Effect of prawn waste as feed ingredient on the growth, nutritional status and feed utilization on the fresh water fish, *Catla catla* (Hamilton, 1822)

Keerthika.R

(Reg.No:17PZO006)

The thesis submitted to

Avinashilingam institute for home science and higher education for women Coimbatore -641043

In partial fulfillment of the requirements for the

Degree of Master of Zoology

April, 2019

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Signature of the

Head of the department

22/4/19

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the Supervisor

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ACKNOWLEDGEMENT

First and foremost I wish to thank **God almighty** for endowing the investigator with immense blessings which helped to overcome the hurdles, paving way for the successful completion of the study.

I express my profound thanks to **Dr .T. S. Avinashilingam,** the founder and first Chancellor, **Dr. Rajammal P. Devadas,** Avinashilingam institute for home science and higher education for women, Coimbatore, for providing the opportunity to undertake the present research programme.

I gratefully acknowledge honorable chancellor Padma **Shri. Dr. P.R.Krishnakumar,** for providing all necessary amenities for the completion of my work.

I also extend my sincere thanks to Vice Chancellor **Dr. (Mrs.) Premavathy Vijayan, M.Sc., M.Ed., and Dip.Spl.Edn. M.Phil., Ph.D.** for providing the needed facilities during the study period.

I wish to extend my thanks to **Dr. (Mrs.) Kowsalya, M.Sc., M. Phil., Ph.D.,** registrar, Avinashilingam institute for home science and higher education for women, Coimbatore, for the encouragement given by her during the investigation.

My sincere thanks to **Dr.P.R. Padma, M.Sc., P.hD., P.G Dip .Adv. Bioinf.PDF at Germany,** Professor , Dean, School of Bioscience, Avinashilingam institute for home science and higher education for women, Coimbatore, for the support given for the successful completion of the work

My sincere thanks to **Dr.(Mrs.)N .Krishnaveni,M.Sc.,B.Ed.,Ph.D.** Professor and Head, Department of Zoology, Avinashilingam institute for home science and higher education for women, Coimbatore, for the academic support given for the successful completion of the work.

The investigator would like to express her sincere thanks to **staff members** and **laboratory assistants** in the Department of Zoology for their constant support.

I wish to thank my **PARENTS, Mr.K.Ramesh, Mrs.R.Sundari, and My brother Sathish.R** for their constant prayers, moral support, and motivation and without their support I would not have been able to reach this height.

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I wish to thank my **DEAR sisters**, **Bharathi.R. M.Sc, M.Phil and Yuvanthi.M, M.Sc,** for their moral support and motivation.

I wish to record my deep sense of gratitude to my **friends** for their motivation and timely help during the course of study.

Keerthika.R

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1. INTRODUCTION

Aquaculture is gaining considerable importance all over the world as a mean of improving world fish production which is currently on decline due to dwindling output from capture fishery. Expansion in aquaculture is strictly related to improvement in nutrition, and up gradation of fish husbandry practices- a challenge for future development in aquaculture. Mass scale fish production heavily depends on the amplification of proper feeding protocols to satisfy nutritional requirements of the cultured species.

Aquaculture has been one of the fastest growing branches of animal protein production during recent decades, and common carp is one of the most frequently farmed fish worldwide and also in Central and Eastern Europe. It is a highly esteemed fish species due to numerous desirable traits such as fast growth rate, good feed conversion ratio of both natural and supplementary feeds and relative resistance to poor environmental conditions and diseases. (Ljubojević1 *et al.,* 2016*)*

India is the second largest aquaculture producer in the world (Mandal and Das 2014) and its production in capture fisheries is about 43,01,534 tonnes and aquaculture is about 45,73,465 tonnes and total production is 8,87,49,999 tonnes in 2011. The success of commercial aquaculture operations depends on a variety of factors relating to the fields of biology, engineering and economics. One key biological component is the availability of suitable diets that are efficiently digested and provide the required nutrients for optimum growth (Mokolensang *et al.,* 2003). Sustainable and successful fish culture mainly depends upon the use of adequate, economically viable and eco friendly feeds. Feed is the single largest expenditure in semi-intensive and intensive fish culture and feed cost accounts about 30-70% of the total for operating expenditure in fish culture. (Yogesh Sachan *et al.,* 2016)

In general, growth of a fish is influenced by the quality and quantity of food materials available and consumed. Thus, variation in quality and quantity of food materials will affect growth rate of the fish. The qualitative and quantitative variations of natural food materials in a water body are under the influence of several a biotic and biotic factors. These variations could be known by qualitative and quantitative analysis of gut contents of a fish and/or by the estimation of gastro somatic index.

The aquaculture development and increase in per unit volume of water depends upon the use of high quality and balance artificial feed (Shaheen *et al.,* 2000). Major carps are an important freshwater fish species normally cultured in Asia particularly in the Indian subcontinent (Khan *et al.,* 2003; Abid and Salim, 2004; Khan *et al.,* 2004).

Aquaculture industry grows, the need for specialized feeds designed for particular production system and species increases proportionately. To date, nutritionists and feed manufacturers have concentrated their efforts on determining the feasibility and selection of wide variety of feedstuffs available to the feed industry for preparation of cost effective feeds. Currently appropriate quality fish feeds in desired quantity are not available. Majority of people use their own farm made feeds in variety of shapes and forms. Mash form is the most popular and convenient for use. Limited knowledge is available about nutrient requirements of different stages of fish and there is very poor know how about the suitable ingredients for balanced and effective feeds. Poor information on different digestive processes in fish alimentary canal is another hindrance to proper feed formulation for particular fish species. On account of which selection of different ingredients from large expanse available in the market is really a challenging job which can ensure viable growth, guaranteed health ultimately maintaining the economics of the fish business.

Fish is highly nutritive and rich source of animal proteins. For the improvement of fisheries and to achieve maximum yields from resources of fresh water, it is necessary to provide artificial feed, by which fish grows rapidly and attains maximum weight in shortest possible time. Among commonly used feed ingredients, fish meal is considered to be the best ingredients, due to its compatibility with the protein requirement of fish (Alam *et al*. 1996). Replacement of fish meal with cheaper ingredients of plant origin in fish feed is necessary because of rising cost and uncertain availability of fish meal (Higgs *et al*. 1995). Inclusion of feedstuffs with relatively high levels of carbohydrate in formulated fish feed is preferred in view of its protein-sparing action that may make the diet more cost effective (Hidalgo *et al*. 1993). According to Rumsey (1993), increased use of plant protein supplements in fish feed can reduce the cost of fish meal. The research has focused on utilizing less expensive and readily available resources to replace fish meal, without reducing the nutritional quality of feed (EI-Sayed 1999).

For commercial culture of fish, the formulation of low-cost balanced diet using locally available waste is needed. Recently fish meal has become the most expensive protein ingredient in aquaculture feeds. Many studies have shown considerable success in partially replacing fishmeal with soybean meal and other soybean products in diet for various fish species (Boonyaratpalin and Tunpibal 1998, Quartararo *et al*. 1998, Hernadez *et al* 2007).

Taxonomic position:

Phylum : Chordata

Class :Osteichthyes

Order : Cypriniformes

Family : Cyprinidae

Genus : *Catla*

Species : *Catla catla*

Compressed body is comparatively large with broad head. Mouth is wide, upper lip is thin and covered by skin of snout. Lower lip is moderately thick, broad and continuous post labial grove. Dorsal profile is more convex than that of abdomen. Gill opening is circular and body deepest at origin of dorsal (*Talwar and Jhingran, 2001*). Pectoral fins are long and extend to pelvic fins. Scales are conspicuously large, lateral line complete with 40-43 scales. In life, its colour is grayish on back and flanks, silvery-white below, fins dusky.

Catla is a fish with large and broad head, a large protruding lower jaw, and upturned mouth. It has large, grayish scales on its dorsal side and whitish on its belly. It reaches up to 182 cm (6.0 ft) in length and 38.6 kg (85 lb) in weight. Catla is a surface and midwater feeder. Adults feed on zooplankton using large gill rakers, but young ones on both zooplankton and phytoplankton. Catla attains sexual maturity at an average age of two years and an average weight.

The catla was formerly listed as the only species in the genus *Catla*, but this was a synonym of the genus Gibelion More recently, Fishes has moved this species to *Labeo*. This species has been frequently confused with the giant_barb (*Catlocarpio siamensis*) of south-east Asia, and the two taxa do bear an extraordinary resemblance to each other, especially in their very large heads.

Catla is one of the renowned and the fastest growing of major carps. This fish is found in ponds, lakes, ox-bow lakes, beels, streems, rivers, canals etc. It is non-predatory and its feeding is restricted to the surface and mid waters. It is abundantly found in the Buriganga, Padma, Meghan and other principal rivers of Bangladesh. It resides in fresh or brackish water, being found within the tidal influences (Day, 1989).

Catla is a preferred species in the carp farming community, owing to its fast-growing nature; hence in Andhra Pradesh it is stocked with rohu at about 10–20 percent and cultured in commercial systems. In other areas of the country, stocking densities for catla and mrigal are increased, with rohu being stocked at around 40–50 percent. Unlike Andhra Pradesh where the stocking percentages for rohu, catla and mrigal are strictly regulated, the availability of seed in other areas influences the species stocking composition and density (Nandeesha *et al*., 2013).

Catla is a cultured and highly growing species, if it get proper food it may be weighted at 4 kg at only one year. Very delicious food and supply a huge amount of protein for Bangladeshis people. So, its demand is extensive. According to Basu *et. al.*, (1993), its flesh contains 19.2% crude protein, 2-5% fat and 70% water. According to Ghosh *et al*. (1933), per garmme of its liver-oil contain 583 IV vitamin-A (*Rahman, 2005).*

Catla (*Catla catla*) is an economically important South Asian freshwater fish. Its higher growth rate and compatibility with other major carps, surface feeding habit, and consumer preference have increased its popularity in carp polyculture systems among fish farmers in India, Bangladesh, Myanmar, Laos, Pakistan and Thailand. The present study was conducted to estimate the digestibility of dry matter and nutrients from three non-conventional ingredients – Azolla, silkworm pupa and soybean meal by catla.

The Indian major carps *Catla catla, Labeo rohita and Cirrhinus mrigala* are the most important, prestigious, commercial fishes in India with a maximum market demand and acceptability as food by the consumers due to their taste. They contribute about 67% of total freshwater fish production (iclarm, 2001) and their total production is about 3.02 million tonnes (FAO, 2006).

Nandeesha, *et al.* (2013) stated that in most of the country, the stocking of undersized fish seed continues to be a major hurdle to increasing fish production. Although a huge amount of carp seed is easily produced by employing the Chinese and/or ‘jar’ hatchery systems, the survival from spawn to fry stage is mostly still less than 20 percent. The fry are then reared to

fingerling size if there is enough space, or else they are used for stocking once they reach 3–4 cm.

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In pond culture, Indian major carps are generally fed on a mixture of rice bran and various varieties of oil cakes. In later form formulated feed containing dietary protein is provided for these species. For increasing productivity, emphasis is being laid on developing cost effective complete supplementary feeds (shodhganga, 2014).

The common carp (*Cyprinus carpio*) has been one of the oldest domesticated species of fish for food.Carps are omnivorous, with high tendency towards the consumption of benthic organisms, such as water insects, larvae of insects, worms, molluscs and zooplankton. Digging in the bottom in search for food items results in turbid water. Zooplankton consumption is dominant in fish ponds where the stocking density is high.

Artificial feed plays an important role in semi intensive fish culture where it is required to maintain a high density of fish than the natural fertility of the water can support (Jhingran, 1991). The role of artificial feed in intensive fish farming cannot be ignored as nutritional requirements of fish depend upon the feed supplied. The quantity and quality of feed consumed have a pronounced effect on growth rate, efficiency of feed conversion and chemical composition of fish (Hassan *et al*., 1996; Jena *et al*., 1998; Erfanullah and Jafri, 1998). Worldwide, approximately 80% of carps and 65% tilapia are cultured without modern compound feeds (Naylor *et al*., 2000).

Global production of both cultured and captured shrimp was 3.34 and 3.12 million tonnes respectively (MPEDA, 2009). In India, shrimp farming has been practiced traditionally in the coastal states and comprises about 1,06,165 metric tonnes of shrimp in the year 2007-08. In West Bengal, 28000 metric tonnes of shrimp was produced during 2007-08 and from India 95% of the production was exported (mpeda, 2009).

Shrimp exported in 2010-11 was 6,12,505 tonnes (mpeda, 2012*).* Shrimp waste meal is a by-product of shrimp processing, consisting of mainly the head, gut and exoskeleton. It has

been processed for export and local consumption (Fanimo *et.al.,* 1996). A huge quantity of bio- waste is produced from these industries because shrimps are normally sold as head less and often in peeled form. This leads to an inevitable increase in waste produced by the shrimp industry as 40-50% of catch are largely of no use (Subasinghe, 1995). Shrimp processing industries are generating huge amount of waste in the country which produces more than 1,00,000 tones of industrial waste (Mathew and Nair, 2006). This bio-waste is then discarded in to the dumping areas resulting the major surface pollution in coastal areas.

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The shrimp waste contains biopolymers like chitin, chitosan and protein with high economical values. "Chitin" (N-acetyl-D-glucosamine) which is the major structural component of the exoskeleton of crustacean is believed to have a growth promoting effect. Shrimp meals have a poor palletizing ability because of high amount of chitin content (Randall and Dearing, 1978). Chitin oligosaccharides, such as chitopentaose and chitohexanoise have been reported to have an effect on the immunostimulation.

The shrimp head wastes are the source of protein, fat and minerals and can offer better energy source ingredients in fish feed industries. Research interest has therefore been awakened in the area of alternative feed resources which have comparative nutritive value but are cheaper than conventional protein sources (Agunbiade *et al., 2003*). The shrimp head waste is particularly rich in lysine, which make it an ideal supplement for cereals (Fanimo *et al.,* 1996). Shrimp head waste contains an average of 46.7% crude protein, 27.8% mineral matter (Frank, 1984) and 27% ash (Fanimo *et al.,* 1996). Shrimp head meal is high in calcium and phosphorus. Calcium carbonate is responsible for the securitization of the exoskeleton and represents most of the mineral matter in shrimp waste.

Fish is highly nutritive and rich source of animal proteins. For the improvement of fisheries and to achieve maximum yields from resources of fresh water, it is necessary to provide artificial feed, by which fish grows rapidly and attains maximum weight in shortest possible time. Among commonly used feed ingredients, fish meal is considered to be the best ingredients, due to its compatibility with the protein requirement of fish (Alam *et al. 1996).*

Protein is the most important nutrient promoting growth in animals. As protein is the costliest among the various ingredients used for the preparation of fish feeds, it is necessary to ascertain the quantitative requirement of dietary protein in order to reduce the cost of feeds.

Alternative protein sources and inclusion levels need to be optimized in aqua feeds to make aquaculture production efficient and cost-effective. Best aqua feeds are not defined by nutritional composition, but the degree to which a fish can digest, absorb and assimilate the nutrients. Therefore, digestibility determinations of various feed ingredients are very important to develop cost-effective diet formulations, evaluate ingredient quality, and limit the excretion of nutrients into the environment, which may cause environmental problems. Digestibility is the quantification of the digestive processes. It gives a relative measure of the extent to which ingested food and its nutrient components are digested and absorbed by the animal. Apparent digestibility coefficients (ADCs) vary between fish species and feed stuffs. Digestibility of individual ingredients in the compounded diet is considered as one of the important factors affecting the growth of fish and hence, it has been recommended to evaluate the digestibility of each ingredient before its incorporation in the diet.(Umalatha.H et al.2018).

Carbohydrate are non-essential in fish diet as the energy they supply can be replaced by protein or lipids and requirements can be met by gluconeogenesis (Hemre et al.,2002) however, carbohydrates and especially starches can be generally utilized well (Wilson,1982) and they can also enhance protein sparing effect for growth(Jauncey,1982). Their digestion and utilization is species dependant with carnivorous fish being less able to utilize them than omnivorous and herbivorous (Krogdhal et al.,2004). Carbohydrate digestion is an extracellular procedure that involves hydrolysis of complex carbohydrates in the stomach, intestine and caecum as well as in the brush-border section of intestines where enzymes such as maltase and sucrose are present (Harpaz *et al.,2001*).

The proportion of fish eating in india increased from 27.2% in 1987-88 to 39.7% in 1996- 97, assuming that this proportion would increase at least to 50%. The total fish eating population in indian by 2020 will be around 700 million.

The American Soybean Association (ASA), under the Soy-in-Aquaculture Program and in cooperation with a local farmer, Mr. Mukhlis, at Lake Maninjau, West Sumatra, Indonesia, conducted an 82-day feeding demonstration with common carp in cages. The objectives of the project were to demonstrate the feasibility of culturing common carp in low volume, high density (LVHD) cages, and to assess their performance on a soy-maximized feed.

Das and Ray (1989) used rice bran for their experiment conducted on growth performance of the *Labeo rohita* on duckweed incorporated pelle ted feed. Bindhu*et al.* (2002) utilized rice bran as feed ingredient in their experiment to report the impact of dietary protein on growth, feed utilization and body composition of *Puntius parraha.* Sharma and Kishan (2006) utilized rice bran as a feed ingredient to find out the growth performance of *Cyprinus carpio* fingerlings fed with various fermented meals. Singh *et al.* (2006) used rice bran for the development of supplementary fish feed from low cost indigenous materials.

Oilcakes are also widely used for substitution of fish meal in diets of fishes. Groundnut oilcake is mostly used in fish diets due to its easy availability in market. Plant protein ingredients especially soyabean and ground nut oil cake have successfully substituted fish meal with nutritional and economic benefits (Fagbenro, *1999*).

Feed taken in by fish undergoes through several mechanical and chemical processes. Once chewed and broken down into small pieces, feed is exposed to various enzymes which include proteases, lipases and amylases (Caruso *et al.,* 2009). The ability of fish to metabolize a diet depends on the availability of appropriate digestive enzymes, which mediate specific degradation pathways modulating both physical and chemical nature of foods (Phillips, 1969). The measurement of specific activities (proteases, amylases and lipases) may provide information about the whole digestive capability and efficiency of fish species under culture (Buddington *et al.,* 1997).

Animal based feed ingredients like fish meal, clam meal, crab meal, et the major source of protein in fish feeds. However, recently the major problem confronting the fish farming industry is the increasing cost of animal based feed ingredients.

The replacement of animal based feed ingredients with locally available and cheaper plant feedstuffs has become highly essential for the future development of the aquaculture sector. For culturing fish in captivity, obviously nothing is more important than the sound

nutrition and adequate feeding. (yogash sachan *et al.*,2016).

Possible uses of these wastes in animal feed preparation have been suggested by Patel *et al.*4. As of now, relatively very little emphasis has been given to the use of these fruit / vegetable processing wastes, which are very cheap and easily available in fish feeds. *(*Yogesh Sachan *et al.*,2016) .

Food and feeding habits of carps have been a field of interest to fisheries researchers since very long. Natarajan and Jhingran (1961) studied the food habits of *Catla catla* and reported a zooplankton dominated food preference for *Catla catla*. Hora and Pillay (1962) reported the feeding habits of *Catla catla*. Khan and Jhingran (1975) have given a report on the food and feeding habits of an Indian major carp *Labeo rohita* (Ham). Rajgopal (1978) described the foods and feeding habits of some commercial fishes from the Tungabhadra reservoir. Sunder *et al.* (1990) studied the food and feeding habit of the *Cyprinus carpio* var. *specularis* from Dal lake (Kashmir) in relation to Gastrosomatic index, condition factor and length-weight of fish and reported that the monthly fluctuations in feeding activity and Gastrosomatic index (GaSI) is in agreement with each other.

The feed ingredients,protein source is one of the expensive ingredients in the formulated feed. Fish meal is still an essential ingredient in the diets and it is also an expensive feed ingredient compared to other protein sources and thus represents a significant cost element in feed and production cost. This has necessitated the search of alternative sources available locally in the country. In this context, use of certain potential aquatic weeds offer excellent scope as the nutrient laden materials are naturally grown in the entire country without much agronomic care. Many aquatic plants such as *Eichornia crassipas, Hydrilla verticillata, Salvinia aculata, Ipomea aquatica, Pistia* spp. etc. contain fairly high amount of protein. Aquatic plants also contain high amount of vitamin E, vitamin C and mineral elements required for normal growth and development of fish.

THE OBJECTIVES OF THE PRESENT STUDY ARE

1. To prepare the fish feed with prawn waste in different composition with basal ingredient. (Control and 3 different feed)
2. To estimate the biochemical compositions such as protein, carbohydrate, lipids and amino acid in the control and experimental feed.
3. To estimate the growth rate (length and weight) of *Catla catla* by feeding the control feed and experimental feed prepared form basal ingredients and prawn waste in different proportions.
4. To estimate and compare growth rate (length and weight) of *Catla catla* in the control and three different treatments during 60 days of the experimental period.
5. To estimate and compare the biochemical composition (protein, carbohydrate, lipids and amino acid) in control and fishes grown in three different feeds.
6. Comparison of feed utilization efficiencies – Feed Efficiency (FE), Feed Conversion Ratio (FCR) and Protein Efficiency Ratio (PER) in the fishes grown in the control and three different treatment.

2.REVIEW OF LITERATURE

The available literature pertaining to the present study entitled **"Effect of prawn waste as feed ingredient on the growth, nutritional status and feed utilization on the fresh water fish *Catla catla* (Hamilton,1822.)**" is given bellow.

FISH FEED FORMULATION AND ITS EFFECT ON GROWTH

. Nadeesha *et al., (2001)* noticed the growth performance of two Indian major carps, *Catla catla and Labio rohita* fed diets containing different level of *Spirulina plantesis* replaced fish meal protein from the standard diet at 25%, 50%, 75% and 100% levels. The result showed that no significant difference in the final weight attained by catla at all level of Spirulina feed compared to the fish meal based control diet. However the replacement of fish meal by more than 25% spirulina resulted in significantly superior growth of rohu.SGR, protein efficiency ratio was improved rohu with higher levels of Spirulina inclusion while in catla did not differ significantly from the control diet. In both species the digestibility of dry matter protein and fat was found to improve marginally with increasing levels of *Spirulina* incorporation. Fat content was significantly higher in *Spirulina* diets. The result of the present study clearly demonstrates that S.*platesis* could be exploited as a protein source for incorporation in Indian major carp diets. Cost effective technologies are available for the cultivation of S.*platesis* using various organic wastes (Venkataraman and Becker, 1986) and these techniques could be effectively used for large scale production of *Spirulina*, they concluded that usefulness of spirulina for partial or complete replacement of fish meal in the diets of Catla and Rohu.

Manjappa *et al., (2002)* studied the growth performance of common carp, fed varying lipid levels through low protein diet, with a note on carcass composition and digestive enzyme. Shearer et al. (2006) reported the effect of growth rate/ body size and low lipid diet on the incident of early sexual maturation in juvenile male spring Chinook salmon (*Oncorhynchus tshwytschar*). Borgeson *et al., (2006)* reported the effect of replacing fishmeal and oil with simple or complex mixtures of vegetable.

Haghbayan and Mehrgan, (2015) studied the effect of replacing fish meal in the diet with enzyme-treated soybean meal (hp310) on growth and body composition of rainbow trout fry. The result showed that diets containing 75% and 100 % HP310 had significantly higher feed

conversion ratio and lower feed intake, weight gain and specific growth rate compared to fish feed diets containing higher levels of fish protein ingredients($p<0.05$).

Chhay et al.,(2010) studied the effect of sun-dried and fresh cassava leaves on growth of Tilapia fish fed based diets of rice bran mixed with cassava root meal. The experiment was conducted for 100 days. Daily gain in weight, length and the ratio weight length after 100 days of growth, did not differ treatment, water quality parameter were not affected by the treatments.

Muzaffar *et al*., (2012) stated that proteins are the major organic materials in most fish tissue, and form an important component of the diet. One of the major requirements of fish culture is the efficient transformation of dietary protein into tissue protein (Webster and Lim, 2002).

Bahrevar and Faghani- Langroudi., (2015) observed the effect of fish meal replacement by blood meal in fingerlings rainbow trout (*Oncorhynchus mykiss*) on growth and /fillet quality traits. These result showed that blood meal is not a suitable protein source as fish meal replacement for fingerlings rainbow trout. Growth, nutrient utilization and body composition were either not improved or were significantly influenced by gradually replacing fish meal by blood meal.

Ajani *et al* (2016) observed the total replacement of fish meal by soybean meal with or without Methionine fortification in the diets of Nile Tilapia, *Oreochromis niloticus.* Result showed that mean Weight Gain (MWG) and increase in length were significantly high ($p<0.05$) in PSM (22.77 g and 16.90cm respectively) and least NSM (17.54 g and 14.63cm,respectively) Specific growth rate (SGR) values were high in PSM(1.62) and only soybean meal (OSM+M) (1.55) and least in NSM(1.48). Fish fed OSM+M had the highest parked cell volume (PCV.20.66%), hemoglobin (Hb,7.87gdl^{-1}) and least White blood cell (WBC,$3.92*10^4mm^{-3}$),while NSM gave the least Hb (5.37gdl^{-1}) and highest WBC($3.92*10^4mm^{-3}$).

Ramamoorthy *et al*.,(2016) studied the proximate ,amino acids and fatty acid composition the marine carbs from the southeast coast of India . The proximate composition (%) results showed significant difference ($p<0.05$) between various species. Higher amount of protein (22.57%) and carbohydrates (1.17%) contents were detected in *C.lucifera* when compared then *P.pelagicus* and *P.glatiator*. while lipids content was higher in *P.pelagicus* (2.15%) and lower recorded in *C.lucifera*.

Eswar *et al*.,(2016) conducted an experiment to investigate the biochemical composition and preliminary qualitative analysis of marine clam *Gafrarium divaricatum* from Mumbai, west coast of India. The result of biochemical analysis showed very high protein content (26.32%), Carbohydrates (11.23%) and lipid (1.29%). the protein consists of ten essential and nine non essential amino acid which are as follows (Lysine 14.36% ,Histidien 9.02%, Methionine 8.92%) and(Alanine 5.94%. aspartic acid 4.98% , Asparagines 3.70%,Tyrosine 3.52% and Proline 3.21%) are the predominant essential amino and non essential amino acids.

Djissou *et al.*,(2016) investigated the complete replacement of fish meal by other animal protein source on growth performance of *Clarias gariepinus* fingerlings . This result showed that the survival and feed utilization were not significantly affected by the ratio between earthworm meal and maggots meal in the diets . Lipid content was higher in carcass and fillet of fishes fed earth worm- and maggots meals-based diets than that of those fed fish meal – based diet.

Soare *et al*.,(2015) observed the replacement of fish meal by protein soybean concentrate in practical diets for pacific white. Result showed that after 42 days, the weight gain of shrimp fed diets with 0 and 25% protein replacement was higher than that observed in shrimp fed 100% replacement , and there were no differences among those fed the other diets. Feed efficiency and survival did not differ among shrimp fed different protein replacements. There was a negative linear trend for growth parameters and feed intake as protein replacement with soybean protein concentrate increased. Fish meal by – product can be replaced by up to 75% soybean protein concentrate, with no harm to the growth of pacific white shrimp.

Kathirvel *et al* ., (2014) studied the, proximate ,amino acids and fatty acid and minerals analysis of box crab, *Calappa lophus* (herbst, 1782) from parangipettai, southeast coast of India. The *Calappa lophus* appendage and body region was average quantity of protein, carbohydrates, fat, ash and moisture in the level of (22.19 gm, 21.45 gm), (8.34 gm, 5.45 gm), (55.18 gm 61.19
gm), (2.56 gm, 2.98 gm), (73.11 gm, 82.98 gm) respectively.

Rad *et al.*,(2013) conducted on experiment to evaluate the effect of different levels of dietary supplementation of *Saccharomyces cerevisiae* on growth performances, feed utilization body biochemical composition of (*Oreochromis niloticus*) fingerlings, four diets containing supplementation at levels 0, 0.5, 1 and 29 kg fed to fingerlings of Nile tilapia (5.01±0.21g) in replicate tanks twice daily to apparent satiation for 56 days. The result of this experiment

indicated clearly that the supplementation of *Saccharomyces cerevisiae* (1.0g yeast Kg diet) enhanced the growth performance and feed utilization of *Oreochromis niloticus* fingerlings.
Yilmaz (2012) investigated the effect of garlic and garlic oils on hematological and biochemical variables of sea bass *Dicentrachus latex.* Result showed that the red blood cell count, hematocrit, hemoglobin concentration, mean corpuscular volume, , mean corpuscular hemoglobin and concentration were not significantly affected by herb oil exposure. And sea bass exposed to the 0.005- ml/l garlic ol- garlic oil mixture (0.005 or 0.01 ml/l garlic oil or to the 0.001-ml/l mixture. The serum lipase level decreased and the cholesterol level increased in fish that were exposed to0.02-ml/l garlic oil.

Jegede (2012)was conducted an experiments to determine the effect of garlic on growth, nutrient utilization resistance ad survival of tilapia zillii fingerlings. Result
Showed that the garlic diet increased the better growth rate and also the survival rate and also the survival rate of tilapia zillii was higher in all the treatments. Apparent protein digestibility (APD) increases with increase in the inclusion level of garlic diet.

Harikrishnan and Balasundaram (2011) studied an impact of plant products on innate and adaptive immune system of cultured finfish and shell fish. He reported that the application of antibiotics and chemical resistance and consumer reluctance. Therefore immunotimulants such as glucan ,chitin, lactoferrin, levamisole ,and some medicinal plant extracts or products have been used to control fish and shellfish disease .In this regard the medical plant extracts and their products act as immunotimulants modulating the immune response to prevent and control fish and shellfish diseases. The immunotimulants mainly facilitie the function of phagocyte cells
,increase their bactericidal activities, and stimulate the natural killer cells, complement, lysozyme activities, and antibody responses in fish and shellfish which confer enhanced protection from infectious diseases. Plants or their byproducts are preferred since they contain several phenolic, polyphenolic, alkaloid, quinine, terpenoid, lectine and polypeptide compounds many of which have been shown to be very effectives alternatives to antibiotics, chemicals, vaccines and other synthetic compounds. In aquaculture the herbal medicines are also know to exhibit anti- microbial activity, facilitate growth, and maturation of cultured species besides under intensive farming the anti-stress characteristics of hergs will be of immense use without posing any environmental hazard. Administration of herbal extracts or their products at various concentrations through oral (diet) or injection route enhance the innate and adaptive immune

response of different freshwater and marine fish and shellfish against bacterial,virual and parasitic diseases. Even an overdose of immunotimulants may induce immunosuppression without side effects but helps to reduce the losses caused by disease in aquaculture.

Jegede (2012) was conducted an experiments to determine the effect of garlic on growth, nutrient utilization resistance ad survival of tilapia zillii fingerlings. Result
Showed that the garlic diet increased the better growth rate and also the survival rate and also the survival rate of tilapia zillii was higher in all the treatments. Apparent protein digestibility (APD) increases with increase in the inclusion level of garlic diet.

Citarasu (2010) reported that hormones antibiotics, vitamins and servile other chemicals have been tested in the aquaculture operations for various remedies and the alternatives herbalbio-medicinal products in the aqua cultural operation, that have the characteristics of growth promoting ability and tonic to improve the immune system acts as appetite stimulants. They increase consumption, include maturation and have antimicrobial capability and also anti stress characteristics that will be of immense use in the culture of shrimps and other fin fishes without any environment and hazardous problems.

Reddy *et al*., (2014) investigated the study on the natural status of the fresh water prawn *macrodrachium rosenbergii* from Nellore coast, India. Fifteen different samples were selected randomly for the estimation of the proximate composition. The average values of the proteins, carbohydrates, lipids, ash and moisture ash in cultured and frozen prawns were recorded ash 74.24± 0.49, 5.50±0.34, 9.09±0.09, 9.71±0.19, 77.14±0.19 and 60.55±0.35, 8.23±0.18, 7.98±0.13, 21.61±0.42 and 74.93±0.23, respectively. The higher amount of proteins, lipids was identified in the cultured prawn.

Varadarajan et al., (2014) observed the proximate composition and minerals content of fresh water crab *spiralothelphusa hydrodroma* (herbst, 1794) from parangipettai, south east coast of India. The result showed that the parameters like protein, carbohydrates, lipids, moisture and ash and minerals of calcium, magnesium, potassium, sodium, iron, copper and zinc were maximum in cephalo thorax and minimum in swimming and walking legs.
The comparative proximate amino acids, fatty acids composition in *Peanuts monodon* (Fabricius, 1798), Fenneropenaeus indices (H.Milne Edwards,1837) and *Aristeus virilis* (Bate, 1881) collected from the nagapattinam landing center (southeast coast of India). The shrimps showed a significant ($p<0.05$) result and varying concentration of protein, lipid, carbohydrates and

moisture. Higher concentration of protein was found in A. virilis (17.25) followed by F. indices and P. monodon (karupasamy et al., 2014).

Puga-lopez (2013) observed the physiochemical proximate composition, microbiology and sensory analysis framed and wild harvested white shrimp *Litopenaeus vannamei* tissue. Both, the framed and wild white shrimp muscles supplied a good source of protein and polyunsaturated fatty acids. The physiochemical composition, microbiological and sensory properties could be associated to their origin and handling. The wild shrimp tended to have a better proximate composition than the framed shrimp, due to the availability of a grater diet variety in their environment.

Siva reddy et al (2013) observed the proximate composition of the prawn, *macrobrachium rosenbergii* from Andhra Pradesh coast, India. Ten different samples were selected from both the sexes for the experimentation. Protein, moisture, fats and ash contents in female and male *macrobrachium rosenbergii* were measured as 25.92±0.40, 75.96±0.15, 5.01±0.42, 1.84±0.05 and 23.14±0.47, 74.16±0.75, 3.35±0.61 and 1.52±0.09 respectively. There
was significant different between male and females in case of proteins, moisture, fat and ash. Gunalan et al (2013) studied the biochemical composition in *Litopenaeus vannamei* the results rewarded that *L. vannamei* species can be considered as a good source of fatty acids as well as protein rich space. Keramah, (2013) investigated the effect of replacement of fish-meal with crab-meal on growth and feed utilization of African giant cattle fish heterobranchus longifilis fingerlings. The result showed that fish feed with 40% crud protein (CP) fish-meal diet had the best growth as indicated in mean weight gain, 6.99±1.70g, specific growth rate, 4.05±0.19% day- 1, feed conservation ratio, 1.12±0.06 and protein efficiency ratio of 2.28±0.12. Diets with fish- meal (FM) performed better than crab meal (CM) diet. Percentage survival rate for FM and CM containing diets varied between 91.0±1.63 and 100% and the condition of fish was not significantly different ($P>0.05$). A part from whole body levels of CP in FM feed fish, moisture and fat contents were unaffected by dietary treatments.

Hakim *et al* ., (2008) studied the effect of replacing soybean meal protein by other plant protein sources on growth performances and economical efficiency of mono sex Nile Tilapia (*Oreochromis niloticus*) cultured tanks. The result showed that replacement of 30% soy bean meal protein by sesame cake protein improved significantly final body weight of Nile Tilapia.

Amar *et al.,* (2008) investigated the efficacy of fermented prawn shell waste as a feed ingredient in Indian white prawn, *Fenneropenaeus indices*. The result showed the enhanced growth could be observed in prawn fed with Feed134 and Feed 124, incorporated with the fermentation products generated using Bacillus spp. The percentage survival rate of prawn after 7 days of challenges was found to be highest for groups feed diet F111 incorporated with fermentation products generated using Bacillus spp.

Soltan and laithy (2008) conducted the experiment to determine the effect of probiotics and some spices as feed additives on the performance and behavior of the Nile Tilapia (*Oreochromis niloticus*). Result showed that supplementation of the basal diets with probiotics *B.subtilis* or Biogen and spices (garlic and fennel) significantly improved survival rate of Nile Tilapia. A combination of *B.subtilis* and garlic and fennel showed the best fish survival rate but did not significantly differ from those recorded for the diets supplemented with probiotics or spices alone. Feed intake .feed utilization and growth performance of Nile Tilapia including final body weight, final body length, weight gain, specific growth rate, feed conversion ratio and protein efficiency ratio were significantly high.

Mandeville *et al.,* (2009) observed the proximate analysis of the commercial shrimp waste indicated the presence of 94.6% protein and 4.2% fat on dry basis. Abdullash *et al.,* (2009) reported the proximate fatty acids (FA) and element compositions of two shrimp species, deep seawater rose shrimp *(Parapenaeusn longirostris)* and red shrimp *(Plesionikamartia).* Amount of lipids in *P.martia* was found as 1.1 and 2.61% proportion of lipids in both shrimp was lower than that of marine fish.

Hasanuzzaman *et al.,* (2009) obtained the optimum replacement of fish meal with soybean meal in the diet for *Macrobrachium rosenbergii* (De Man 1879) cultured in low saline water. The result showed that maximum average weight (21.72±0.20g) was observed in prawn fed on diet-3, while the minimum weight (21.72±0.61g) was measured for diet – 1. The prawn fed on diet -3 displayed higher specific growth rate (1.31±0.27%. /day) and survival (71.97%) than the control group. Protein efficiency ratio (1.51, PER) and feed conversion ratio (2.52, FCR) in prawn fed diet -3 were higher and lower respectively, but the difference was insignificant compared to the other treatments.

Bhavan *et al.*, (2010) investigated the proximate composition and biochemical constituents in the muscle of adult male and female prawn of *Macrobrachium rosenbergii* collected from two different culture sites. The pro[portion of total protein amino acids lipid, fatty acids , carbohydrates and RNA was found to be higher in the female prawns than in the males. In contrast, the proportions of moisture and ash contents were higher in male prawns when compared with females. The level of DNA was found to be unchanged in both male and female prawns.

Yang *et al.,(*2004) investigated the effect of replacement of dietary fish meal by meat and poultry by-product meal on growth and feed utilization of gibel carp, caresses auratus.The results showed that feeding rate for that meat and bone meal (MBM_{50})group was significantly higher than for other groups expect the poultry by-product meal (PBM_{50})group ($p<0.05$).Growth rate in the MBM_{15} group was significantly higher than that in the control ($p<0.05$) , whole there was no significant difference in the growth between the control and other groups($p>0.05$). Feed efficiency and protein efficiency ratio in MBM_{15} was significantly lower while that in MBM $_{15}$ was significantly higher ($p<0.05$). replacement of fish meal by MBM at 500 gkg^{-1} protein significantly decreased apparent dry matter digestibility (ADCD) and gross energy (ADCE) while apparent protein digestibility(ADCP) was significantly decreased by the replacement of MBM or PBM($p<0.05$). The result suggest that MBM and PBM could replaced up to 500g kg^{-1} of fish meal protein in diets for gibel carp without negative effects on growth while 150 g kg^{-1} replacement by MBM protein improved feed utilization.

Ali Aberoumand (2011) conducted the study to compare the proximate composition and energetic value of three selected marine fishes and prawn. Prawn contained the highest protein compared to other fishes. The highest fat content among the three studied fish species was in 10 yellow fin Tuna fish at 6.89+2.76% while the fat content of prawn was1.06%. Yellow fin Tuna fish contained the highest energetic value of 33.6kcal/g of the entire sample.
Analysis of the trends in the state-wise production of primary agricultural commodities and in the state-wise aquaculture or carp-culture production shows that those states that are the major producers of agricultural commodities have also established themselves as major producers of aquaculture commodities. This clearly shows that feed ingredient and feed availability play a pivotal role in the development of freshwater aquaculture, mainly the carp-farming sector of India (Nandeesha, M.C., 2013).

Kumar Lalit *et al* .,*(2015)* investigate the food and feeding habits of an Indian major carp *Catla catla* from Udai Saga, Udaipur. On the basis of qualitative and quantitative analysis of gut contents, *Catla catla* has been categorised as planktivorous (zooplankton feeder).

3. MATERIALS AND METHODS

Experimental animal

Freshwater fish, *Catla catla* were collected from the Pollachi district, Aliyar. The fishes were kept in large aquarium tanks and acclimatized to laboratory conditions for two weeks prior to the beginning of the experiment. During this period they were fed with control (.Rice bran, Soya bean meal, Coconut oilcake, Egg albumin, Sunflower oil, Tapioca flour, Vitamin E). The tank was cleaned periodically and water was infused at regular intervals to ensure sufficient oxygen supply to fish.

Experimental feed and feed formulation

The feed ingredients fish meal, soy meal, groundnut oilcake and rice bran was purchased from local market, sun dried and separately powdered using lab mixture grinder. The diets were prepared using such as FM, soybean meal, groundnut oilcake, rice bran, sunflower oil, egg albumin, tapioca flour and vitamin E and served as control feed. The FM in the basal diet was replaced with dried prawn waste at the following concentrations, 50%, 75%, and 100% and served as experimental feeds. The FM served as control feed. Egg albumin and tapioca flour were used as binding agents. Vitamin E and a pinch of salt were also added (Table 1).These substances were mixed with hot water and made into dough and boiled for 30 minutes. Then the boiled feed was taken out side, cooled and finally made into noodles. The moist noodles were dried for 3-5 days in the sun to prevent fungal attack and were broken into pieces of 0.5cm in length. Three different diets and control were prepared for the present investigation

TABLE-1

Experimental Treatments and control

Ingredient	Control (BI+FM)	Diet-1 (BI+FM+R50)	Diet-2 (BI+FM+R75)	Diet-3 (BI+FM+R100)
Groundnut oil cake	25	25	25	25
Rice bran	10	10	10	10
Soybean meal	25	25	25	25
Egg albumin	6	6	6	6
Sunflower oil	2	2	2	2
Tapioca flour	6	6	6	6
Vitamin E	1	1	1	1
Fish meal	25	12.5	6.25	0
Prawn waste	-	12.5	18.75	25
Total	100	100	100	100

BI- Basal Ingredient, FM- Fishmeal, R- Replacements

Experimental design

The experiment was carried out in three different treatments in three replication and control. Each trough contained about eight individuals in the control and in three treatments. Prior to stocking, biochemical composition were analyzed in the fish *Catla catla.* The water level in each trough was maintained throughout the experimental period.

During this time, experimentally prepared prawn waste feed was given to the fishes. Before feeding biochemical composition such as protein, carbohydrate, fat, amino acid, moisture and ash were calculated in prepared fish feeds. Every fifteen days, growth performance were analyzed in terms of length and weight (%) and biochemical composition such as protein, carbohydrate, fat, amino acid moisture and ash were calculated in the muscle tissues of the *Catla catla* fishes in control and three different treatment before and after the experiments. The medium was changed two days once in order to remove the faucal and unconsumed wastes.

Evaluation of growth performance and feed utilization efficiency

The evaluation of growth performance and feed utilization efficiencies in *Catla catla* was performed by using a number of criteria. The growth and feed utilization efficiencies such as feed efficiency (FE) , feed conversion Ratio (FCR) and Protein Efficient Ratio(PER) was determined in the present investigation.

Growth measurement

Growth performance of fish was determined in terms of initial and final individual fish weight (mg/g) and length (cm/m)

1. Growth performance

Calculation

$$\text{Percentage of weight gain} = \frac{\text{Final weight} - \text{Initial weight}}{\text{Initial weight}} \times 100$$

$$\text{Percentage of length gain} = \frac{\text{Final length} - \text{Initial length}}{\text{Initial length}} \times 100$$

2. Feed utilization efficiencies

$$\text{Feed Conservation Ratio (FCR)} = \frac{\text{Food fed *}}{\text{Weight gain**}}$$

*- as fed basis i.e. Dry weight

** - Wet or fresh weight gain

$$\text{Protein Efficiency Ratio (PER)} = \frac{\text{Live weight gain (g)}}{\text{Protein consumed (g)}} \times 100$$

$$\text{Protein Value (PPV)} = \frac{\text{Body protein gain Productive}}{\text{Protein consumed}} \times 100$$

$$\text{(g) Feed Efficiency (FE)} = \frac{\text{Weight gain}}{\text{Food fed (g)}}$$

Biochemical composition

The moisture content (%) was estimated by subtracting the dry weight of the sample from the wet weight (APHA, 2005). Total protein (mg/g) content of the samples was estimated using bovine serum albumin as a standard (Lowry *et al*., 1951). Carbohydrate (mg/g) content was estimated using the methodology of Dubois *et al*., 1956 with glucose as a standard. Lipid (%) content was accessed by adopting Folch *et al*., 1957 method. Amino acid (mg/g) content was estimated using methodology of Moore and Stein, 1948 method. To determine the ash content (APHA, 2005), samples were incinerated in muffle furnace at 600°C for 4 hours and. All the parameters were determined (in triplicates) on dry weight basis.

1. Estimation of total protein (Lowry *et al.*,1951)

Principle

Protein reacts with folin ciocalteus reagent to give a colored complex. The colour formed is due to the reaction of alkaline copper with the protein at the reduction of phosphomolybdate by tyrosine and tryptophan present in the protein. The intensity of the colour depends upon the amount of these aromatic acids present and thus varies for different proteins.

Reagents

1) 80% ethanol: 80 ml of ethyl alcohol was dissolved in 20 ml distilled water.

2) NaOH (0.1N): 400 mg of NaOH was dissolved in 10 ml of distilled water.

3) NaOH (1N): 4 g of NaOH was dissolved in 100 ml of distilled water.

4) Solution A : 2 g of sodium carbonate was dissolved in 100 ml of 0.1N NaOH.

5)Solution B: Solution B was prepared by dissolving 500 mg of copper sulphate in 1% sodium potassium tartarate (1 g of sodium potassium tartarate in 100 ml of distilled water).

6) Solution C: Solution C was prepared by mixing 50 ml of solution A with 1 ml of solution B.

7) Folin ciocalteus reagent: Folin ciocalteus reagent was prepared by mixing 1 ml of folin ciocalteus reagent with 1 ml of distilled water.

8) Blank: 5 ml of solution C,0.5 ml of 1N NaOH and 0.5 ml of folin ciocalteus reagent served as the blank solution served as the standard.

9) Standard: Bovine serum albumin (BSA) at the concentration of mg/ml and different dilutions form this stock solution

Procedure

Standard preparation

For plotting the standard curve, a set of standard were run 0.1, 0.2, 0.3, 0.4,…..1 ml of standard solution was taken in a series of test tubes. The volume of each test tube was made up to 1 ml with distilled water. 5 ml of alkaline copper reagent was added, mixed and allowed to stand for 10 minutes at room temperature. 0.5 ml of 1N folin-ciocalteus reagent was then added to each tube and shaken well, the blue colour developed was read at 720 nm after 30 minutes along with the reagent blank in a colorimeter.

A standard graph for this was plotted with corresponding OD value on Y-axis and standard concentration on X-axis.

Sample preparation

100 mg of feed was homogenized with 2 ml of 80% ethanol. Then it was centrifuged at 5000 rpm at 4°C for 15 minutes. The precipitate was dissolved in 1N NaOH and made up to 5 ml. from this, 0.5 ml was taken and then 5 ml of folin ciocalteus reagent was added and the intensity of the colour developed was read at 660 nm in a spectrophotometer.

Calculation

$$\text{Protein present in the sample (\%)} = \frac{\dfrac{\text{OD of the sample}}{\text{OD of the standard}} \times \text{Conc. of the standard (mg)}}{\text{Weight of the sample (mg}} \times 100$$

2. Estimation of carbohydrate (Roe *et al*.,1995)

Principle

Sulphuric acid hydrolyzes the die and oligosaccharides into monosaccharide's and converts the monosaccharide's into furfural or furfural derivatives, which react with anthrone and produces a complex colored product.

Reagents

80% ethanol: 80 ml of ethanol was dissolved in 20 ml of distilled water.

Anthrone reagent: 200mg of anthrone powder was dissolved in 50 ml cold concentrated sulphuric acid. To this, 0.5 ml of thiourea was added to stabilize the colour.

Standard: 100 mg of D-glucose was dissolved in 100 ml of saturated benzoic acid and different dilutions from this stock solution served as a standard.

Procedure

Standard preparation

For plotting the standard curve, a set of standard were run 0.1, 0.2, 0.3,……,1 ml in a series of test tubes. The volumes in each test tube were made up to 1 ml with distilled water. 5 ml of anthrone reagent was added. A blank containing 1 ml of distilled water and 5 ml of anthrone reagent was also kept.

Sample procedure

100 mg of feed / tissue was taken and it was homogenized well using 2 ml of 80% ethanol. Then it was centrifuged at 5000 rpm for 15 minutes at 4°C. To the clear supernatant (0.5ml), 4 ml of anthrone reagent was added and the test tubes were kept in a boiling water bath for 15 minutes. The test tubes were taken out and kept in a dark room for 10 minutes and finally the colour developed was measured at 620 nm in a spectrophotometer.

Calculation

$$\text{Carbohydrates present in the sample (\%)} = \frac{\text{mg of glucose}}{\text{Volume of test sample (mg)}} \times 100$$

3. Estimation of lipid (Folch *et al*.,1957) Principle

The quantitative determination of lipid by gravimetric method using chloroform methanol mixture (3:1).

Reagent

Chloroform methanol mixture (3:1)

Procedure

100mg of fish sample was weighed separately and ground well 5 ml of chloroform methanol mixture. The homogenate was centrifuged, taken in a small weighted beaker and this beaker was placed inside a large beaker filled with water along the side and kept overnight in hot air oven without any disturbance. In between the methanol (with dissolved protein layer) and chloroform (with dissolved fat) a white precipitate was formed. The methanol layer was removed without disturbing the chloroform was evaporated in oven at above 60°C. The beaker was weighted and the different between the final and initial weight of the beaker gives the lipid content of the tissue.

Calculation

Lipid present in the Sample (%) = Final weight – Initial weight

4. Estimation of ash and moisture (APHA, 2005)

Principle

The sample was allowed to dry by kept in desiccators. The different between the wet weight of the tissue and its dry weight give the amount of water present in the fresh tissue. On heating the dry material to higher temperature all the organic constituents were burnt leaving only the inorganic constituents in the form as ash.

Procedure

Known amount of wet sample was taken individually on previously weight concave glass and they were kept in desiccators, maintaining 0.5% relative humidity. Dry the tissues in the desiccators till they reached a constant weight. Then the dried materials were transferred individually in silica crucible and kept in a muffle furnace and heated at 550-600°c for 4 h.

Calculation

$$\text{Moisture} = \frac{\text{Wet weight (g)} - \text{Dry weight (g)}}{\text{Wet weight (g)}} \times 100$$

$$\text{Ash} = \frac{\text{Weight of ash (g)}}{\text{Weight of sample taken (g)}} \times 100$$

5. Estimation of Amino acid (Moore and Stein, 1948)

Principle

When amino acid is heated with ninhydrin, they undergo deamination and a beautiful blue or purple colour developed, which is of value in both qualitative and quantitative determination of amino acid. The keto acid formed in the oxidative deamination is decomposed by heat into an aldehyde and carbon dioxide.

Ninhydrin + amino acid $\rightarrow$ Hydrantin + aldehyde + CO_2 + NH_3

Hydrantin reacts with some of the ninhydrin to from Rothmans

purple. Reagents

1) 10% Sodium tungstate: Prepared by dissolving 10 g Sodium tungstate in 100 ml-distilled water.
2) 2/3N.H_2SO_4: 6.6 ml of concentrated sulphuric acid was diluted to 350 ml with distilled water.
3) Ninhydrin reagent: It was prepared by dissolving 0.5 g of ninhydrin in 12.5 ml ethanol and stored in refrigerator.
4) 1% Leucine solution (standard solution): 100 mg leucine was dissolved in 10 ml of 80% ethanol.

Procedure

From each group 1 g of tissue/feed sample was accurately weighed and homogenized individually with 2ml distilled water, to this 1 ml of sodium tungstate and 1 ml 2/3 N H_2SO_4 were added. This mixture was then centrifuged at 3000 rpm for 10 minutes and the supernatant were collected. Three test tubes were taken and labeled as blank, test and standard. 0.5 ml supernatant was added to the test tube ‘test’, 0.5 to ‘standard’ and 4.5 ml distilled water was added to both test tubes. 5ml distilled water was added to the blank. 0.5 ml ninhydrin was pipetted to all test tubes and were cotton plugged. The tubes were kept in boiling water bath until blue colour developed. The tubes were cooled and the intensity of the colour developed was measured with colorimeter at 540 nm.

Calculation

$$\text{Amino acids present in the sample (\%)} = \frac{\frac{\text{OD of the sample}}{\text{OD of the standard}} \times \text{Conc. of the standard (mg)}}{\text{Weight of the sample (mg)}} \times 100$$

Statistical analysis

Results were expressed as Mean ±SD. The data obtained were analyzed for significant differences($p<0.05$) by two way analysis of Variance(ANOVA) and mean separation were accomplished by Duncan's Multiple Range Test(DMRT) using STATISTICAL software (Stat soft Inc.1999).

Figure -1: Fresh water fish Catla catla

Figure-2: Acclimatization of experimental fish, Catla catla in laboratory condition

Figure-3: Fish feed prepared from basal ingredients with fish meal and prawn in different concentration

Basal ingredient – soybean meal, groundnut oilcake, wheat bran, sunflower oil, egg albumin, tapioca flour and vitamin E.

C-Control (BI+FM)

T1-Treatment 1(BI+FM+R50)

T2-Treatment 2(BI+FM+R75)

T3-Treatment 3(BI+R100)

Figure-4: Experimental set up shows fresh water fish *Catla catla* in control and three different treatments

C-Control (BI+FM)

T1-Treatment 1(BI+FM+R50)

T2-Treatment 2(BI+FM+R75)

T3-Treatment 3(BI+R100)

4. Results and discussion

The results of the present investigation on the "Effect of prawn waste as feed ingredient on the growth, nutritional status and feed utilization on the fresh water fish *Catla catla* (Hamilton,1822.)" are presented below.

4.1. Growth performances

4.4.1. Weight gain (g)

Weight gain in *Catla catla* during different days of the experiment in the control and three different treatments are presented in table 2 and figure 5.

Among the three different treatments (T1, T2 and T3) and control, the maximum weight gain was observed in the fishes grown in T2 (2.70g) followed by T1 (2.38g), T3 (1.47g) on the 15th day of the experiment and the minimum value (1.40g) was recorded in the control fishes. On the 30th day, maximum weight gain (4.67g) was noticed in the fishes grown in T3 followed by T2 (3.30g) and T1 (2.54g) and minimum value (2.16g) was observed in the *Catla catla* grown in the control.

On the 45th day, maximum weight gain was observed in the fishes grown in T2 (5.60g) followed by T3 (4.37g), T1 showed the 3.73g weight gain and the minimum value (3.73g) was recorded in control. On the 60th day of the experiment maximum value was observed in T2 (6.10g) followed by T1 (5.80g) and T3 (5.87g), minimum value (4.13g) was noticed in the *Catla catla* grown in the control.

During the experiment period (15, 30, 45 and 60 days), among the three different treatments and control the maximum weight gain was noticed in T2 fishes when compare to the other treatments and minimum weight gain was observed in the control. The growth indices showed significantly differences from those produced by the diet. Guo *et al., (2007)* noticed that the highest protein (36.3%) and fat content in the poultry by-product meal increased the weight gain, specific growth rate and muscle proximate composition (moisture, protein and lipid) in cineaste drum, *Nibea miichthioides.*

4.1.2. Length gain (cm)

Length gain in *Catla catla* during different days of the experiment in the control and three different treatments are presented in table 3 and figure 6.

On the 15th day of the experiment, the maximum length gain was observed in the grown fishes in T2 (1.54cm) followed by T1 and t3 observed the equal length gain of (1.07cm) and the minimum value (1.00cm) was estimation in the control fishes. On the 30th day, maximum value was observed in the T2 (3.80cm) followed by T1 (2.76cm) and T3 (2.50cm). Minimum value (2.33 cm) was noticed in the *Catla catla* grown in the control fishes.

On the 45th day of the experiment, the maximum value was estimated in T2 (4.39cm) followed by T1 (3.43cm) and T3 (3.21cm) and the minimum value (2.56cm) was recorded in control fishes. On the 60th day, maximum length gain was recorded in T2 (5.45cm) followed by T1 (3.90cm) and T3 (3.70cm) and the minimum value (3.43cm) was recorded in control fishes.

Among the three different treatments the maximum length gain was observed in T2 fishes during 15, 30, 45 and 60 days and minimum length gain was observed in the *Catla catla* grown in control fishes.

Feed utilization efficiencies

Feed conversion ratio (FCR)

Feed conversion ratio (g) in *Catla catla* during days of the experiment in the control and three different treatments are presented in table 4 and figure 7.

On the 15th day of the experiment, the maximum value observed in T2 (1.05g) fishes followed by T1 (0.96g), T3 (0.93cm) and the minimum values (0.90g) was recorded in the control.

On the 30th day, maximum value was noticed in the fishes grown in T2 (0.93g) followed by T1 (0.86g) and T3 (0.78g) and the minimum value (0.71g) was observed in the control.

On the 45th day, maximum value was observed in the fishes grown in T2 (0.79g) followed by T3 (0.72g) and T1 (0.50g). Minimum value (0.71g) was recorded in the control.

On the 60th day of the experiment maximum value was observed in the T2 (0.68g) followed T3 (0.66g) and T1 (0.60g). Minimum value (0.53g) was noticed in *Catla catla* grown in the control.

During the experimental period (15, 30, 45 and 60 days), among the three different treatments and control the maximum value was noticed in t2 feed and a minimum value was observed in the control. Feed with poor water stability disintegrate rapidly and cause feed waste, water pollution and give poor feed conversion ratio(fcr) (sanhotra,1994). Feeding rate is important for the growth, feed conversion, nutrient retention efficiency and chemical

composition of body tissue (Storebakken and Austreng, 1987). The high-energy diet produced the lowest fcr and the highest nutrient retention (Coyle *et al.*, 2004: Zamal *et al.*, 2009)

Feed efficiency (FE)

Feed efficiency (g) in catla catla during days of the experiment in the control and three different treatments are presented in table 5 and figure 8.

On the 15th day of the experiment, the maximum value was observed in T2 (1.19g) fishes followed by T3 (1.05g) T1 (1.03g) and the minimum values (0.91g) was recorded in the control.

On the 30th day, maximum value was noticed in the fishes grown in T2 (1.38g) followed by T1 (1.17g) and t3 (1.27g) and the minimum value (1.07g) was observed in the control.

On the 45th day, maximum value was observed in the fishes grown in T2 (1.69g) followed by T3 (1.39g) and T1 (1.23g). Minimum value (1.01g) was recorded in the control.

On the 60th day of the experiment maximum value was observed in the T2 (1.89g) followed by T3 (1.50g) and T1 (1.62g). Minimum value (1.46g) was noticed in *Catla catla* grown in the control.

During the experimental period (15, 30, 45 and 60 days), among the three different treatments and control the maximum value was noticed in T2 feed and a minimum value was observed in the control feed. Per and fcr are also generally related to digestibility of nutrients (jabir *et al.*, 2012).

Estimation of biochemical composition

(protein, carbohydrate, lipids, moisture and amino acids) in control and three different feeds are presented.

The protein, carbohydrate, lipids and moisture in control and three different feeds are shown in table 7 and figure 10.

Protein (mg/g)

The maximum protein content was observed in T2 feed (6.63mg/g) followed by T3 (5.63mg/g) and T1 (5.50mg/g). Minimum protein content was noticed (4.97mg/g) in control feed. The protein content was high in T2 feed when compared to other two feeds and control.

Protein plays a very important role in the maintenance of blood glucose level (jrueger *et al.*,1968). Proteins serve as building blocks for cellular and organic structure. Proteins form components of catalysts called enzymes which are essential for the metabolic reactions

Carbohydrate (mg/g)

The maximum carbohydrates content was recorded in T2 feed (7.73mg/g) followed by T3 (6.43mg/g), T1 (5,53mg/g) and the minimum carbohydrates content was noticed by control feed (5.06mg/g). Carbohydrates are utilized by the cells mainly in the form of glucose.

Lipids (mg/g)

Lipid content was high in T2 feed (0.33mg/g) when compared to the other feeds T3 (0.11mg/g) and T1 (0.08mg/g) respectively. Lipid content was low in control feed (0.07mg/g). The greater protein and lipid content represents higher energy density (Dempson *et al.,* 2004).

Amino acids (mg/g)

Amino acids was maximum in T3 (mg/g) followed by T2 (5.23mg/g) and T1 (4.90mg/g). The minimum amino acid content was noticed in control feed (4.60mg/g).

Estimation of biochemical composition

(protein, carbohydrate, lipids, moisture and amino acids) in *catla catla* is presented below.

Protein (mg/g)

Protein content in the muscle tissue of *Catla catla* in the control and three different treatments are presented in table 8 figures 11.

Protein content in the muscle tissue of *Catla catla* during 15th day, was maximum in T2 (4.03mg/g) fishes followed by T3 (3.93mg/g), T1 (3.53mg/g) and control (3.20mg/g).

During 30th day, maximum protein content was observed in T2 (4.40mg/g) followed by T3 and T1 observed equal value (4.37mg/g) of protein content. The minimum value (3.73mg/g) was observed in *Catla catla* grown in the control.

During 45th day, maximum value was observed in T2 (5.07mg/g) followed by T3 (4.80mg/g) and T1 (4.57mg/g) and minimum value (4.27mg/g) was observed in control fishes.

During 60th day, maximum value was observed in T2 (5.77mg/g) followed by T3 (5.33mg/g) and T1 (4.73mg/g). The minimum value (4.60mg/g) was observed in control fishes.

Among the three different treatments and control the maximum value was observed in the T2 fishes during 15, 30, 45 and 60 days and minimum protein content was observed in the control fishes.

Protein is essential sustenance of life and exists in largest quantity of nutrients as a component of the human body. It is very important to provide all essential amino acids to human in an appropriate amount for optional protein synthesis (Elshehawy *et al.,* 2016). Protein is essential for normal tissue function, growth and maintenance of body tissue (Weatherly and gills 1987). Proteins are molecular tools that perform an astonishing Varity of functions. In addition, to serving as structural materials in all living organisms, they are involved in such diverse functions in catalysis, metabolic regulation, transport and defense (Trudy and James, 2010).

Protein requirement for optimal growth and feed efficiency of fingerling fish usually ranged from 40 to 45 % depending upon species (harpaz *et al.,* 2001). Protein is one of the most prominent biochemical compounds of caceans and its quantity in this class of organism is largely influenced by the extent of fat. Soybean meal incorporation improved the palatability of feeds and also enhanced the protein digestibility (jose *et al.,* 2006)

4.4.2 Carbohydrate (mg/g)

Carbohydrate content in the muscle tissue catla catla in the control and three different treatments are presented table 9 figures 12.

Carbohydrate content in the muscle tissue of *Catla catla* during 15th day of the experiment was maximum value in T2 (4.23mg/g) followed by T3 (4.13mg/g) and T1 recorded the (3.80mg/g) of carbohydrate content. The minimum value (3.43mg/g) was observed in the *Catla catla* grown in the control.

During 30th day, maximum value was observed in T2 (5.10mg/g) followed by T3 (5.00mg/g) and T1 (4.40mg/g). The minimum value (4.13mg/g) was observed in control fishes.

During 45th day, maximum value was observed in T2 (5.97mg/g) followed by T3 (5.67mg/g) and T1 (5.03mg/g). The minimum value (4.67mg/g) was recorded in control fishes.

During 60th day, maximum value was observed in T2 (7.20mg/g) followed by T3 (6.30mg/g), T1 (5.67mg/g). The minimum value (4.70mg/g) was noticed in control fishes.

Among the three different treatments and control the maximum carbohydrate content was observed in T2 fishes during 15, 30, 45 and 60 days and minimum carbohydrate content was observed in the control fishes.

Carbohydrate are an important and an inexpensive source of energy supplying nutrients which serve as precursors for the dispensable amino acids, and some nutrients also act as metabolic intermediates necessary for growth (health,1987).The carbohydrate metabolism is distribution when the animals are subjected to stress (McLeay and Brown,1975). Thus carbohydrate is considered to be the first among the organic nutrients to be utilized generate required energy (Health, 1987).

Lipids (mg/g)

Lipid content in the muscle tissue of *Catla catla* in the control and three different treatments are presented in table 10. Figure13.

Lipid content in the muscle tissue of *Catla catla* during 15th day of the experiment, the maximum value was observed in T1 (0.07mg/g) followed by T3 (0.04mg/g) and in T2 (0.03mg/g) of lipid content was noticed. The minimum value (0.02mg/g) was observed in the *Catla catla* grown in the control.

During 30th day, maximum value was observed in T2 (0.10mg/g) followed by T3 (0.05mg/g) and T1 (0.04mg/g) and in control minimum value (0.02mg/g) was analyzed.

During 45th day, maximum value was observed in T2 (0.06mg/g) followed by T3 (0.04mg/g) and T1 (0.03mg/g) and in control minimum value (0.02mg/g) was estimated.

During 60th day, maximum value was observed in T3 (0.06mg/g) followed by T2and T1 which contain the equal level of lipid (0.56mg/g) and in t1 0.06mg/g of lipid and in control minimum value (0.03mg/g) of lipid was observed.

Among the three treatments and control the maximum lipid content was observed in the T2 fishes during 15, 30, 45 and 60 days and minimum lipid content was observed in the *Catla catla* grown in the control.

Lipids are highly efficient as sources of energy and they contain more than twice the energy of carbohydrates and proteins (Okuzumi and Fujii 2000). Deficiency of lipid in the diets has been reported to induce mortality of larvae and pl of shrimp (Chandge and Ral, 1990).

The level of phospholipids in refined fish oil, on the other hand is low. The triglycerides/ phospholipids ratio in the residual lipid content in fish meal is about 2:1 of total lipids (Tocher *et al*,.2008). Phospholipids acts as surfactants in the intestine during lipid digestion and may thus improve lipid emulsification and increase lipid digestibility in fish (Hung et al., 1997; Tocher *et al.,* 2008). Carcass lipid content positively correlated with dietary lipid level irrespective of

protein level and carcass protein, fat and ash content increased with increasing dietary lipid (Subhadra *et al.,* 2006).

Moisture (%)

Moisture content in the muscle tissue of *Catla catla* in the control and three different treatments are present in table 11 figures 14.

Moisture content in the muscle tissue of *Catla catla* during 15th day, maximum value was recorded in T1 (49.33%) followed by T2 (44.00%), T3 (42.67%) and the in control the maximum value (30%) was noticed.

During 30th day, maximum content was observed in T3 (57%) followed by T2 (49.67%) and T1 (41.67%). The minimum value (36%) was observed in control fishes.

During 45th day, maximum value was observed in T2 (55.67%) followed by T3 and T1 where equal values of moisture content (47.67%) was noticed, in T1 (46.33%) and control was minimum value (45.67%) was estimated.

During 60th day, maximum value was observed in T3 (55%) followed by T2 (53.67%) and t1 (45.33%). The minimum value (34.67%) was observed in control fishes.

Among the three different treatments and control the maximum value was observed in T3 fishes during 15, 30, 45 and 60 days minimum moisture content was observed in the control fishes.

Moisture and fat usually vary inversely in fish flesh; while the protein is more constant (Bela and Assem, 1995).moisture is required for normal functioning of many biological molecules. It is present in two forms, bound to the protein and in the free from (Stanchena, 2003). There exists an inverse relationship between the moisture and lipid content. The percentage of water is a good indicator of its relative content of energy, proteins and lipids (Olagunju *et al.,* 2012).

Amino acids (mg/g)

Amino acid content in the muscle tissue of *Catla catla* in the control and three different treatments are presented in table 12 figures 15,

Amino acid content in the muscle tissue of *Catla catla* during 15th day of the experiment maximum value was obtained in T2 (4.43mg/g) followed by T3 (43.3mg/g) and T1 (3.67mg/g) and the minimum value (3.53mg/g) was recorded in the *Catla catla* grown in the control.

During 30th day, maximum amino acid content was observed in T2 (5.23mg/g) followed by T3 (5.20mg/g) and T1 (4.50mg/g) and minimum value (4.33mg/g) was observed.

During 60th day, maximum value was observed in T2 (5.96mg/g) followed by T3 (5.80mg/g) and T1 (4.83mg/g) and control in the minimum value (4.33mg/g) was observed.

During 60th day, maximum value was observed in T2 (6.77mg/g) followed by T3 (6.25mg/g) and T1 (5.50mg/g). The minimum value (5.1omg/g) was observed in control fishes.

Among the three different treatments and control the maximum value was observed in T2 fishes during 15, 30, 45 and 60 days and minimum amino acid content was observed in the control. Amino acid is the buildings blocks of proteins and also plays a central role as intermediates in metabolism (Baldwin, 2003)

Ash (%)

Ash content in the muscle tissue of *Catla catla* in the control and three different treatments are presented in table 13 figures 16.

Ash content in the muscle tissue of C*atla catla* during 15th day of the experiment maximum value was obtained in T2 (50.33%) followed by T3 (48.67%) and T1 (37%) and control in minimum value (36.33%) was observed.

During 30th day, maximum amino acid content was observed in T2 (50.33%) followed by T1 (49.67%), T3 (38.33%) and minimum value (37%) was observed in control fishes.

During 45th day, maximum value was observed in T2 (59.67%) followed by T3 (49.33%) and T1 (40.33%) was observed in the *Catla catla* grown in the control.

During 60th day, maximum value was observed in T2 (56.33%) followed by T1 (48%) and T3 (44.67%). The minimum value (44.33%) was observed in control fishes.

Among the three different treatments and control the maximum value was observed in T3 fishes during 15, 30, 45 and 60 days and minimum value was observed in the control fishes. Ash is the measure of the mineral content of a food item, this results indicates that the prawn waste are good sources of minerals.

Table-2

Weight gain (g) in *Catla catla* during different days of the experiment in control and three different treatments

Treatments	**No of days during experiment**			
	15 days	**30 days**	**45 days**	**60 days**
Control	1.40±0.95	2.16±0.16	3.73±0.18	4.13±1.46
T1	2.38±0.25	2.54±0.56	4.30±0.54	5.80±0.45
T2	2.70±0.75	3.30±0.29	5.60±0.99	6.10±1.22
T3	1.47±0.86	4.67±0.35	4.37±0.22	5.87±0.96
SEd	0.1467			
Cd(p<0.05)	20.5076			

Values are mean ± SD of the three samples in each group

C-Control (BI+FM)

T1-Treatment 1(BI+FM+R50)

T2-Treatment 2(BI+FM+R75)

T3-Treatment 3(BI+R100)

Table-3

Length gain (cm) in *Catla catla* during different days of the experiment in control and three different treatments

Treatments	**No of days during experiment**			
	15 days	**30 days**	**45 days**	**60 days**
Control	1.00±0.65	2.33±0.53	2.56±0.18	3.43±0.21
T1	1.07±0.41	2.76±0.54	3.43±0.54	3.90±0.21
T2	1.54±0.27	3.80±0.29	4.39±0.26	5.45±0.12
T3	1.06 ±0.41	2.50±0.56	3.21±0.89	3.70±0.04
SEd	2.6351			
Cd(p<0.05)	3.7572			

Values are mean ± SD of the three samples in each group

C-Control (BI+FM)

T1-Treatment 1(BI+FM+R50)

T2-Treatment 2(BI+FM+R75)

T3-Treatment 3(BI+R100)

Table - 4

Feed conversion ratio (FCR (g g^{-1}) in Catla catla during different days of the experiment in the control and three different treatments

Treatments	**No of days during experiment**			
	15 days	**30 days**	**45 days**	**60 days**
Control	0.90±0.12	0.71±0.04	0.71±0.08	0.53±0.01
T1	0.96±0.01	0.86±0.09	0.50±0.02	0.60±0.02
T2	1.05±0.05	0.93±0.02	0.79±0.02	0.68±0.03
T3	0.93 ±0.06	0.78±0.04	0.72±0.02	0.66±0.01
SEd	0.0335			
Cd(p<0.05)	0.09512			

Values are mean ± SD of the three samples in each group

C-Control (BI+FM)

T1-Treatment 1(BI+FM+R50)

T2-Treatment 2(BI+FM+R75)

T3-Treatment 3(BI+R100)

Table-5

Feed efficiency (FE (gg^{-1})) in Catla catla during different days of the experiment in the control and three different treatments

Treatments	**No of days during experiment**			
	15 days	**30 days**	**45 days**	**60 days**
Control	0.91±0.04	1.07±0.03	1.01±0.75	1.46±0.06
T1	1.03±0.01	1.17±1.10	1.69±0.06	1.62±0.04
T2	1.19±0.02	1.38±0.04	1.27±0.03	1.89±0.04
T3	1.05 ±0.07	1.27±0.06	1.39±0.10	1.50±0.08
SEd	0.14767			
Cd(p<0.05)	0.29850			

Values are mean ± SD of the three samples in each group

C-Control (BI+FM)

T1-Treatment 1(BI+FM+R50)

T2-Treatment 2(BI+FM+R75)

T3-Treatment 3(BI+R100)

Table- 6

Protein efficiency ratio (PER (g gain/g protein intake)) in the muscle tissue of *Catla catla* during different days of the experiment in the control and three different treatments

Treatments	**No of days during experiment**			
	15 days	**30 days**	**45 days**	**60 days**
Control	27.07±1.21	47.97±3.76	53.87±1.69	54.64±7.45
T1	47.03±2.82	62.67±2.56	66.43±4.03	90.33±11.33
T2	64.67±1.88	69.87±5.06	74.97±5.71	69.47±5.64
T3	54.33±4.18	58.47±5.11	57.77±1.99	58.60±1.51
SEd	3.95018			
Cd($p<0.05$)	7.98377			

Values are mean ± SD of the three samples in each group

C-Control (BI+FM)

T1-Treatment 1(BI+FM+R50)

T2-Treatment 2(BI+FM+R75)

T3-Treatment 3(BI+R100)

Table-7

Protein, carbohydrates, lipids and amino acids (mg/g) in the control and three different feeds

Treatments	**Feed**			
	protein	**Carbohydrates**	**lipids**	**amino acids**
Control	4.97±0.21	5.06±0.15	0.07±0.02	4.60±0.36
T1	5.50±0.11	5.53±0.06	0.08±0.03	4.90±0.49
T2	6.63±0.15	7.73±0.59	0.33±0.41	5.23±0.47
T3	5.63±0.31	6.43±0.50	0.11±0.01	6.10±0.35

Values are mean ± SD of the three samples in each group

C-Control (BI+FM)

T1-Treatment 1(BI+FM+R50)

T2-Treatment 2(BI+FM+R75)

T3-Treatment 3(BI+R100)

Table- 8

Protein (mg/g) in the muscle tissue of *Catla catla* during different days of the experiment in the control and three different treatments

Treatments	**No of days during experiment**			
	15 days	**30 days**	**45 days**	**60 days**
Control	3.20±0.10	3.73±0.25	4.27±0.15	4.60±0.20
T1	3.53±0.15	3.97±0.21	4.57±0.23	4.73±0.12
T2	4.03±0.15	4.40±0.26	5.07±0.25	5.77±0.25
T3	3.93±0.15	4.37±0.15	4.80±0.20	5.33±0.06
SEd **Cd($p<0.05$)**				

Values are mean ± SD of the three samples in each group

C-Control (BI+FM)

T1-Treatment 1(BI+FM+R50)

T2-Treatment 2(BI+FM+R75)

T3-Treatment 3(BI+R100)

Table-9

Carbohydrate (mg/g) in the muscle tissue of *Catla catla* during different days of the experiment in the control and three different treatments

Treatments	**No of days during experiment**			
	15 days	**30 days**	**45 days**	**60 days**
Control	3.43±0.25	4.13±0.32	4.67±0.25	4.70±1.40
T1	3.80±0.10	4.40±0.26	5.03±0.42	5.67±0.15
T2	4.23±0.15	5.10±0.26	5.97±0.31	7.20±0.30
T3	4.13±0.15	5.00±0.17	5.67±0.25	6.30±0.20
SEd **Cd(p<0.05)**				

Values are mean ± SD of the three samples in each group

C-Control (BI+FM)

T1-Treatment 1(BI+FM+R50)

T2-Treatment 2(BI+FM+R75)

T3-Treatment 3(BI+R100)

Table-10

Lipid (mg/g) in the muscle tissue of *Catla catla* during different days of the experiment in the control and three different treatments

Treatments	**No of days during experiment**			
	15 days	**30 days**	**45 days**	**60 days**
Control	0.02±0.01	0.02±0.04	0.02±0.01	0.03±0.02
T1	0.05±0.66	0.04±0.01	0.03±0.01	0.06±0.01
T2	0.03±0.02	0.10±0.15	0.06±0.04	0.56±0.01
T3	0.04±0.01	0.05±0.01	0.04±0.01	0.97±0.03
SEd **Cd($p<0.05$)**				

Values are mean ± SD of the three sample in each group C-

Control (BI+FM)

T1-Treatment 1(BI+FM+R50)

T2-Treatment 2(BI+FM+R75)

T3-Treatment 3(BI+R100)

Table-11

Amino acids (mg/g) in the muscle tissue of *Catla catla* during different days of the experiment in the control and three different treatments

Treatments	**No of days during experiment**			
	15 days	**30 days**	**45 days**	**60 days**
Control	3.53±1.53	4.03±1.53	4.33±1.53	5.10±2.64
T1	3.67±1.15	4.50±2.00	4.83±2.08	5.50±1.00
T2	4.43±1.53	5.27±4.04	5.96±3.21	6.67±2.52
T3	4.20±2.00	5.20±3.00	5.80±1.73	6.26±0.59
SEd	1.68000			
Cd(p<0.05)	3.37526			

Values are mean ± SD of the three samples in each group

C-Control (BI+FM)

T1-Treatment 1(BI+FM+R50)

T2-Treatment 2(BI+FM+R75)

T3-Treatment 3(BI+R100)

Table-12

Moisture in the muscle tissue of *Catla catla* during different days of the experiment in the control and three different treatments

Treatments	**No of days during experiment**			
	15 days	**30 days**	**45 days**	**60 days**
Control	30.00±2.00	36.00±1.00	45.67±9.02	34.67±2.52
T1	40.00±12.53	41.67±10.69	46.33±1.15	45.33±10.50
T2	44.00±3.61	49.67±6.66	55.67±9.24	53.67±9.07
T3	42.67±6.81	57.00±10.39	47.67±7.51	55.00±4.36
SEd	6.91295			
Cd(p<0.05)	13.97189			

Values are mean ± SD of the three samples in each group

C-Control (BI+FM)

T1-Treatment 1(BI+FM+R50)

T2-Treatment 2(BI+FM+R75)

T3-Treatment 3(BI+R100)

Table- 13

Ash (%) in the muscle tissue of *Catla catla* during different days of the experiment in the control and three different treatments

Treatments	**No of days during experiment**			
	15 days	**30 days**	**45 days**	**60 days**
Control	36.33±1.53	37.00±5.57	40.30±7.37	44.33±2.09
T1	50.33±6.66	49.67±6.66	40.33±4.16	48.00±7.81
T2	37.00±3.00	53.33±1.53	59.67±5.13	56.33±3.21
T3	48.67±6.66	38.33±8.02	49.33±5.13	44.67±5.86
SEd	5.72228			
Cd($p<0.05$)	11.56540			

Values are mean ± SD of the three samples in each group

C-Control (BI+FM)

T1-Treatment 1(BI+FM+R50)

T2-Treatment 2(BI+FM+R75)

T3-Treatment 3(BI+R100)

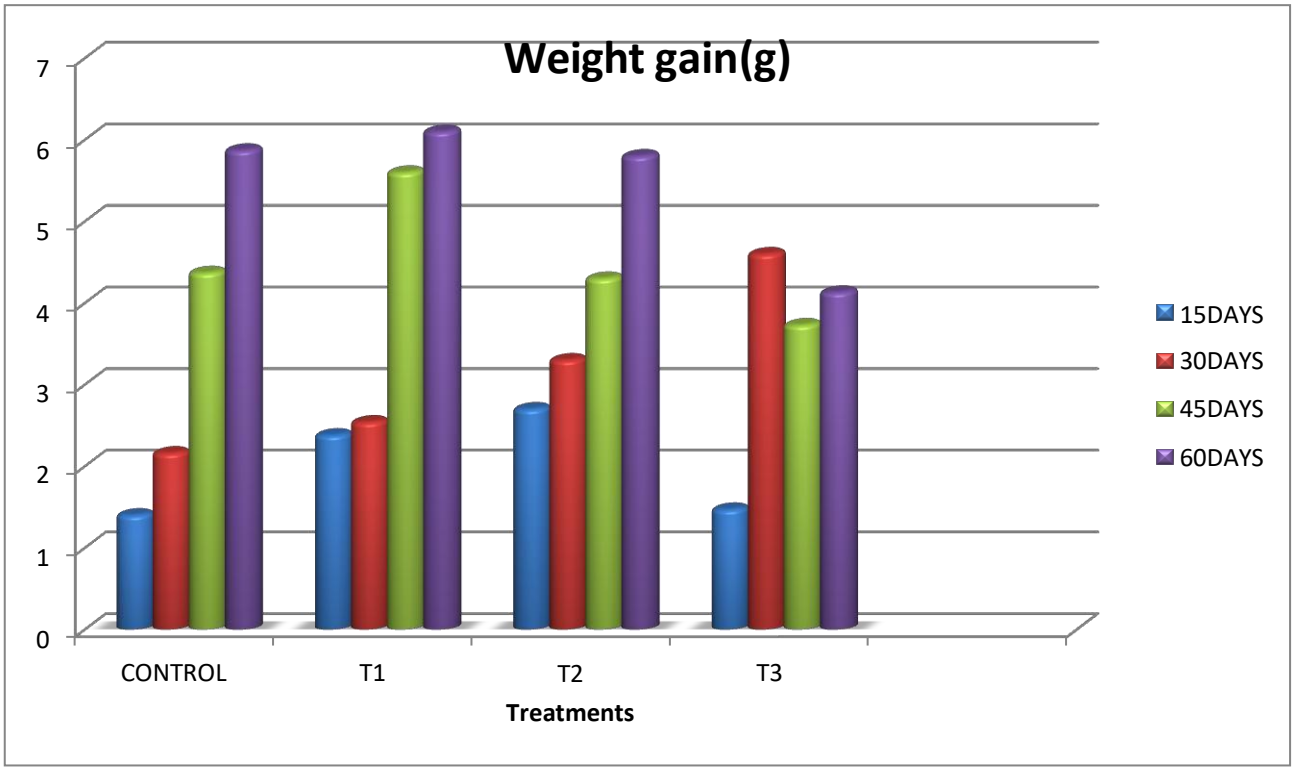

Figure-5

Weight gain (g) in *Catla catla* during different days of the experiment in control and three different treatments

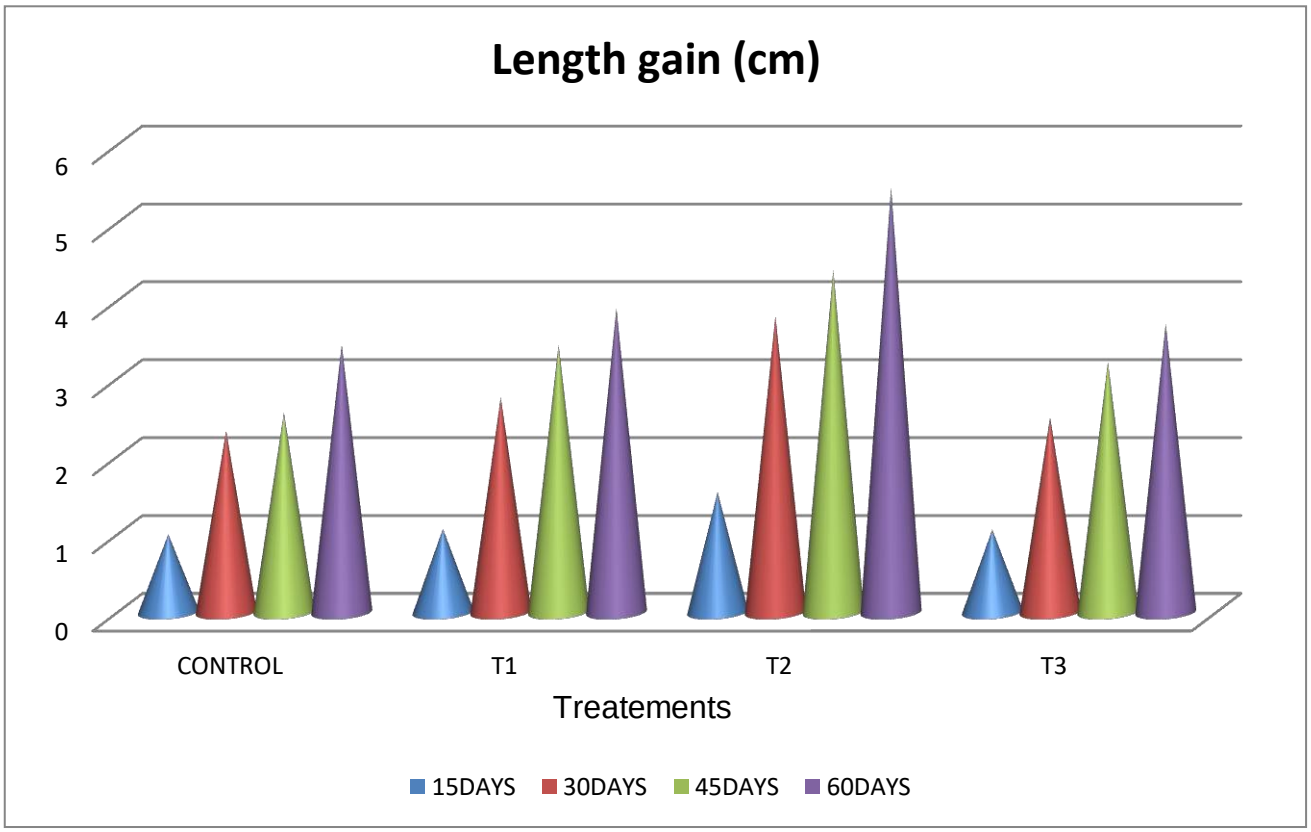

Figure-6

Length gain (cm) in *Catla catla* during different days of the experiment in control and three different treatments

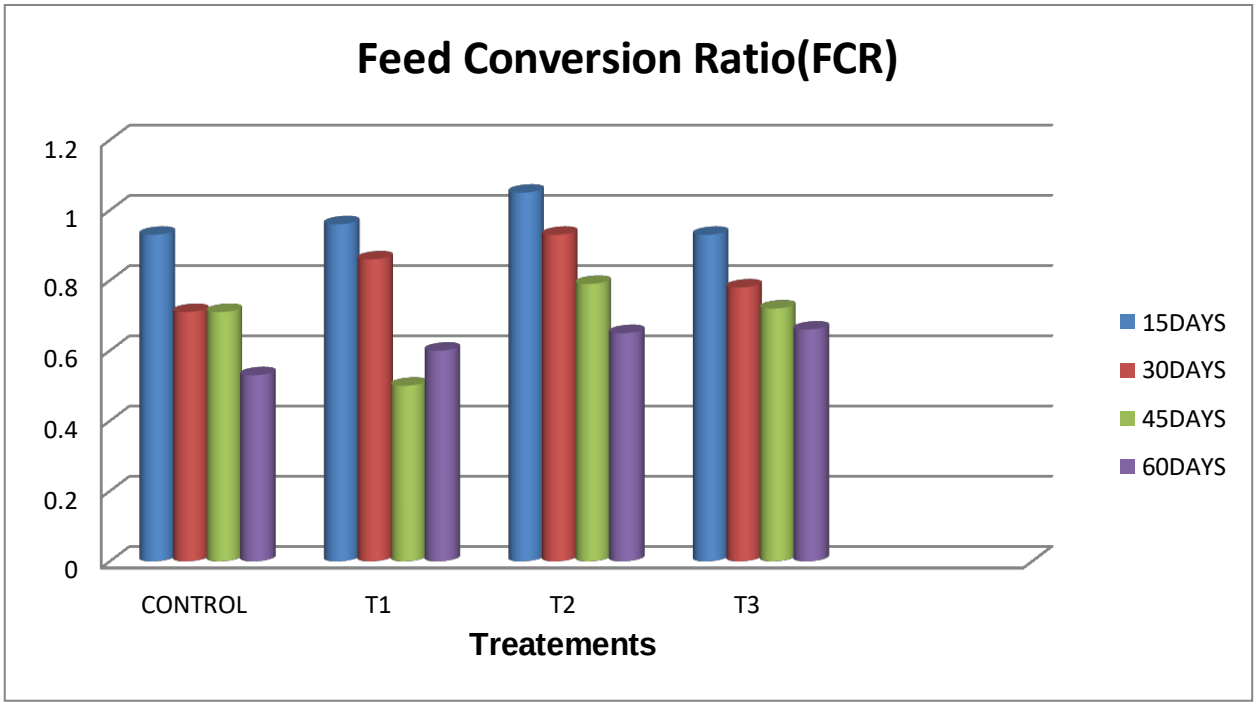

Figure-7

Feed conversion ratio (FCR (g g^{-1}) in Catla catla during different days of the experiment in the control and three different treatments

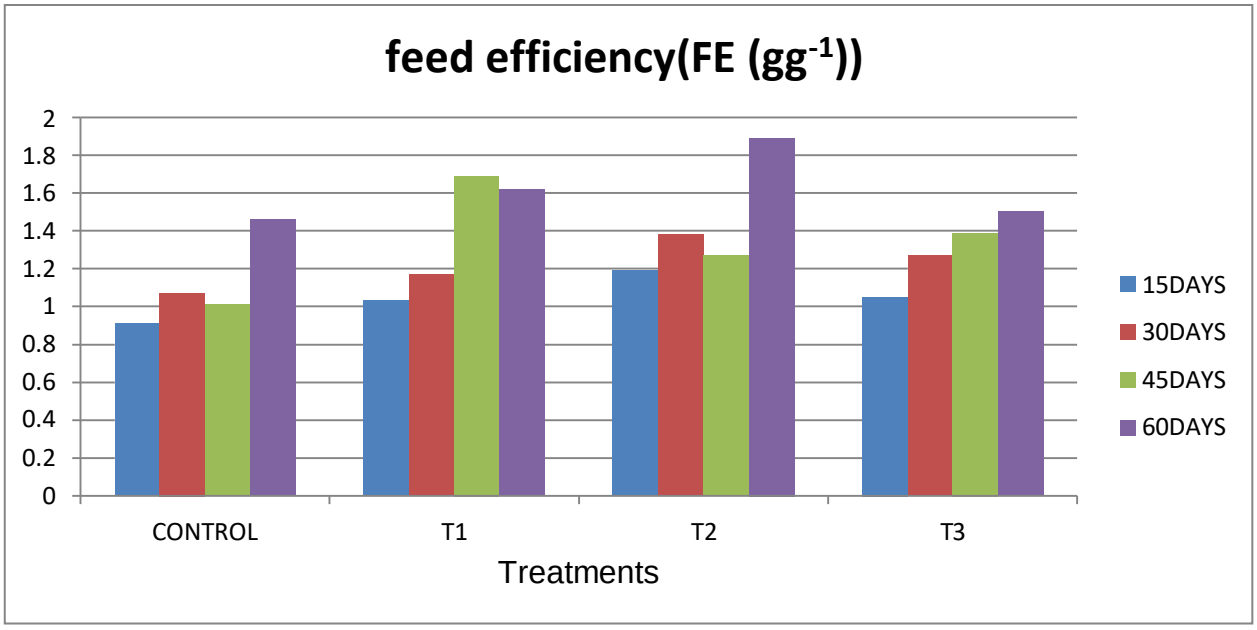

Figure-8

Feed efficiency (FE (gg^{-1})) in Catla catla during different days of the experiment in the control and three different treatments

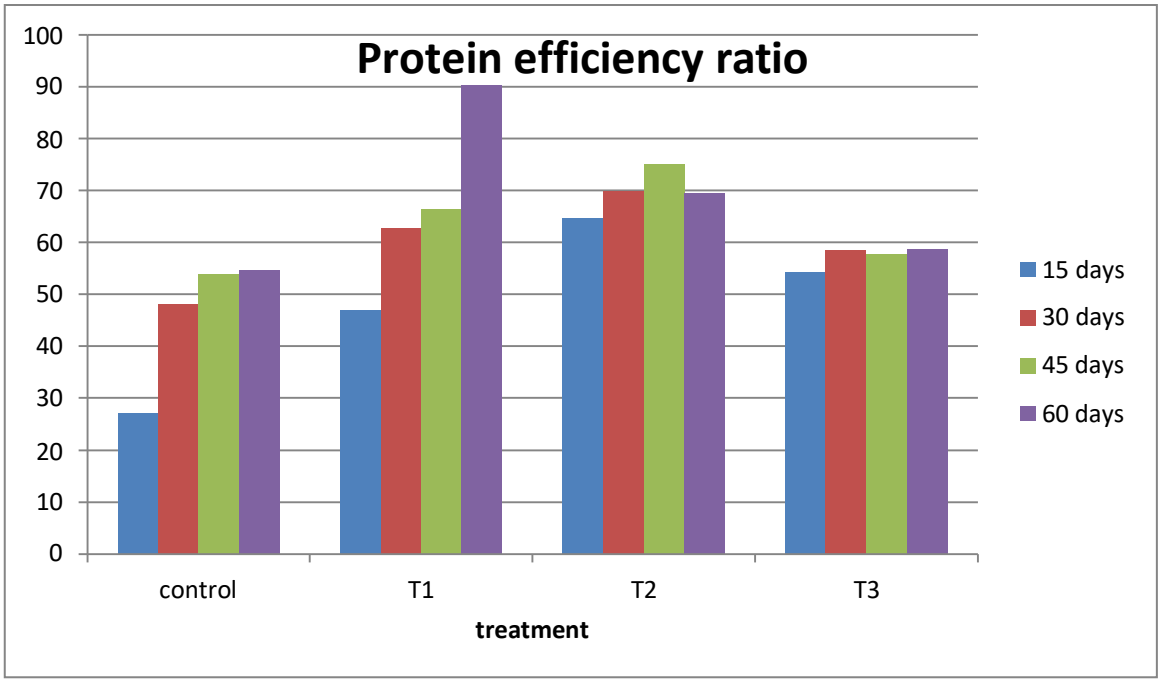

Figure-9

Protein efficiency ratio (PER (g gain/g protein intake)) in the muscle tissue of *Catla catla* during different days of the experiment in the control and three different treatment

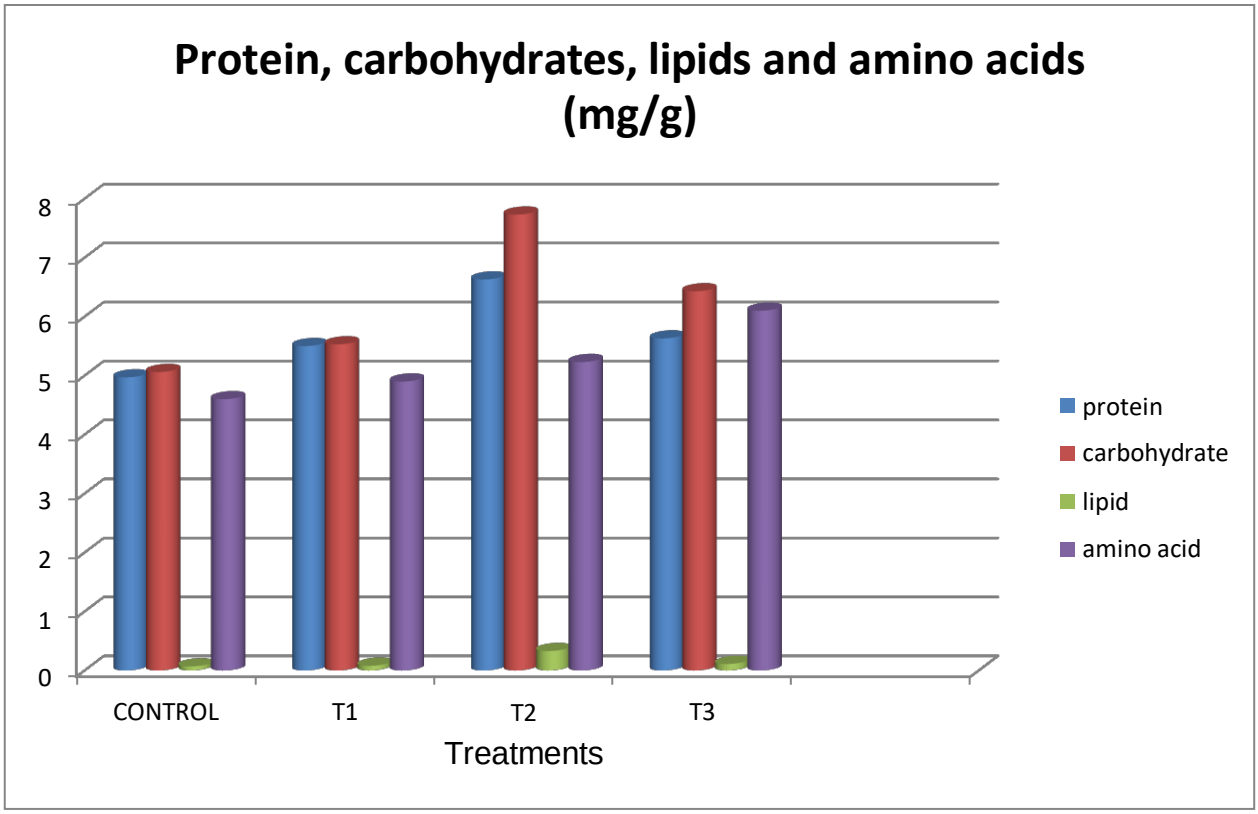

Figure-10

Protein, carbohydrates, lipids and amino acids (mg/g) in the control and three different feeds

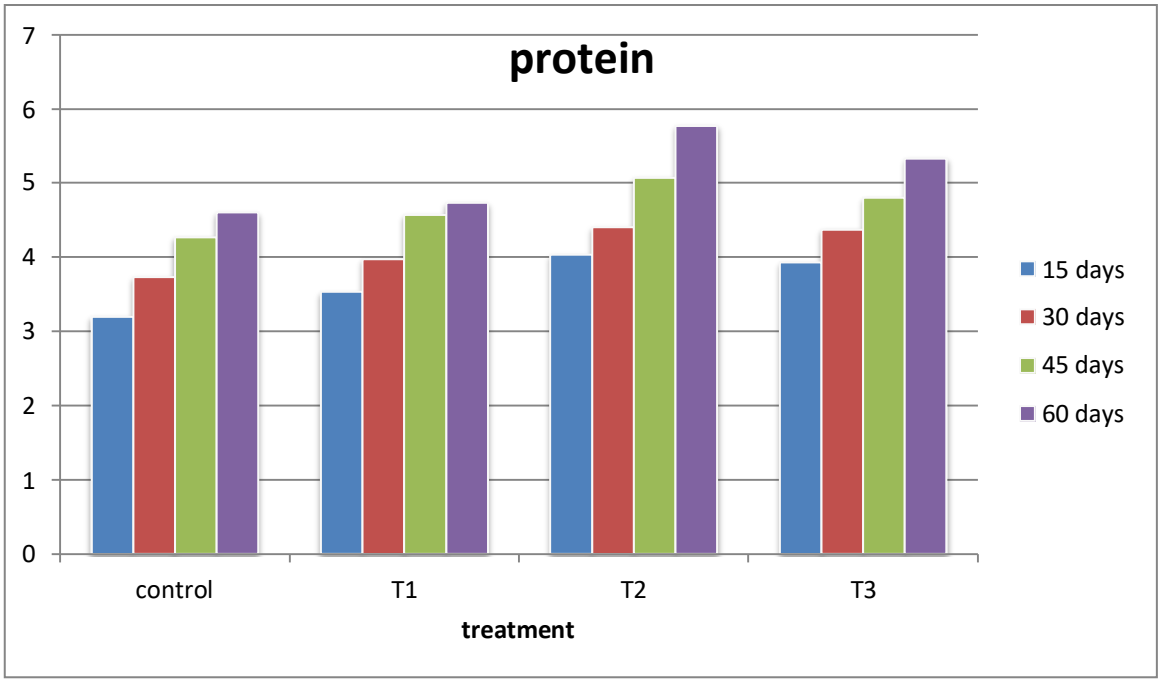

Figure-11

Protein (mg/g) in the muscle tissue of *Catla catla* during different days of the experiment in the control and three different treatments

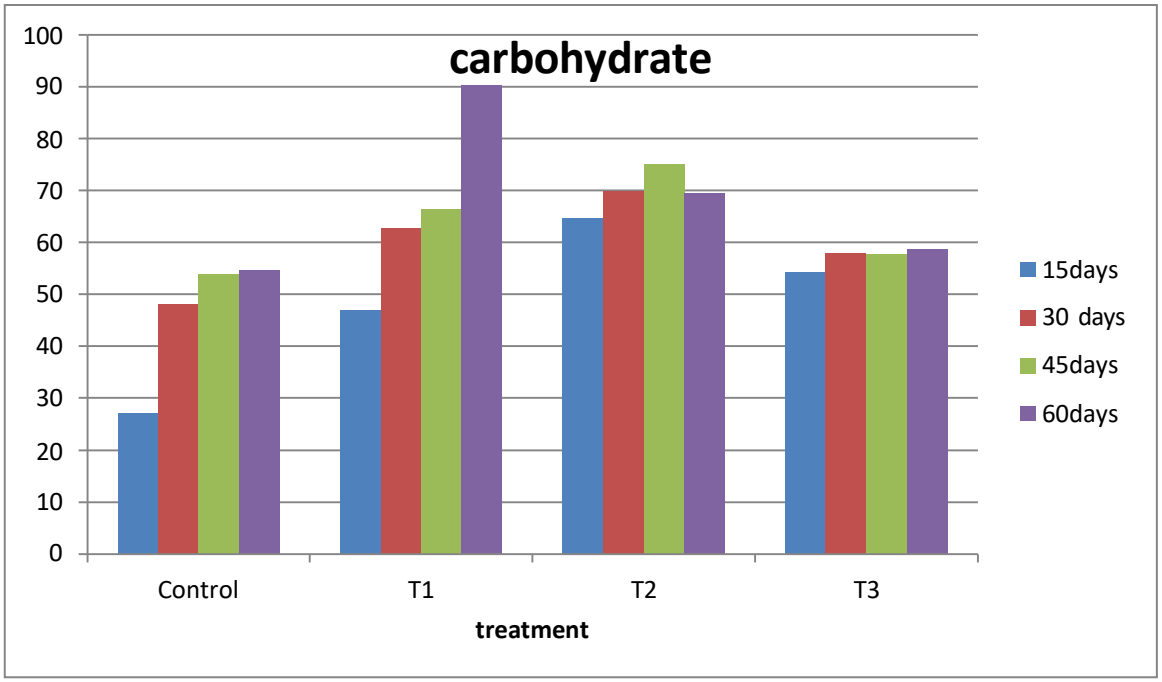

Figure-12

Carbohydrates (mg/g) in the muscle tissue of *Catla catla* during different days of the experiment in the control and three different treatments

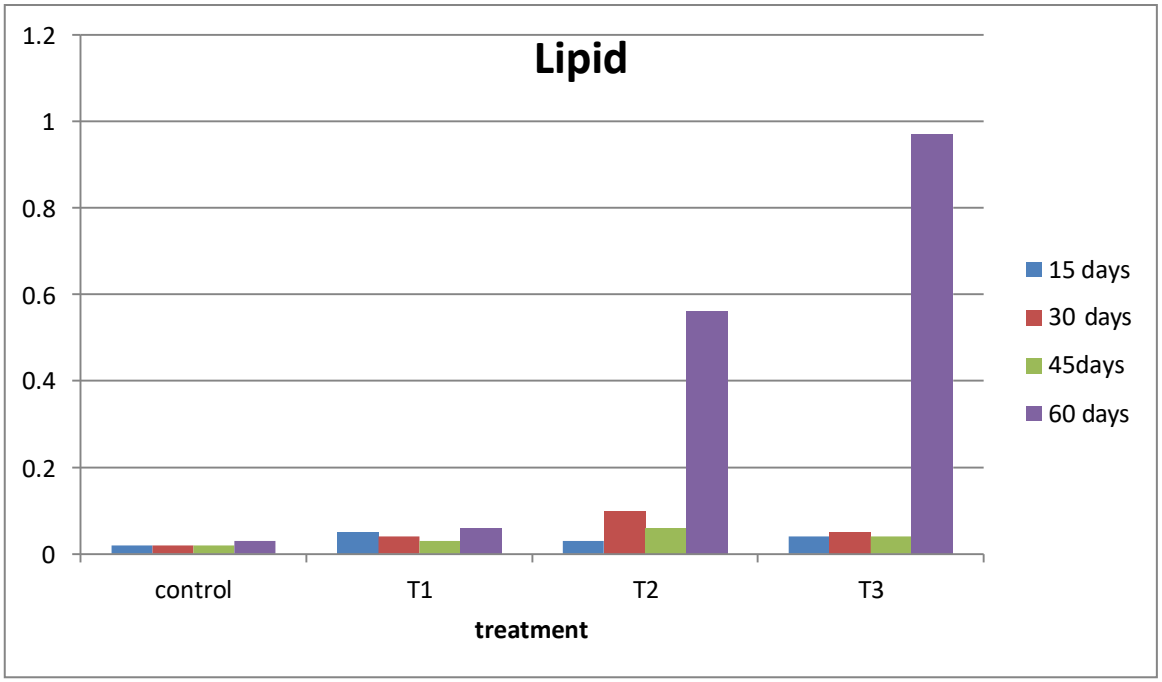

Figure-13

Lipid (mg/g) in the muscle tissue of *Catla catla* during different days of the experiment in the control and three different treatments

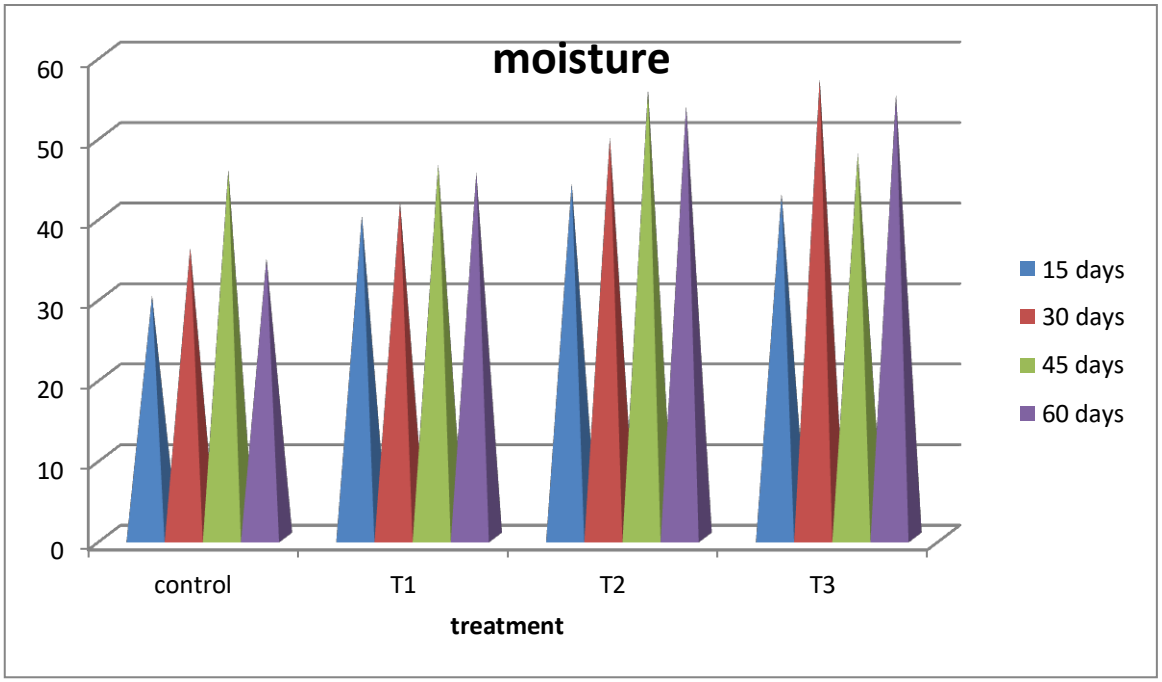

Figure-14

Moisture (%) in the muscle tissue of *Catla catla* during different days of the experiment in the control and three different treatments

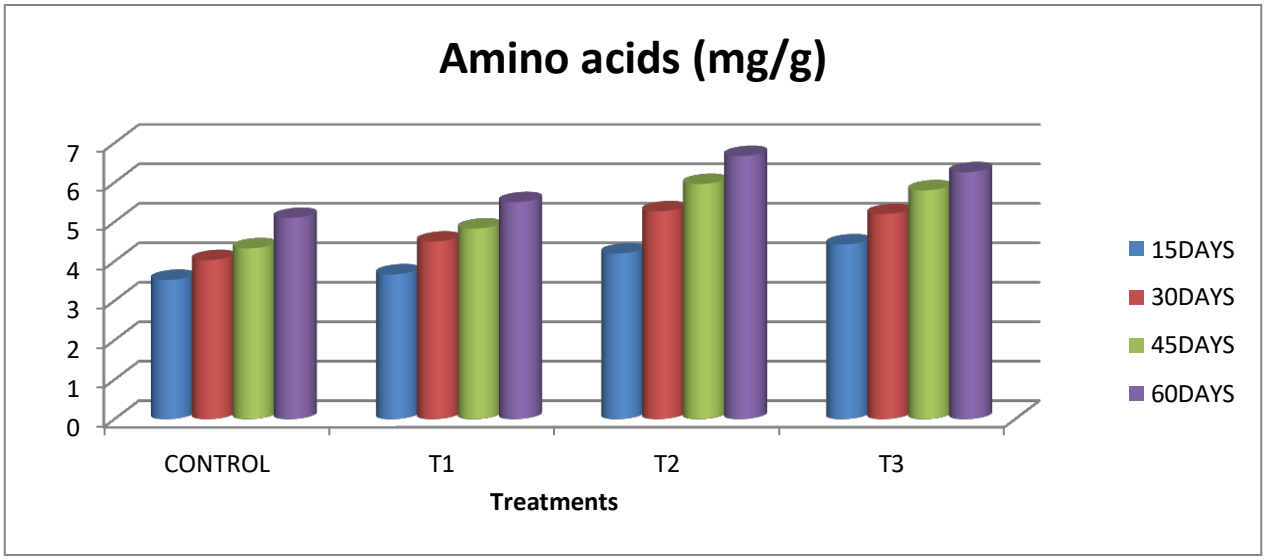

Figure-15

Amino acids (mg/g) in the muscle tissue of *Catla catla* during different days of the experiment in the control and three different treatments

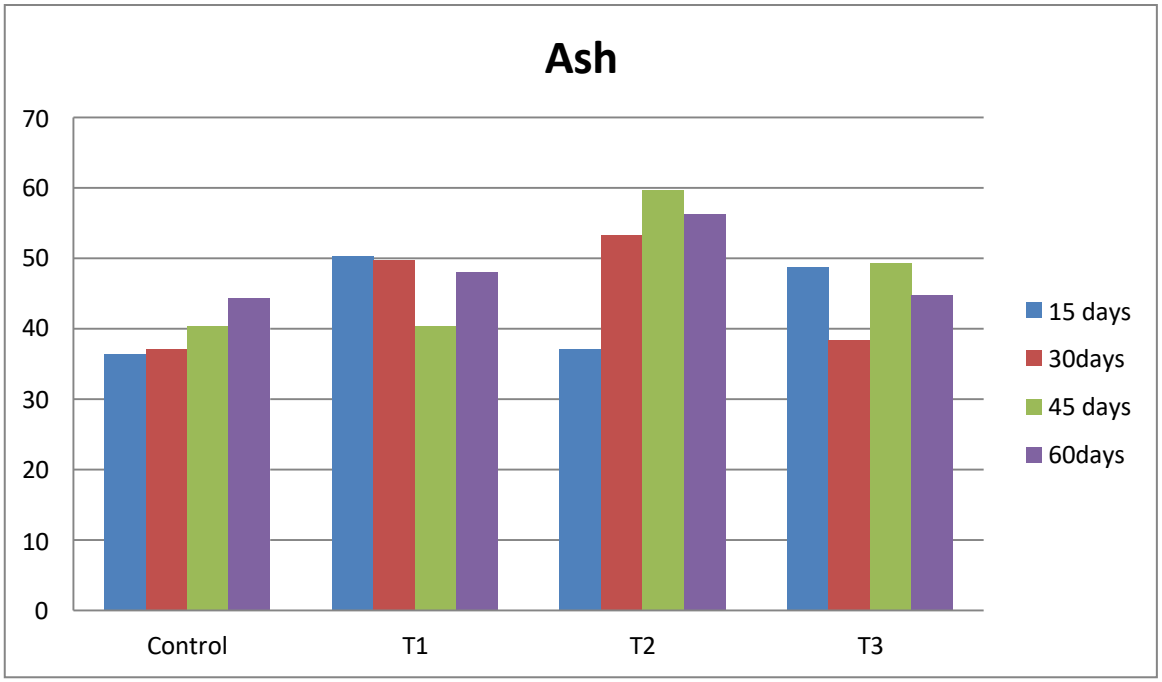

Figure-16

Ash (%) in the muscle tissue of *Catla catla* during different days of the experiment in the control and three different treatments

Summary and conclusion

Effect of prawn waste as feed ingredient on the growth ,nutritional indices and feed utilization efficacy on the fresh water fish *catla catla* has been studied .In the present investigation fishes were fed with the feed prepared from different concentration of prawn waste as replacement diet for 60 days and the result were observed for 15,30,45and 60 days.

The bio-growth parameters such as weight gain, length gain, and also recorded the feed utilization efficiencies such as feed efficiency(FE),feed conservation ratio(FCR) in *catla catla* for 15,30,45and 60 days were studied. The bio chemical composition such as protein, carbohydrates, lipids, moisture, ash and amino acids content were analyzed in the control and three different treatments. Bio chemical composition includes protein, carbohydrates, lipids, moisture, ash and amino acids in the control and three different treatments.

The detail of the study is summarized below

- Prawn waste feed improved the growth performance, feed utilization efficacy and biochemical composition in the *Catla catla.*
- During the experiment period (15, 30, 45 and 60 days), the maximum weight gain (6.10g) was observed in T2 fishes and minimum weight gain (4.13g) was recorded in the control fishes.
- During the experiment period (15, 30, 45 and 60 days), the maximum length gain (5.45cm) was observed in T2 fishes and minimum weight gain (3.43cm) was recorded in the control fishes.
- Among all the treatments maximum weight gain, length gain were reported in fishes grown in T2 treatments and minimum values was observed in control fishes.
- The feed utilization efficiency such as feed efficiency (FE), feed conversion ratio (FCR) and protein efficiency ratio (PER) in *Catla catla* was analyzed in the control and three different treatments.
- The highest (90.33) PER recorded in T2 fishes and lowest (27.07) was obtained in control.

- Feed conversion ratio (FCR) showed significant variations. The maximum FCR (1.09g) was obtained in T2 feed and minimum (1.05g) was recorded in the control feed.
- Feed efficiency (FE) showed significant variations. The maximum FE (1.19g) was obtained in T2 feed and minimum (1.07g) was recorded in the control feed.
- Maximum protein content (6.63mg/g), carbohydrates content (7.73mg/g) and lipid content (0.33mg/g) was found in T2 feed and amino acid ((5.23mg/g) was observed in T3 feed. Minimum protein content ((4.97mg/g), carbohydrates content (5.06mg/g) and lipid content (0.07mg/g) was found in T2 feed and amino acid (4.60mg/g) were recorded in the control feed
- On 15^{th}, 30^{th}, 45^{th} and 60^{th} days of the experiment ,maximum protein content (4.03 to 5.77) was observed in T2 fishes followed by T3(3.93 to 5.33) and T1(3.53 to 4.73). Minimum protein content (3.73 to 4.40mg/g) was observed in control feed. During the experimental period the maximum protein content was recorded in T2 (4.03mg/g) fed fishes.
- On 15^{th}, 30^{th}, 45^{th} and 60^{th} days of the experiment, maximum carbohydrates content (4.23 to 7.20) was observed in T2 fishes followed by T3(4.13 to 6.30) and T1(3.80 to 5.67). Minimum protein content (3.43 to 4.20) was observed in control feed.
- On 1^{5th}, 3^{0th}, 4^{5th} and 6^{0th} days of the experiment ,maximum lipid content (0.03 to 0.56) was observed in T2 fishes followed by T3 (0.04 to 0.96) and T1 (0.05 to 0.06). Minimum lipid content (0.02 to 0.03) was observed in control feed.
- On 15^{th}, 30^{th}, 45^{th} and 60^{th} days of the experiment ,maximum moisture content (44 to 53.67) was observed in T2 fishes followed by T3(42.67 to 55) and T1(40 to45.33). Minimum moisture content (30 to 34.67) in control feed.
- On 15^{th}, 30^{th}, 45^{th} and 60^{th} days of the experiment ,maximum ash content (37 to 56.33) was observed in T2 fishes followed by T3 (48.67 to 44.67) andT1(50.33 to 48). Minimum ash content () was observed in the control feed.
- The result were subjected to two-way ANOVA to show the significant n=value of biochemical parameter, feed utilization efficiency and biochemical composition in *Catla catla* grown in T2 feed.

In conclusion of the present study, it was observed that 50% supplementation of prawn waste had significantly improved the growth rate in terms of weight gain, length

gain and feed utilization efficiency (FCR,FE and PER). Furthermore the supplementation of prawn waste also increased the biochemical composition which facilitated digestion process. In coherence to the obtained results, supplemented nutrients from prawn waste provided sustainable growth for the fish *Catla catla*.

The present investigation provides adequate information on the formulation of fish feed with prawn waste in different concentration to feed the fish, *Catla catla.* Hence in aquaculture practices it is preferable to use prawn waste as fish feed for the enhancement of growth nutrient level and feed utilization efficiency. The finding would be formulating a cost effective and eco-friendly farm made feed.

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