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Effect of food restriction and realimentation on the growth performance & body composition of Common carp (*Cyprinus carpio* var. *communis*)

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Abstract
The present study was conducted to investigate the effect of food restriction and realimentation on compensatory growth and body composition of common carp (*Cyprinus carpio* var. *communis)*. The fish were subjected to four different feeding regimes: Control or T_0 (fed regularly two times a day, 0S1F), T_1 (one day food restriction followed by one day refeeding, 1S1F), T_2 (two day food restriction followed by one day refeeding, 2S1F) and T_3 (three day food restriction followed by one day refeeding, 3S1F). It was observed that the fish subjected to T_2 (2S1F) and T_3 (3S1F) feeding regimes showed significantly (p<0.05) lower body weight gain, and specific growth rate (SGR) as compared to the control (T_0) group. However, no significant difference was observed between T_0 (0S1F) and T_1 (1S1F) groups, thus indicating complete compensatory growth in one day food restriction followed by one day refeeding group. The FCR was significantly lower in all the food restriction regimes. With increase in food restriction, the body moisture content increased but crude protein and total lipid content decreased significantly (p<0.05). No significant difference was observed in the ash content of the fish. The results indicated that one day food restriction followed by one day refeeding could be an efficient strategy for rearing of common carp fingerlings.

Keywords: Food restriction, *Cyprinus carpio*, body composition

1. Introduction

Aquaculture is the fastest food production sector in the world and it is expanding at an average annual rate of 8-11% since 1984 (FAO, 2006) [22]. Aquaculture is expected to be the largest source of food by the year 2030 (Brugère and Ridler, 2004; Carella and Sirri, 2017) [13, 14]. In 2018, total aquaculture production attained 82.1 million tonnes while as total world fisheries and aquaculture production attained 178.5 million tonnes (FAO, 2020) [24]. The second largest producer of fish in the world is India and its contribution to the global fish production is 5.68% (FAO, 2014) [23]. In order to meet the ever increasing demand for easily digestible protein and food security, aquaculture is being advocated as the possible solution, thus making it among the future food production sectors. Aquaculture stimulates growth of a number of subsidiary industries besides being a foreign exchange earner and a powerful income and employment generator.

The highest recurrent cost in the aquaculture is the feed cost and it often ranges from 60-70% (Singh *et al.*, 2006) [54]. So for the development and well-being of the industry, any reduction in feed costs either through diet development, improved husbandry or other direct or indirect means is therefore crucial.

Fish shows an impressive capacity to withstand prolonged periods of food limitation, particularly in comparison to mammals (Miriam *et al.*, 2011). An extraordinary resilience to prolonged starvation is exhibited by many fish species which undergo three phases during starvation: First is a short transient phase, second is a long, protein conservation steady state phase where fat oxidation is the primary energy source and shift to protein mobilization as a main energy source (Bar, 2014) [10]. In aquaculture an important method to reduce feed costs is through developing appropriate feeding management and culture strategies and to use the periods of starvation (food restriction) and re-feeding as one such appropriate feeding method (Azodi *et al.*, 2013) [9]. The process of food restriction followed by strategic refeeding results in better growth in some species. Such type of growth is called as compensatory growth. It is a period of accelerated growth that occurs after re-feeding following a period of food restriction or exposure to unfavourable conditions like low temperature, low oxygen and reproductive effort (Ali *et al.*, 2003) [5].

2. Materials and methods

The experiment was conducted for a period of sixty days from 10th February to 9th April, 2020 at wet laboratory Faculty of Fisheries, Rangil, Ganderbal (34^0.21', 74^0.80').

2.1. Collection and acclimatization of experimental fishes

The experiment was carried out in plastic tubs of 40 liter capacity in the indoor wet laboratory. Each tub was filled with stabilized and treated clean water. The setup was supplied with proper aeration using aerators through plastic tubes connected with aeration stones. The water quality was maintained in the optimum range by cleaning the residual feed and excreta through siphoning and partial water exchange. The fish were acclimatized for a period of ten days in laboratory conditions before the start of experiment. The basal diet @3% body weight per day was given during this period. Feeding was avoided twenty-four hours before stocking. After this, ten fingerlings of *Cyprinus carpio* were placed in each of these plastic tubs.

2.2 Feed formulation

The feed formulation was done by Pearson's square method using known values of protein content of ingredients (Table 1). The ingredients were weighed and mixed in appropriate ratios, as determined by Pearson's square method. Feed was prepared in the form of dry pellets with an optimum protein concentration of 32%. The contents were mixed uniformly with adequate quantities of water to ensure good consistency of doughs for autoclave. Pelleting of each diet was carried out by passing the dough of blended mixture through hand pelletizer with (1mm) diameter mesh size.

Table 1: Basal Feed Formulation with an optimum protein concentration of 32%

Ingredient	Inclusion level	Inclusion level for 500g feed
Rice bran (%)	24.25	124.9
Wheat bran (%)	24.25	124.9
Fish meal (%)	22.25	110.08
Mustard oil cake (%)	22.25	110.08
Vegetable oil (%)	5	25
Vitamin & Mineral mixture (%)	2	10
Total	100	500

2.3 Experimental design and feeding regime

The experimental setup was divided into four groups each with four replicates following a completely randomized design. The experimental groups were fed with their respective diets @3% of body weight twice a day on the feeding day (Table 2). In one group designated as Control (T_0), the fishes were fed regularly without any food restriction. Subsequently, the other groups were restricted for food for one day (T_1) followed by one day refeeding, two day food restriction (T_2) followed by one day refeeding, three day food restriction (T_3) followed by one day refeeding.

Table 2: Treatments and feeding regime

Treatments	Feeding Regime
Control (T_0)	Regular / Daily feeding (F0S)
Treatment one (T_1)	One day food restriction & one day re-feeding (1F1S)
Treatment two (T_2)	Two day food restriction & one day re-feeding (1F2S)
Treatment three (T_3)	Three day food restriction & one day re-feeding (1F3S)

2.4 Growth parameters

Initially the weight of fishes was recorded. To study the growth parameters, the fishes were sampled after every ten (10) days which contributed to a total of six (6) samplings during the sixty (60) days of study period. The feeding rate was adjusted corresponding to the body weight of the fishes. The weight was recorded using an electronic weighing balance. The following growth parameters were recorded:

2.5 Percentage weight gain

Percentage weight gain is defined as the amount of weight gained over a period of time in relation to the total weight.
The percentage weight gain was calculated using the following formula:

$$\text{Weight gain (\%)} = \frac{[\text{final fish weight (g)} - \text{initial fish weight (g)}]}{\text{initial fish weight (g)}} \times 100$$

2.6 Feed conversion ratio (FCR)

Feed conversion ratio is the mathematical relationship between the input of feed that has been fed and the weight gain of a population.
The Feed Conversion Ratio was calculated by the following formula:

$$\text{FCR} = \frac{\text{Feed given (g)}}{\text{Body weight gain (g)}}$$

2.7 Specific growth rate (SGR)

Specific growth rate is a term used in aquaculture to estimate the production of fish after a certain period.
The Specific Growth rate was calculated by the following formula:

Specific growth rate (SGR, % d^{-1}) =

$$\frac{[\text{Ln final wet weight (g)} - \text{Ln initial wet weight(g)}]}{\text{Experimental days}} \times 100$$

2.8 Body composition analysis

The body composition analysis of fish samples was carried out at the Disease Investigation Laboratory, Sheep Husbandry Department, Government of Jammu and Kashmir, Srinagar.

Initially, four fish samples were taken for body composition analysis and at the end of the experiment four samples from each replicates were taken for the analysis. The body composition included the following: Moisture content, Crude Protein content, Total Lipid content, and Total Ash content.

3. Results and Discussion

3.1 Growth performance

The growth performance of fish was expressed in terms of body weight gain; weight gain percentage, feed conversion efficiency (FCR), and specific growth rate (SGR), as shown in Table 3.

A significant difference (p<0.05) was recorded in the body weight gain and in the weight gain percentage among different treatments. However, no significant variation (p>0.05) was observed between T_1 (1F1S) group and the T_0 (control). Highest body weight gain as well as the highest weight gain percentage was observed in T_0 (control) followed by T_1 (1F1S) group while the lowest was observed in T_3 (1F3S) group.

The FCR of different experimental groups, as depicted in Table 3 and Figure 3, varied significantly (p<0.05) among different treatments. Highest FCR was observed in T_0 (control) while the lowest FCR was shown by T_2 (1F2S) group followed by T_3 (1F3S) group.

The SGR of different experimental groups is indicated in the Table 3 and Figure 4. The SGR varied significantly (p<0.05) among different treatments. However, no significant difference (p>0.05) was observed between T_1 (1F1S) group and the T_0 (control). The treatment T_0 (control) showed the highest SGR followed by T_1 (1F1S) group while the lowest SGR was observed in the T_3 (1F3S) group.

Table 3: Growth performance of experimental groups subjected to food restriction and realimentation regimes

Treatments	IW (g)	FW (g)	WG (g)	WG (%)	FCR	SGR
T_0	2.4±0.04	4.41±0.03^c	2.01±0.03^c	83.68±2.38^c	2.85±0.06^c	1.01±0.02^c
T_1	2.43±0.05	4.4±0.06^c	1.97±0.02^c	80.89±1.35^c	1.36±0.01^b	0.99±0.01^c
T_2	2.42±0.04	3.91±0.06^b	1.49±0.03^b	61.65±0.65^b	1.16±0.01^a	0.8±0.01^b
T_3	2.44±0.06	3.46±0.07^a	1.02±0^a	41.92±0.96^a	1.22±0.03^a	0.58±0.01^a
p value	0.96	<0.05	<0.05	<0.05	<0.05	<0.05

T_0: Control (fed regularly, F0S); T_1: Treatment one (one day food restriction and one day refeeding, 1F1S); T_2: Treatment two (two day food restriction and one day refeeding, 1F2S); T_3: Treatment three (three day food restriction and one day refeeding, 1F3S). IW = Initial weight, FW = Final weight, WG = Weight gain, FCR = Feed Conversion Ratio, SGR = Specific Growth Rate. The data were expressed by Mean ± Standard Error (n = 4). Different superscript letters in the same column denote significant differences (p<0.05) between the experimental groups.

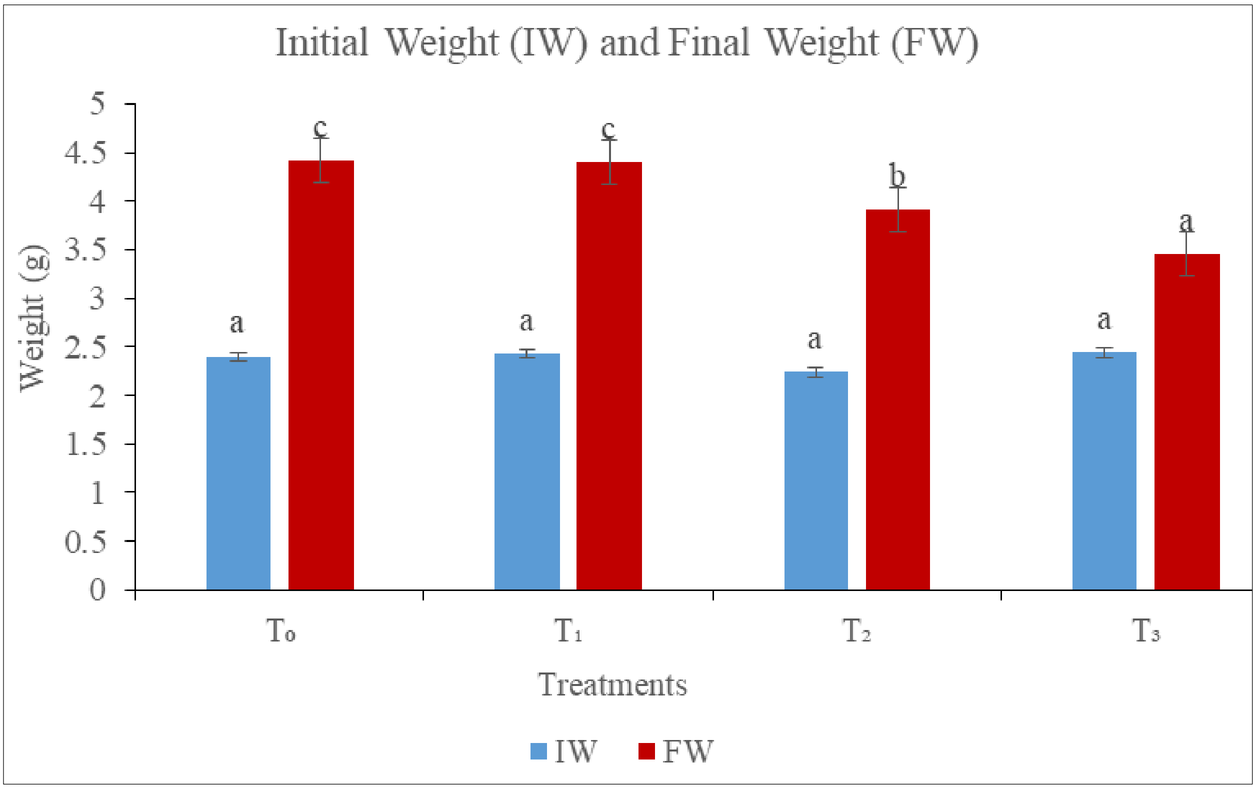

Fig 1: Comparison between initial and final weight of *Cyprinus carpio* fingerlings subjected to different feeding regimes

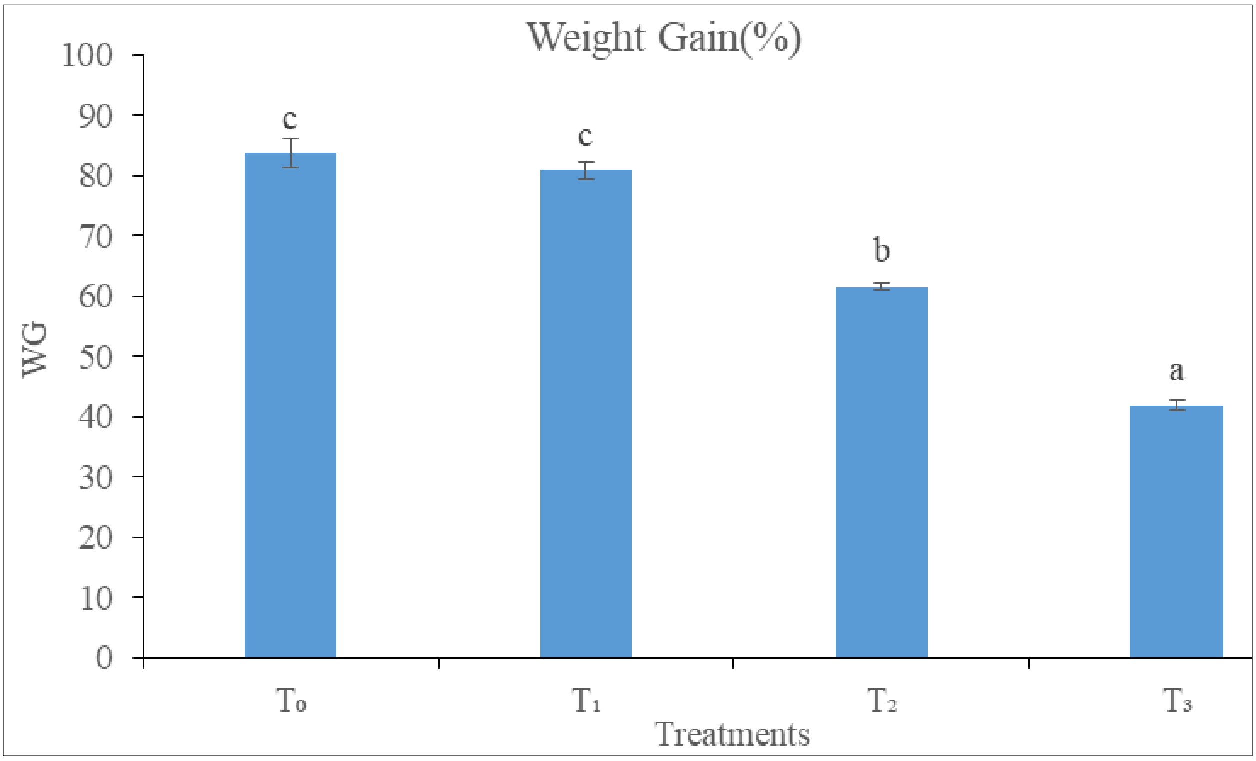

Fig 2: Weight gain percentage of *Cyprinus carpio* fingerlings subjected to different feeding regimes

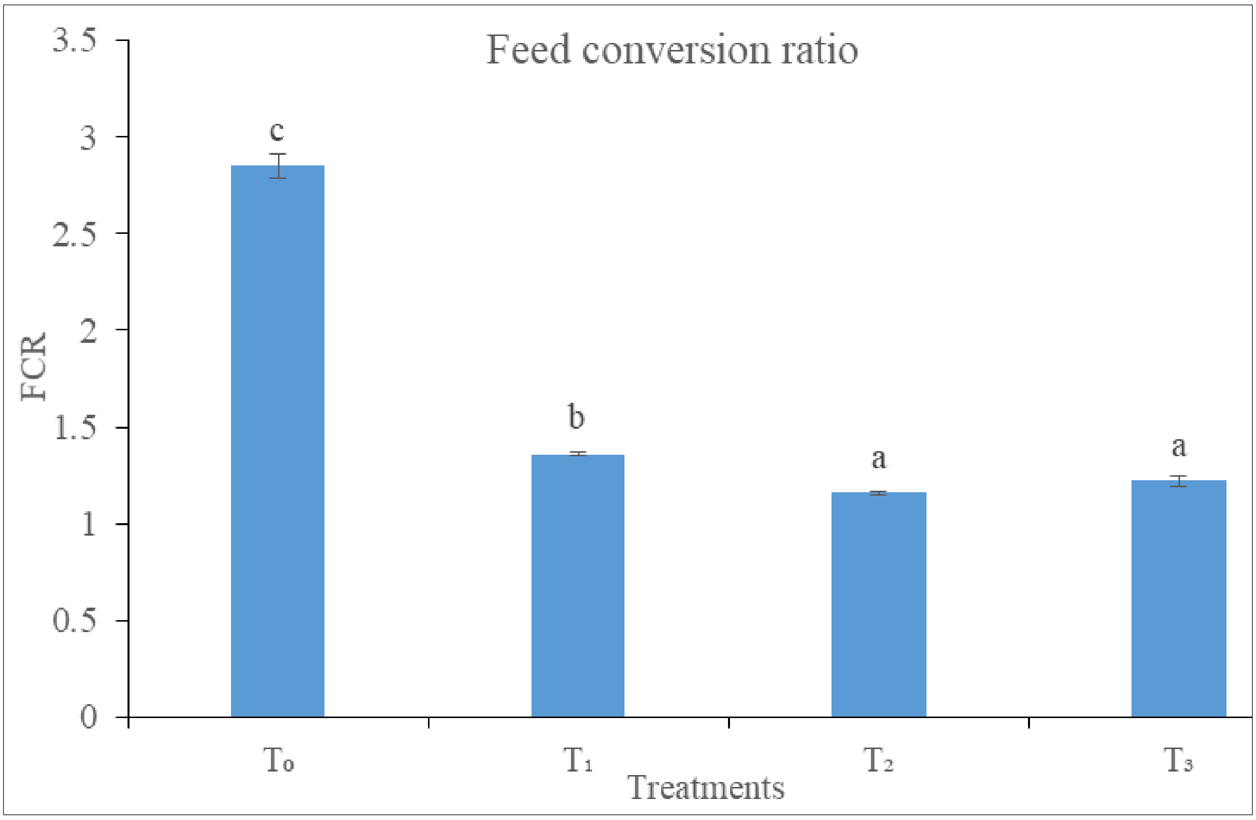

Fig 3: Feed Conversion Ratio of *Cyprinus carpio* fingerlings subjected to different feeding regimes

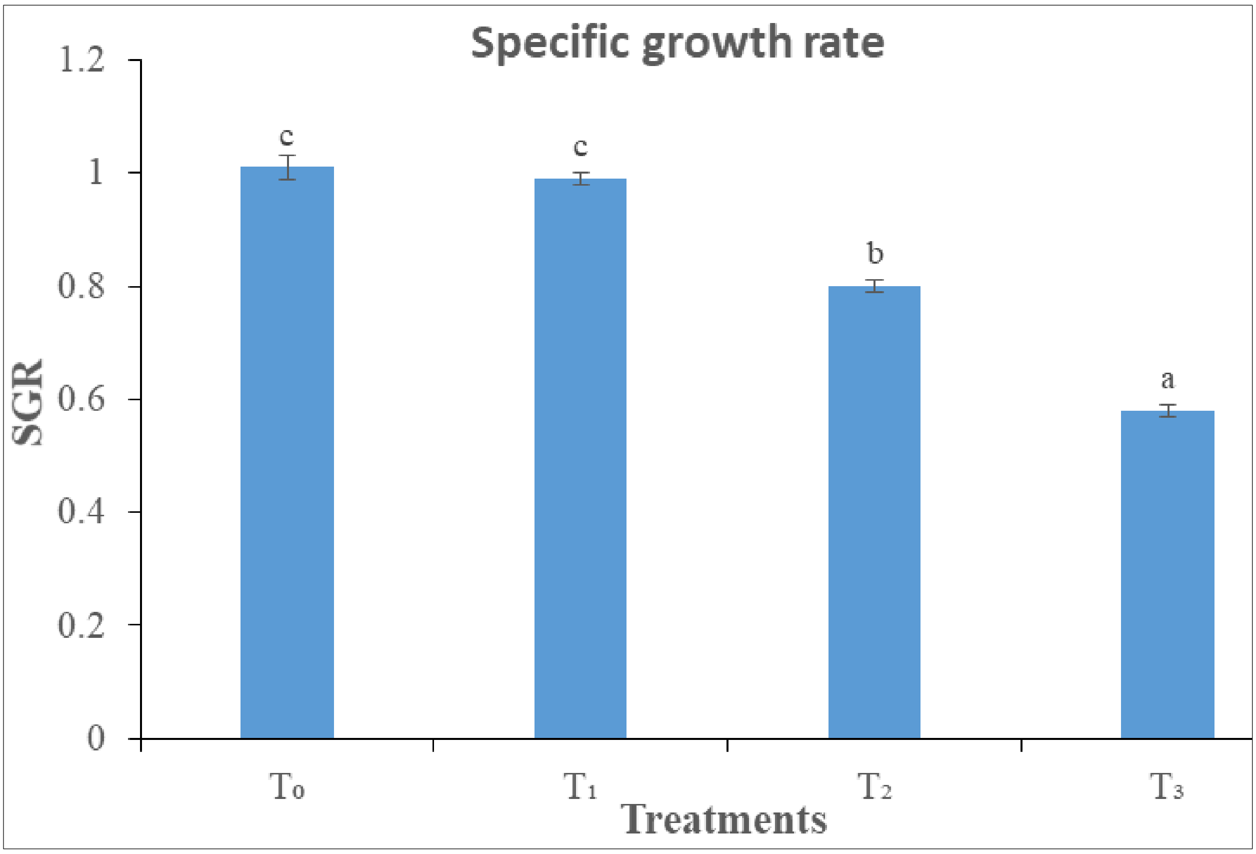

Fig 4: Specific growth rate of *Cyprinus carpio* fingerlings subjected to different feeding regimes

3.2 Body composition analysis

The fish body composition analysis of fish was expressed in terms of moisture (Figure 5), crude protein (Figure 6), total lipid (Figure 7) and total ash content as shown in Table 4.

A significant difference ($p<0.05$) was recorded in the moisture content among different treatments. However, no significant variation ($p>0.05$) was observed between T_0 (control) and the T_1 (1F1S) group. Highest moisture content was observed in initial samples followed by T_3 (1F3S) and lowest in T_0 (control) and T_2 (1F2S) group respectively.

The crude protein of different experimental groups, as depicted in Table 4 and Figure 6, varied significantly ($p<0.05$) among different treatments. Highest FCR was observed in T_0 (control) while the lowest crude protein content was observed in T_3 (1F3S) group.

The total lipid content of different experimental groups is indicated in the Table 4 and Figure 5. The total lipid content varied significantly ($p<0.05$) among different treatments.

Highest lipid content was observed in T_0 (control) and T_1 (1F1S) group while lowest lipid content was observed in T_2 (1F2S) group and T_3 (1F3S) group.

Total ash content of the fish samples is shown in Table 4. No significant difference (p=0.773) was observed in the fish samples in their total ash content.

Table 4: Body composition (% dry weight) of Common carp fingerlings subjected to different feeding regimes at the end of sixty (60) days experimental trial

Treatments	Moisture	Crude Protein	Total Lipid	Total Ash
Initial	75.27 ± 0.03^d	16.68 ± 0.03^b	6.5 ± 0.02^b	2.38±0.02
T_0	73.61 ± 0.03^a	16.74 ± 0.01^{cd}	6.5 ± 0.02^b	2.43±0.03
T_1	73.62 ± 0.01^a	16.72 ± 0^{bc}	6.47 ± 0.01^b	2.43±0.08
T_2	74.22 ± 0.01^b	16.78 ± 0.01^d	6.35 ± 0.01^a	2.38±0.01
T_3	74.42 ± 0.02^c	16.14 ± 0.01^a	6.3 ± 0.01^a	2.43±0.01
p value	<0.05	<0.05	<0.05	0.773

T0: Control (fed regularly, F0S); T1: Treatment one (one day food restriction and one day refeeding, 1F1S); T2: Treatment two (two day food restriction and one day refeeding, 1F2S); T3: Treatment three (three day food restriction and one day refeeding, 1F3S). The data were expressed by Mean ± Standard Error (n = 3). Different superscript letters in the same column denote significant differences ($p<0.05$) between the experimental groups.

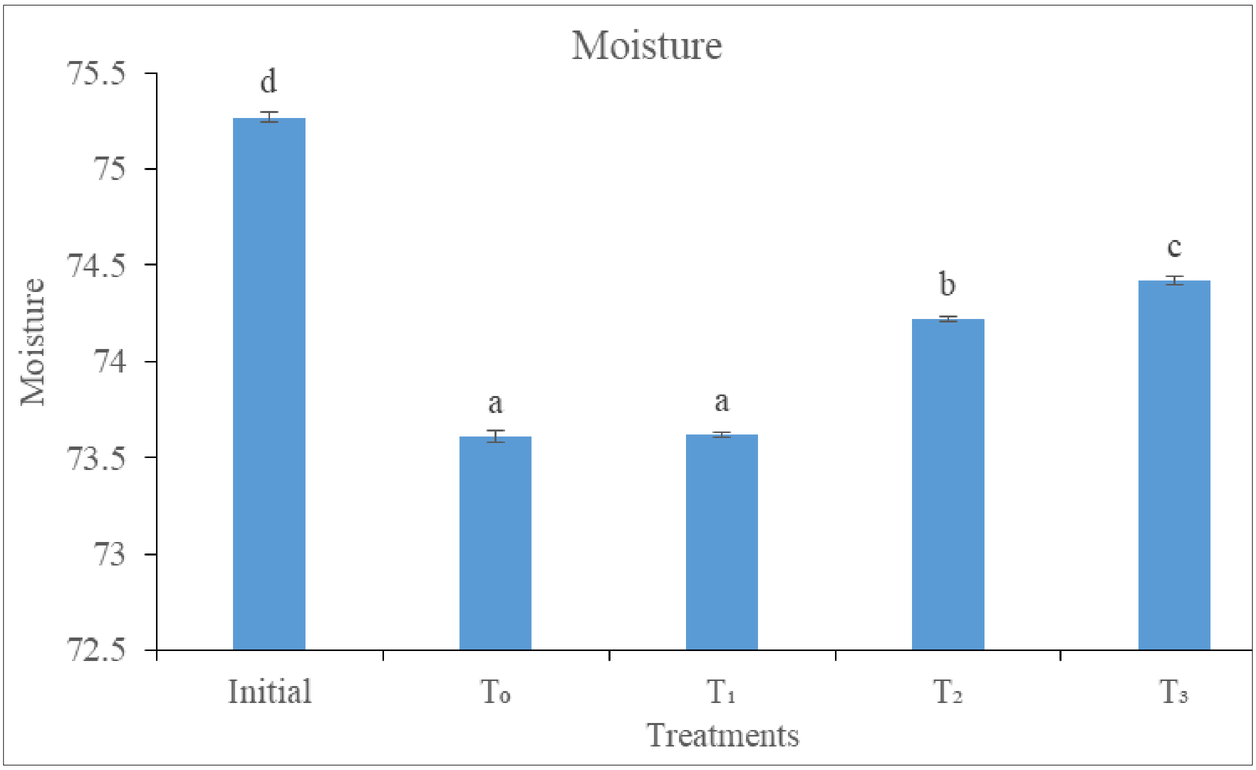

Fig 5: Moisture content (%) of *Cyprinus carpio* fingerlings subjected to different feeding regimes

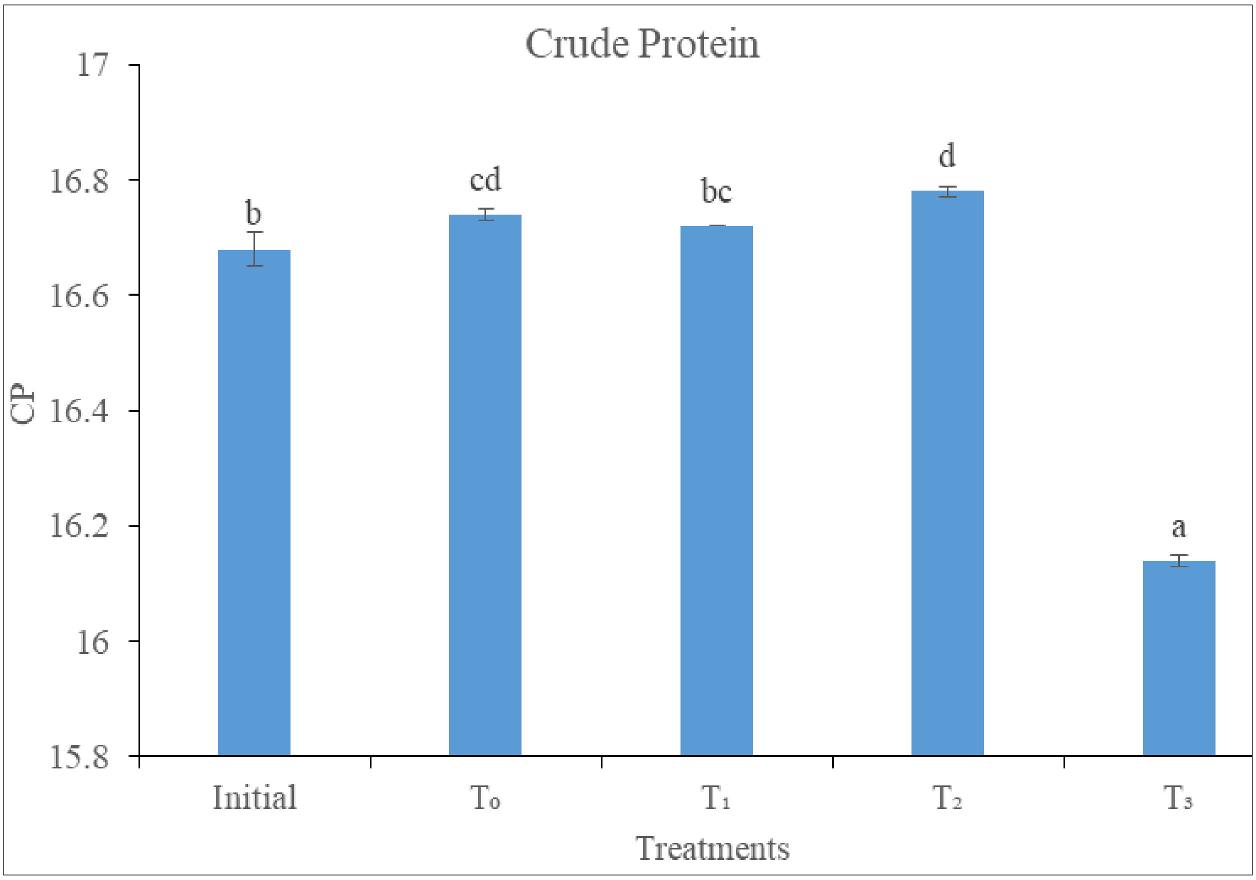

Fig 6: Crude protein content (%) of *Cyprinus carpio* fingerlings subjected to different feeding regimes

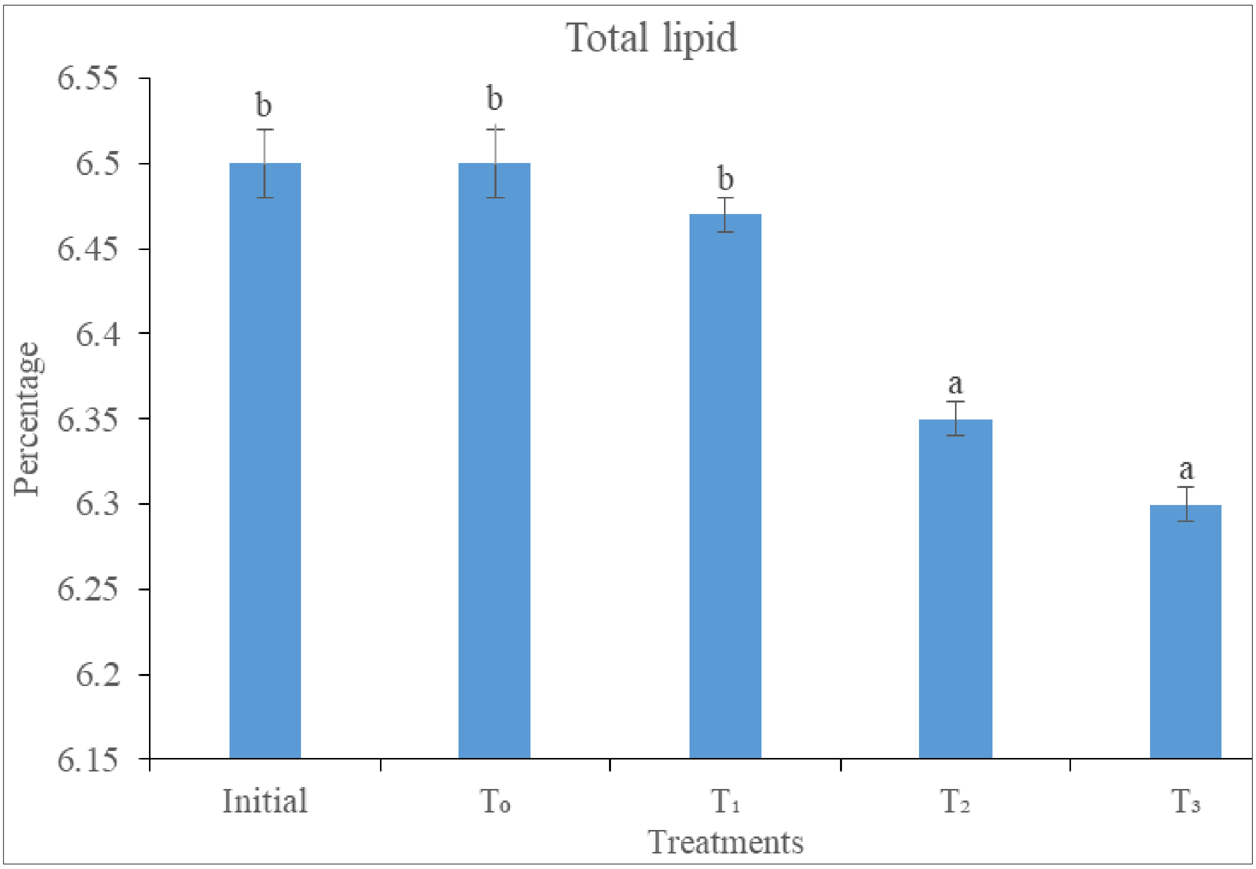

Fig 7: Total lipid content (%) of *Cyprinus carpio* fingerlings subjected to different feeding regimes

4. Discussion

4.1. Growth parameters

The results of the study indicated that Common carp (*Cyprinus carpio*) fingerlings subjected to different food restriction and refeeding regimes showed significant differences in their growth performances. Highest body weight gain and specific growth rate was observed in the control (fed daily) and T_1 group (one day food restriction and one day refeeding) as they showed no significant difference.

One day food restriction appears to be readily overcome by *C. carpio* fingerlings upon realimentation. Weight lost during the one day period of food deprivation was recovered such that weight gain of restricted and regular fed fish was significantly same at the end of experiment. In other words we can say that when the amount of feed consumed by fingerlings was decreased by 50% in the T_1 group (i.e., 1F1S), the final values didn't significantly differ from control, indicating full compensation.

However, *C. carpio* fingerlings could not attain the equal weight to control or one day food restricted fish in more extended food restricted period (1F2S & 1F3S). The body weight gain and specific growth rate of fish subjected to two day food restriction, one day refeeding (T_2) & three day food restriction, one day refeeding (T_3) were significantly lower than fish fed regularly and the one fed alternatively over the experimental period. The inability of *C. carpio* fingerlings deprived for two and three days to catch up in body weight probably resulted from the weak capacity for compensatory growth coupled with greater weight losses during feed deprivation. One more explanation could be that the period of food restriction was too long for the fingerlings to recover or attain compensatory growth.

Similar trends were observed by Kim & Lovell (1995) [35] in channel catfish *(Ictalurus punctatus)* held in ponds. They observed that the fish could recoup weight loss and attain final weights equal to control (satiate fed fish) when deprived of feed for three weeks, but more extended feed deprivation prohibited the channel catfish from attaining a final weight equal to control fish. A similar finding has been reported in *Ictalurus punctatus* by Gaylord & Gatlin (2001) [28].

Hybrid tilapia deprived for one week had similar body weights to the controls, whereas fish deprived for two & four weeks had significantly lower body weights than control (Wang *et al.*, 2000) [58]. As the results of our study demonstrate full compensatory growth in the one day food restricted / one day refed group and a poor compensatory growth with the increase in the food restriction which is in similarity with the findings of Hybrid Tilapia (*Oreochromis mossambicus* × *Oreochromis niloticus*) in which full compensatory growth was observed in two day deprived / four day refed group while as poor compensatory growth was reported with the increase in feed deprivation. Their results are in agreement with our present study. *C. catla* showed full compensatory growth when subjected to short duration of deprivation (2D2F regime) where as it was unable to compensate as the duration of deprivation increased (Nagar and Patidar, 2015) [43] which shows the similarity with our study. Abdel-Hakim *et al.* (2009) [1] also reported similar results in Hybrid tilapia (*Oreochromis niloticus* × *Oreochromis aureus*) subjected to one, two and three days food deprivation per week, respectively for four months indicating that moderate feed deprivation (one, two days/ week) enabled tilapia to compensate the growth retardation and the decrease was more pronounced at higher deprivation days.

Similar results were obtained for complete compensatory growth in terms of weight gain and specific growth rate in *Labeo rohita* (Prabhakar *et al.*, 2008) [47]; Giebel carp

(*Carrrasius auratus gibelio*) and Chinese long snout catfish (*Leiocassilon grostris)* by Zhu *et al.* (2004) [60]; Thai pangas (*Pangassius hypophthalmus)* by Amin *et al.* (2005) [7], *Sparus aurata* (Eroldgan *et al.*, 2006) [19]; pike perch (Mattila *et al.*, 2009) [36]; Nile tilapia (Passinato *et al.*, 2015; Cuvin-Aralar *et al.*, 2012) [45, 18]; *Lates calcirifer* (Tian & Qin, 2003) [57] subjected to different food restriction and refeeding regimes. However, Ali *et al.*, 2006 [30] and Iqbal *et al.*, 2006 [30] observed weak compensatory growth in *L. rohita* and *C. mrigala* respectively when subjected to five and ten days deprivation and refeeding. This indicates that response of compensatory growth varies with the duration of deprivation. In the present study, it was observed that compensatory growth depends on duration of food restriction and short term food restriction thus elicited compensatory growth in *C. carpio* fingerlings.

Poor compensatory growth in fish exposed to longer deprivation periods were reported in Nile tilapia (Passinato *et al.*, 2015) [45]; *O. mossambicus* (Christensen and Mclean, 1998) and other species such as *Centropomus parallelus* (Riberio and Tsuzuki, 2010) [51] and *Sparus aurata* (Peres *et al.*, 2011) [46]. These findings are in agreement with our study.

In our study we also achieved complete compensation when the ratio of refeeding: food restriction period was kept at 1:1. As the food restriction period was smaller and relatively less severe complete compensatory growth was elicited. Futher, when the period of food restriction was kept longer (1:2 in T_2 and 1:3 in T_3) than the refeeding period, poor compensatory growth was observed.

The mechanism of growth compensation can be partly explained by the increase in feed intake per feeding (i.e., hyperphagia) but also by the regularity of the ingested amount of feed (Mattila *et al.*, 2009) [36]. Hyperphagia is considered the major mechanism causing compensatory growth as reported by many researchers (Miglavs & Jobling 1989; Wang *et al.*, 2000) [38, 58], it can be assumed that in our study this phenomena resulted in the compensatory growth of one day food restricted and refed group.

In the present study, feed conversion ratio was reported to decrease with the increase in the duration of deprivation. A similar trend was observed in *Catla catla* subjected to different feed cycling regimes (Savliya-Patidar, 2015) [43]. Eroldoğan *et al.* (2008) [20] and Amin *et al.* (2005) [7] reported similar trend in juveniles of *S. aurata* and thai pangas, *P. hypophthalmus* respectively. The highest FCR was reported in the regularly fed treatment and the lowest FCR was reported in the T_2 and T_3 groups. We also observed that it needed approximately 50% more feed in case of control than the feed needed in one day food restricted group i.e., T_1 to produce almost same quantity of fish. This is an indication that short term food restriction and realimentation cycles could indeed be a useful tool in reducing feed amount without compromising fish farm production output which is in similarity with the study of Gabriel *et al.*, 2017 [26]. The lowest FCR in feed deprived & refed fish may be due to the less number of days during refeeding, ultimately reducing feed quantity and feed cost. As we observed the decreased FCR in T_2 and T_3 without increasing feed intake. A similar trend was observed by Russell & Wootton (1992) [52]; Jobling (1994) [34], Wang *et al.* (2000) [58]; Eroldoğan *et al.*, 2006 [19] following different restriction and refeeding regimes. Decreased FCR observed in the present study may be because of the enhancement in digestive enzyme activities which likely occurred in restricted feeding regimes as also suggested by Eroldoğan (2008) [20] and Jobling (1994) [34].

5. Body composition

The present study observed a significant difference in the body composition of Common carp fingerlings subjected to four different feeding regimes i.e. (T_0: Control (fed regularly, F0S); T_1 (one day food restriction and one day refeeding, 1F1S); T_2 (two day food restriction and one day refeeding, 1F2S); T_3 (three day food restriction and one day refeeding, 1F3S). Significant differences ($p<0.05$) in the body composition were observed in many previous studies when the fish were subjected to different food restriction and refeeding protocols thus showing similarity with our results (*Catla catla*, Nagar and Patidar (2015) [43]; *Acipenser baeri*, Morshedi *et al.* (2012) [21], *Barbonymus schwanenfeldii*, Eslamloo *et al.* (2012) [21], Mohantta *et al.*, (2015).

In T_2 (1F2S) and T_3 (1F3S) lower level of protein and lipid was observed as compared to T_1 (1F1S) and T_0 (F0S) which is in similarity with the results of Golden perch, *Macquaria ambigua* (Collins & Anderson, 1995) [17]; *Tilapia mossambica* (Chalapathi-Rao *et al.*, 1987) [15]. The depletion of lipid and protein content in T_2 & T_3 may be due to their utilization as an energy source during food restriction period. Moisture content was found to be significantly higher in T_2 and T_3. Inverse relationship was found between the moisture & lipid content of the fish. This is in accordance with the results of Ali *et al.* (2006) [30]. Increase in moisture content with increased duration of food restriction may be associated with tissue rehydration resulting due to depletion of body constituents during food restriction. This is in accordance with the results of *Catla catla*, Savliya Patidar (2015) [43] Ash content showed no significant difference ($p>0.05$) which is in accordance with the observations of Ali *et al.* (2006) [30].

6. Acknowledgements

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