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Effect of Richmin and Vanimin on Carbohydrate metabolism in selected fish species – *C. catla, L. rohita, C. mrigala*

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ABSTRACT

Carbohydrates are one such group of carbon compounds which are essential to life. Almost all organisms use carbohydrates as building blocks of cells and as a matter of fact, exploit their rich supply of potential energy to maintain the life. The effect of Richmin and Vanimin on carbohydrate metabolism in different fish species were analysed in this study. The experimental group of fishes shall further be divided into two groups. Richmin and Vanimin which are commercially available are been selected for the study. To assess the carbohydrate metabolism effect of Richmin and Vanimin in *C. catla, L. rohita, C. mrigala*, the total carbohydrates, serum glucose levels, liver and muscle glycogen concentration were measured. The richmin and vanimin fed fish species muscle and liver showed enhanced levels of their total carbohydrate content over their corresponding control values and the increment was found to be statistically significant ($P<0.001$) over the control values. Statistically significant ($P<0.001$) increase in the liver and muscle and liver tissues glycogen content over the control feed fed fish tissues was registered in the present study for the fish tissues fed with richmin and vanimin. Richmin and Vanimin fed fishes serum showed increased levels of their glucose content. The study revealed that synthetic feed Additives have accelerative upon the carbohydrate metabolism of fish species.

Key words: Richmin, Vanimin, *C. catla, L. rohita, C. mrigala,* Glucose, Glycogen.

INTRODUCTION

Carbohydrates are one such group of carbon compounds which are essential to life. Almost all organisms use carbohydrates as building blocks of cells and as a matter of fact, exploit their rich supply of potential energy to maintain the life. Carbohydrates constitute by far the greatest proportion of organic material on the face of the earth and the most abundant carbohydrate is cellulose which forms the main supporting structure of plants. They constitute important materials for the necessities of life such as food and clothing, housing and health. Glycosides of the digitalis-strophanthus group are used in medicine and it is recognized that the invasiveness and antigenic nature of some bacteria is due to their polysaccharide envelope. A great many of the wondrous colours of the plant kingdom are due to the glycoside pigments of the anthocyanin and flavones groups. The nourishment of the cell come from the circulating glucose of the extra cellular environment and the activities of the cell may be profoundly altered by the mucopolysaccharides of the surrounding extra cellular tissue or the blood borne glycoprotein hormones of the anterior pituitary. Mucopolysaccharides are also biological lubricants in the form of saliva, mucus and synovial fluid. (Prakash, Arora, 1998).

Impairment of carbohydrate metabolism has been observed in a variety of physiological disorders and pathological conditions (Harper *et al*., 1979). This may prove to be of negative survival value for the affected organisms.

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Investigations were conducted earlier on carbohydrate metabolism during pathological conditions in different animals following exposure of some kinds of pesticides (Dikshit *et al*., 1975 Wolnicki *et al.,* 2003). Glucose in the blood which shows most striking alternations in its concentration in response to change in environmental factors (Umminger, 1975). More over in several fishes blood glucose level has been correlate to their level of activity (Gangadhar, 1998) and thus ultimately the level of blood glucose is attributed to and indirect level of B.M.R. in fishes (Umminger, 1977).

Glucose is the principal sugar in blood of fishes, serving the tissues as a major metabolic fuel. Besides yielding energy through glycolysis and TCA cycle, pentose sugars are also formed in the hexose monophosphate shunt from glucose, which are important constituents of nucleotides, nucleic acids and many coenzymes. In general glucose level in the blood of circulating fluid is maintained in an animal through active absorption of glucose from the digested food stuffs, and also is formed from glycogen, amino acids and glyceral through glycogenolysis and gluconeogenesis under certain stress conditions. In several fishes, blood glucose level has been correlated to their level of activity and hence to their level of metabolism. There are evidences that in fish's blood glucose level shows most striking alterations in response to the change in environmental factors (Umminger, 1975; Hattinght, 1977). The levels of it may even be affected under toxic stress, which reflects the variations in the entire carbohydrate metabolism, (Tewari *et al*., 1987). Blood glucose level has been reported as a reliable and sensitive indicator of environmental stress in fishes (Silbergeld, 1974).

Glycogen, commonly called as animal starch, is the main storage polysaccharide and a great source for blood glucose. Maintenance of glycogen reserves is one of the important features of the normal metabolism (Mong and Poland, 1981). Alterations in liver and muscle glycogen under situations of stress have been reported, and a significant depletion if tissue glycogen is said to reflect the state of strenuous activity on the part of the fish (Tewari *et al*., 1987). In many of the fishes red muscle is known to be predominantly oxidative where as white muscle is known to be predominantly glycolytic (Gordon, 1968). Hence the white muscle which is more active anaerobically could accumulate more inert metabolic glycogen than the red muscle, capture, handling, nutritional status all have profound effects on the carbohydrate metabolism and blood electrolytic balance. (Donaldson, 1981). Further, depletion of glycogen indicates the rapid utilization of energy stores to meet the energy demands warranted by the environment (Basha Mohideen and Sudharshan Reddy, 2003). Donaldson *et al.,* (1981) studied seasonal variations in tissue glycogen content in exotic fish *Cyprinuscarpio*. But, studies involving carbohydrate energy reserves in fishes exposed to different nutritional media are highly scanty. Therefore, the objective of this study is to evaluate the effect of Richmin and Vanimin on Carbohydrate metabolism in selected fish species - *C. catla, L. rohita, C. mrigala.*

MATERIALS AND METHODS

Synthetic Feed Additives

For the present study stocking / Breeders pond, breeding tubs, Hatching tub and Nursery cum Rearing ponds were used at the Government fish farm at Warangal (District) Telangana, India. The breeders were fed with shell, rice bran and ground nut oil cake regularly at the rate of 2% body weight of the fish. The fishes selected for the study shall be divided into two groups viz. control group and experimental groups .The control group of fishes shall be fed with control feed i.e. Groundnut cake, rice bran. The experimental group of fishes shall further be divided into two groups. Richmin and Vanimin, which are commercially available have been selected for the study. All other chemicals used are of technical grade from PVS laboratories, Vijayawada, Andhra Pradesh (India).

The first group of experimental fish shall be fed with control feed mixed with Richmin. The second group of experimental fish shall be fed with control feed mixed with vanimin. The two groups of experimental fish shall be fed twice a day at 10 a.m. and at 5 p.m. The exposure period selected for the study is 30 days. After 30 days the fishes were killed and isolated the tissues like muscle and liver at 4^0C and stored at - 80^0C.

General Experimental Conditions:

For the present study, the following experimental ponds and tubs were used at the Government fish farm, at Warangal, District (Telangana). The breeders were fed with shellar rice bran and ground nut oil cake regularly at the rate of 2% body weight of the fish.

Pond types:

Stocking / Breeders pond: There are two breeders' ponds; these are called as segregation ponds for maintaining and rearing of breeding species. The shape of the pond is rectangle of size 100'x30'x4'. Each pond is provided with inlet, outlet and overflow pipe. The bottom of pond is katcha to enable the breeders, to grow well and for buffer action. Every week the stagnant water is replaced with fresh water through exchange method.

Breeding tubs: 4 cement breeding tubs of size 15'x10'x4' were used for breeding of major carps. Each pond is provided with an inlet of 2" GI pipe and 2" outlet is provided at the bottom for bailing out water. Prior to, the water is released into the pond upto the over flow pipe, care is taken to maintain the water level with continuous water flow.

Hatching tub: The hatching tub is echo-hatchery type and movable. The tub is made up of zinc sheet and is cylindrical in shape. The tub height is 2.10′ and dia of 3.2′. There are two chambers inside the tub since there is a round mesh of 1.80′ height and 30 cm dia. There is one outlet in this tub to drain out the spawn after hatching. There is one semicircular chamber ½″ pipe with nozzle to circulate water flow and to wash the eggs with freshwater continuously. The nylon cloth is inserted over the second chamber to arrest the over flow of eggs into the second chamber.

Nursery cum rearing pond: These ponds are built with brick and cement and the shape of nursery pond is rectangle, having the size of 50′x15′x4′. Each nursery is provided with an inlet, outlet and overflow pipe. The bottom and side walls of the nursery are plastered with cement to make them smooth. The inlet is connected to the pipeline to draw water. The inlet is provided with 3 inch gate valve to regulate the flow of water.

Experimental Methods

Estimation of Total Carbohydrates:

The Carbohydrate content was estimated by the method of Carrol et al., (1956). The tissues 0.2 ml homogenates were individually homogenized in 10% trichloroacetic acid and centrifuged at 3000 rpm for 15 minutes. To 1.0 ml of TCA supernatant, 4 ml of anthrone reagent was added and the colour was read against the reagent blank at 600 nm in a spectrophotometer. From the optical density, the total carbohydrate content was calculated in comparison with the standard and the values were expressed as mg carbohydrates/gm/wet weight of the tissue.

Estimation of Glucose:

Glucose in the samples was determined by colorimetric method as described by Nelson and Somogyi (1952). 0.1 ml of blood was collected. 3.9 ml of deproteinizing solution (5% Zinc sulphate and 0.3 N Sodium hydroxide in 1:1 ratio) was added to it and the mixture was centrifuged at 3000 rpm for 10 minutes. To 1 ml of the supernatant from each of these mixtures, 1 ml of alkaline copper reagent was added and it was shaken vigorously and heated in a boiling water bath exactly for 20 minutes. Then it was cooled and 1 ml of arsenomolybdate colour reagent was added to it. Distilled water was added to the entire solution till it measured 10 ml and the optical density of the colour developed was measured in a spectrophotometer at a wave length of 540 nm. A blank and glucose standards were also run simultaneously. Glucose content was expressed as mg of glucose / 100 ml of blood.

Estimation of Glycogen:

Glycogen content in liver and muscle of fish was estimated using the anthrone reagent method as described by carroll et al., (1956). Since glycogen concentration in muscle is known to vary in different regions of body (Nandeesha et al, 2002) care was taken in dissecting out this sample from the same region of body of fishes i.e., The antero dorso lateral region of the trunk. The organs were digested with 3.0 ml of hot 30% potassium hydroxide (Oda et al, 1958). The digestate was cooled and 3.75 ml of absolute ethanol was added to it. The entire mixture was kept over night in a refrigerator. Then the mixture was centrifuged for 15 minutes at 2500 rpm. Decanted the supertanant and 10.0 ml of warm distilled water was added to the residue to dissolve the precipitated glycogen. To 0.2 ml of this 1.8 ml

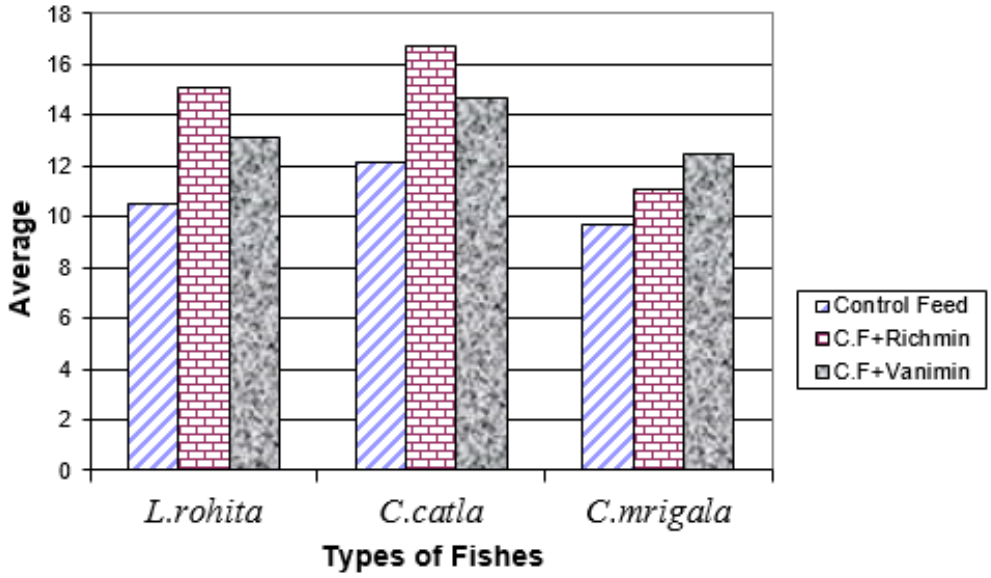

Figure-1. Impact of Richmin and Vanimin on the *Total Carbohydrates in Muscle* of Selected Fish species *C.catla, L.rohita, C.mrigala*

at distilled water and 0.5 ml of 2% anthrone reagent dissolved in 72% concentrated sulphuric acid were added and heated in a boiling water bath exactly for 10 minutes. The mixture was cooled and the optical density of the colour developed was measured in a spectrophotometer at a wavelength of 620 nm. A blank and glucose standards were also run similarly. The glycogen content is expressed as mg/g wet wt of the organ.

RESULTS AND DISCUSSION

The richmin and vanimin fed fish species muscle and liver showed enhanced levels of their total carbohydrate content over their corresponding control values and the increment was found to be statistically significant ($P<0.001$) over the control values. Liver appeared to possess more carbohydrate content compared to the muscle tissue (Fig. 1, 2).

Statistically significant ($P<0.001$) increase in the liver and muscle and liver tissues glycogen content over the control feed fed fish tissues was registered in the present study for the fish tissues fed with richmin and vanimin. Liver tissue of the three fish species appeared to possess high glycogen content in the present study (Fig.3, 4).

Richmin and vanimin fed fishes serum showed increased levels of their glucose content (Fig. 5) and the changes were found to be statistically significant ($P<0.001$) over their control values.

Carbohydrate metabolism is essentially the metabolism of glucose and substances related to glucose. Glucose occupies central position of carbohydrate metabolism in an organism, representing complex groups, sequences and cycle of reactions which integrate at various points. The reactions concerned with metabolism of lipids and proteins as these molecules serve the source of carbon in the synthesis of cellular components (Nelson and Cox 2000). Glycogen is the chief carbohydrate present in tissues, while glucose is of the blood and other body fluids. Glycogen a storage carbohydrate from intestinal absorption is inadequate. Glycogen breakdown into glucose is governed by the extrinsic and intrinsic factors which also controls the physiology of an organism.

Carbohydrate metabolism essentially constitutes two segments namely synthesis of Carbohydrates which includes gluconeogenesis and catabolism which includes glycolysis, glycogenolysis, pentose pathway and Krebs cycle. The catabolic pathways not only fulfill the needs of energy demands but also supply the amphibolic intermediates and reduced neuotides (NADPH) required for protein and lipid metabolism (Nelson and Cox, 2000).

The mechanism by which glycogen is synthesized and broken down in tissues is initiated by phosphorylase enzymes. The process of glycolysis in tissues commences with the consent of glycogen breakdown and the glucose released is fragmented into three carbon compounds, pyruvic acid and lactic acid by a series of enzymes under anaerobic conditions. The end products lactate and pyruvate are interconvertible by the enzyme lactate dehydrogenese (LDH). Pryuvate undergoes oxidative de carboxylation by pyruvate dehydrogenese to provide acetyl Co-A.

Acetyl-Co-A is an essential substrate for Kreb's cycle, which generates reduced nucleotides for the ultimate generation of ATP molecules through electron transport. Amphibiolic intermediates formed in the Krebs cycle may be channeled into amino acid or fatty acid synthesis.

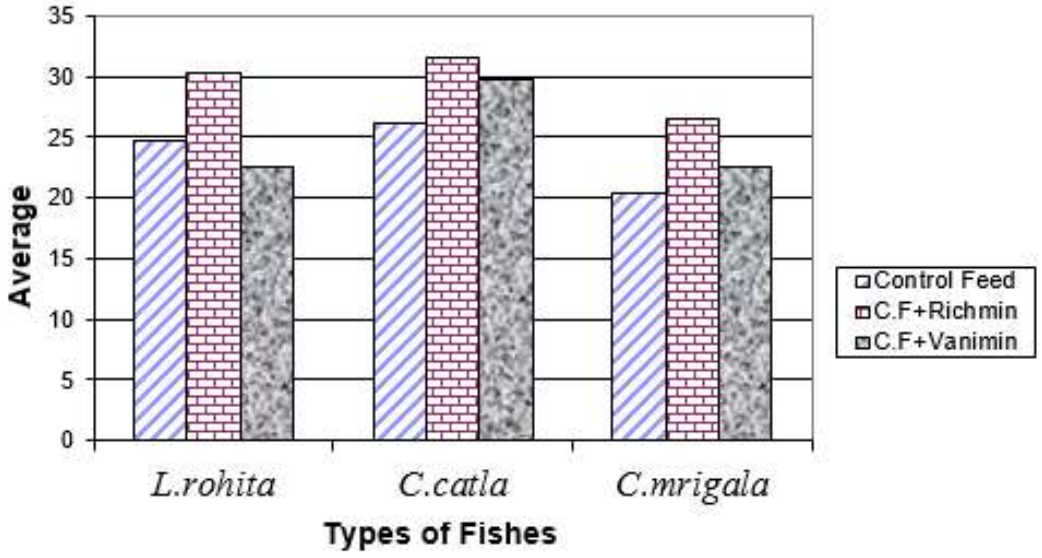

Figure-2. Impact of Richmin and Vanimin on the *Total Carbohydrates in Liver* of Selected Fish species *C.catla, L.rohita, C.mrigala*

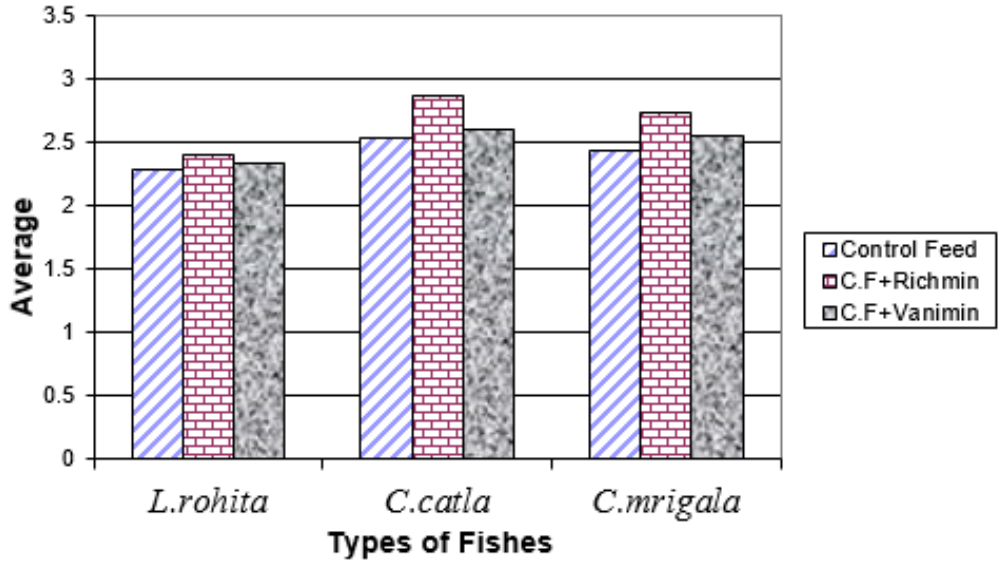

Figure-3. Impact of Richmin and Vanimin on the *Glycogen in Muscle* of Selected Fish species *C.catla, L.rohita, C.mrigala*

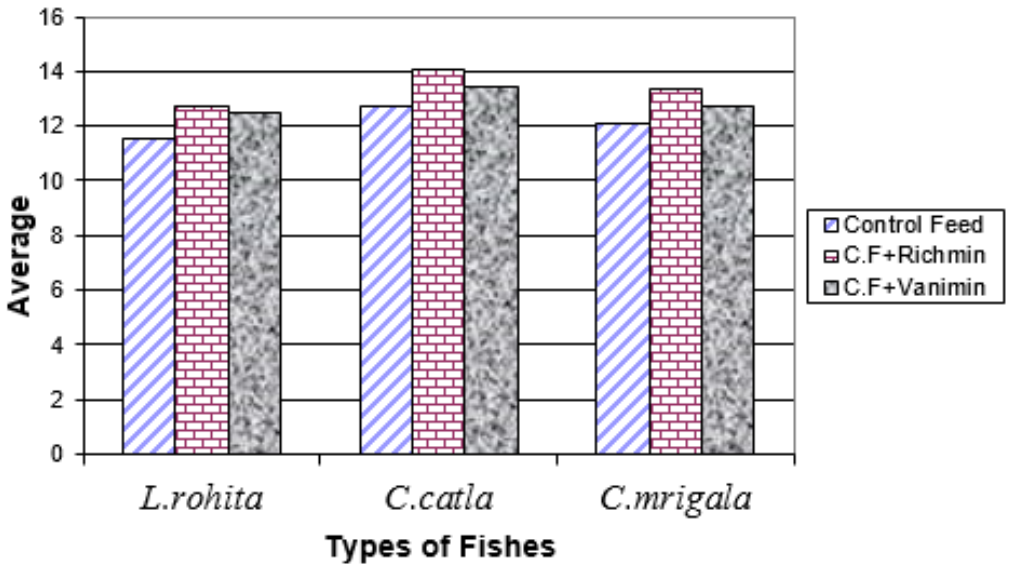

Figure-4. Impact of Richmin and Vanimin on the *Glycogen in Liver* of Selected Fish species *C.catla, L.rohita, C.mrigala.*

Channeling of carbohydrate precursors into energy yielding reactions or synthetic reactions depends on the biochemical make up of an organ system concerned or physiological alterations dependant on changed environmental conditions. Pyruvate utilisation for energy requirements is tissue specific and varies according to environmental conditions imposed on the animal.

Since biological system has some flexibility, animals use this capacity to divert metabolic pathways to an alternate source to synthesize energy to overcome the energy crisis created by the stress. The increased levels of total carbohydrates, glycogen (Fig. 1,2,3,4) in the muscle and liver of richmin and vanimin fed fishes indicate that these tissues and blood show an upsurge in glycogenesis and glycogenolysis and this situation was more in the tissues and blood of richmin and vanimin fed fishes. This trend further may be due to accumulation of feeding capacity of the fish species selected for the study. The overall results on carbohydrates glycogen and glucose in the present study supports an upsurge of carbohydrate metabolism in the fish tissues and blood fed on either richmin and vanimin.

The hepatocytes contain glycogen and the amount of which in the liver cells depends upon the nutritional state of the animal (Junqueira et al., 1995). Also, hepatocytes contain varying amounts of lipid in electron-dense

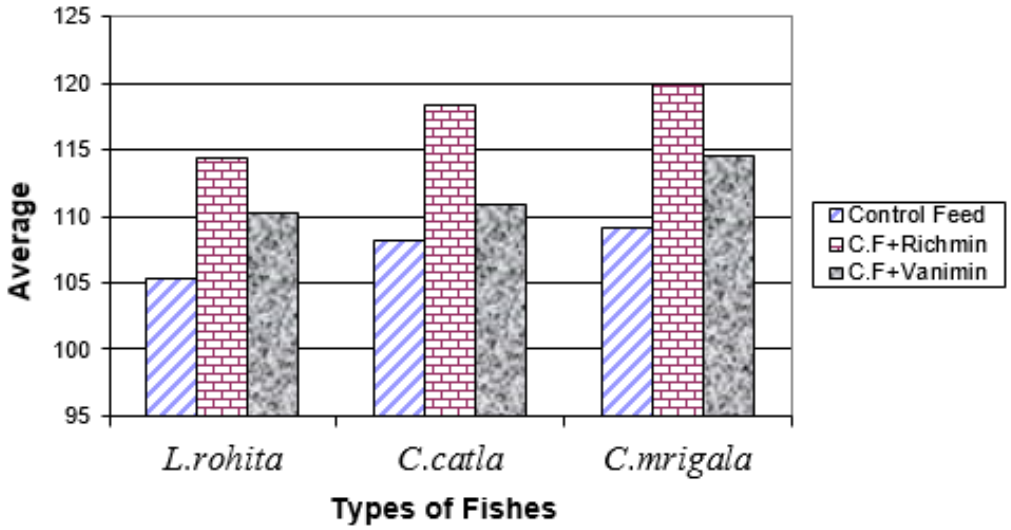

Figure-5. Impact of Richmin and Vanimin on the *Blood Glucose* of Selected Fish species *C.catla, L.rohita, C.mrigala.*

droplets; they are few in number in the normal liver but are dramatically increased after consumption of hepatotoxic substances. Hence, fish hepatocytes are good indicator of dietary quality (Kugler and Pequignot, 1988). Mixed diets induced better liver histology and bigger hepatocytes of fish fed on feed are may be due to the increased glycogen inclusion. Lipid vacuolations were prominent and abundant in fish fed with T2 diet especially during the early developmental stages (10-30 dpff). Increased liver lipid deposits may indicate diet of insufficient vitamin content, too much carbohydrate and high-unsaturated fatty acids (Kugler and Pequignot, 1988).

CONCLUSION

The study revealed that synthetic feed Additives have accelerative upon the carbohydrate metabolism of fish species. Thus, Richmin and Vanimin additives have enormous nutritional components which enhance the biomass of the fishes and this result shows in improving the yield and productivity of fresh water carps. Overall in this study the feed additives appeared to be more beneficial in improving the metabolism and fish yield to farmers. From the present experimental work, the author concludes that both Richmin and Vanimin increase the productivity over the control hence they may be used in Aquaculture practices.

group HDL was 36.2 mg/dl and HDL increased to 28.36% when compared to the diabetic control group rats.

LDL in normal control group rats was 40.2 mg/dl and in diabetic control (Alloxan treated) group 86.2mg/dl, in standard control (Glybenclamide) group 39.4 mg/dlLDL was decreased to 51.27% when compare to the diabetic Control group rats in *Chloroxylon swietenia* plant extract treated group Wistar rats and it was 38.2 mg/dl.

Measurement of triglycerides, total cholesterol, HDL and LDL profiling helps to understand the risk of diabetes. Impairment of insulin secretion or it absence leads to an excessive and prolonged increased and decreased concentration of triglycerides, total cholesterol, HDL and LDL reduces triglyceride, total cholesterol, and LDL cholesterol levels in people with diabetes (Alam khan, *et al* 2003). After consumption of Jaman fruit extract, there was decrease in serum TGL in some individuals but not significant. Total cholesterol was decreased, but not-significantly. Also LDL was not-significantly decreased. HDL was not affected (Mahpara Safdar *et.al.*,2006).

Cissus sicyoides significant decreases in triglyceride levels (Glauc*etal*., 2004; Mamidala et al, 2020). The treatment with *Chloroxylo swietenia* similar was the situation with *M. vulgare*reduced the hypercholesterolemia. In relation with triglycerides, *C.obtu sifolia* decreased the basal hyper triglyceridemia from 340.5 to 197.48 mg/dl and the extract from *M. vulgare* reduced the basal values of triglycerides by 5.78%. But the differences were not significant (Herrera-Arellano *et al*., 2004). The hypolipidemic action of flavonoid rich extract obtained from seeds of *Eugenia jambolana*was confirmed by significant decrease in the levels of LDL(27%-29%), triglycerides (about 35%- 37%) and increase in HDL (21%-34%) over untreated diabetic rats (Sharma *et al*. 2008). HDL cholesterol, a friendly lipoprotein, was decreased in both the diabetic groups in respect to the control (Mamidala et al, 2020; Sharma *et al*. ,2008).

However, in all these cases, the cholesterol levels were found to be in the normal range. Similar trend was noticed in the levels of triglycerides and LDL-cholesterol (LDL-C) in all the groups. This present research study demonstrated the effects of *Chloroxylon swieteniaon* the reduction of glucose, triglyceride, LDL cholesterol, and total cholesterol levels in Wistar rats.

Conflicts of Interest

Authors declare that there is no conflict of interests regarding the publication of this paper.

References

[1] Bashamohideen, Md. And Sudharshan Reddy, K. 2003. Effect of sublethal exposure of malathion on carbohydrate metabolism in field crab *O.senex senex* with special emphasis on body size. *J. Invertebrate. Aquatic Biol*. 1:54-58.

[2] Carrol, N.V., Longley, R.W. and Row, J.H. 1956. Glycogen determination in liver and muscle by use of anthrone reagent. *J. Biol. Chemi.* 22:583-593.

[3] Dikshit, T.S.S., Datta, K.K. and Pandya, K.P. 1975. Effect of DDT on the free amino acid pool of brain and kidney in guinea pigs. Environ. Physiol.Biochem. 5:404-407.

[4] Donaldson, E.M. 1981. The pituitary internal axis as an indicator of stress in fish. In stress and fish (A.D. Pickering, Ed.) : 11-47.

[5] Gangadhar, B., M. C. Nandeesha, T. J. Varghese and P. Keshavanath 1998. Effect of feeding 19 - Norethisterone on growth and body composition of rohu, *Labeo rohita. Asian Fish. Sci.,* **11**: 51-58.

[6] Gordon, M.S. 1968. Oxygen consumption of red and white muscle from tuna fishes. Science, 159: 87-90.

[7] Harper, H.A., Rodwell, V.M. and Mayer, P.A. 1979. In: Review of physiological chemistry, 17th edition, *Longe Medical Publications*, Maruzer company limited, California.

[8] Hattingh, J. 1977. Blood sugar as an indicator of stressing the fresh water fish, *Labeo rohita* (Smith.). *J.Fish.Biol*. 10:191-195.

[9] Kugler C.O.J. and J. Pequignot 1988. Growth and Histological Studies on the Liver and Anterior Intestine of Nile Tilapia (Oreochromis niloticus) Fed on Mixed Foods: Daphnia magna Chlorella vulgaris and Commercial Carp Pellets. In : R.S.V. Pullin,

[10] Mamidala, E., Davella, R., Gurrapu, S., & Shivakrishna, P. (2020). In silico identification of clinically approved medicines against the main protease of SARS-CoV-2, causative agent of covid-19. arXiv preprint arXiv:2004.12055.

[11] Mong, F.S.F. and Poland, J.L. 1981. Substrate restoration in muscle grafts of rat. Ires. Med. Sci. 916:508-512.

[12] Nandeesha, M.C., Gangadhara. B. and Manissery, J.K., (2002). Further studies on the use of mixed feeding schedules with plant- and animal-based diets for common carp *Cyprinus carpio* (Linnaeus). Vol.33 : 14 pp.1157

[13] Nelson, N. and Somogyi, N. 1952. A photometric adaptation of somogyi method for the determination of glucose. (L.). Applied Ichthyology, **19:**157-160.

[14] Oda, T., Seki, S. and Okazaki, H. 1958. Colorimetric methods for the estimation cytochrome – C – oxidase and cytochrome-c-oxidase systems. Acta. Med, Okayama. 12 : 293-301.

[15] Padmavathy, P., Ramanathan, N. and Francies, T. 2002. Oxygen utilization in *Cirrhinus mrigala* and *Labeo rohita* under normoxic and hypoxic conditions. Indian. J. Fish. 49(2) : 115-121.

[16] Prakash, M. and Arora, K. 1998. Physiological biochemistry. Annual publications. 1-3.

[17] Silberged, E.K. 1974. Environmental stress in fishes. Bull. Environ. Contam. Toxicol. 11:20.

[18] Tewari, H., Gill, T.S. and Pant, J. 1987. Impact of chronic lead poisoning on the hematological and biochemical profiles of a fish, *Barbus conchonius* (Ham.). Bull. Environ.Contam.Toxicol.38:748-752.

[19] Umminger, B.L., 1975. Physiological studies on super cooled kill fish, *Fundulus heteroclitus,* III carbohydrate metabolism and survival at subzero temperature. J.Exp.Zool.173:159-174.

[20] Umminger, B.L., 1977. Relation of the whole blood sugar concentration in vertebrates to standard metabolic rate. Com.Biochem.Physiol. 56a: 457-460.

[21] Wolnicki, J., Myszkowski, L. and Kaminski, R., 2003. Effect of supplementation of a dry feed with natural food on growth, condition and size distribution of juvenile tench *Tinca tinca* (L.). Applied Ichthyology, **19:**157-160.