Original Research Article

Effect of protein diet on growth and body composition of *Labeorohita* reared in glass tank

Abstract

Fish is used as a great source of food and the quality of its meat can be indicated by fat and protein contents as well as low quantity of water in fish meat. Therefore, the present study was performed for improvement in weight, feed-conversion ratio, protein-efficiency ratio, and growth. The experiment was performed at commercial level in field (pond) andthe influence of fish meal protein on growth of *Labeorohita* was investigated in 60 days culture trials. 90 fingerlings were maintained in well aerated 280 L three glass tanks in triplicate. These glass tanks were named as T1, T2 and T3. T1 was consists of 30% fish meal protein with control diet (containing proteins, fats vitamins and fibers), T2 consist of 35% protein with control diet,while T3 consist on only control diet. The fish were fed of 4% percent of their body weight twice a day at twelve hours interval for 6 weeks. There was a significant increase ($p<0.05$) in body weight, feed-conversion ratio, protein-efficiency ratio, and gross fish production of fish having 35% fish meal protein with control diet (T2) when compared with 30% fish meal protein with control diet T1 and control diet T3. These findings suggest that 35% fishmeal protein appear to be sufficient for obtaining optimum growth in fish. Thus, in future fishmeal, protein will also use for many other fish for producing large number of fish.

Keywords: fish meal protein,fingerlings, protein-efficiency ratio, growth performance, *Labeo rohita*

1. Introduction

For production of fishes aquaculture environment is very important. In whole world, different types of fish are introduced. In different countries, fish trade is used for different purpose like sport fishing [1]. Fish is also used for ornamental purpose. For human being and other animal in different countries, fish is used as a great source of food. By water proportion the quality of fish meat can be indicated. The quality of fish meat can be indicated by fat contents and protein. The quality of fish meat will be good if low quantity of water is present in fish meat. In whole world fish body is used as food because it contain large amount of protein.In body of fish the amount of carbohydrates is very low. The amount of water present in fish body is 75 to 80 percent. Protein percentage in fish body is 25 to 30% and the lipid percentage in fish body is 2 to 12% [2]. Fish is a rich source of vitamin A. By using the fish as diet dietary requirement of human can be fulfilled. Large amount of calcium is present in skeleton of fish. The calcium present in skeleton of fish is very beneficial form human health. Smaller fish are the great source of calcium for human[3].

Fish is cultured in farm as well due to the elevated command of fish as an important human food. Farmers face the difficulty in collection of fish variety because of deficient the information about the pond ecology. The absence of vitamin D in old age people and children can be accomplished by using fish as a source of vitamin D. As compare to all other meat doctors prefer the fish meat. If diet not contains fish meat the diet than it is considered to be deficient in vitamin D.In diet of Japanese people fish is present in great amount. Fish meat is used for preventing the deficiency of vitamin D[4].

Protein food is very important for maximum production of fish. The effect of four different kinds of feed ingredient on growth of *Labeo rohita*was checked. At the rate of 4% of body

weight of fish Fingerlings of *Labeorohita* are provided with 30%, 35%, 40% and 45% with ground nut cake, azolla, rice ban and tapioca powder[5]. The growth and digestive enzyme activities of fingerlings of *Labeorohita* can be controlled by phytase, plant ingredients and cellulose enzyme. By using plants ingredients two diets for fishes are prepared. For protein utilization the phytase and cellulose are two most important enzymes. Amylase, protease, and lipase are most important for proper functioning of fish[6].

The acceptance for feed in fish increased due to plant protein mixture. Plant protein mixture is very important for fish health and growth[7].Nutritional composition, biological availability, energy content and digestibility of feedstuff ingredients are important criteria for inclusion of any plant or animal protein ingredient to formulate practical diets for fish[8].Different plants ingredients used for survival, feed-conversion ratio, and for survival of *Labeorohita*. From plants corn gluten meal, sunflower meal, canola meal, and rapeseed meal obtained. These play important role in growth of fish and with these ingredients the length and weight of fish is increased[9].

For growing population, we have to balance the urgent needs for food. Corn is present in all plant foods. IT is rich source of complex carbohydrate, which consists on simple sugars starch, and carbohydrates. Large amount of fats and carbohydrates reduced the protein breakdown. *Labeorohita* has ability to use 43% carbohydrates in diets. This carbohydrate has no bad effects on fish health. These feed minimize the cost of fish and improve the growth and meat composition of fish. Feed represent 40 -50% of production costs so nutrition is very important in fish farming[10].

This experiment was performed to conclude the consequence of plant protein, on fingerlings of *Labeorohita*. Asparagus, giricidia, eichhornia, guar gum binger, mineral vitamin mixture, and rice bran used as plant protein mixture. 20, 30, 40, 50, 60 and 70 % plant protein mixture used. This experiment performed for 120 days. The highest growth performance

recorded in 50% diet as compared to all other diets. This experiment showed the importance of plant protein mixture on fish body.

2. Materials and Methods

2.1.Site Selection

This research was performed to investigate different kinds of proteins on fingerlings of *Labeorohita*. Nighty (90) fingerlings of *Labeo rohita*with different body size were grown under extensive culture. These specimens were taken from Multan hatchery and were transported into the research Laboratory of Fisheries at Bahauddin Zakariya University, Multan (BZU) in a plastic container.

2.2.Experimental Design

Three water tanks (T1, T2, & T3)contain thirty (30) specimen of *Labeo rohita*in each tank. At the start of experiment, individual weight of fingerlings of *Labeorohita* was determined. After two weeks weight gain by fingerlings of *Labeo rohita* were also determined. The fingerlings were feed with control diet on erected iron stand fit glass aquaria which contain 280 liter water. The water was flow at a speed of 300 ml /min in all aquariums. By using digital pH meter, water pH was calculated and by means of a glass thermometer water temperature was recorded. Water temperature was varied from 20 to 25 degree centigrade, while water pH was 7.4. Oxygen was continuously supply in each glass aquaria. The experiment was done for sixty (60) days in which variable ratio of proteins and control diet (3.7 Kcal/g of metabolizable energy, 3.4% fiber and 7.03% fat as well as vitamins and minerals in the form of dry pellets) was used. At the rate of 4% body weight fingerlings were fed with three different level of crude protein (CP). The amounts of diets were adjusted according to individual weight of fingerlings of *Labeo rohita*. Feed sizes were 2nm because the mouth of fingerlings can swallow only the pellet of 2nm. The ratios of three

different feeds were consisting on T1 (feed with CP 30% + Control Diet), T2 (feed with CP 35%+ Control Diet) and T3 (feed with CP 100% control diet). The weight of fish was determined with respect to different level of monthly interval.

2.3.Analysis

Body length, weight gain, and feed-conversion ratio (FCR) was recorded at the end of feeding trial using the following formulas. Survival rate of fingerlings was also determined at the end of feeding trial.

Weight gain = final weight –initial weight Length gain = final length –initial length

FCR = Total dry feed fed /total wet weight gain

For water percentage, the initial weight for moisture substance determination was taken from fish body and then was placed in oven at about 105 degree centigrade for 10 hours,until constant weight were reached. The sample was minced in grinder and moisture content was determined. For dry weight calculation, about 02gm sample from fish body was taken. The sample was dried and placed in oven at 90 to 101 degree centigrade for 1 to 2 hours. The powdered was obtained in an electric Molinix blender at about 60 to 65 degree centigrade. The residue substance in fishbody was determined by taking 2g sample from fish body. In muffle furnace, the sample was heated at about 300 to 1100 degree centigrade,then for 6 hours cooling process was occurred at 5 degree centigrade. White substance or ash was obtained and through vacuum distillation organic substances from fish body were measured. Two gram sample from fish body was reacted with strong acid like HCl almost 2ml at high temperature 300 to 400 degree centigrade. Then this material was taken in flask at 40 degree centigrade and adds 50m of distilled water. In Protein content analysis by kjeldahl method the amount of protein was determined. Both wet and dry samples were processed for protein content analysis. To release nitrogen meat was digested with strong acid which was determined by a titration technique. From the nitrogen

concentration of the fish meat amount of protein was calculated. For conversion of nitrogen content to protein content a conversion factor of 6.25(equivalent to 0.16g nitrogen per gram of protein) was used. Fat was determined by the method described by (AOAC, 1990) by using Soxhlet apparatus. For estimation of fat content, the dried sample left after moisture determinations were finely grinded and, the fat was extracted from sample size 02gram time 4 to 5 hours. After extraction the solvent was evaporated and extracted material were weighed.

2.4.Statistical Tool and Data Analysis

Data on specific growth rate, percentage survival and feed-conversion ratio of*Labeorohita* given with different diets were subjected to One Way Analysis of Variance (ANOVA) to determine the significant differences among the treatment means (5 % level of significance). The Intelbased Statistical Package for the Social Science (IBM SPSS) Version 20 Program ws used in the statistical analysis. Correlation analysis was used to determine the relationship of temperature with the growth and survival of the experimental stocks.

3. Results

Results of the various body composition parameters among the three groups (Group A: fish meal 30%, Group B: fish meal 35% and Group C: control group) following ANOVA are shown in Table 1 and 2.

3.1.Water percentage

The mean percentage of water content in T1,T2, and T3 were 82.099, 81.380 and 81.470, respectively. As shown in fig.1 and table 1, the percentage of water content in group A was greater as compared to group B and group C. There was a statistically noteworthy distinction $p<0.05$ of group A when compared to group B and group C.

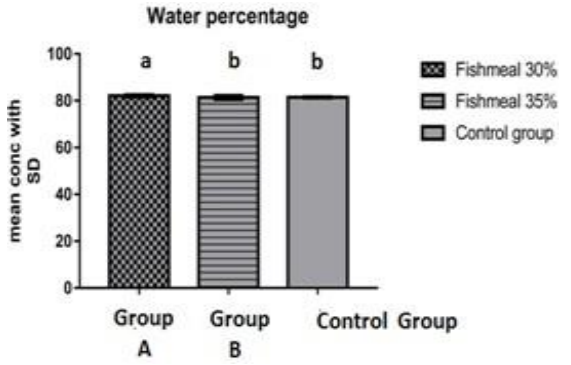

Figure 1 Comparisonof groups regarding water percentage

3.2.Dry weight percentage

The dry weight percentages in T1, T2, and T3 were 17.900, 18.698,and 18.530. The value of dry weight in group B was greater than the value of dry weight in group A. As shown in fig.2 the value of dry weight at group B was statistically similar to value of dry weight in groupC. There was not a significant difference P> 0.05 between the value of dry weight in group B and group C. But the dry weight in group A has a clear significant differencep<0.05 from the value of dry weight in group B and group C (Table 1).

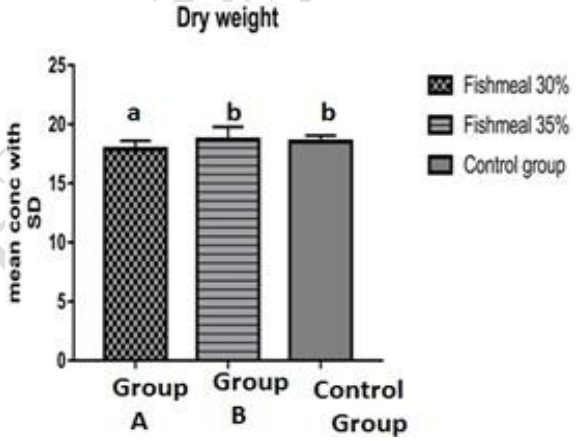

Figure 2 Comparison of groups regarding dry weight

3.3.Ash dry weight

The percentages of ash dry weight in T1,T2 and T3 were 17.469, 14.630, and 11.481. All these value were different from each other. So there were a significant difference p<0.05 was present among these entire three group (Table 1 and Fig. 3).

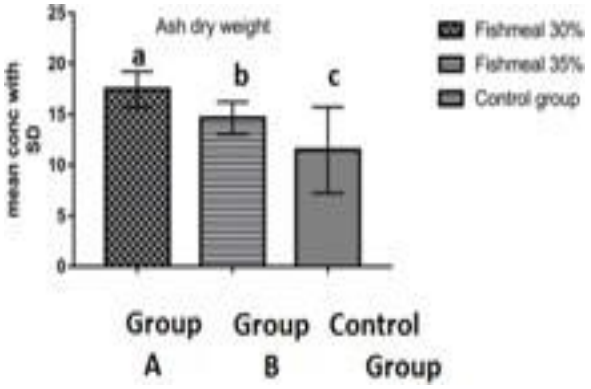

Figure 3 Comparison of groups regarding Ash content dry weight

Table 1 Analysis of morph metric parameters among the groups

Sr. No	Parameter	Fishmeal 30% (Mean ± Std)	Fishmeal 35% (Mean ± Std)	Control group (Mean ± Std)
1	Water percentage	82.099±0.705[a]	81.380±1.075[b]	81.470±0.521[b]
2	Dry weight	17.900±0.705[a]	18.698±1.075[b]	18.530±0.521[b]
3	Ash dry weight	17.469±1.779[a]	14.630±1.582[b]	11.481±4.235[c]
4	Ash (wet weight)	3.119±0.212[a]	2.727±0.372[b]	2.125±0.787[c]
5	Organic content dry	82.530±1.779[a]	85.370±1.582[b]	88.518±4.235[c]
6	Organic content wet	17.834±0.712[a]	18.570±1.077[b]	18.495±0.525[b]
7	Fat (dry weight)	45.061±4.391[a]	41.543±9.285	45.061±2.730
8	Fat (wet)	8.068±0.870	7.770±2.036	8.354±0.661
9	Protein (dry)	54.569±4.395	58.187±9.296	54.750±2.779
10	Protein (wet)	9.765±0.817[a]	10.799±1.554[b]	10.140±0.462[a]
11	Condition factor	0.575±0.008[a]	0.567±0.029[a]	0.585±0.011[ab]

3.4.Organic content dry

Mean value observed in T1, T2 and T3 was 82.530, 85.370 and 88.518. All these values are different from each other. So there were a significant difference $p < 0.05$ was present among all these three groups (Table 1 and Fig. 4).

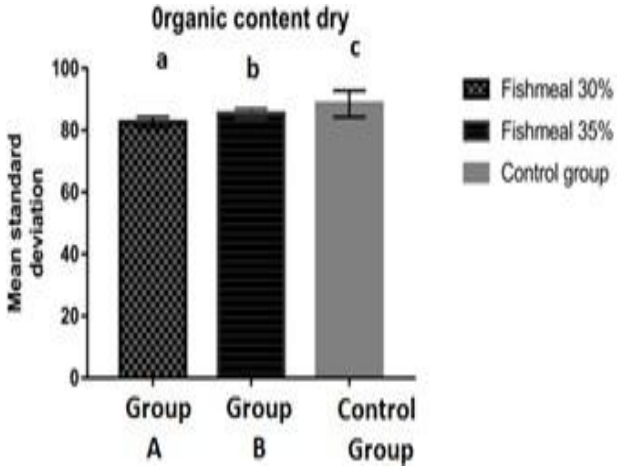

Figure 4 Comparison of groups regarding organic content of dry weight

3.5.Fat dry weight

The percentages of fat dry weight content in T1, T2 and T3 were 45.061, 41.543 and 45.061. All the groups have same value. There was not a significant difference observed among these groups (Table 1 and Fig. 5).

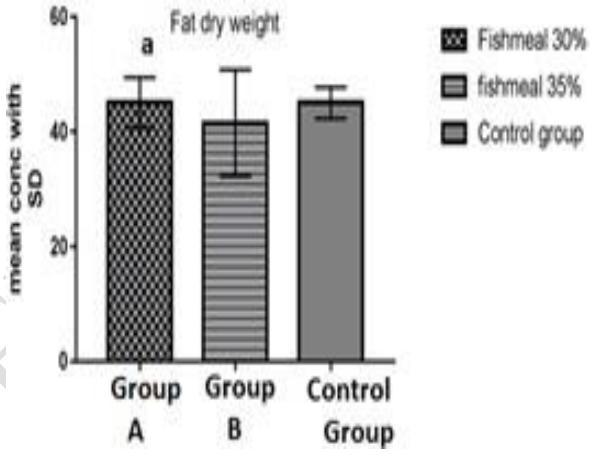

Figure 5 Comparison of groups regardingpercentage of fat dry weight

3.6.Protein dry weight

The mean percentage of protein content in dry weight in group A, group B and group C was 54.569, 58.187 and 54.750, respectively. As represent in fig.6 values of the group B (fed on fish meal 35%) were comparatively higher. However, results of ANOVA did not reveal any significant difference ($p>0.05$) among the mean values of the three groups.

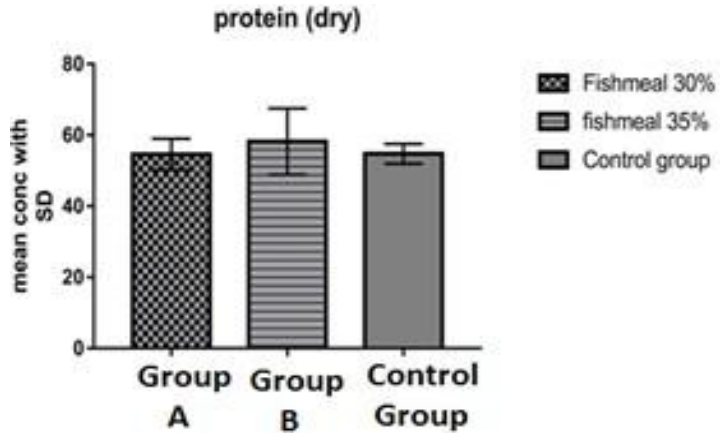

Figure 6 Comparison of groups regardingpercentage of Protein dry weight

3.7.Weight Gain

As shown in fig.7and table 2 comparisons of groups regarding weight gain revealed no significant difference among the groups. The values of T1, T2 and T3 observed were 112.08, 117.90 and 117.20, respectively.

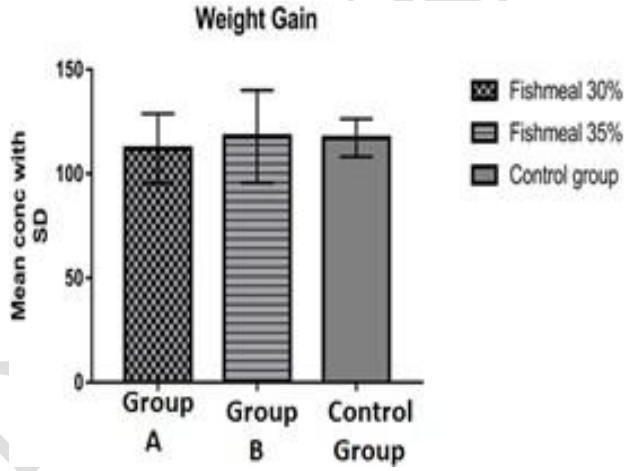

Figure7 Comparison of groups regardingpercentage of weight gain

3.8.Increased in body width

Mean values observed in the group T1, T2 and T3 were 4.97, 5.28 and 6.04, respectively. As shown in fig. 8 and table 2, the difference was non-significant among the group means.

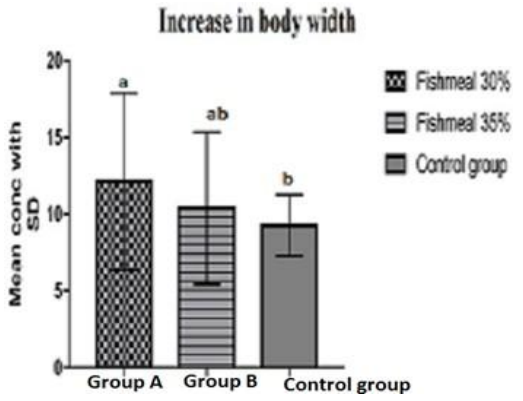

Figure8 Comparison of groups regardingpercentage of increased in body width

Table 2 Mean values of various growth parameters

Parameters	Fish meal 30% Group A (Mean ± Std)	Fish meal 35% Group B (Mean ± Std)	Control group Group C (Mean ± Std)
Increase in body width	12.12±5.78[a]	10.38±4.95[ab]	9.26±1.96[b]
Weight gain	112.08±16.69	117.90±22.17	117.20±9.06
Increase in body length	4.97±0.35[a]	5.28±0.94[a]	6.04±0.28[b]
Specific growth rate	1.249±0.129[a]	1.291±0.160[a]	1.292±0.069[b]

4. Discussion

Fishes hold huge importance in the economy of our country. Fisheries departments at various research and development organizations are actively contributing in developing high meat yielding fish varieties. A good number of fish meats are exported from Pakistan to various continents of the world giving a huge boost to the GDP of the country[11]. Main focus of scientists and researchers is to produce varieties of fishes which are more yielding and can better serve the needs of the population. Their size ranges from 12mm to15m[12].

Labeo rohita is one of the finest meats producing fish of the country and is significant in our overall meat production and revenue generation. More and more efforts are being put to increase its quality through good nourishment and proper feed. With optimum qualitative

characteristic of meat higher yield of fish in term of growth can be obtained[13]. The main target of aquaculture process is obtaining higher yield of fish. Fish growth means the increase in fish weight and fish length[14]. Physicochemical parameters like temperature, dissolved oxygen, pH and hardness of water in tanks for different feed treatments did not show any significantvariations[15].Present study consists of 11 parameters to see the response of Fish meal at different concentrations. These readings were analyzed to interpret the optimum level of growth of fingerlings. Results were statistically correlated with the previous studies to find a connection between our study and previous studies done. These results are pretty much in line with some of the previously conducted studies where more or less similar results were observed by various researchers. There results were statistically similar to those of ours result [16]. Water percentage response to Fishmeal 30% was minutely greater to that of Fishmeal 35% and our control group. Body composition of fish is used for best indication of physiological condition of fish. For the selection of appropriate fish it is very important for human to determine the information about these parameters[17].The protein and lipids are present in high concentrations if water is present in low percentage. Our result similar with the result of previous reports[18].

Similarly when we compared the Dry weight in three subgroups, we found that the dry weight was statistically significant in Fishmeal 35% and our control group as compared to Fishmeal 30% also found similar sort of results where fishmeal at 35% gave significant results[19].When Ash dry weight was critically compared, we found a significant difference in the subgroups. Fishmeal 30% gave a much higher yield of Ash dry weight as compared to two other sub groups. Similarly Ash wet weight was also statistically significant in Fishmeal 30% as compared to Fishmeal 35% and Control group. This result also matches with the previous result[20].

Furthermore, the organic content dry was higher in Fishmeal 35% as compared to Fishmeal

30 similar to previous studies[21]. Wet was more or less similar in all three subgroups having a non-significant P-valuesimilar to previous result [22]. In the first 6 parameters, Ash dry weight and organic content dry was statistically different between the three subgroups whereas other parameters showed more or less the same response in the parameters studied in three different subgroups. These findings were more or less the same in the previously conducted studies[23]and our results did not show any contradiction to the results. The percentage of dry Fat in fish meal 30% was greater than the percentage of fat at 35% and control group. So a significant difference is present between these groups. A statistically significant difference is present. The Fat percentage at fishmeal 30% was statistically significant. The percentage of fat at 30% was greater than other groups. This result was also similar with the previous result [16]. In present study, the percentage of dry protein at 35% fishmeal was higher in 35% fish meal. In control group and in 35% fishmeal similar percentage of protein dry weight was observed. So a significant protein dry weight percentage was present in 35% fish meal. But in other two less difference was observed so a non-significant difference was observed in 30% fish meal and in control group. At 30% dietary protein level Adult Indian carp showed maximum body composition. Fingerlings show greatest body composition at 35% dietary protein. But fry show maximum body composition at 40% dietary protein level. In present study at control group and at 30% diet show non-significant affects. Similar studies have been conducted by different scientists and some of the studies have shown similar results to what we observed while others had contradictory results as well. Our result also in aligns with the previous result[24]. Similar explanations were made by [25]who registered that fingerlings of *Catla catla* (Ham.) which is another herbivorous fish, efficiently utilize carbohydrates up to 35% but show poor growth on diets with dietary fat contents up to 12%. The protein sparing effect was only observed with carbohydrates. Our results are also in line with the previous findings which showed that the growth of walking catfish fed diets

containing different CHO/LIP ratios (0.02 to 43.00) differed significantly. They reported that the highest weight gain, significant growth ratio, feed conversion ratio, protein efficiency ratio, and energy retention values were observed in fish fed 27% dietary carbohydrates and 8% lipids corresponding to a CHO/LIP ratio of 3.38. Morph metric characters remain a key marker in the estimation of growth parameters of Fish when fed on different diets. Morph metric characters are best defined when seen in the context of Length, Weight and Size[26].

We compared the initial and final weight between the three sub groups. The difference between final and initial weight in Subgroup A was significantly higher to that of Sub group B and C.Length parameters were not significantly different between the three subgroups showing a non-significant P value. The duration of the study also defines the differences observed.Since ours research was a controlled study with fixed time duration so we need to interpret our results in a larger context and see if a more prolonged study would have shown different results. Varying degree of differences were observed in some of the sub groups but overall the five parameters studied for morph metric characters were more or less similar to each other. These results are in line with majority of the studies conducted on Morph metric characters of *Labeo rohita*[27]. Determination of age and growth contribute significantly to assess the yield potential of fishes in a water body and its characteristics. Harmonic development of important tissues results from simulation and retention of chemical components such as protein, lipids *etc*.

Conclusion

Proximate body composition analysis of *Labeo rohita* showed highest average value of fat ash and protein (in wet weight) T1 and T2 respectively and minimum average water percentage in T3 when fed with the 35% dietary crude protein. Minimum percentage of fat, ash and protein wet weight were 7.770, 2.125, 9.765 respectively and maximum average water percentage 82.099 was recorded at T1.In conclusion, the present study determined that

body composition of *Labeo rohita* fingerlings was best on T2 or 35 percent dietary protein level. This showed that 35 percent dietary protein level is more suitable and compatible for meeting the body requirement of fish.

COMPETING INTERESTS DISCLAIMER:

Authors have declared that no competing interests exist. The products used for this research are commonly and predominantly use products in our area of research and country. There is absolutely no conflict of interest between the authors and producers of the products because we do not intend to use these products as an avenue for any litigation but for the advancement of knowledge. Also, the research was not funded by the producing company rather it was funded by personal efforts of the authors.

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