)

Ingredients	Crude Protein (%)	Crude Fat (%)	Ash (%)	Moisture (%)
Fish meal	52.50	6.50	8.85	8.30
GNOC	38.15	8.07	8.78	5.90
Wheat flour	8.70	1.99	1.70	11.57
Tapioca	0.19	0.10	0.06	10.6
Chlorella powder	34.12	3.28	31.24	18.20

Note: Data expressed as Mean \pm SE (n = 3)

Table 2: Composition of experimental diets (%)

Ingredients (%)	Diets (30 % protein)				
	T ₀ (Control)	T ₂ (5%)	T ₃ (10%)	T ₄ (15%)	T ₅ (20%)
Sterilized fishmeal	42.00	40.00	38.00	36.00	34.00
GNOC	37.00	37.00	37.00	37.00	37.00
Chlorella sp.	0.00	5.00	10.00	15.00	20.00
Wheat Flour	7.50	7.50	7.50	7.50	7.50
Tapioca Powder	7.50	7.00	6.50	6.00	5.50
Fish Oil	4.00	4.00	4.00	4.00	4.00
Vitamin & Minerals	2.00	2.00	2.00	2.00	2.00
Total	100	100	100	100	100
Proximate analysis of experimental diet					
Nutrients	T ₀ (Control)	T ₁ (5%)	T ₂ (10%)	T ₃ (15%)	T ₄ (20%)
Crude Protein	30.10	30.15	29.85	29.89	30.12
Crude Fat	7.85	7.24	7.18	7.55	6.98
Moisture	6.81	7.72	8.56	8.30	8.21
Ash	15.71	15.78	16.15	16.54	16.88

Note: Data for proximate analysis is expressed as Mean \pm SE (n = 3)

Table 3: Range of Physico-chemical parameters of the water during the experimental period

Weeks	Temperature (°C)	pH	DO (ppm)	Alkalinity (ppm)
1 st	21.4-21.6	7.5-7.6	6.99-7.00	163-178
2 nd	21.5-21.8	7.6-7.7	6.83-6.85	173-179
3 rd	22.3-22.5	7.4-7.5	6.73-6.83	180-184
4 th	22.7-22.9	7.5-7.6	6.52-6.62	156-160
5 th	23.5-23.8	7.6-7.7	6.60-6.73	165-171
6 th	22.9-23.3	7.4-7.6	6.59-6.70	160-168
7 th	24.1-24.5	7.6-7.7	6.29-6.31	170-179
8 th	24.2-24.9	7.5-7.6	6.89-6.99	177-182

Note: Data expressed as Mean \pm SE (n = 8)

Table 4: Performance for *Labeo catla* fry fed with different experimental diets during culture period

Diets	T ₀ (control)	T ₁ (5%)	T ₂ (10%)	T ₃ (15%)	T ₄ (20%)
Initial body weight (g)	6.55 \pm 0.065 ^a	6.70 \pm 0.041 ^{ab}	6.60 \pm 0.091 ^{ab}	6.50 \pm 0.108 ^a	6.80 \pm 0.041 ^b
Final body weight (g)	20.53 \pm 0.075 ^a	20.58 \pm 0.085 ^a	22.90 \pm 0.238 ^b	23.88 \pm 0.165 ^b	33.50 \pm 1.132 ^c
Mean weight Gain (g)	13.98 \pm 0.103 ^a	13.88 \pm 0.118 ^a	16.30 \pm 0.158 ^b	17.38 \pm 0.131 ^b	26.70 \pm 1.093 ^c
Specific growth rate	1.90 \pm 0.018 ^a	1.87 \pm 0.016 ^a	2.07 \pm 0.010 ^b	2.17 \pm 0.022 ^c	2.66 \pm 0.046 ^d
Survival rate (%)	100 \pm 00 ^a				
Feed conversion ratio	2.92 \pm 0.023 ^e	2.71 \pm 0.022 ^d	2.60 \pm 0.020 ^c	2.43 \pm 0.028 ^b	2.09 \pm 0.032 ^a
Protein efficiency ratio	1.14 \pm 0.009 ^a	1.23 \pm 0.010 ^b	1.28 \pm 0.010 ^c	1.37 \pm 0.016 ^d	1.60 \pm 0.025 ^e

Note: Data expressed as Mean \pm SE (n = 4); Different alphabetical superscripts indicate there is a significant difference (p<0.05)

Table 5: Proximate analysis of fish carcass (dry weight basis)

Experimental Fish	Protein %	Fat %	Ash %	Moisture %
T ₀	12.85 \pm 0.067 ^a	3.38 \pm 0.021 ^c	2.79 \pm 0.032 ^a	80.98 \pm 0.015 ^e
T ₁	13.95 \pm 0.044 ^c	3.41 \pm 0.017 ^c	3.61 \pm 0.017 ^b	79.03 \pm 0.010 ^c
T ₂	14.71 \pm 0.021 ^d	3.00 \pm 0.015 ^a	3.93 \pm 0.017 ^c	78.36 \pm 0.017 ^b
T ₃	13.11 \pm 0.078 ^b	3.23 \pm 0.021 ^b	4.03 \pm 0.029 ^d	79.63 \pm 0.069 ^d
T ₄	16.92 \pm 0.066 ^e	3.44 \pm 0.017 ^c	4.39 \pm 0.021 ^e	75.25 \pm 0.031 ^a

Note: Data expressed as Mean \pm SE (n = 3)

Conclusion

Recently the majority of juveniles of fish and waste fish are used for fishmeal in fishmeal production. Such activities are harmful for sustainable fisheries, so that fishmeal uses as sources of protein is critical. Micro-algae meal

supplementation improved growth, feed consumption, lipid metabolism, body structure, disease tolerance, and carcass consistency of a variety of fish. It is observed that higher growth; higher SGR; higher PER; better FCR and survival in *L. catla* fry can be obtained by feeding the (T₄ treatment)

20% *chlorella* powder on the basis of the results obtained from the experiment. Apart from these observations, micro alga has significant effect on disease resistance antioxidant potentials, in future the experimental diets can also be utilize to check the efficiency of *Chlorella* for disease resistance and better survival rate.

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