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Full Length Research Paper

Effect of dietary protein, lipid and carbohydrate contents on the liver composition and enzyme activity of *Cyprinus carpio communis* fingerlings

Muzaffar Ahmad.*, Qureshi T. A. and Singh A. B.

Department of Zoology and Applied Aquaculture, Barkatullah University, Bhopal, M .P. (462026), India.

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This study aimed to determine a feed formulation with best protein to energy ratio which would result in better liver composition and enzyme activity of *Cyprinus carpio communis*. Fingerlings having average weight 1.64 g ± 0.13 and length 5.26 ± 0.10 cm were fed on four different formulated feeds and a control feed (each in a triplicate set), 6% of their body weight, three times a day, during 90 days. Feeds were formulated using ground nut oil cake, mustard oil cake, rice bran, wheat bran, fish meal and soybean meal in order to suffice the balanced need of protein and energy of the common carp. Liver composition and acid and alkaline phosphatase activity of fingerlings were measured. There was a significant increase in liver lipid content with the increase in dietary carbohydrate level. The ACP and ALP activity was the maximum in the fingerlings fed on the diet having optimum protein to energy ratio of 20.54 mg protein/Kj. This work concluded that a diet containing 40% protein, 9.31% lipid and 10.08% carbohydrate is the best one for a more profitable and successful culture of the common carp.

Key words: Protein to energy ratio, carp production, acid phosphatase, alkaline phosphatase.

INTRODUCTION

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 (Weatherley and Gill, 1987). However, protein is essential for normal tissue function, for the maintenance and renewal of fish body protein and for growth. Because of the cost of the protein the feed will be more cost effective if all the protein is used for tissue repair and growth and little catobolized for energy (Jauncey, 1998). From a practical point of view, the ideal situation should tend to maximize the use of dietary protein for growth, minimizing the use of proteins for functional protein synthesis, gluconeogenesis, lipogenesis and energy (Lovell, 1998). If adequate protein is not provided in the diet, there is a rapid reduction or cessation of growth and a loss of weight due to withdrawal of protein from less vital tissues to maintain the functions of more vital tissues. On the other hand, if too much protein is supplied in the diet, only part of it will be used to make new proteins and the remainder will be catabolized to produce energy (Wilson, 1989; NRC, 1993; Gullaine, 2001). Although the utilization of proteins for basal energy metabolism is a well-established phenomenon, conventional "energy-yielding" nutrients like fats and carbohydrates can reduce the oxidation of protein to satisfy the energy needs of fish and thus improve the utilization of dietary protein (Cho, 1987).

Lipids are an extremely diverse group of compounds many of which function as important sources of metabolic energy. Among the various types of lipid it is the simple, glycerol based, fats and oils that are of most interest in terms of general nutrition (Jauncey, 1998). Lipids normally occur in foodstuffs and in the fat deposits of most animals in the form of triglycerides, which are esters of fatty acids and glycerol (McDonald et al., 1988). Thus, dietary lipids provide a source of indispensable nutrients, the essential fatty acids. In addition, they also act as carriers of certain non - fat nutrients, notably the fat-soluble vitamins A, D, E and K and they are also an important source of energy (New, 1986). Lipids contain

*Corresponding author. E-mail: muzaffarahmad8@gmail.com. Tel: 9018877446, 9086768832.

more energy per unit weight than any other biological compound for example, one gram of lipid contains almost twice as much total energy as either one gram of carbohydrate or one gram of protein (Jauncey, 1998; Gullaine, 2001). Dietary lipids, mainly in the form of triglycerides, are hydrolyzed to free fatty acids and glycerol by pancreatic lipase, aided by the saponifying and emulsifying action of bile acids in the digestive tract. Absorption generally occurs primarily in the anterior ileum including the caecum (Cowey and Walton, 1989). Energy that is not utilized immediately is stored for future use as glycogen and carcass fat. Since glycogen reserves in fish are usually low, the main energy stored is fat. Experiments show that during starvation or food restriction in fish most of the metabolic energy is derived from lipid and, to a more limited degree from protein and carbohydrate (De Silva and Anderson, 1995). Lipid is digested and metabolized with greater relative ease and so serves as a much better source of energy for protein sparing than carbohydrate.

Unlike protein and fat, carbohydrate as a nutrient was not considered essential to fish because of their ability to synthesize carbohydrate metabolites (glucose/glycogen etc.) from excess dietary protein and fat. Compared to the farmed terrestrial animals, the utilization of dietary carbohydrates in fish is limited, but the inclusion of carbohydrate in fish feeds has certain beneficial effects (Wilsosn, 1994). The utilization of carbohydrate in fish varies depending on its complexity, source, level in the diet, pre-treatment and degree of gelatinization. The ability of fish to utilize carbohydrate also differs greatly between species and life stage as a consequence of the marked variations in the anatomy of the digestive tract and in the food habits (Steffens, 1989; Wilson, 1994; Lovell, 1998; Mustafizur et al., 2008). It is also thought that herbivorous and omnivorous fish species utilize carbohydrate better than carnivorous fishes (Shimeno et al., 1979; El-Sayed and Garling, 1988). The inability of fish to utilize dietary carbohydrate has been illustrated by glucose tolerance tests. Oral administration of glucose to different fish species led to linear increase of blood glucose concentration, with a poor response of plasma insulin levels. This implies that glucose levels in blood are poorly regulated by fish, their response being frequently similar to diabetic mammals (Palmer and Ryman, 1972; Bergot, 1979; Cowey and Walton, 1989; Chakraborty et al., 1992; Kaushik, 1995). Other carbohydrates such as fibres, hemicellulose, lignin and pentosans generally form indigestible fractions in the feed, often act as pellet binders. Some fish species can tolerate up to 8% of dietary fibre and depressed growth may occur when the fibre content reaches 20% (NRC, 1993; Wang et al., 1985; Davies, 1985).

Enzyme activity is a measure of the quantity of active enzyme present and is thus dependent on conditions, which should be specified. Amounts of enzymes can either be expressed as molar amounts, as with any other chemical, or measured in terms of activity, in enzyme units. Biologists studying fish in their natural habitats estimate growth rate using bony structures and mark/recapture studies. These methods do not estimate short-term changes in growth rate. A close link between the activity of some of the enzymes of energy metabolism and growth rate has been shown for several fish species (Sullivan and Somero, 1983; Kiessling et al., 1989; Houlihan, 1991; Yang and Somero, 1993). In the present study alkaline and acid phosphatase enzyme activities were measured to investigate the effect of control and formulated feeds on them. Both acid and alkaline phosphatases are hydrolase enzymes responsible for removing the phosphate groups from the nucleotides, proteins and alkaloids. Acid phosphatase (ACP) is most effective in an acidic medium (pH below 7) while as alkaline phosphatase (ALP) is effective in an alkaline medium (pH= 8.5). The ACP and ALP are widely distributed in the organs and tissues of vertebrates in general and fish in particular (Hollands and Smith, 1964; Shrivasthava, 1966; Hinton et al., 1972, Wiihmore and Goldberg, 1972; Goel and Shastry, 1973; Oide, 1973). The ACP and ALP participate in the processes of digestion, absorbtion, ossification and in numerous pathological processes (Chen et al., 1997).

The aim of the present study was to carry out orderly nutritional research with common carp by using different dietary protein, lipid and carbohydrate contents for determination of a feed formulation with optimum protein to energy ratio (P/E ratio) which would result in better liver composition and enzyme activity so as to make production of Common carp economical.

MATERIALS AND METHODS

Composition of control and formulated feeds experimented

Four feeds (Feed A, B, C and D) were formulated using ground nut oil cake, mustard oil cake, rice bran, wheat bran, fish meal and soybean meal. The ingredients were selected so as to suffice the balanced need of protein and energy of the common carp. Feeds were formulated using “Pearson-Square method” with different protein, carbohydrate and lipid contents. Control feed consisted of 50% mustard oil cake and 50% rice bran. Feed A consisted of ground nut oil cake (15%), mustard oil cake (15%), rice bran (10%), wheat bran (10%), fish meal (25%) and soybean meal (25%). The combination aimed at the supply of maximum protein component than energy. Feed B consisted of ground nut oil cake (18%), mustard oil cake (60%), rice bran (2%), wheat bran (8%), fish meal (4%) and soybean meal (8%). This combination, instead of having fish meal as a source of protein had mustard oil cake. Feed C consisted of ground nut oil cake (8%), mustard oil cake (12%), rice bran (40%), wheat bran (30%), fish meal (6%) and soybean meal (4%). This combination aimed at the use of carbohydrate rich diet for the growth. Feed D consisted of the mixture of equal quantity (16.66%) of all the ingredients. Vegetable oil (1.5 ml per 100 g of feed) and cod liver oil (1.5 ml per 100 g of feed) were incorporated in each formulated feed to ensure adequate supply of fatty acids of both n - 6 and n - 3 series, assumed to be essential for common carp. Vitamin - mineral mixture (2 g per 100 g of feed) was added to each formulated feed for the maintenance of fish health. Sodium

alginate (5 g per 100 g of feed) was used as binder and oxytetracycline (500 mg per 100 g of feed) as antibiotic for control and formulated feeds. The amount of sodium alginate used as binder for feed preparation was in accordance with Dias et al. (1998). Composition of control and formulated feeds (% in dry weight basis) experimented is given in Table 2.

Dry ingredients were mixed for about 30 min in a Hobart mixer (Belle, Mini 150; England) to ensure that the mixture was well homogenized and then blended with oil for about 15 min. Water was added at 20 to 30% v/w to give a pelletable mixture. A pelleting machine (Hobart, model, A 200) was used to pellet the feeds. An appropriate die was used to form pellets of desired sizes (1.0 to 3.0 mm). Pellets were oven dried and fed to the fishes, 6% of the body weight, three times a day at 10 A.M., 2.0 P.M. and 5.0 P.M. every day.

Cyprinus carpio communis fingerlings having average weight 1.64 g ± 0.13 and length 5.26 cm ± 0.10 were used for the experiment. Prior to the initiation of the feeding trail, fingerlings were acclimatized for one week. During this period, traditional mixture of mustard oil cake and rice bran (1:1) was fed to the fingerlings. Each formulated feed and control feed was fed to triplicate group of fingerlings for 90 days. Fifty fingerlings were reared in each fiber glass tank. Water was supplied to each tank at the rate of 1 l min^{-1}. About thirty percent of water was replaced weekly with freshwater to adjust water quality. Water analysis of the experimental tanks was done regularly to monitor any unusual changes. The tanks were aerated throughout the experiments with aquarium air pumps (RS-180, Zhongshan Risheng Company Limited., China). Biochemical analysis (dry matter, moisture, crude protein, crude lipid, carbohydrate and ash of feed ingredients, feeds and liver) was determined by using standard procedures (AOAC, 1995). The energy content of feed ingredients and feeds were calculated calorimetrically.

Enzyme activity

The acid and alkaline phosphatase activity in the liver of fingerlings was measured to investigate the effect of control and formulated feeds on them.

Collection of liver

Initially and at the end of the experiment, 5 fingerlings from each tank were dissected and livers were removed. The livers were blotted dry with tissue paper and kept in small plastic bags, individually, before freezing at –60°C until used for enzyme determination.

Enzyme extraction

Enzyme extraction was carried following the procedure of Jaroli and Sharma (2005). Frozen livers were homogenized in an electrical homogenizer (Utra - India) with ice-cold physiological saline (9 g sodium chloride/l). Samples were homogenized at 200 rpm for 3 min. 10 ml of physiological saline was used for 1 g of frozen liver sample. Homogenised samples were centrifuged (Mistral 3000, UK) at 3600 rpm for 15 min at 4°C to remove cell debris and nuclei. The resulting supernatant (sample) was used for measurement of enzyme activity.

Measurement of enzyme activity

The acid and alkaline phosphatase enzyme activity was measured following the procedure of Jafee and Badansky (1943). For the estimation of ACP activity 2 ml of reagent solution containing citrate buffer, napthylphosphate, fast red TR-salt, pentane-di-ol, tatarate and acetic acid was taken in a clean and sterile test tube. Then 200 µl sample was added to it. The mixture was shaken thoroughly and incubated at 37°C for 5 min and initial absorbance was recorded at 405 nm using a spectrophotometer (CECIL CE 2031) and subsequently three readings were taken at one minute interval. The ACP activity was expressed as IU/l and was calculated as:

ACP activity (IU/l) = 122 A

Where, A=change in absorbance per minute

For the estimation of ALP activity, reagent solution (3 ml) containing diethanolamine buffer, $MgCl_2$ and P-nitrophenol phosphate was taken in a clean and sterile test tube. To it, 50 µl sample was added and mixed thoroughly. The mixture was incubated at 37°C for 5 min and initial absorbance was recorded at 405 nm using a spectrophotometer (CECIL CE 2031) and subsequently three readings were taken at one minute interval. The ALP activity was expressed as IU/l and was calculated as:

ALP activity (IU/l) = 3300 A

Where, A=change in absorbance per minute

RESULTS

Biochemical composition of fish feed ingredients

Biochemical composition of fish feed ingredients (% in dry weight basis) used for the present research work is given in Table 1. The dry matter content of fish feed ingredients is the highest (95.37 ± 0.17%) in mustard oil cake and the least (91.55 ± 0.28%) in rice bran. The moisture content of fish feed ingredients is the highest (8.45 ± 0.21%) in rice bran and the least (4.63 ± 0.13%) in mustard oil cake. The crude protein of fish feed ingredients is the highest (53.60 ± 0.21%) in fish meal and the least (13.45 ± 0.13%) in rice bran. The crude lipid of fish feed ingredients is the highest (9.73 ± 0.19%) in mustard oil cake and the least (3.37 ± 0.17%) in rice bran. The carbohydrate content of fish feed ingredients is the highest (19.61 ± 0.17%) in rice bran and the least (4.33 ± 0.14%) in fish meal. The ash content of fish feed ingredients is the highest (12.50 ± 0.16%) in rice bran and the least (4.12 ± 0.17%) in mustard oil cake. The energy content of fish feed ingredients is the highest (4.92 ± 0.21 Kcal/g) in mustard oil cake and the least (1.86 ± 0.22 Kcal/g) in rice bran. Out of six ingredients, ground nut oil cake and mustard oil cake were used as the source of lipid to provide energy of 4.74 ± 0.13 Kcal/g and 4.92 ± 0.21 Kcal/g, respectively. Fish meal and soybean meal were used as protein source, providing 53.60 ± 0.21% and 50.12 ± 0.17% crude protein, respectively. Rice bran and wheat bran were used as the source of carbohydrate to provide instant energy of 1.86 ± 0.22 Kcal/g and 1.99 ± 0.26 Kcal/g, respectively. There is no significant difference ($P > 0.05$) in the biochemical composition of ground nut oil cake and mustard oil cake; rice bran and wheat bran; fish meal and soybean meal.

Table 1. Biochemical composition of fish feed ingredients **(**% in dry weight basis).

S. No.	Ingredients	Dry Matter	Moisture	Crude protein	Crude lipid	Carbohydrate	Ash	Energy (Kcal/g)
1	Ground nut oil cake	$95.09^{c} \pm 0.21$	$4.91^{a} \pm 0.18$	$42.21^{b} \pm 0.17$	$9.05^{c} \pm 0.28$	$8.62^{b} \pm 0.13$	$4.62^{a} \pm 0.21$	$4.74^{b} \pm 0.13$
2	Mustard oil cake	$95.37^{c} \pm 0.17$	$4.63^{a} \pm 0.13$	$39.56^{b} \pm 0.18$	$9.73^{c} \pm 0.19$	$7.32^{b} \pm 0.12$	$4.12^{a} \pm 0.17$	$4.92^{b} \pm 0.21$
3	Rice bran	$91.55^{a} \pm 0.28$	$8.45^{c} \pm 0.21$	$13.45^{a} \pm 0.13$	$3.37^{a} \pm 0.17$	$19.61^{c} \pm 0.17$	$12.50^{c} \pm 0.16$	$1.86^{a} \pm 0.22$
4	Wheat bran	$91.84^{a} \pm 0.23$	$8.16^{c} \pm 0.26$	$16.10^{a} \pm 0.12$	$4.58^{a} \pm 0.13$	$16.26^{c} \pm 0.19$	$11.92^{c} \pm 0.21$	$1.99^{a} \pm 0.26$
5	Fish meal	$93.82^{b} \pm 0.19$	$6.18^{b} \pm 0.16$	$53.60^{c} \pm 0.21$	$7.78^{b} \pm 0.26$	$4.33^{a} \pm 0.14$	$10.60^{b} \pm 0.20$	$3.92^{c} \pm 0.23$
6	Soybean meal	$93.63^{b} \pm 0.12$	$6.37^{b} \pm 0.15$	$50.12^{c} \pm 0.17$	$7.56^{b} \pm 0.24$	$4.72^{a} \pm 0.10$	$10.05^{b} \pm 0.18$	$3.63^{c} \pm 0.13$

Values are means ± SD. Means in the same column having different superscripts are significantly different ($P < 0.05$) and means in the same column with same superscript are not significantly different ($P > 0.05$).

Table 2. Composition of control and formulated feeds experimented (% in dry weight basis).

Ingredients	Control	Feed A	Feed B	Feed C	Feed D
Ground nut oil cake	Nil	15	18	8	16.66
Mustard oil cake	50	15	60	12	16.66
Rice bran	50	10	2	40	16.66
Wheat bran	Nil	10	8	30	16.66
Fish meal	Nil	25	4	6	16.66
Soybean meal	Nil	25	8	4	16.66
Sodium alginate (g)	5	5	5	5	5
Vitamin[1] mineral mixture (g)	Nil	2	2	2	2
Vegetable oil (ml)	Nil	1.5	1.5	1.5	1.5
Cod liver oil[2] (ml)	Nil	1.5	1.5	1.5	1.5
Oxytetracycline (mg)	500	500	500	500	500

[1]Supplevite – M (Sarabhai Chemicals India); [2]Cod liver oil (Sea cod, M/S Universal Medicare Ltd. Mumbai).

Biochemical composition of control and formulated feeds experimented

Biochemical composition of control and formulated feeds experimented (% in dry weight basis) is given in Table 3. The highest dry matter content (94.01 ± 0.19%) was recorded in feed B and the least (92.73 ± 0.28%) in feed C. The highest moisture content (7.27 ± 0.23%) was recorded in feed C and the least (5.99 ± 0.17%) in feed B. The highest crude protein (42 ± 0.26%) was recorded in feed A and the least (25.98 ± 0.19%) in feed C. The highest crude lipid (9.31 ± 0.25%) was recorded in feed B and the least(5.49± 0.18%) in feed C. The highest carbohydrate content (34.63 ± 0.19%) was recorded in feed C and the least (10.08 ± 0.10%) in feed B. The highest ash content (9.45 ± 0.16%) was recorded in feed B and the least (8.59 ± 0.26%) in feed C. The highest energy content (4.65 ± 0.13 Kcal/g) was recorded in feed B and the leas(3.48 ± 0.16 Kcal/g) in feed C. The highest P/E ratio (22.64 mg protein /Kj ± 0.36) was recorded in feed A and the least (17.18 mg

Table 3. Biochemical composition of control and formulated feeds experimented (% in dry weight basis).

Biochemical composition	Control	Feed A	Feed B	Feed C	Feed D
Dry Matter	$92.89^{a} \pm 0.17$	$93.77^{b} \pm 0.21$	$94.01^{b} \pm 0.19$	$92.73^{a} \pm 0.28$	$93.44^{b} \pm 0.16$
Moisture	$7.11^{b} \pm 0.21$	$6.23^{a} \pm 0.16$	$5.99^{a} \pm 0.17$	$7.27^{b} \pm 0.23$	$6.56^{a} \pm 0.19$
Crude protein	$26.50^{a} \pm 0.31$	$42.00^{c} \pm 0.26$	$40.00^{b} \pm 0.21$	$25.98^{a} \pm 0.19$	$34.75^{ab} \pm 0.17$
Crude Lipid	$5.80^{a} \pm 0.26$	$8.94^{b} \pm 0.19$	$9.31^{b} \pm 0.25$	$5.49^{a} \pm 0.18$	$8.22^{b} \pm 0.16$
Carbohydrate	$32.95^{b} \pm 0.18$	$12.92^{a} \pm 0.16$	$10.08^{a} \pm 0.10$	$34.63^{b} \pm 0.19$	$15.07^{a} \pm 0.22$
Ash	$8.68^{a} \pm 0.21$	$9.39^{b} \pm 0.19$	$9.45^{b} \pm 0.16$	$8.59^{a} \pm 0.26$	$9.15^{b} \pm 0.15$
Energy (Kcal/g)	$3.66^{a} \pm 0.15$	$4.44^{b} \pm 0.11$	$4.65^{b} \pm 0.13$	$3.48^{a} \pm 0.16$	$4.26^{b} \pm 0.19$
P/E (mg protein/Kj)	$17.33^{a} \pm 0.22$	$22.64^{c} \pm 0.36$	$20.54^{b} \pm 0.21$	$17.18^{a} \pm 0.19$	$19.53^{ab} \pm 0.15$

Values are means ± SD. Means in the same row having different superscripts are significantly different ($P < 0.05$) and means in the same row with same superscript are not significantly different ($P > 0.05$).

Table 4. Liver composition of fingerlings fed on control and formulated feeds after 30 and 90 days of experiment.

Observations after 30 days of experiment							
	Initial	**Control**	**Feed A**	**Feed B**	**Feed C**	**Feed D**	**± SEM**
Moisture	72.86 ± 0.31	$71.43^{a} \pm 0.18$	$71.83^{a} \pm 0.21$	$71.92^{a} \pm 0.18$	$71.32^{a} \pm 0.16$	$71.69^{a} \pm 0.14$	0.18
Crude protein	7.18 ± 0.28	$6.31^{a} \pm 0.13$	$6.59^{a} \pm 0.12$	$6.63^{a} \pm 0.21$	$6.22^{a} \pm 0.17$	$6.48^{a} \pm 0.16$	0.15
Crude lipid	3.16 ± 0.29	$4.19^{a} \pm 0.21$	$3.87^{a} \pm 0.23$	$3.79^{a} \pm 0.16$	$4.28^{a} \pm 0.17$	$3.96^{a} \pm 0.15$	0.20
Observations after 90 days of experiment							
Parameters (%)	**Control**	**Feed A**	**Feed B**	**Feed C**	**Feed D**	**± SEM**	
Moisture	$64.78^{a} \pm 0.24$	$69.73^{b} \pm 0.18$	$69.89^{b} \pm 0.23$	$64.62^{a} \pm 0.17$	$69.61^{b} \pm 0.27$	0.21	
Crude protein	$4.63^{a} \pm 0.18$	$5.06^{a} \pm 0.14$	$5.12^{a} \pm 0.16$	$4.52^{a} \pm 0.22$	$4.94^{a} \pm 0.26$	0.20	
Crude lipid	$6.63^{b} \pm 0.28$	$4.54^{a} \pm 0.19$	$4.43^{a} \pm 0.23$	$6.79^{b} \pm 0.16$	$4.67^{a} \pm 0.14$	0.22	

Values are means ± SD of five replications (d.f. 5, 35). Means in the same row in the same block having different superscripts are significantly different ($P < 0.05$) and means in the same row in the same block with same superscript are not significantly different ($P > 0.05$). SEM = Standard error of mean.

protein/Kj ± 0.19) in feed C.

Liver C

Liver composition of fingerlings (% mean wet Weight basis) fed on control and formulated feeds after 30 and 90 days of experiment is given in Table 4.

Moisture

The initial liver moisture content of the fingerlings was recorded 72.86% ± 0.31. After 30 days, there was no significant difference ($P > 0.05$) in the liver moisture content of the fingerlings fed on control feed, Feed A, B, C and D.

After 90 days, the liver moisture content was recorded the highest (69.89 ± 0.23%) in the fingerlings fed on feed B and the least (64.62 ± 0.17%) in the fingerlings fed on feed C. There was no significant difference ($P > 0.05$) in the liver moisture content of the fingerlings fed on control

Table 5. Initial and final acid and alkaline phosphatase activity (IU/l) in the liver of fingerlings fed on control and formulated feeds for 90 days.

Acid-phosphatase (ACP) activity (IU/l)							
	Initial	**Control**	**Feed A**	**Feed B**	**Feed C**	**Feed D**	**± SEM**
Enzyme activity	3.33 ± 0.21	5.56^{a} ± 0.17	8.16^{b} ± 0.11	8.67^{c} ± 0.10	5.46^{a} ± 0.15	7.48^{ab} ± 0.14	0.13
Alkaline-phosphatase (ALP) activity (IU/l)							
	Initial	**Control**	**Feed A**	**Feed B**	**Feed C**	**Feed D**	**± SEM**
Enzyme activity	0.64 ± 0.19	1.69^{a} ± 0.13	3.23^{b} ± 0.12	3.74^{c} ± 0.16	1.60^{a} ± 0.17	2.41^{ab} ± 0.14	0.16

Values are means ± SD of five replications (d.f. 5, 35). Means in the same row in the same block having different superscripts are significantly different ($P < 0.05$) and means in the same row in the same block with same superscript are not significantly different ($P > 0.05$); SEM = Standard error of mean.

feed and Feed C, A, B and D.

Crude protein

The initial liver crude protein of the fingerlings was recorded 7.18% ± 0.28. After 30 days, there was no significant difference ($P > 0.05$) in the liver crude protein of the fingerlings fed on control feed, Feed A, B, C and D.

After 90 days, the liver crude protein was recorded the highest (5.12% ± 0.16) in the fingerlings fed on feed B and the least (4.52% ± 0.22) in the fingerlings fed on feed C. There was no significant difference ($P > 0.05$) in the liver crude protein of the fingerlings fed on control feed, Feed A, B, C and D.

Crude lipid

The initial liver crude lipid of the fingerlings was recorded 3.16 ± 0.29%. After 30 days, there was no significant difference ($P > 0.05$) in the liver crude lipid of the fingerlings fed on control feed, Feed A, B, C and D.

After 90 days, the liver crude lipid was recorded the highest (6.79 ± 0.16%) in the fingerlings fed on feed C and the least (4.43 ± 0.23%) in the fingerlings fed on feed B. There was no significant difference ($P > 0.05$) in the liver crude lipid of the fingerlings fed on control feed and Feed C; A, B and D.

Enzyme activity

Initial and final acid and alkaline phosphatase activity (IU/l) in the liver of fingerlings fed on control and formulated feeds for 90 days are given in Table 5.

Acid - phosphatase (ACP) activity

The initial ACP activity of the fingerlings was recorded 3.33 (IU/l) ± 0.21. After 90 days, the ACP activity was recorded the highest (8.67 (IU/l) ± 0.10) in the fingerlings fed on feed B and the least (5.46 (IU/l) ± 0.15) in the fingerlings fed on feed C. There was no significant difference ($P > 0.05$) in the ACP activity of the fingerlings fed on control feed and feed C. The ACP activity of the fingerlings fed on feed A was significantly lower ($P < 0.05$) as compared to the ACP activity of the fingerlings fed on feed B.

Alkaline - phosphatase (ALP) activity

The initial ALP activity of the fingerlings was recorded 0.64 (IU/l) ± 0.19. After 90 days, the ALP activity was recorded the highest (3.74 (IU/l) ± 0.16) in the fingerlings fed on feed B and the least (1.60 (IU/l) ± 0.17) in the fingerlings fed on feed C. There was no significant difference ($P > 0.05$) in the ALP activity of the fingerlings fed on control feed and feed C. The ALP activity of the fingerlings fed on feed A was significantly lower ($P < 0.05$) as compared to the ALP activity of the fingerlings fed on feed B.

DISCUSSION

Liver composition

The effect of carbohydrate on liver composition has been investigated by a number of researchers (Kaushik and Oliva-Teles, 1985; Brauge et al., 1994; Khattab et al., 2000). In the present study after 90 days, the liver crude lipid of the fingerlings increased significantly and liver moisture content of the fingerlings decreased significantly with the increase in dietary carbohydrate level from 10.08 to 34.63%, while as the liver protein content of the fingerlings did not vary significantly among the fingerlings fed on control and formulated feeds. These results show, like seen by Hilton and Atkinson (1982), Wee (1986), Kim and Kaushik (1992), Mazur et al. (1992), Brauge et al. (1994) and Habib et al. (1994) in several fish species, an increase in liver lipid content with the increase in dietary carbohydrate level.

The higher liver lipid content in fingerlings fed on feed

C and control feed is due to the inability of the common carp to utilize excess dietary carbohydrate level (above 15%) due to omnivorous feeding habit, which get converted into lipid in the liver thereby resulting in fatty liver, which is not desirable for economic production of fish. The inability of the omnivorous fish to utilize the dietary carbohydrate levels above 15% has been reported by several researchers (Furuichi and Yone, 1982 in common carp and red seabream; (Anderson et al., 1984 in tilapia; Furuichi et al., 1986 in yellow tail; Wilson and Poe, 1987 in channel catfish). The least lipid content in the liver of the fingerlings fed on feed B is due to the optimum dietary carbohydrate level (10.08%) required for common carp, which does not result in fatty liver.

Enzyme activity

The ACP and ALP activity of the fingerlings fed on feed A was significantly lower when compared to the ACP and ALP activity of the fingerlings fed on feed B. The ACP and ALP activity of the fingerlings increased significantly with the increase in the dietary P/E ratio up to 20.54 mg protein/Kj. Above 20. 54 mg protein/Kj dietary P/E ratio, the ACP and ALP activity of the fingerlings decreased significantly. The ACP and ALP activity was the maximum in the fingerlings fed on feed B having optimum dietary P/E ratio of 20.54 mg protein/Kj because the proteins are not catabolized for energy at optimum dietary P/E ratio. At low dietary P/E ratio and above optimum dietary P/E ratio, the dietary proteins are catabolized for energy hence the ACP and ALP activity was least in the fingerlings fed on feed C and decreased above optimum dietary P/E ratio. The utilization of dietary proteins for energy at low dietary P/E ratio has been observed in several fish species (Company et al., 1999; McGoogan and Gatlin III, 2000; Yamamoto et al., 2000). Since the fingerlings fed on feed B are having the maximum ACP and ALP activity, it could be said that they are having better metabolic activity because metabolic activity is correlated with ACP and ALP activity (Chen et al., 1997). The maximum acid and alkaline phosphatase enzyme activity at optimum P/E ratio coincide with the works of Shimeno et al. (1997), Erdal and Genc (2006), Wang and Zirong (2008), James et al. (2009) and Siakpere et al. (2010) in several fish species.

Conclusion

Based on liver composition and enzyme activity this work concludes that a dietary containing 40% protein, 9.31% lipid, 10.08% carbohydrate and having P/E ratio 20.54 mg protein/Kj is the best one for a more profitable and successful culture of the common carp.

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