IJISET - International Journal of Innovative Science, Engineering & Technology, Vol. 2 Issue 10, October 2015.
www.ijiset.com
ISSN 2348 – 7968

Changes in Glycogen Content of Brain in Starving Catfish *Clarias batrachus* [Linn.]

Nayan K. Prasad
Reader, Department of Zoology, R.R.M. Campus, Tribhuvan University, Nepal.

Abstract

Glycogen is a multi-branched polysaccharide of glucose that serves as a form of energy storage in animals. The present paper deals with changes in the level of glycogen in *Clarias batrachus*, which was subjected to prolonged fasting of 40 days. Tissues of brain were selected for the study. Glycogen content was estimated by the calorimetric method of Kemp *et al.* (1954) as modified by Krishnaswamy & Srinivasan (1961). The starvation showed a non-significant depletion of glycogen content in brain tissues up to 20 days of starvation. However, the male showed more depletion of glycogen content than that of female. After 40 days of starvation, the glycogen depleted about 51% in male and 50% in female of their normal values. *Clarias* adapted well to starvation stress and survived all throughout the experimental period.
Key words: *Brain, Clarias batrachus, Glycogen, Starvation.*

Introduction

Starvation is experienced in most species of fish during certain periods of every year largely due to environmental conditions. It affects the normal metabolism of the body and prolonged starvation may even cause death of the animal. In fact, organism facing the starvation fights it at the cost of its own body-reserves till death. A decline in various body constituents of fish, following experimental starvation, have been reported by various authors. This paper deals with the results obtained for the fresh-water catfish, *Clarias batrachus*, following starvation up to 40 days.

The present study is aimed to know the facts and causes of starvation and their consequent impacts on animals. A number of similar studies have been carried out by many workers but most of the works have been confined to mammalian fauna. In Nepal, very little works have been done to study the starvation-induced effects in fishes. In fact, starvation affects the physiological status and biochemical constituents of fish (Rajyasree & Naidu 1989 and Tripathi & Verma 2003). So, it is pertinent to see and reveal the effect of starvation on fish as there is a unique feature of fish to withstand prolonged starvation through physiological and biochemical changes (Mustafa, 1983).

In the light of above fact, the present study has been designed to know the level of glycogen content of brain of *Clarias batrachus* during prolonged period of starvation of 40 days starting from 0 day to 40 days by estimating glycogen constituents at an interval of 10 days.

Materials & Methods

For the present investigation, healthy live fish were collected from a local fish pond with the help of fishermen. The fish were brought to the laboratory in large earthen pots covered with mosquito net. They were identified with the help of the book entitled “Fishes of UP and Bihar” (Srivastava, 2006) and were treated with 0.1% $KMnO_4$ solution for five minutes to get rid of any dermal infection. Healthy fish of an average length (18.8 cm) and weight (34.4 g) were transferred one by one with the help of small hand net to a large glass aquarium of about 110 litres capacity measuring about 75cm x 30cm x 45cm in size. They were allowed to acclimatize under laboratory condition for 20 days. During this period, the fish were fed twice daily with commercial fish food to avoid their starvation. Twenty four hours before starting the experiment, the food was stopped to clear off the alimentary canal. The study was carried out from May 2009 to June 2013.

ISSN 2348 – 7968

Biochemical estimations were made by taking the samples from each sex of acclimatized and well fed fish and the values obtained were taken as normal value for *C. batrachus.* A control group was kept in tap water. The remaining fish were divided into four batches – A, B, C and D keeping 10 fishes each (5 males and 5 females). The fishes of batch A were kept without food at room temperature for 10 days, that of batch B for 20 days, C for 30 days and D for 40 days.

Starting from 0 days up to 40 days at an interval of 10 days each, fish were dissected and their brains were removed immediately. Starting from 0 day up to 40 days, the fish were dissected at an interval of 10 days. Their brains were removed and placed immediately in the ice-cold fish saline for different biochemical studies. Before transferring the brains into saline, they were properly cleaned by removing other attached tissues. Before use, the tissues were nicely blotted with filter papers.

The total glycogen content of brain was estimated to the calorimetric method of Kemp *et al.* (1954) as modified by Krishnaswamy & Srinivasan (1961). A weighted quantity of the brain tissue was homogenized in 5 ml of ice-cold 10% TCA in tissue homogenizer. The homogenate was centrifuged for 20 minutes at 500 x g. The sediment was rehomogenized in 5 ml TCA and centrifuged again. The supernatants of both the fraction were pooled. 2 ml of the supernatant was mixed with 6 ml of concentrated sulphuric acid. The tube was kept in a boiling water bath for 6.5 minutes. After the development of the colour, the optical density of the pink colour was measured in photoelectric colorimeter at 515 mμ filter. The glycogen content (mg/g wet weight) in the unknown sample was determined from standard curve which was drawn by taking glucose and was linear.

Result

In present study, the level of glycogen content in the brain tissues of *Clarias* was found higher in female than that of male both under normal and starved conditions. Up to 20 days of starvation, the brain showed non-significant depletion of glycogen content in both the male and female, but thereafter it was sharp. After 40 days of starvation, the glycogen depletion in brain tissues was about 51% in male and 50% in female of their normal values. (Table-1; Fig.-1).

The analysis of variance and the bar notation of the recorded data showed that it decreased between each of the succeeding days intervals i.e. between 0 & 10 days, 0 & 20 days, 0 & 30 days, 0 & 40 days. In brain, the glycogen depletion between 30 days & 40 days of starvation was highly significant at 1% of P.

Table 1: Brain Glycogen in *C. batrachus* (mg/gm wet weight) during different period of starvation

Clarias	**Days of Starvation**				
	0	**10**	**20**	**30**	**40**
Male	16.17 ± 0.19	15.83 ± 0.22	15.41 ±0.18	12.66** ± 0.20	7.88** ±0.19
Female	19.25 ±0.28	18.87 ±0.20	18.63 ±0.45	16.23** ±0.34	9.90** ±0.15

Values are mean of 8 samples of both male & female ± SE
** Significant

Discussion

The fishes get maximum support from their environment which enables them to conduct their activities without recourse to their body constituents and so their basal energy consumption is very low. For this reason, fish are able to withstand astonishingly long periods of starvation such as, *Amia calva* survived for 20 months without food (Smallwood 1916).

The carbohydrate metabolism plays a primordial part during food deprivation as carbohydrates are the principal fuel for energy production. Glucose molecules are constantly oxidized to release energy. During starvation, glycogen is broken to produce glucose which in turn is supplied through blood to the needy organs. The increased level of glucagon during starvation stimulates the synthesis of glucose from glycogen (Cahill, 1970; Bell et al., 1976 and Chaudhary & Mandal, 1981). The glucose production occurs directly from the glycogen

ISSN 2348 – 7968

stores of liver and indirectly through other tissues like muscle, gonad and brain through the Coricycle and alanine glucose cycle (Cori, 1931 & Felig *et al*., 1969).

Effect of prolonged as well as short term starvation has been extensively studied, mostly on temperate fish. Fontaine and Hatey (1953) observed 54% drop in liver glycogen content of *Salmo salar* during spawning migration starvation. Inui & Oshima (1966) observed slower rate of glycogen depletion in muscle than that of liver in starving *Anguilla japonica*.

The decrease in the carbohydrate content in the starving organism was observed by several workers in different tissues of different animals such as Fontaine & Hatey (1953) in the liver of *Salmo salar*, Matsumoto & Tatsuo (1980), Prasad (1980), Chaudhary & Mandal (1981) in the brain of *Schizodactylus monstrosus*, Saini & Purohit (1981) in *Mus musculus*, Haranath *et al.* (1984) in *Tilapia mossambica*, and Prasad (2013, 2014) in the liver and gonads of *C. batrachus*.

In the present investigation, the glycogen stores of the brain did not show significant drop up to 20 days of starvation. Thereafter, it started to drop sharply. These findings are in conformity with the findings of Fontain & Hatey (1953), Rivera & Jesus (1974), Chaudhary & Mandal (1981), Ottolenghi *et al.* (1981) and Freminet & Lilliane (1981).

The value of glycogen and glucose was found higher in females than males under both normal and starved condition. This finding of sex difference is in conformity with Shreni (1979) and Singhal *et al.* (1981).

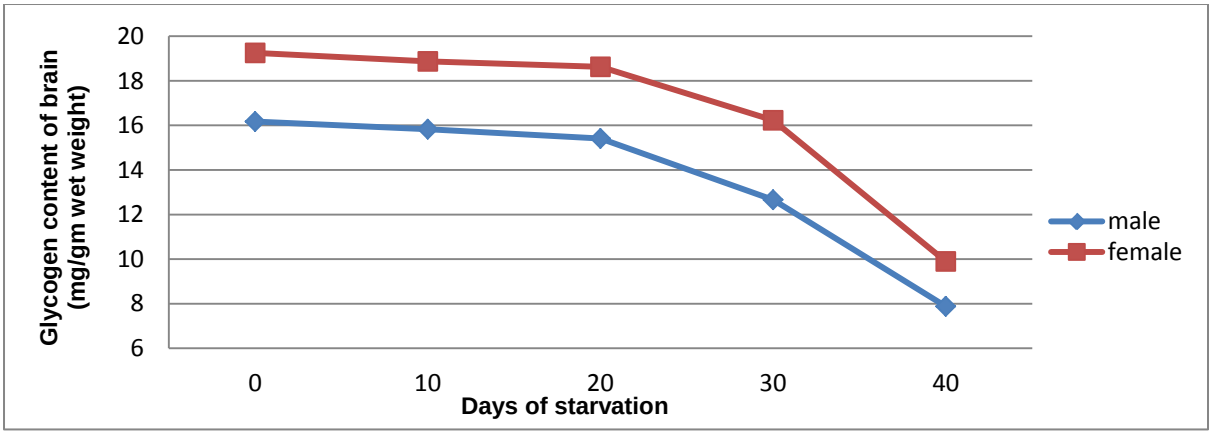

Fig. 1: Glycogen content in the brain of male and female *Clarias.*

Conclusion

The fresh water catfish, *Clarias batrachus* can sustain starvation for prolonged period. Starvation influences the biology of the body at various levels, especially the biochemical composition of various organs. In the present investigation, the collected data clearly indicates that the level of glycogen in brain remains higher in female than that of male *Clarias batrachus*, both in normal and starved conditions. This level decreases sharply after 20 days of starvation. In other words, starvation is the root reason for the decrease of glycogen in brain of both the sexes of *Clarias batrachus*. Though this decrease may have severe negative consequences for the biology of the body, further studies are required.

Acknowledgement

The author renders his heartfelt gratitude to Dr. R. P. Sinha, & Dr. A. K. Verma, Professors, Department of Zoology, C. M. Science College, LNMU, Darbhanga, India, for their generous and constant guidance as well as for providing lab facilities during the research work.

References:

ISSN 2348 – 7968

[1] A. Freminet, and L. C. Lilliane. “Effects of fasting on liver and muscle glycogen in rats and pigs”, J. Physio. (Pans), Vol. 76, no. 8, 1981, pp. 877-880.

[2] A. Kemp, and V. K. Heijningen, “A colorimetric micromethod for the determination of glycogen in tissues”, Biochem. J., Vol. 56, 1954, pp. 646-646.

[3] A. Rivera, and J. M. Jesus, “Starvation and glycogen of the brain and vital organs of the rhesus monkey”, J. Nutr., Vol. 104, no. 9, 1974, pp. 1189-1194.

[4] B. K. Prasad, “Effect of starvation on the carbohydrate content of tissue of some teleosts”, Ph. D. Thesis, Patna Univ., Patna, India, 1980.

[5] C. F. Cori, “Mammalian carbohydrate metabolism”, Physio. Rev., Vol. 11, 1931, pp. 143-175.

[6] C. Ottolenghi, A. C. Puviani, and L. Brighenti, “Glycogen in liver and other organs of *Ictalurus melas*: seasonal changes and fasting effects”, Comp. biochem. Physiol., Vol. 68, no. 3, 1981, pp. 313-322.

[7] D. K. Chaudhary, and S. Mandal, “Fluctuations of glycogen content in brain during post-embryonic development in *Schizodactylus monstrousus:* D. Effect of starvation stress and corpora allatectomy”, I. J. Exp. Biol., Vol. 19, 1981, pp. 532-534.

[8] D. K. Shreni, “Influence of starvation on the brain and liver cholesterol levels of catfish, *Heteropneustes fossilis* (Bloch)”, Proc. Animal Sci., Vol. 88, no. 3, 1979. Pp. 205–208.

[9] G. F. Jr. Cahill, Starvation in man, New. Eng. J. Mad., 1970.

[10] G. H. Bell, D. E. Smith, and C. R. Paterson, Text Book of Physiology and biochemistry, 9th edition, E.L.B.S. and C. Livingstone, England. 1976.

[11] G. J. Srivastava, Fishes of U.P. and Bihar. 11th ed. Vishwavidyalaya Prakashan, Varanasi, India. 2006.

[12] G. Tripathi, and P. Verma. “Starvation-Induced Impairment of Metabolism in a Freshwater Catfish”, Z. Naturforsch., Vol.58c, 2003, pp. 446-451.

[13] J. Saini, and K. G. Purohit. “Effect of starvation stress on tissue glycogen and blood glucose levels in the house mouse, *Mus musculus*”, Comp. Physiol. Ecal., Vol. 6, no. 2, 1981, pp. 115-116.

[14] M. Fontaine, and J. Hatey, “Contribution to the study of glucose metabolism in the Salmon (*Salmo salar*) at different stages of its development and of its migration”, Physiologia comp. oceal. Vol. 3, 1953, pp. 37-52.

[15] M. Matsumoto, and H. Tatsuo, “Effects of starvation on glycogen contents of heart, skeletal muscle and liver in several mammals”, Jpn. J. Zootech. Sci., Vol. 51, no. 2, 1980, pp. 860-865.

[16] M. Rajyasree, and K. R. P. Naidu, “Starvation induced changes in biochemical aspects of hepatic tissue of fish, *Labeo rohita*”, Indian J. Fish., Vol. 36, 1989, pp. 339-341.

[17] N. Prasad, “Starvation-induced effects on glycogen content of liver in *Clarias batrachus*”, Gun Sagar, Vol. 3, no. 3, 2013, pp. 87-90.

[18] N. Prasad, “Effects of prolonged starvation on glycogen content of gonads in *Clarias batrachus*”, Ideal Science Review, Vol. 6, no. 1, 2014, pp. 34-37.

[19] P. C. Singhal, R. K. Gupta, and L. D. Joshi, “Effect of sex on blood sugar levels on experimental animals”, I. J. Exp. Biol., Vol. 19, 1981, pp. 1201-1202.

ISSN 2348 – 7968

[20] P. Felig, O. E. Owen, J. Wahren, and G.F. Jr. Cahill, "Amino acid metabolism during prolonged starvation", J. CIin. Invest., Vol. 48, 1969, pp. 584.

[21] S. Krishnaswamy, and V.V. Srinivasan, "A simple method of determination of glycogen content of marine animals", Curr. Sci. Vol. 30, 1961, pp. 353-353.

[22] S. Mustafa, "Changes in Biochemical composition in Starving Catfish, *Heteropneustes fossilis*", Japanese Journal of Ichthyology, Vol. 29, 1983, pp. 4.

[23] V. B. Haranath, M. Bhaskar, M. V. C. Rao, P. S. Rani, P. Reddanna, and G. Govindappa, "Effect of long-term starvation on branchial tissue glycolysis of freshwater fish, *Tilapia mossambica* (Peters)", Indian J. Fish., Vol. 30, no. 2, 1983, pp. 340-343.

[24] W. M. Smallwood, "Twenty month of starvation in *Amia calva*", Biol. Bull. Mar. Biol. Lab. Woods hols., Vol. 31, 1916, pp. 453-464.

[25] Y. Inui, and Y. Oshima, "Effect of starvation on metabolism and chemical composition of sols", Bull. Jap. Soc. Scient. Fish., Vol. 32, 1966, pp. 492 -501.