ISSN 2250-3153

Synthesis and Accumulation of Tissue Triglycerides as an Index of Starvation in Anabas testudineus(Bloch)

Padmavathi Godavarthy[*], Y.SunilaKumari[**] and E.Bikshapathy[***]

[*] Department of Zoology, Osmania University College for Women, Koti, Hyderabad-500095, India
[**] Department of Zoology, Osmania University College for Women, Koti, Hyderabad-500095, India
[**] Centre for Cellular and Molecular Biology (CCMB) Uppal, Hyderabad, 500007, India

Abstract- Triglycerides are stored lipids with high calorific value, and so may be the preferred choice of fuel during starvation, which may be partially utilized and also may be a simultaneously synthesized along the degradation, probably to balance the lipid over load and also to prevent lipotoxicity of cells caused due to accumulation of FFA(free fatty acids). There may not be any absolute depletion of lipids, during starvation as lipid is considered to be the element constant. To study and understand the relevance of triglycerides during starvation we selected, Anabas, testudineus, a sturdy fish from coastal waters of Andhra Pradesh (INDIA) and subjected it to brief (15days) and prolonged fasting (60days).Triglyceride concentration was estimated by the method of Raghuramulu et.al (1983).Six non adipose tissues, like liver, kidney, brain accessory respiratory organ, pectoral and lateral line muscle were chosen for the study. There was an overall upsurge observed in the triglyceride levels. During the short term fasting stress, the increase was found to be a significant one in tissues such as liver ($P<0.01$), kidney,($P<0.001$) brain,($P<0.001$) accessory respiratory organ($P<0.001$) and lateral line muscle($P<0.001$). Pectoral muscle (NS) however showed an insignificant rise. Long term starvation results were different from that of the short term. Tissues such as kidney($P<0.05$) and brain($P<0.05$) showed a significant rise and other non adipose tissues such as kidney, pectoral and lateral line muscle, all showed an insignificant increase. Accessory respiratory organ did not show any change in the triglyceride content. Anabas, adapted well to the starvation stress and survived all through the experimental period.We suggests that the triglyceride synthesis and accumulation during fasting may be used as an index to describe starvation status of anabas.

Index Terms- Triglycerides, Anabas testudineus, starvation stress, lipotoxicity, FA (fatty acid) overload, triglyceride/FFA flux

I. INTRODUCTION

Starvation has certainly been a major factor in natural selection, since the beginning of life. Among the vertebrates, fishes in particularly, teleosts are known to undergo prolonged periods of starvation (Larson and Lewander, 1973; Loughna and GoldSpink, 1984).Many species of fish tolerate starvation both in their natural environment during migrations and also during reproduction (Hinch *et.al* 2005; Miller *et.al*, 2009).Fishes have evolved the capability to endure long term food shortage, by reducing the metabolic rates and energy expenditure (Mayz, 1996 and Conner *et.al*, 2000). During these starvation periods, fishes reduce their energy expenditure which is largely derived from protein turnover (Salem *et.al*,2000).However no uniform pattern has been observed in the nutrient utilization and there is a species dependent variation in the utilization of energy reserves, as some species utilize glycogen as first energy reserve (Hung *et.al*, 1997;Figuerdo-Garutti *et.al*,2002; Meton *et.al*,2003) while in certain fishes, glycogen is spared and lipid or protein or both are utilized as energy substrates (Sheriden and Mommenson,1991; Navvorro and Guietrezz,1995; Gillis and Ballantyne;1996). Authors such as Ida Coordt Elle *et.al* (2012), opined that during times of nutrient scarcity, fat depots such as triglycerides can be catabolized to yield glycerol and FFA. During fasting, metabolic alterations seem to be rather complicated with a fluctuating utilization of all three main groups of energy reserves; proteins, lipids and carbohydrates(Dave *et.al*;1975).Starvation study by Keyes *et.al*, (1950) on humans and on emperor penguins, by Robin *et.al*,(1988) suggests that fat is the energy source of metabolism and this may be because of lipid being a far more superior form of stored fuel from gravimetric view point and the energy obtained by lipid utilization is ~ 9.4 cal/ gm of lipid stores (Cahill,1986). During many a times, when organisms are deprived of food, they engage in a set of evolutionarily conserved behavioral, physiological and structural responses to reduce overall metabolism which involves the activation of lipolysis and fatty acid degradation (Wang, *et.al*, 2006), and hence it is reasonable to assume that most of the fasting animals may depend upon lipid reserves for their energy requirements. From these studies it is quite evident that lipid seems to be a preferred form of fuel for the starving animal and so the present investigation is an attempt to understand and analyze the lipid(triglycerides) mobilization and utilization during brief and prolonged fasting. For this purpose we selected a sturdy fish, *Anabas testudineus*, found in the local ponds and rivers of Andhra Pradesh (INDIA) which is known to withstand starvation approximately for a period of about 2-4 months. To understand if there was any tissue specific selectivity observed for the utilization of lipids in the non adipose tissues, we selected six tissues such as liver, kidney brain, accessory respiratory organ, pectoral and lateral line muscle, for the study.

II. MATERIAL AND METHOD

Fish weighing of 20-25gm were obtained from Kolleru Lake of Eluru, Andhra Pradesh (INDIA). Care was taken to ensure quick transport to the laboratory. Overcrowding was avoided during packing to minimize the mortality rate. They were carefully transferred into Durex storage tank of capacity 500 liters, made of material corrosive resistant polypropylene. The closed plastic lid of the tank was replaced by a grill lid made of iron. This helped in proper ventilation and aeration of the tank. Fish which were injured or dead were removed from the tank from time to time. Disinfectant (KMnO4) was used to avoid infection. Fish were fed with boiled egg, rice bran meal and commercial fish feed *ad. libitum*. Any leftover feed and fecal matter were removed daily. Water in the tank was changed every day. Fish were brought to the laboratory and sufficient time was allowed for acclimatization. Experimentation was done thereafter. Fish measuring about 3'-4" in length and in the same range of weight were selected carefully and were grouped together and kept in circular tubs made of plastic. The mouth of these tubs was covered with fine mesh and appropriately placed such that they were properly ventilated and well aerated. Two types of experimental set ups were designed.

In the first set up, fishes were allowed to starve for 15 days and a parallel control was also maintained. The control animals were fed regularly both in the morning and evening. On the 16th day both experimental and control animals were sacrificed by concussion, and the tissues were removed for biochemical analysis. The second experimental set up consisted of fishes which were allowed to starve for two months (long term). Experimental group and a corresponding control group were maintained. Control group was fed regularly as in the case of the short term. On the 61st day, the animals were sacrificed, for the experimentation. Six animals in the control and six in the experimental group were killed by concussion and the tissues were removed and used for further experimentation. The tissues selected for the experiment were liver, kidney, brain, pectoral muscle, lateral line muscle and accessory respiratory organ. The tissues after being removed were quickly transferred to ice. They were mildly dried using tissue paper, before weighing. The weight of the tissues was estimated using electric balance. The weighed tissues were thoroughly homogenized using mortar and pestle with isopropanol as the medium. Triglycerides were estimated by the method of Raghuramulu, *et.al* (1983)

Principle: Glycerol moiety is oxidized to formaldehyde and the latter condensed with ammonia and 2, 4, Pentanedione to produce 3, 5 diacyl, 1, 4, dihydrotoludene which is yellow in colour and has absorption at 405 nm.

III. METHOD

A known quantity of tissue was homogenized with 4 ml of Isopropanol. 0.4 grams of alumina was added to the tubes. These tubes were placed on mechanical shaker for 15 minutes, and later subjected to centrifugation. After centrifugation, 2 ml of supernatant was transferred into separate tubes. 0.6ml of saponification reagent (alcoholic KOH) was added to the tubes and incubated at 60-700C for 15 minutes. After cooling, 1ml of Sodium meta-periodate solution was added and mixed thoroughly. 0.5 ml of acetyl acetone reagent was added and again mixed. The tubes were incubated at 500C for 30 mints. After cooling the optical density was read at 405 nm in a spectrophotometer, against the distilled water blank. A working standard was prepared using triolein, and the triglyceride content was expressed as mg/gm wt of tissue.

IV. RESULTS

Short term food deprival of *Anabas*, led to a rise in triglyceride content in all of the tissues studied. There was a significant elevation found in most of tissues except for the pectoral muscle. Liver showed an increase of 196.8% ($P<0.01$) kidney showed a rise of 196.9% ($P<0.001$). The increase in brain was 183.5% ($P<0.001$). The rise in accessory respiratory organ was 110.6% ($P<0.001$) and the lateral line muscle showed an increase of 128.5% ($P<0.001$) (Tables-I, II, III; Figs.-1, 2, &3).**Long term starvation** of two months (60 days), of *Anabas*, showed an elevation in the triglyceride content in all of the tissues except for accessory respiratory organ. The increase was statistically significant in tissues like kidney and brain, where as in other tissues like liver, pectoral muscle and lateral line muscle the rise was not statistically significant. Liver showed an increase of 43.4% (NS). Kidney showed an increase of 59.4% ($P<0.05$). The increase in brain was 34.6% ($P<0.05$).Accessory respiratory organ did not show any change in the triglyceride content. The value remained the same, in both the control and experimental groups. The % rise in pectoral muscle was 14.9% (NS) and the increase in lateral line muscle was 32% (NS) (Tables-I, II, III; Figs.-1, 2, &3).

Table - I
Triglyceride Levels in liver and kidney tissues during short term and Long term starvation in *A.testudineus*

S.No.	Tissue analyzed	Short Term (15 Days)		Long Term (60 Days)	
		Control	Experimental	Control	Experimental
1.	Liver	5.333 SE = ± 1.173	15.83* SE = ± 1.376	3.833 SE = ± 0.872	5.5NS SE = ± 0.562
		% Variation = +196.831		% Variation = +43.490	
2.	Kidney	1.291 SE = ± 0.1	3.833*** SE = ± 0.247	1.333 SE = ± 0.255	2.125** SE = ± 0.085
		% Variation = +196.90		% Variation = +59.414	

Values expressed in mg of triglyceride / gm wt. of tissue
Each value is mean of SE ± of 6 individual observations
P < 0.01*, P < 0.05**, P < 0.001***, NS = Not Significant

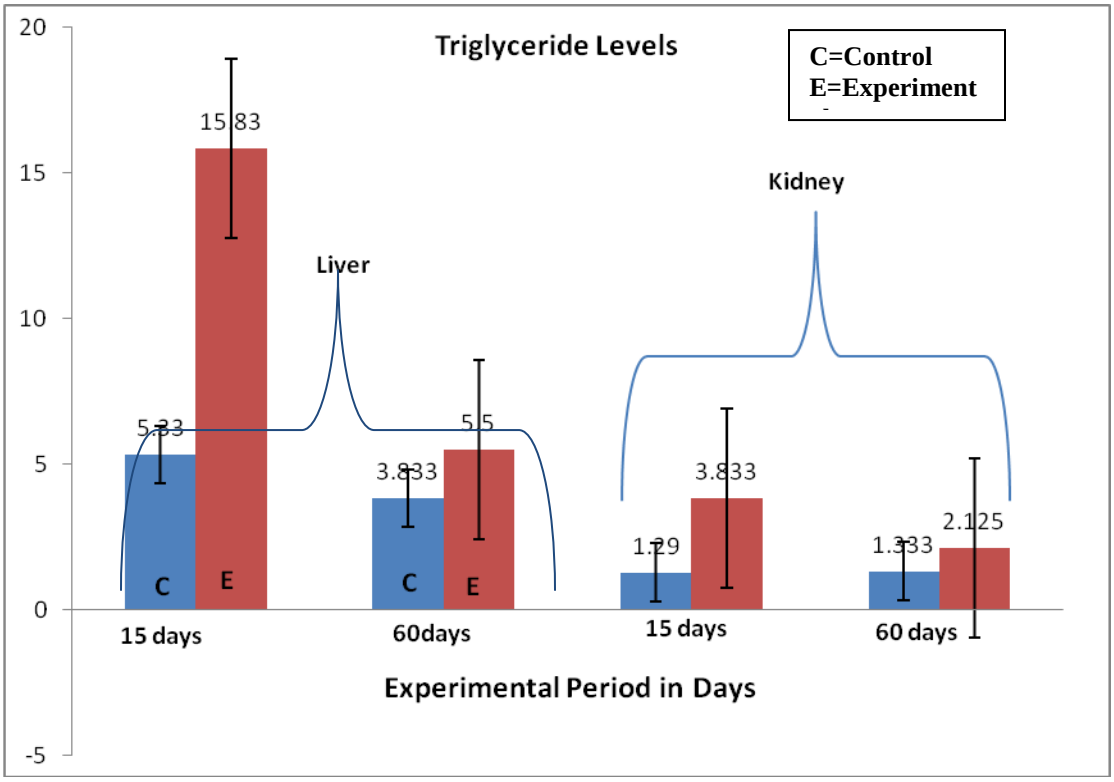

Fig-1
X -axis: mg of triglyceride/gm weight of tissue
Y-axis: Experimental (starvation) period in days

Table - II
Triglyceride levels in brain and accessory respiratory organ during short term and long term starvation in *A.testudineus*

S.No.	Tissue analyzed	Short Term (15 Days)		Long Term (60 Days)	
		Control	Experimental	Control	Experimental
1.	Brain	0.999 SE = ± 0.121	2.833*** SE = ± 0.074	1.444 SE = ± 0.69	1.944** SE = ± 0.180
		% Variation = +183.583		% Variation = +34.626	
2.	Accessory	2.110	4.444***	2.333	2.333

	Respiratory Organ	SE = ± 0.28	SE = ± 0.164	SE = ± 0.148	SE = ± 0.243
		% Variation = +110.616		% Variation = nil	

Values expressed in mg of triglyceride / gm wt. of tissue
Each value is mean of SE ± of 6 individual observations
P < 0.001***, P < 0.05**

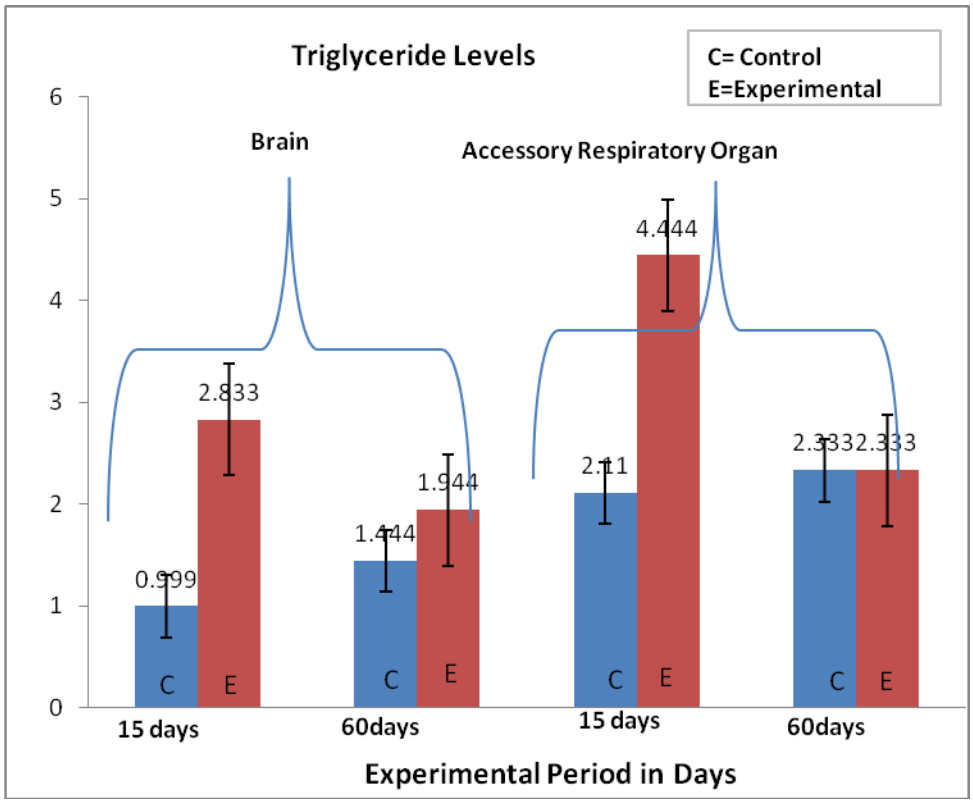

Fig-2
X -axis: mg of triglyceride/gm weight of tissue
Y-axis: Experimental (starvation) period in days

Table - III
Triglyceride Levels in Pectoral and Lateral Line Muscles during Short Term and Long Term starvation in *A.testudineus*

S.No.	Tissue Analyzed	Short Term (15 Days)		Long Term (60 Days)	
		Control	Experimental	Control	Experimental
1.	Pectoral Muscle	1.083 SE = ± 0.105	2.166NS SE = ± 0.052	1.375 SE = ± 0.201	1.58NS SE = ± 0.20
		% Variation = 100		% Variation = +14.909	
2	Lateral Line Muscle	0.875 SE = ± 0.106	2*** SE = ± 0.129	1.041 SE = ± 0.076	1.375NS SE = ± 0.211
		% Variation = +128.571		% Variation = +32.084	

Values expressed in mg of triglyceride / gm wt. of tissue
Each value is mean of SE ± of 6 individual observations
P < 0.001***, NS = Not Significant

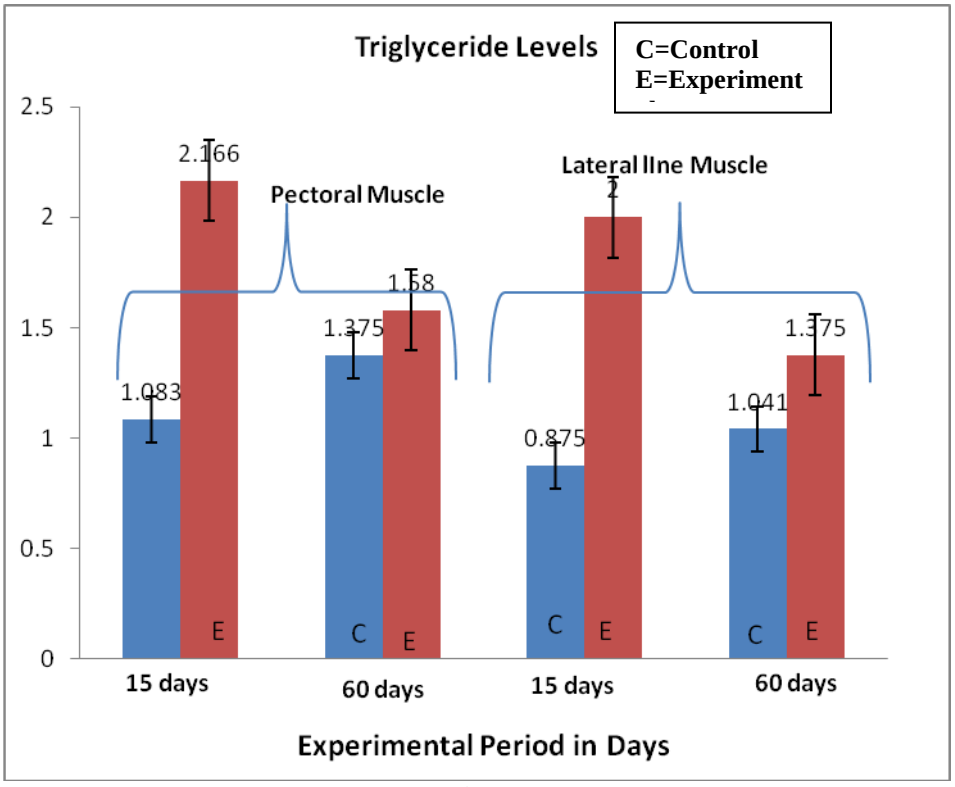

Fig-3
X -axis: mg of triglyceride/gm weight of tissue
Y-axis: Experimental (starvation) period in days

V. DISCUSSION

In the present study, when *anabas* was subjected to brief fasting of 15days and prolonged fasting of 60 days, there was an overall upsurge observed in the triglyceride content in all of the non adipose tissues analyzed. The animal adapted well for the stress and survived all throughout the starvation period. Short term fasting of a fortnight lead to a significant elevation of triglycerides in tissues like liver, kidney, brain, and accessory respiratory organ and lateral line muscle, however, pectoral muscle showed an insignificant increase. Long term fasting of 60 days in *Anabas*, showed a slightly different pattern. There was a significant rise observed in triglyceride levels of kidney and brain, while the other non adipose tissues such as liver, pectoral and lateral line muscle showed an insignificant rise in triglyceride content. Interestingly the accessory respiratory organ showed no change when compared to that of the fed group. Probably if the starvation period would have been prolonged further, may be a significant change would be expected. These results, of triglyceride upsurge coincides with the observations of Yar Mohmmadi *et.al* (2012), who have observed increased levels of total tissue lipids and triglycerides, in the plasma of the starving juvenile Persian sturgeon, *Acipencer persicus* and attributed that triglycerides may be the preferred fuel over the other nutrients for mobilization, during fasting. The upsurge in the triglycerides observed in the tissues of *anabas* may be interpreted as follows. Triglycerides may be partially utilized as a fuel during food deprivation and the remaining of unused FFA, may be recycled back by reesterification process, so as to maintain the triglyceride/FFA flux between the tissues. This explanation is based upon the studies and observations of Jensen and Chandramouli,(2001),who have suggested that during fasting as much as 65% of FFA are reesterifried in WAT (white adipose tissue) and the glycerol 3 phosphate needed for glycerol backbone, are obtained from the gluconeogenic precursors such as lactate or pyruvate,(Reshef, Hanson *et.al*,(1970), Reshef, L.,O. Meyuhas, *et .al*(1972) and this pathway is known as glyceroneogenesis(abbreviated form of gluconeogenesis) which is known to be essential for the regulation of triglyceride /FFA flux, between the tissues during fasting as suggested by Botion and Kettle Hut *et.al* (1995).Starvation studies by Terrrione (1920), suggested that, fat that remained in the body of the starved animal was an integral part of the cell and he named it as the "element constant" and also suggested that even when there is degradation of superficial fat, during extended periods of starvation the fat still remains in a state of "dynamic equilibrium". Therefore it may be assumed that there can never be an absolute degradation of fat in the body of a starving animal, and there may be a certain quantity of fat being maintained even during acute fasting, as it said to be the "element constant". Based upon these observations it may be said that the overall upsurge in the triglyceride levels of *anabas*, may be due to reesterification of FFA occurring along their utilization, so as to maintain the triglyceride/FFA flux and also to prevent lipotoxicity of cells caused to due to accumulation of FA as proposed by Listerberg (2003).This concentrated accumulation and synthesis of triglycerides during acute starvation may serve as an index to describe the starved state of the animal.

Starvation is an interesting physiological condition, characterized by several metabolic changes, such as low insulin production, reduced metabolic activity, protein/lipid utilization or tissues such as brain adapting to ketone utilization, etc. In our own observations with *Anabas*, which when was subjected to starvation stress showed changes such as reduced secretion of plasma insulin (P.Godavarthy and Y.Sunila Kumari (2011a), hyperglycemia (P.Godavarthy and Y.Sunila Kumari (2011b) and also increased circulating ketones.(P.Godavarthy and Y.Sunila Kumari(2012).All of these changes suggest that during starvation, because of reduced insulin levels, there is an impairment of glucose uptake, by the tissues and hence resulting in increased plasma glucose levels and due to reduced or non availability of glucose, starving *anabas* seems to adapt itself to mobilize the lipids (triglycerides), thereby causing their oxidation, resulting in FFA release and their oxidation leading to ketone formation, which may be utilized by the starving brain. In this context it is relevant to suggest that the released FFA may not be efficiently utilized, and so may have resulted in a mismatch between cellular lipid influx and lipid utilization, and thus causing FFA accumulation in tissues, which may lead to lipotoxicity (Listen Berger *et.al,* 2003) and so reesterification of FFA becomes essential, which may be accomplished *via* glyceroneogenesis with lactate and pyruvate acting as precursors for this process.

VI. Conclusion

In conclusion it may be said that when *anabas* was subjected to brief and prolonged fasting, a physiological state of stress characterized by insulin insufficiency, which hampers the capacity of glucose uptake by the tissues and thus leading to hyperglycemia. To compensate the reduced or non availability of glucose, the animal heavily relies upon lipid sources for survival. In this process the triglycerides are mobilized, utilized and thus releasing FFA. But as there seems to be an unequal balance between availability and utilization, which may lead to unwanted accumulation, and so the unused FFA are therefore reesterifried and recycled back, to form triglycerides so as to maintain the balance between the triglyceride /FFA flux and also to prevent the FFA induced lipotoxicity of cells, which may be caused due to their accumulation. The glycerol 3 phosphate needed for the glycerol back bone, for the triglyceride synthesis, may be provided *via* glyceroneogenesis, with lactate and pyruvate acting as precursors. In an insulinopenic state, such as starvation, characterized by hyperglycemia, and ketone upsurge, synthesis and accumulation of triglycerides may serve as an index to describe the starvation status of *anabas.*

References

[1] Botion, L. M., I. C. Kettelhut, Migliorini, R.H (1995). "Increased adipose tissue glyceroneogenesis in rats adapted to a high protein, carbohydrate-free diet." Horm Metab Res 27(7): 310-3.

[2] Cahill, F. George, Jr. (1986) "Physiology of Gluconeogenesis." In "Hormonal control of Gluconeogenesis" By Naomi Kraus - Friedman, Vol. I. CRC Press. Inc. BOCA Raton, Florida

[3] Dave Goran, Maj-Lis Johansson - Sjobeck, Ake Larsson, Kerstin Lewander and Ulf Lidman. (1975) Metabolic and Haematological effects of starvation in the European EeL, *Anguilla Anguilla* L. - I. Carbohydrate, Lipid Protein and inorganic ion metabolism. Comp. Biochem. Physiology. Vol.52A. pp.423 to 430.

[4] Figueiredo-garutti M.L,NavarroI,Capilla E,Souza,R,Moraes G,Gutierrez,J.,Vicentini-Paouline M.L; (2002) Metabolic changes in *Brycoon Cephalaus* (teleostei Characidae) during post feeding and fasting, Comp Biochem Physiol A 132: 467-476

[5] Godavarthy P and Y.Sunila Kumari, (2011) "An Investigation of Plasma Insulin Levels During Starvation Stress in *Anabas testudineus* (Bloch); Bulletin of Pure and Applied Sciences, Vol30A (zoology) Issue No:2; pp-79-84

[6] Godavarthy P and Y. Sunila Kumari, (2011) "Hyperglycemia- A Stress Response to Starvation in Climbing Perch, *Anabas testudineus* (Bloch),International Journal of Biological Sciences and Engineering, Vol-2;No;4pp-282-286

[7] Godavarthy P and Y. Sunila Kumari, (2012) "Starvation Induced in Plasma Ketone (Acetoacetate) levels in Climbing Perch, *Anabas testudineus"*(Bloch); International Journal of Fisheries and Aquaculture Sciences, Vol, 2 No: 1 pp-21-26

[8] Gillis TE, Ballentyne .J.S (1996). The Effects of starvation on plasma free Amino acids and glucose concentrations in Lake Sturgeon, *Acipenser fulvescens*. Journal of Fish Biology .49: 1306-1316

[9] Hinch, SG, Cooke SJ, Haeley MC, Farrel AP.; (2005) Behavioral physiology of fish migrations; salmom as model approach. Fish Physiol. 24: 239-295

[10] Hung SSo, Liu W, LiH, Storebakken. T,Cui Y (1997) Effect of Starvation on some morphological and biochemical parameters in white sturgeon, *Acipenser transmonatamus* Aquaculture 151; 357-363

[11] Ida Coordt Elle, Steven Vestergaard. Rodkaer, Julius Fredens and Nils Joakin Faegeman, (2012); A method for measuring fatty acid oxidation in *C.elegans. Worm: 1:1*, landes Bioscience, *pp-1-5*

[12] Jensen, M. D., V. Chandramouli,. William C. Schumann, Karin Ekberg, Stephen F. Previs, Sameer Gupta, and Bernard R. Landau (2001). "Sources of blood glycerol during fasting." Am J Physiol Endocrinol Metab ; 281(5): E998-1004.

[13] Keys Ancel Josef Brozek, Austin Henschel, Olaf Mickelsen Henry Longstreet Taylor. (1950) The Biology of Human Starvation Vol.I, Vol.II. The University of Minnesota Press. Minneapolis, London, Geoffrey Cumberlege. Oxford University Press, 289-300; 575, 576.

[14] Larsson Ake and Lewander Kerstin (1973) "Metabolic effects of starvation in the eel, *Anguilla Anguilla* L." Comp. Biochem. Physiol. Vol.44A pp.367-374.

[15] Laura L. Listenberger , Xianlin Han, Sarah E. Lewis , Sylvaine Cases , Robert V. Farese, Jr. Daniel S. Ory ,and Jean E. Schaffer ;(2003), Triglyceride accumulation protects against fatty acid-induced lipotoxicity; *PNAS*; Vol. 100, No. 6 -3077–3082

[16] Loughna, P.T. and Goldspink, G. (1984) "The effects of starvation upon protein turnover in red and white myotomal muscle of the rainbow trout *Salmo gairdneri* Richardson. Journal of Fish Biology 25, 223-230.

[17] Mayzaud, P., (1976). Respiration and nitrogen excretion of zooplankton. IV. The influence of starvation on the metabolism and the biochemical composition of some species. Marine Biology 37, 47–58.

[18] Meton.I, fernanadez.F,Bannante IV (2003) Short and long term effects of refeeding on key enzyme activities in glicolisis-gluconeogenesis in the liver of gilt head sea bream (*Sparus aurata*) Aquaculture 225; 99-107

[19] Miller KM, Schulze AD, Ginther N, Li S, Patterson DA, Farrell AP, Hinch SG (2009) Salmon spawning migrations: metabolic shifts and environmental triggers. Comp Biochem Physiol 4D;75-89

[20] Navvorro.I, Guitrezz.J, (1995) fasting and starvation. In: Hochachka PW, Mommsen TP (eds) Biochemistry and molecular Biology of fishes, Metabolic Chemistry, Vol-4 Elsevier, Amsterdam, pp-393-434

[21] O'Connor, K.I., Taylor, A.C., Metcalfe, N.B., (2000). The stability of standard metabolic rate during a period of food deprivation in juvenile Atlantic salmon. Journal of Fish Biology ; 57 (1), 41–51.

[22] Raghuramulu, N.K., Madhavan Nair, Kalyana Sundram, S. (1983) A Manual of Laboratory Techniques by National Institute of Nutrition. Indian Council of Medical Research, Jamai-Osmania, Hyderabad - 50001, A.P., India.

[23] " Reshef L, Hanson RW, Ballard FJ. (1970); A possible physiological role for glyceroneogenesis in rat adipose tissue. J Biol Chem ; 245:5979–5984.

[24] Reshef, L., Meyuhas, O., Boshwitz, C.H., Hanson, R.W. and Ballard, F.J. (1972). "Physiological role and regulation of glyceroneogenesis in rat adipose tissue." Israeli J Med Sci 8(3): 372-81.

[25] Robin Patrice Jean, MichelFrain, Claude Sardet, Rene Groscolas, and Yvn Le Maho.(1988) "Protein and lipid utilization during long term fasting in emperor penguins. American J. Physiology. 254. R61-R68.

[26] Salem M, Silverstein,J. Rexroad CE, Yao J (2007); Effect of starvation on global gene expression and proteolysis in rainbow trout,(*Oncorhynchus myksis*) BMC Genomics 8: 328-343

[27] Sheridan, M.A. and Mommsen, T.P. (1991) "Effects of nutritional status on in vivo lipid and carbohydrate metabolism of Coho Salmon, *Oncorhynchus Kisutch."* General and Comparative Endocrinology. 81, 473-483.

[28] Terroine, EF. (1920) "Contribution a la connaissance de la physiologie des substances grasses et lipoidiques." Ann. Nat. Zool. Paris, 10, Ser. 4, 1-397.

[29] Wang T, Hung CC, Randall D.J,(2006) The comparative physiology of Food deprivation from feast to famine; Annu Rev Physiol 68;223-51;PMID; 16460272

[30] YarMohammadi, M; A. Shabani, M. Pourkazemi, H. Soltanloo, M. R. Imanpour; (2012), Effect of starvation and re-feeding on growth performance and content of plasma lipids, glucose and insulin in cultured juvenile Persian sturgeon (*Acipenser persicus* Borodin, 1897) Journal of Applied Ichthyology : 10.1111/j.1439-0426.2012.01969.x

AUTHORS

First Author – Padmavathi Godavarthy M.Sc, Ph.D, M.S(Cellular and Molecular Biology), Department of Zoology, Osmania University College for Women, Koti, Hyderabad-500095, India, Email: anjanipadmavathi@gmail.com
Second Author – Y.SunilaKumari M.Sc, Ph.D, Department of Zoology, Osmania University College for Women, Koti, Hyderabad-500095, India, Email: sunilapamara@yahoo.com
Third Author – E.Bikshapathy, M.Sc, Ph.D, Centre for Cellular and Molecular Biology (CCMB) Uppal, Hyderabad, 500007, India, Email: ebp@ccmb.res.in,

Correspondence Author – Padmavathi Godavarthy M.Sc, Ph.D, M.S(Cellular and Molecular Biology), Department of Zoology, Osmania University College for Women, Koti, Hyderabad-500095, India, Email: anjanipadmavathi@gmail.com , ,padmagodavarthy@gmail.com, Contact mobile: 9140-9059681804