International Journal of Advance Research in Science and Engineering
Volume No.06, Issue No. 12, December 2017
www.ijarse.com
IJARSE
ISSN: 2319-8354

EFFECT OF ALTERED P^H ON THE LIPID METABOLISM OF PRAWN LITOPENAEUS VANNAMEI

Ramanamma.T.[1] , Suneetha.T.[2], Sailaja.V[3]

[1,2,3]*Department of Zoology,Vikrama Simhapuri University,(India)*

ABSTRACT

Litopenaeus vannamei of body weight 6.5. ± 0.5 gms were selected to study the lipid peroxidation, phospholipids, total Cholesterol, triglycerides and xanthine oxidase on exposure to altered P^H media i.e., 6.5 at acidic and 9.0 at alkaline P^H he lipid peroxidation was more in hepatopancreas than muscle both in acidic (6.5) and alkaline P^H. However the phospholipid, Litopenaeus vannamei of body weight 6.5. ± 0.5 gms were selected to study the lipid peroxidation, phospholipids, total Cholesterol, triglycerides and xanthine oxidase on exposure to altered P^H media i.e., 6.5 at acidic and total cholesterol, triglycerides and xanthine oxidase contents are more in muscle when compared with hepatopancreas at altered P^H medium. The disorders in lipid organization of biological membranes results in alterations in the activity of a number of membrane bound enzymes in the presence of altered P^H.

Keywords:Altered P^H,Litopenaeus vannamei,Phosphelipids,Lipid peroxidation,total cholesterol.

I.INTRODUCTION

In aquatic habitats usually variations occurs in abiotic factors such as temperature, salinity, photoperiod, P^H, turbidity and gaseous contents daily and seasonally. Each of these factors, single or together it altered can impose a considerable load of stress on the physiology of aquatic animals.

Lipids play an important role as source of energy for prawn. P^H of the living environment has considerable effect on the prawn, *Litopenaeus vannamei*. The altered P^H may affect the oxidative reactions cellular membrane functions, activity of membrane enzymes and the transportation of specific molecules. On the other hand, the formation of malondialdehyde (MAD) during peroxidation process of fatty acids having double bonds can create covalent bonds and polymerize cellular membrane components (Sureda et al., 2006; Tejada et al; 2007).

In several crustacean species the hemolymph lipid exists as a complex lipoprotein moiety. Electrophoretic studies have demonstrated the existence of lipoproteins in UCA pugilator (Fielder et al ; 1971) callinectes sapidus (Horn and kerr, 1969; kerr, 1969) carcinus maenas (Ceccaldi and Martin, 1969), *Paratelphusa*

hydrodromous (Adiyodi, 1968), *Homarus americanus* (Barlow and Ridgway, 1969), Cancer magister (Allen, 1972) *Panulirus interruptus* (Lee and puppione, 1978).

Lipids of prawn *Litopenaeus vannamei* play a vital role in the moulting hormone activity, especially ecdysteroids. Lipids of prawn also play a vital role in the reproduction such as oocyte growth (Blanchet et al.,1979), embryonic development and hence associate with developmental metabolic processes.

Although several studies are there on the study of carbohydrate and protein metabolism of prawn in the presence of altered P^H (Bhaskar 1982, Sobha Rani, 1984, Sailaja 2001) very less information is there on lipid metabolism. Hence the present study has been undertaken.

II.MATERIAL AND METHODS

The prawn *Litopenaeus vannamei* were obtained from Otturu hatcheries, Otturu nearer to Kavali at SPSR Nellore Dist., and they were maintained under laboratory conditions in normal brackish water at room temperature (27.5^0C) Salinity (25 ppm) P^H (7.4 ± 0.1) and exposed to 12 hrs photo period. The prawns were fed daily with a standard commercial diet. After acclimatization to laboratory condition the prawns were exposed to short term (24 hrs) acidic and alkaline sublethal P^H conditions (6.5 & 9.0). The maintenance of P^H was done according to the method given by Bhaskar and Govindappa (1982).

Basically all experimental animals were divided into three groups First group of animals was considered as control and maintained them in normal brackish water with P^H 7.4 ± 0.1. Second group (10 prawns in 5 liters of water) was maintained in 6.5 P^H medium. Third group (10 prawns in 5 litres of water) was maintained in 9.0 P^H medium.

In view of differential metabolic responses of various tissues under different stress conditions two important tissues of prawn such as muscle and hepatopancreas were selected for the present study. Since the hepatopancreas is useful in detoxification of water pollutant (Poels and stick, 1975 ; Stick et al 1977 ; Jacobson, 1977) and acts as a key organ in metabolic regulation. like liver of chordates. Similarly muscles are involved in the locomotory activities, intermediary metabolic adjustments and provide skeletal strength. Throughout the investigation these two tissues were taken always from the same region of prawn to maintain the experimental uniformity. After 24 hrs of exposure the prawns were taken out of water, then the hepatopancreas and muscle tissues were isolated. The tissues were chilled immediately by keeping them in the ice chambers and utilized them for the lipid metabolic studies like phospholipids (Zilversmidth and Davis,1950),total cholesterol (Liebermann Burchard as described by Natclson 1971), triglycerides (Natclson 1971),lipid peroxidation(Ohkawa et al.,1979),xanthine oxidase (srikanthan and Krishnamurthy 1955).

III.RESULT

Hepatopancreas is having high lipids when compared with muscle. The lipids of the muscle mainly composed of phospholipid, triglyceride and cholesterol. Head lipids have high triglyceride levels, lower levels of phospholipids, higher levels of triglyceride and cholesterol. All these lipids were very important in the process of reproduction and moulting.

In the presence of altered P^H disorders occurs in the lipid organization of biological membranes which result in alterations in the activity of a number of membrane bound enzymes (ohkawa et al., 1979). Lipid peroxidation products are constantly involved in some of the pathophysiological effects associated with oxidative stress in cell and tissues. Lipid peroxidation of cell membranes causes a loss of the fluid properties of the membrane as well as increase in membrance permeability. (Packer 1984, Pradhan et al., 1990) As a result of lipid peroxidation free radicals steal electrons from the lipids in cell membranes resulting in cell damage.

In acidic and alkaline P^H the lipid peroxidation content was more in hepatopancreas when compared with muscle.

The oxidative degradation of lipids was more in hepatopancreas. The reason may be as the hepatopancreas is the site for all metabolic reactions the hepatic cells may easily undergo the process of oxidation in acidic condition. The same is repeated in alkaline condition also.This might be due to altering membrane fluidity protein structure and cell signaling process and finally enzymatic inactivation of membranes(Dean et al ; 1991).

The percent change of depletion in the phospholipids content can be represented as

PL acidic condition : Muscle < Hepatopancreas

PL alkaline condition : Muscle < Hepatopancreas

IV.DISCUSSION

Membrane phospholipids have a dual role as structural building blocks of cell membranes and as precursor molecules involved in signal transduction such as the lipid second messengers diacylglycerol, phosphatidicacid, lysophosphatidic acid and arachidonic acid (Hodgkin *et al.*,1998). The decreased PL content may serve as a metabolic alarm to the animal in the sense that the membrane integrity is lost. The phospholipid content was decreased in the present study in all the tissues of prawn which might be implicated to the enzymatic hydrolysis of membrane phospholipids by phospholipases leading to loss of membrane integrity. From the observation of present study coupled with the above reports, it can be speculated that both the inhibition of phospholipid synthesis and activation of phospholipases have been involved in the reduced levels of phospholipids in tissues of *P. vannamei* after exposure of different concentrations of P^H.

From the results it was clear that pH might be implicated to the either reduced synthesis / augmented degradation by lipoprotein lipase activity. The results are also in congruence with the previous reports where

substantial loses of lipids including cholesterol and occurrence of peroxidative damage during arsenic induced oxidative damage (Haider and Najar, 2008) and during exposure to environmental pollutants and heavy metals (Haider and Hasan, 1984; Haider *et al.*,1981; Pandey *et al.*, 1989).

The decreased levels of triglycerides in different organs of prawn might be due to enhanced lipolysis through lipase activity. Hence, decreased triglyceride content observed in the present study might be implicated to the activation of phospholipases and lipases due to excitotoxin-induced calcium flux resulting in accumulation of free fatty acids, diacylglycerols, eicosonoids and lipid peroxides.

Naveed and Janaiah (2011) reported that the reduction in XOD activity in hepatopancreas of prawn, *Channa punctatus* exposed to triazophos leads to increase in cellular damage and may be due to non-availability of Iron to the prawn during toxic period. Free radicals are formed during the normal metabolic processes (Yoshikawa *et al*., 1990), in addition to being generated by exposure to toxic agents (Halliwell and Gutteridge 1985) and several other disease states (Kellog and Fridovich 1977; Lambert and Bondy 1989). It has been well established that uric acid is the most abundant antioxidant and a powerful free radical scavenger (Waring, 2002) and particularly effective in quenching hydroxyl, superoxide and peroxynitrite radicals and may serve a protective physiological role by preventing lipid peroxidation (Squadrito *et al.*,2000) and increased uricacid concentrations during oxidative stress might be considered as a compensatory mechanism that confers protection against increased free radical injury (Nieto e*t al.*,2013) that occurs in acidic and alkaline conditions in the present study.

V.CONCLUSION

Finally it can be concluded that altered P^H effect the levels of different types of lipids. This is because lipids plays a vital role in the structure of membranes. In the altered P^H lipids can undergo the process of stress which leads to the changes in the functional properties of membranes.

TABLE-1.1

Alterations in the Lipid metabolism in muscle of *L.vannamei* in acidic and alkaline conditions.

Values are expressed as –lipid peroxidation (LP) : µ moles of malondialdehyde formed / gram wet wt of the tissue. Phospholipids (PL): mg of phospholipids/g wet wt of the tissue. Total cholesterol (TC): mg of total cholesterol / g wet wt of the tissue. Triglycerides (TG): mg of triglycerides / g wet wt of the tissue. Xanthineoxidase(XOD): µ moles of formazan formed / mg of protein / hour.

MUSCLE	CONTROL	ACIDIC	ALKALINE
LP	33.013	40.181*	46.486*
	±0.567	±0.536	±0.341
		(21.26)	(39.2)
PL	41.824	34.809*	27.324*
	±0.501	±0.768	±0.413
		(-19)	(-35.9)
TC	25.693	20.216*	14.923*

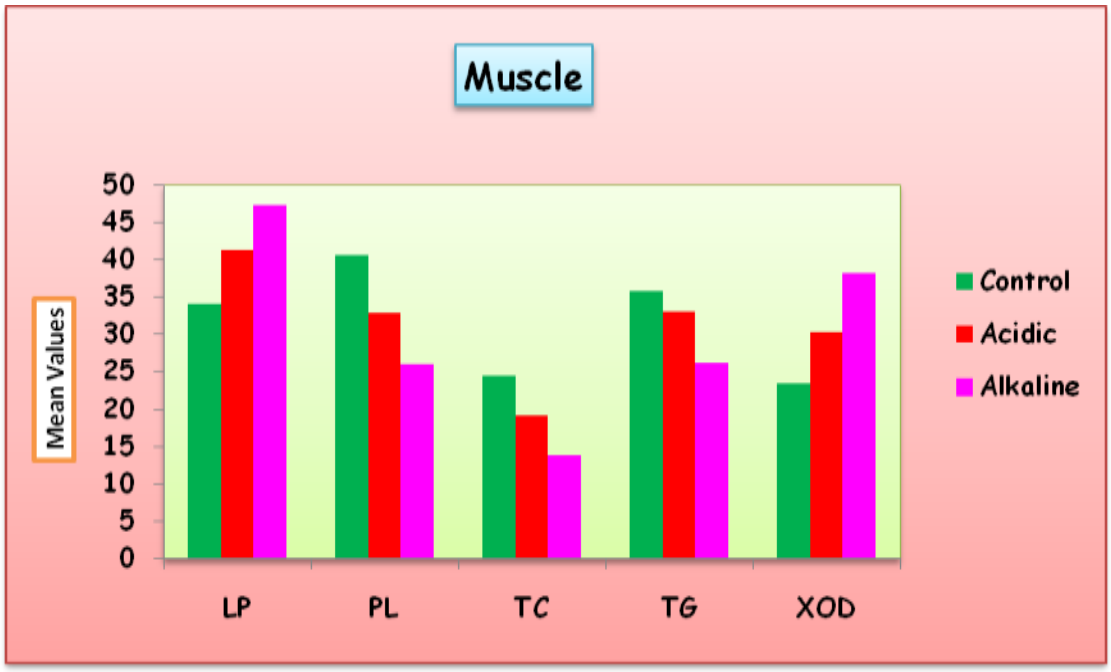
	±0.326	±0.226	±0.055
		(-21.44)	(-43.34)
TG	35.962	31.269	27.139*
	±0.306	±0.852	±0.824
		(-7.73)	(-26.74)
XOD	24.519	31.174*	38.555*
	±0.214	±0.388	±0.497
		(28.96)	(63.07)

All the values are mean, ±SE of eight individual observations.

Values in '( )'parentheses are % change over control.

*Values are significant at P < 0.05 in Scheffe test.

Graph.1.2. Alterations in the Lipid metabolism in Muscle of *L.vannamei* in acidic and alkaline conditions.



Values are expressed as

LP : µ moles of malondialdehyde formed / gram wet wt of the tissue.

PL: mg of phospholipids/g wet wt of the tissue.

TC: mg of total cholesterol / g wet wt of the tissue.

TG: mg of triglycerides / g wet wt of the tissue.

XOD: µ moles of formazan formed / mg of protein / hour.

International Journal of Advance Research in Science and Engineering
Volume No.06, Issue No. 12, December 2017
www.ijarse.com
IJARSE
ISSN: 2319-8354

1.3 ANOVA TABLE. MUSCLE OF VANNAMEI:

Parameters	Between groups			Within groups			(a)+(b)	(x)+(y)	F
	df(a)	Sum of Squares (x)	Mean SS	df(b)	Sum of Squares (y)	Mean SS			
LP	2	561.375	280.687	15	4.043	0.27	17	565.418	141.454
PL	2	631.030	315.515	15	21.208	1.414	17	652.237	223.158
TC	2	348.023	174.012	15	3.626	0.242	17	351.649	719.774
TG	2	263.132	131.566	15	45.527	3.035	17	308.66	143.348
XOD	2	591.543	295.771	15	13.26	0.884	17	604.802	334.595

One way ANOVA results in Lipid metabolism of Muscle of *L.vannamei*. The analysis is between different concentrations of pH and control.

TABLE 2.1

Alterations in the Lipid metabolism in Hepatopancreas of *L.vannamei* in acidic and alkaline conditions.

Values are expressed as –lipid peroxidation (LP) : µ moles of malondialdehyde formed / gram wet wt of the tissue. Phospholipids (PL): mg of phospholipids/g wet wt of the tissue. Total cholesterol (TC): mg of total cholesterol / g wet wt of the tissue. Triglycerides (TG): mg of triglycerides / g wet wt of the tissue. Xanthineoxidase(XOD): µ moles of formazan formed / mg of protein / hour.

HEPATOPANCREAS	CONTROL	ACIDIC	ALKALINE
LP	29.128	37.772*	43.224*
	±0.054	±0.582	±0.201
		(28.02)	(47.02)
PL	34.415	26.859*	21.785*
	±0.264	±0.374	±0.349
		(-20.89)	(-36.19)
TC	22.341	15.948*	10.447*
	±0.237	±0.212	±0.074

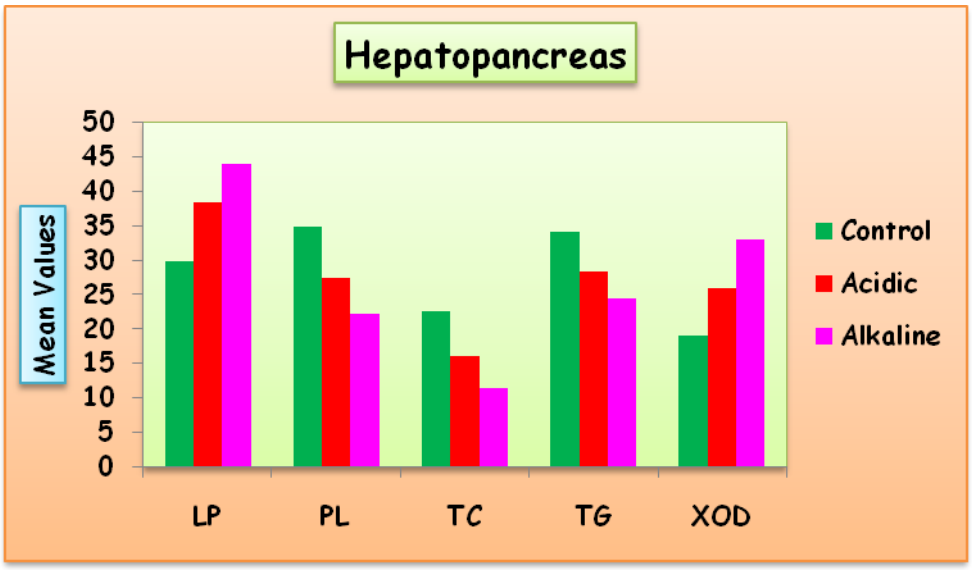
		(-28.6)	(-49.4)
TG	35.547	27.232*	23.489*
	±0.366	±0.426	±0.45
		(-16.86)	(-28.2)
XOD	18.226	25.025*	31.963*
	±0.166	±0.238	±0.122
		(35.66)	(72.38)

All the values are mean, ±SE of six individual observations.

Values in '( )'parentheses are % change over control.

*Values are significant at $P < 0.05$ in Scheffe test.

GRAPH 2.2. Alterations in the Lipid metabolism in Hepatopancreas of *L.vannamei* in acidic and alkaline conditions.



Values are expressed as

LP : µ moles of malondialdehyde formed / gram wet wt of the tissue.

PL: mg of phospholipids/g wet wt of the tissue.

TC: mg of total cholesterol / g wet wt of the tissue.

TG: mg of triglycerides / g wet wt of the tissue.

XOD: µ moles of formazan formed / mg of protein / hour.

2.3 ANOVA TABLE. Hepatopancreas of *L.vannamei.*

Parameters	Between groups			Within groups			(a)+(b)	(x)+(y)	F
	df(a)	Sum of Squares (x)	Mean SS	df(b)	Sum of Squares (y)	Mean SS			
LP	2	606.679	303.34	15	1.529	0.102	17	608.208	975.635
PL	2	484.711	242.356	15	9.179	0.612	17	493.89	396.047
TC	2	360.076	180.038	15	3.205	0.214	17	363.281	842.622
TG	2	284.418	142.209	15	15.564	1.038	17	299.982	137.054
XOD	2	566.107	283.054	15	2.603	0.174	17	568.71	631.184

One way ANOVA results in Lipid metabolism of **Hepatopancreas** of *L.vannamei.* The analysis is between different concentrations of pH and control.

REFRENCES

[1]Sureda, A, Box, A, Ensenat, M, Alou, E, Tauler, P, Deudero, S, & Pons, A.. Enzymatic antioxidant response of a labrid fish (*Coris julis*) liver to environmental caulerpenyne. *Comparative Biochemistry and Physiology, Part C, , 144*, 2006, 191-196.

[2]Tejada, S, Sureda, A, Roca, C, Gamundí, A, & Esteban, S. Antioxidant response and oxidative damage in brain cortex after high dose of pilocarpine. *Brain Research Bulletin*, doi:10.1016/j.brain resbull .2006. 10.005., 71, 2007,372-375.

[3]Fielder, D.R., Rao, K.K., Fingerman, M., A female-limited lipoprotein and the diversity of hemocyanin components in the dimorphic variants of the fiddler crab, Uca pugilator, as revealed by disc electrophoresis. *Comp. Biochem. Physiol. 39B*, 1971, 291-297.

[4]Horn, E.C., Kerr, M.S., The haemolymph proteins of the blue crabs, Callinectes sapidus. I. Hemocyanins and certain pther major protein constituents. Comp. Biochem. Physiol. 29, 1969,493-508.

[5]Kerr, M.S., The haemolymph proteins of the blue crab, Callinectes sapidus. II. A lipoprotein serologically identical to oocyte lipovitellin. *Develop. Biol. 20*, 1969,1-17.

[6]Cecaldi, H.J., Martin, J.L.M., Evolution des proteins de l'haemolymphe chez Carcinus maenas (L.) Durant P ovogenese. C.R. Seanc. *Soc. Biol. 163*, 1969, 2638-2641.

[7]Adiyodi, R.G., Protein metabolism in relation to reproduction and moulting in the crab,Paratelphusa hydrodromous (Herbst): Part II. Fate of conjugated proteins during vitellogenesis. *Indian J. Exp. Biol. 6*, 1968, 200-203.

[8]Barlow, J., and J. Ridgway.. Changes in serum protein during the molt and reproductive cycles of the American Lobster (Homarus amencanus). J. Fish.Res. Board Can. 26, 1969, 2101-2109.

[9]Allen, W.V.,. Lipid transport in the dungeness crab, Cancer magister Dana. *Comp. Biochem. Physiol.43B*, 1972, 193–207.

[10]Lee, R.F., Puppione, D.L.,. Serum lipoproteins in the spiny lobster, Panulirus interruptus. *Comp. Biochem. Physiol. 59B*, 1978, 239–243.

[11]Blanchet, M.-F., P. Porcheron, and F. Dray.Variations du taux des ecdysteroides au cours d descycles de mue et de vitellogenese chez le Crustace Amphipode, *Orchestia gammarella. Int. J. Invert. Reprod. 1*, 1979, 133-139.

[12]Bhaskar, M, Krishna Murthy, V., Reddanna, P. and Govindappa S., Effect of environmental acidity and alkalinity on white muscle protein fractions of fresh water fish, *Tilapia mossambica* (Peters); *J. Environ, Biol., 4(3)*, 1983,155 -160.

[13]Sobha Rani, P., Madhuri, E., Sailaja, V., Bhaskar, M. and Govindappa, S., Changes in the excretory products of fish red muscle on acclimation to low pH; *J. Aqua. Biol., 14 (1 & 2)*, 1999, 87 -89.

[14]Sailaja, V., Murthy, V.K., Madhuri, E., Bhaskar, M. and Govindappa, S., Studies on the red muscle lipid. metabolism of the fish, *Sarotherodon mossambicus* (Trewavs) on exposure to sub -lethal acidic media; *J. Aqua. Biol., 14 (1&2)*, 2001,83 -85.

[15]Bhaskar, M. and Govindappa, S., Tissue compensatory metabolic profiles in *Tilapia mossambiea* (Peters) on acclimation to sublethal acidic and alkaline media. Gill glycogen metabolism; *Archi. Internat. de physiol., Bioch., 93*, 1985, 59-63.

[16]Poels, C.L.M. and Strik, J.J.T.W.A., In sublethal effects of toxicchemicals on aquatic animals; J.H.Koeman & JJTWA. Strick ed. *EIsevier scientific publishing company, Amsterdam. 81*. 1975.

[17]Strik, J.J., Dejongh, T.W.A., Vanrijnvan, H.H., Alkemade, J.W.A. and Wmite T.P. Sublethal effects of toxic chemicals on aquatic animals J.H., Keoman & Strik (ed) J J T.W.A; *Elsevier Scientific Publising Company, Amsterdam, 31*,1977

[18]Jacobson-Kram, R.R. Tice, and A.V. Carrano, Sister- chromatid exchange: Second report of the Gene-Tox program. *Mutat. Res., 297*, 1997,101–180.

[19]Ohkawa, H., Ohishi, N. and Yagi, K. Assay for lipid peroxide in animals and tissues by thiobarbituric acid reaction. *Anal Biochem. 95*, 1979, 351-358.

[20]Packer, L.Vitamin E, Physical exercises and tissue damage in animals. *Med. Biol. 62*, 1984, 105.

[21]Pradhan, D., Weiser, M., Lunley - Sapanski, K., Frazier, D., Kemper, S., Williamson, P. and Schlegel, R.A. Peroxidation induced perturbation of erythrocyte lipid organization Biochim Biophys. *Acta, Biomembranes. 1023*, 1990,398-404.

[22]Dean, R.T., Hunt, J.V., Grant, A.J., Yamamoto, Y. and Niki, E. Free radical damage proteins: the influence of the relative localization of radical generation, antioxidants, and target proteins. *Free Radical Biology and Medicine 11*, 1991, 161-168.

[23]Hodgkin, M.N., Pettitt, T.R., Martin, A., Michell, R.H., Pemberton, A.J. and Wakelam. M.J. Diacylglycerols and phosphatidates: which molecular species are intracellular messengers: *Trends Biochem. Sci. 23*, 1998, 200-204.

[24]Haider, SS., Najar, M .S.A. Arsenic induces oxidative stress, sphingolipidosis, depletes proteins and some antioxidants in various regions of rat brain. *Kathmandu University Medical Journal, Vol. 6, No. 1, Issue 21*, 2008, 60-69.

[25]Haider SS, Hasan M. Neurochemical changes by inhalation of environmental pollutants sulfur dioxide an hydrogensulfide: Degradation of total lipids, elevation of lipid peroxidation and enzyme activity in the discreate regions of the guinea pig brain and spinal cord. *Industrial health 22*, 1984,23-31.

[26]Haider SS, Hasan M, Hassan SN, Khan SR and Ali SF. Regional effects of sulfur dioxide exposure on the guinea pig brain lipids, lipid peroxidation and lipase activity. *Neurotoxicology; 2*, 1981,443-50.

[27]Pandey, N.N. Report on 'Immediate and Residual Effects of MIC gas Exposure on Animals of Bhopal Gas Tragedy'. *Indian Veterinary Research Institute, lzzatnagar, India. 1989*

[28]Naveed, A and C. Janaiah . Effect of Triazophos on Protein Metabolism in the Fish, Channa punctatus (Bloch). *Current Research Journal of Biological Sciences. 3(2)*, 2011,124-128,

[29]Yoshikawa, T., Toyokuni, S., Yamamoto, Y., Naito Y(eds). Free radicals in Chemistry Biology and Medicine, *OICA International, London. 1990*

[30]Halliwell, B. and J.M.C. Gutteridge. Hypoxia induced metabolic and antioxidant enzymatic activities in the estuarine fish Leiostomus xanthurus. *J. Exp. Mar. Biol. Ecol., 279*, 1985, 1-20.

[31]Kellog, E.W and Fridovich, I. Liposome oxidation and erythrocyte lysis by enzymatically generated superoxide and hydrogen peroxide. *J. Biol chem, 252*, 1977,6721-6728.

[32]Lambert, C.E and Bondy, S.C. (). Effects of MPTP, MPP and paraquat on mitochondrial membrane potential and oxidative stress. *Life Sci 44*,1989,1277-1284.

[33]Waring, W.S. Uric acid: an important antioxidant in acute ischaemic stroke. *QJM. 95(10):691*, 2002,3.

[34]Squadrito,G.L., Cueto, R., Spleneser, A.E., Valavanidis, A., Zhang, H. and Pryor, W.A. Reaction of uric acid with peroxynitrite and implications for the mechanism of neuroprotection by uric acid. *Arch Biochem Biophys. 376:* 2000,333-7.

[35]Nieto, F.J., Lribarren, C., Gross, M.D., Comstock, G.W., Culter, R.G. Uric acid and serum antioxidant capacity: a reaction to atherosclerosis; Atherosclerosis. 148: 2000,131-9.