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Biochemical changes in the selected tissues of *Channa punctatus* exposed to Arsenic and its possible revival with turmeric.

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Abstract

Arsenic (As) is a pollutant widely distributed in nature and released into the environment through both geogenic process and anthropogenic disturbances. High concentration of arsenic has been found in many areas of Assam. Fishes are ideal organism for studying the toxicological effects and since they respond to toxicants in a similar way as higher vertebrates including human, hence *Channa punctatus* was taken as a laboratory animal model. In fishes, arsenic (As) is absorbed via the gills and through contaminated food which is capable of causing disturbance to the antioxidant system. Fish liver is the target organ of arsenic toxicity as it can uptake accumulate and excrete the arsenic. Enzymes are biochemical macromolecule which control metabolic processes of organisms. Since a slight variation in enzyme activities would affect the organism, hence by estimating the enzyme activities in an organism, metabolic disturbances in its body can be easily found. Significant alterations in the total protein and in the marker enzymes AST and ALT were observed in arsenic treated fishes. But when the arsenic treated fishes were exposed to phytochemical products of turmeric extract, a well marked revival from arsenicosis has been observed.

Keywords: Arsenic, *Channa punctatus*, Phytochemical, arsenicosis, AST, ALT.

1. Introduction

As the human race witnesses spectacular changes through rapid industrialization and modernization of human lives and easy and comfortable livings, it has resulted in more and more application of chemicals in every field of human activities for their welfare. These results in the application of various chemicals for controlling different agricultural and household pests/insects. Under recommended conditions of use and application rates, it is unlikely that insecticides/pesticides and their degraded products will attain significant levels in the environment. But the indiscriminate use has resulted in the presence of harmful chemicals in the living systems (Shivakumar, 2005).

The natural physiological functioning of an organism gets disturbed on exposure to toxicant stress. It induces its effect first at cellular or even at molecular level, but ultimately causes physiological, pathological and biochemical alterations. It is, therefore necessary to focus attention on changes in biochemical composition of organisms, which are constantly under pollutant threat. Since aquatic environment is the ultimate sink for all pollutants, aquatic toxicity testing has became an integral part of the process of environmental hazard evaluation of the toxic chemicals. Generally, the potential impact of pollutants is more on the aquatic organisms because in the hydrosphere, pesticides and such other substances are transported to a greater distance and hence many more non-target organisms are likely to be exposed to them than in the terrestrial environment (Murty,1986).

Arsenic is an element which is present at low concentrations in air, soil and water. Arsenic, a non-

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essential trace element, a potent toxin, mutagen and xenobiotic metalloid has recently appeared as a major pollutant of drinking water in several districts of Assam, West Bengal, Tamilnadu and Andhra Pradesh. At present, one of the most worldwide environmental problems is that the drinking water has been polluted by arsenic. Arsenic poisoning from underground drinking water in Bangladesh was first identified in 1993 in the Nawabgonj District. The detection of arsenic in milk and meat is a new finding.

Fish is a major and easily available source of food in nature for mankind. It provides a significant amount of animal protein intake in the diet of a large population. It has been advised that fish should be consumed two or three times weekly because of the pharmaceutical effects of omega 3 polyunsaturated fatty acids which exist abundantly in fish oil. But, wide ranges of contaminants are continuously introduced into the aquatic environments and fish from polluted waters seriously threaten human health due to the bioaccumulation of metals in muscle and other tissues. Fish, as a living bio indicator organism, play an increasingly important role in monitoring of water pollution since they respond with great sensitivity to changes in the aquatic environment.

However, all the metals taken up by fish are not accumulated because fishes have the ability to regulate their body metal concentration to some extent showed that excretion of metals could occur through the gills, bile (*via* faeces), kidney and skin. There are five potential routes for a pollutant to enter a fish. These routes are through the food, non-food particles, gills, oral consumption of water and the skin. Once the pollutants are absorbed, they are transported by the blood to either a storage point (that is, bone) or to the liver for transformation and storage. If the pollutants are transformed by the liver, they may be stored there or excreted in the bile or passed back into the blood for possible excretion by the gills or kidneys, or stored in fat, which is an extra hepatic tissue. Proteins are basic molecules to any living system. In cells, they function as enzymes, structural materials, lubricants and carrier molecules.

The use of biochemical measurements in organisms as indicators of pollution, give information about the adaptive or deleterious responses in organism exposed to a certain amount of chemicals. Such analysis provides early warning signals before other toxicological points, including death are evident (Livingstone, 1998).Alterations in biochemical components like protein, carbohydrate and lipid as response to environmental stress are authenticated by many investigators; Ramakrishna and Sivakumar (1993) in *Oreochromis mossambicus*, Malla Reddy and Bashamohideen (1995) in *Cyprinus carpio* , Singh, *et al.* (1996) in *Heteropneustes fossilis*,Tilak *et al*., (2001) in *Labeo rohita* , Kumar and Saradhamani (2004) in *Cirrhinus mrigal*, Saraswathi (2004) in *Labeo rohita*, Arockia Rita and John Mitton (2006) in *Oreochromis mossambicus* and Prabhakara Rao and Radhakrishnaiah (2006) in *Cyprinus carpio.*

Understanding of the protein components of cell becomes necessary in the light of the radical changes taking place in protein profiles during pesticide intoxication. Both the protein degradation and synthesis are sensitive over a wide range of conditions and show changes to a variety of physical and chemical modulators. The physiological and biochemical alterations observed in an animal under any physiological stress can be correlated with the structural and functional changes of cellular proteins. Proteins occupy a unique position in the metabolism of cell because of the proteinaceous nature of all the enzymes which mediate at various metabolic pathways (Lehninger, 2004; Harper, 2003).

The induced stress and pathological conditions on protein metabolism showed alterations in sub-cellular proteins, enzyme activity levels which are found to be dependent on the protein making up to the cytosol fraction. The general nature of protein make up was studied to ascertain whether such relations do exist under induced stress. Enzyme bioassays however remain a useful technique in studying or diagnosing sublethal effects of toxic pollutants.

Aminotransferases mobilise the aminoacids into carbohydrate and lipid metabolism. There exists a rapid turnover of free aminoacids from cell to cell, tissue to tissue through the circulating fluid and utilize for various purposes through interconversions. Transaminases form an important group of enzymes mediating carbohydrates, protein and lipid metabolism. Transamination represents the mechanism causing eventual deposition of nitrogenous waste products like ammonia and urea resulting in the production of carbon compounds, which contribute towards gluconeogenesis and fatty acid formation. AST and ALT are two important enzymes mainly involved in the inter-conversion of important compounds such as pyruvate,oxaloacetate, á-ketoglutarate and aminoacids thus bringing the protein and carbohydrate metabolism on one hand and alanine, aspartic acid and glutamic acid on the other (Moore, 1964; Knox and Greengard,

1965). Aminotransferases also act as precursors of gluconeogenesis and probably during the period of stress, they meet the energy demands by channeling aminoacids into carbohydrate metabolism (Watts and Watts, 1974; Martin *et al*., 1983). The aspartate aminotransferase catalyses the interconversions of aspartic acid, and á-ketoglutaric acid to oxaloacetic acid and glutamic acid, while alanine, amino transferase catalyses the interconversion of alanine and á-ketoglutaric acid to pyruvate and glutamic acid.

Considering the role of above biomarkers in the field of eco-toxicology, the present study has been undertaken to understand the biochemical alterations induced by arsenic on exposure to sub-lethal concentrations to fish *Channa punctatus* in different tissues exposed. The present study also aims to study the possible remedial effect of turmeric on arsenic treated fish. The bioassays include Total protein level, Aspartate Aminotransferase (AST) and Alanine Aminotransferase (ALT) activities.

2. Material and methods

The fish *Channa punctatus* measuring 10 to 12 cm in length and 7.5 to 8 gm in weight were used in the experiment. Fish were washed with 0.1% KMnO4 solution to avoid dermal infection. All the precautions laid down by APHA *et al*., (1998) are followed, for maintaining the fish. The fishes were divided into three groups. The first group of fishes were exposed to 1/ 10 th of 96 hr LC50 i.e., 4.6 mg L-1 of sodium arsenite . The second group of fishes were exposed to turmeric (1%) extract. Whereas the third group of fishes were exposed to sodium arsenite for 96 hours and then exposed to turmeric extract. If mortality occurred during the experimental period, dead fish were removed immediately. At the end of the exposure time gill, liver and muscles were dissected out of the fish for the estimation of Total Protein level, Aspartate Aminotransferase (AST) and Alanine AminoTransferase (ALT) activities.

2.1 Estimation of total protein content

Total protein content was estimated by the modified method of Lowry *et al.,* (1951). 5% homogenates of gill, muscle and brain and 2% homogenates of liver and kidney were prepared in 5% trichloroacetic acid and centrifuged at 3000 rpm for 10 minutes. The supernatant was discarded. The suspended protein residue was dissolved in 1 ml of 1N NaOH. From this 0.2 ml of the extract was taken into the test tube and 5 ml of alkaline copper solution (50 ml of 2% Na2CO3 and 1ml of 0.5% CuSO4. 5H2O in 1% sodium potassium tartrate) was added. The contents were mixed well and allowed to stand for 10 minutes. To this 0.5 ml of 50% folin phenol reagent (diluted with distilled water in 1:1 ratio) was added. After 30 minutes, the optical density was measured at 540 nm in a spectrophotometer against a blank. The values were expressed as mg/gr wet weight of the tissue.

2.2 Estimation of aminotransferases activity

The activity of AAT and ALAT were determined by the method of Reitman and Frankel(1957). The selected tissues were homogenized in 5% ice-cold 0.25 M sucrose solution. The supernatants were used for the analysis of the enzyme activities.

2.3 Estimation of AST activity

The reaction mixture of 1.5 ml contains: 1 ml of phosphate buffer (pH 7.4), 0.1 ml of L-aspartate (L-Aspartic acid), 0.1 ml of á-ketoglutaric acid and 0.3 ml of supernatant as enzyme source. The reaction mixture was incubated at 370 C for 30 minutes. The reaction was stopped by adding 1 ml of 2, 4-dinitrophenyl hydrazine solution prepared in 0.1 N HCl and was allowed to stand for 20 minutes at room temperature. The rest of the details were the same as for alanine aminotransferase.

2.4 Estimation of ALT activity

The reaction mixture of 1.5 ml contains 1 ml phosphate buffer (pH 7.4), 0.1 ml of L- alanine, 0.1 ml of á-ketoglutarate and 0.3 ml of supernatant as enzyme source. The contents were incubated at 370 C for 30 minutes. The reaction was stopped by the addition of 1 ml of 2, 4- dinitrophenyl hydrazine solutions. After 20 minutes, 10 ml of 0.4 N sodium hydroxide was added and the colour developed was read at 545 nm in a spectrophotometer.

3. Result and discussion

3.1 Total protein

The results of the present study indicate that arsenic intoxication induced in decrease level of protein content in gill, liver and muscle of the fresh-water fish *Channa punctatus.* But the protein level were almost maintained like that of control when the fishes were treated with turmeric control. Whereas the protein level were increased when the fishes were treated with

turmeric after arsenic exposure. The results are summerised in Table 1.1 and graphically represented in Fig 1.1

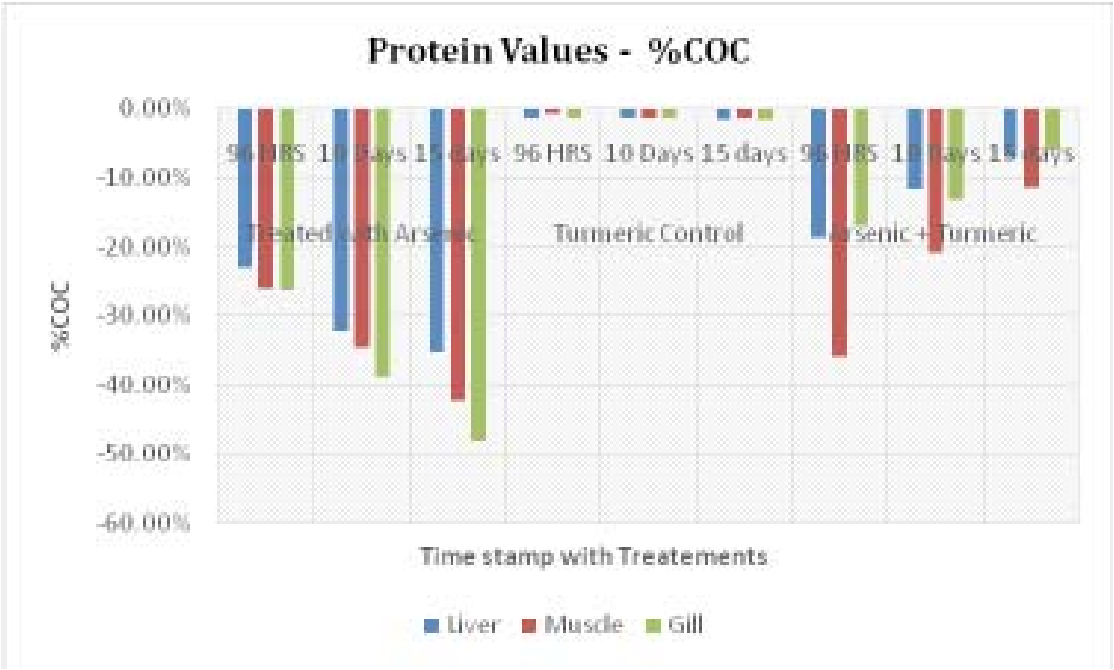
Table1.1: Change in the specific activity levels of Total Protein content (mg/gr wet tissue) in different tissues of fish Channa punctatus exposed to sublethal concentrations of sodium arsenite (1/10th LC50) and turmeric extract.

Tissue	Control	Treated with Arsenic			Turmeric Control			Arsenic + Turmeric		
Time stamp→		96 HRS	10 Days	15 days	96 HRS	10 Days	15 days	96 HRS	10 Days	15 days
Liver	76.09 ±0.30	58.64 ±0.38 a	51.54 ±0.40 A	49.3 ±0.33 a	75.14 ±0.30	75.12 ±0.26	74.99 ±0.11.	61.86 ±0.26 B	67.32 ±0.51 B	70.66 ±0.47 B
Muscle	93.32 ±0.16	69.01 ±0.70 a	60.97 ±0.50 A	53.72 ±0.26 a	92.65 ±0.19	92.21 ±0.53	92.13 ±0.45	59.85 ±0.21 B	73.91 ±0.21 B	82.63 ±0.13 B
Gill	60.02 ±0.45	44.31 ±0.19 a	36.61 ±0.42 A	31.11 ±0.35 a	59.29 ±0.58	59.21 ±0.44	59.16 ±0.45	49.78 ±0.84 B	52.12 ±0.78 B	56.36 ±0.77 B

Protein Values %COC

Tissue	Control ↓	Treated with Arsenic			Turmeric Control			Arsenic + Turmeric		
Time stamp→		96 HRS	10 Days	15 days	96 HRS	10 Days	15 days	96 HRS	10 Days	15 days
Liver	76.09	-22.93%	-32.26%	-35.21%	-1.25%	-1.27%	-1.45%	-18.70%	-11.53%	-7.14%
Muscle	93.32	-26.05%	-34.67%	-42.43%	-0.72%	-1.19%	-1.28%	-35.87%	-20.80%	-11.46%
Gill	60.02	-26.17%	-39.00%	-48.17%	-1.22%	-1.35%	-1.43%	-17.06%	-13.16%	-6.10%

Fig 1.1: Change in the % change over control of Total Protein content (mg/gr wet tissue) in different tissues of fish Channa punctatus exposed to sublethal concentrations of sodium arsenite (1/10th LC50) and turmeric extract.



2. Amino transferases

Aspartate Amino Transferase (AST) and Alanine Amino Transferase (ALT) activity :

The changes in the levels of aspartate amino transferases (AST) and alanine amino transferases(ALT) were studied in different tissues like liver, muscle and gill in the test fish *Channa punctatus* under sublethal concentrations of Sodium arsenite. The values are expressed as IU/L.

2(a) Aspartate Amino Transferase (AST)

The results of the present study indicate that arsenic intoxication induced increase activity of AST in gill, liver and muscle of the fresh-water fish *Channa punctatus*. But the AST activites were almost maintained like that of control when the fishes were treated with turmeric control . Whereas the AST activities showed a decreasing trend towards control values when the fishes were treated with turmeric after arsenic exposure. The calculated values of AST and percentage change over control along with standard error were given in Table 2.1.a, 2.1.b and are graphically represented in Fig 2.1

Table 2.1.a : Change in the specific activity levels of Aspartate Amino Transferase (AST) (IU/L) in different tissues of fish Channa punctatus exposed to sublethal concentrations of sodium arsenite (1/10th LC50) and turmeric extract.

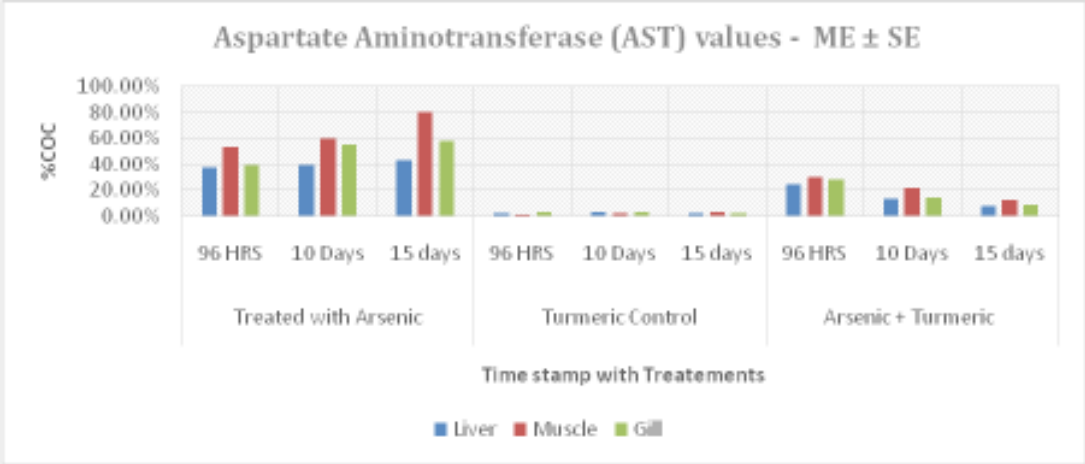
Tissue	Control ↓	Treated with Arsenic			Turmeric Control			Arsenic + Turmeric		
Time stamp→		96 HRS	10 Days	15 days	96 HRS	10 Days	15 days	96 HRS	10 Days	15 days
Liver	4.13 ±0.44	5.66 ±0.50 a	5.74 ±0.52 a	5.92 ±0.54 A	4.19 ±0.48	4.24 ±0.20	4.23 ±0.30	5.13 ±0.45 b	4.66 ±0.43 B	4.45 ±0.34 b
Muscle	3.79 ±0.25	5.79 ±0.36 a	6.06 ±0.52 a	6.81 ±0.60 A	3.81 ±0.53	3.84 ±0.44	3.85 ±0.39	4.92 ±0.50 b	4.58 ±0.48 B	4.22 ±0.46 b
Gill	4.41 ±0.16	6.12 ±0.4 a	6.86 ±0.45 a	6.97 ±0.46 A	4.56 ±0.37	4.53 ±0.40	4.48 ±0.27	5.66 ±0.24 b	5.01 ±0.21 B	4.79 ±0.39 b

Data represented as mean ±SE, n=5. aP<0.05 versus normal control, bP<0.05 versus sodium arsenite treated group.

Table 2.1.b: Change in the % change over control of Aspartate Amino Transferase (AST) activity in different tissues of fish Channa punctatus exposed to sublethal concentrations of sodium arsenite (1/10th LC50) and turmeric extract.

Aspartate Aminotransferase (AST) values - %COC										
Tissue	Control ↓	Treated with Arsenic			Turmeric Control			Arsenic + Turmeric		
Liver	4.13	37.00%	39.00%	43.00%	1.45%	3.00%	2.00%	24.00%	12.83%	7.75%
Muscle	3.79	53.00%	60.00%	80.00%	0.53%	1.32%	2.00%	29.82%	20.84%	11.35%
Gill	4.41	39.00%	55.56%	58.05%	3.00%	3.00%	2.00%	28.34%	13.61%	8.62%

Fig 2.1: Change in the % change over control of Aspertate Amino Transferase(AST) activity in different tissues of fish Channa punctatus exposed to sublethal concentrations of sodium arsenite (1/10th LC50) and turmeric extract.



2 (b) Alanine Amino Transferase (ALT)

The results of the present study indicate that arsenic intoxication induced increased activity of ALT in gill, liver and muscle of the fresh-water fish *Channa punctatus*. But the ALT activites were almost maintained like that of control when the fishes were treated with turmeric control .Whereas the ALT activities showed a decreasing trend towards control values when the fishes were treated with turmeric after arsenic exposure. The calculated values of ALT and percentage change over control along with standard error were given in Table 2.2.a, 2.2.b and are graphically represented in Fig 2.2

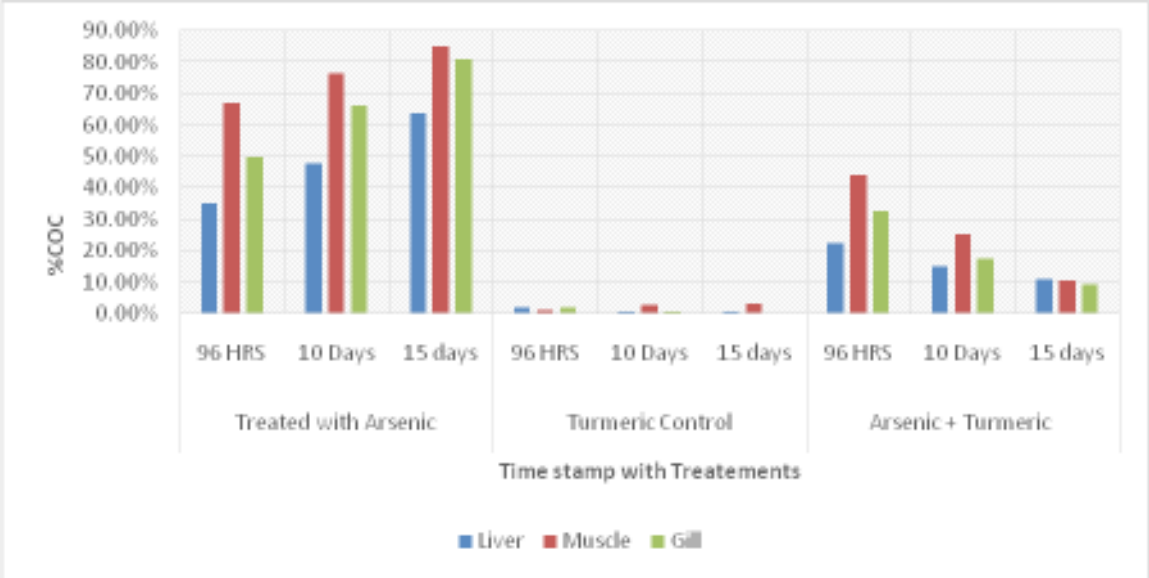
Table 2.2.a: Change in the specific activity levels of Alanine Amino Transferase (ALT) (IU/L) in different tissues of fish Channa punctatus exposed to sublethal concentrations of sodium arsenite (1/10th LC50) and turmeric extract.

Tissue	Control ↓	Treated with Arsenic			Turmeric Control			Arsenic + Turmeric		
Time stamp→		96 HRS	10 Days	15 days	96 HRS	10 Days	15 days	96 HRS	10 Days	15 days
Liver	10.79 ±0.19	14.60 ±0.09 a	15.96 ±0.16 a	17.65 ±0.32 a	10.95 ±0.19	10.88 ±0.30	10.87 ±0.27	13.21 ±0.40 b	12.43 ±0.20 b	11.99 ±0.2 B
Muscle	5.53 ±0.14	9.21 ±0.35 a	9.76 ±0.14 a	10.23 ±0.38 a	5.59 ±0.34	5.69 ±0.23	5.70 ±0.51	7.98 ±0.1 b	6.93 ±0.36 b	6.11 ±0.2 B
Gill	12.72 ±0.52	19.04 ±0.26 a	21.16 ±0.23 a	23.08 ±0.56 a	12.95 ±0.31	12.82 ±0.37	12.78 ±0.45	16.87 ±0.20 b	14.98 ±0.20 b	13.92 ±0.25 B

Data represented as mean ±SE, n=5. aP<0.05 versus normal control, bP<0.05 versus sodium arsenite treated group.

ALT values - %COC										
Tissue	Control ↓	Treated with Arsenic			Turmeric Control			Arsenic + Turmeric		
Time stamp→		96 HRS	10 Days	15 days	96 HRS	10 Days	15 days	96 HRS	10 Days	15 days
Liver	10.79	14.6	15.96	17.65	10.95	10.88	10.87	13.21	12.43	11.99
Liver	10.79	35.00%	48.00%	64.00%	2.00%	0.83%	1.00%	22.43%	15.20%	11.12%
Muscle	5.53	67.00%	76.49%	84.99%	1.08%	2.89%	3.07%	44.30%	25.32%	10.49%
Gill	12.72	50.00%	66.35%	81.00%	2.00%	1.00%	0.50%	32.63%	17.77%	9.43%

Fig 2.2: Change in the % change over control of Alanine Amino Transferase(ALT) activity in different tissues of fish Channa punctatus exposed to sublethal concentrations of sodium arsenite (1/10th LC50) and turmeric extract.



4. Discussion

Protein is the body builder. The decrease in protein level observed in the arsenic treated fish in the present study may be due to their degradation and also due to their possible utilization for metabolic purposes. Habib *et al*., (2007). have reported that arsenic toxicity is related to its high ability of reacting with protein and non-protein thiol groups leading to an alteration of cellular pathways. Vutukuru (2005) have reported a decrease in protein constituents in various tissues in fresh water fish *Labeo rohita* induced by chromium toxicity. Eva, (1990) reported a reduction of gill protein content in *Anabas testudineus* when exposed to sub-lethal concentration of Cuman L. Banerjee(2007) reported that the main reason for protein decrease in the gill tissues might be due to excessive synthesis followed by sloughing of the slime (made up of glycoprotein) induced by the arsenic toxicity .The other reason for the loss of proteins might be due to rejection of damaged cellular components of the gills resulted due to the contact of different toxicants , including an arsenic salt (Singh and Banerjee., 2008). The protein contents in the liver of chromium treated *Catla catla* was also found to be decreased(Vincent *et al*.,1995). Palanichamy and Baskaran (1995) reported a decrease in muscle and liver protein in *Channa punctatus* exposed to the heavy metals mercury, cadmium and lead for periods of 30 days. A decrease in protein content of liver, muscle and kidney in *Channa punctatus* were documented when exposed to sub-lethal concentration of sugar mill waste (Karupasamy, 1990). The reduction of total protein might be because of necrosis of cells and consecutive destruction in protein synthesizing machinery (Premdas and Anderson, 1963).

Since the heavy metal stress was known to induce significant change in protein metabolism, it is likely that the aminotransferases were also considerably affected. Increased activities of AST and ALT in different tissues of fish suggest either increased operation of transamination or increased synthesis of amino acids from other sources like glucose or fatty acids during Sodium arsenite intoxication. The AST and ALT are liver specific enzymes and they are more sensitive measure of hepatotoxicity and histophathalogic changes and can be assessed within a shorter time (Balint *et al*., 1997). The increase in AST and ALT indicate the tissue damages in liver, kidney and gill (Rajyasree and Neeraja, 1989; Oluah, 1999). Aminotransferases are important as they convert amino acids into keto acids and incorporate them in to TCA Cycle. Both AST and ALT level increased in tissues of fish suggesting the conversion of aminoacids released by the proteolysis into keto acids for energy production. The increase in AST and ALT activities in our study supports earlier findings and serves as indicator of tissue damage (Oluah, 1998; Oluah, 1999; Zikic *et al*., 2001 Satyaparameshwar *et al*., 2006).

AST and ALT are located in both mitochondrial and cytosol fractions of the cell. A close relation appears to exist between the mitochondrial integrity and transaminase levels(Bonitenko, 1974) and any modification in the organization of mitochondria is bound to alter the enzyme systems associated with it. The alteration in the activities of AST and ALT as observed in the present study may also be due to the mitrocondrial distruption and damage as a result of quinalphos induced stress. Tilak *et al.,* (2005) reported elevation in the levels of AST and ALT in different tissues of brain, liver, muscle, gill and kidney of the fishes *Catla catla, Labeo rohita* and *Cirrhinus mrigala* exposed to chlorpyrifos. Anita susan *et al.,* (1999) and Tilak *et al.,* (2003a) also reported increase in activities of AST and ALT in different tissues of fish *Catla catla* and *Channa punctatus* exposed to fenvalerate.

To summarize, the result of our test of Total protein , Aspartate Amino Transferase (AST), Alanine Amino Transferase (ALT) showed a tremendous and encouraging effect of Curcumin**,** a polyphenol; the active ingredient of the spice turmeric in resisting the ill effect of toxic materials like Sodium arsenite etc. on fresh-water lives like fishes which ultimately effect the human lives who consume the fishes as food supplement for protein.

References

Anita Susan, T., Veeraiah, K. and Tilak, K. S.,1999. Biochemical and enzymatic changes in the tissues of *Catla catla* exposed to the pyrethroid fenvalerate. J. Ecobiol. 11: 109-116.

APHA, AWWA and WEF 1998: Standard methods for the examination of water and waste water, 20th edition, Clesceri, L.S. Greenberg, A.E. and Eaton, A.D. (Eds.), American Public Health Association, American Water Work Association, Water Environment Federation, Washington DC.

Arockia, J. J. and John Mitton, M. C., 2006. Effect of carbamate pesticide lannate (methomy1) on the biochemical components of the freshwater cichlid (*Oreochromis mossambicus*). Indian J. Environ. Ecoplan. 12 (1): 263-268.

Banerjee, T. K., 2007. Histopathology of respiratory organs of certain air-breathing fishes of India. Fish Physiology and Biochemistry, 33: 441-454.

Balint, T., Ferenczy, J., Katai, F., Kiss, I., Kraczer, L., Kufcsak, O.,1997. Similarities and differences between the massive eel (*Anguilla anguilla* L.) devastations that occurred in lake Ablation in 1991 and 1995. Ecotoxicol. Environ. Saf. 37: 17–23.

Bonitenko, Y. U.,1974. Isoenzymes of aspartate amino-transferase in acute dichloroethane poisoning, Gig. Tr. Prof. Zabo. 7: 46-47.

Eva, C., 1990. Toxicity and impact of carbonate fungicide cumin L. on respiratory and protein metabolism of an air breathing fish *Anabas testudineus* (Bloch). M.Phil. Thesis, Annamalai University.

Habib, G.M., Shi, Z., Lieberman, M.W.,2007. Glutathione protects cells against arsenite- induced toxicity. Free Radic Biol Med. 42: 191-201.

Harper, H. A., 2003. In: Harper's Review of Biochemistry (eds.) D.W. Martin, P.A. Mayes and V.W. Rodwell. Lange Medical Publications, Marusen, Asia (Singapore).

Karupasamy, S., 1990. Toxicity impact of sugar mill effluent on freshwater fish *Channa punctatus (Bloch*). M.Phil. Thesis, Annamalai university, Annamalai nagar, India.

Knox, W. E. and Greengard, O., 1965. Advances in Enzyme regulation (Weber G. ed.,) Pergmon Press, New York, 3, 247.

Kumar, K. and Saradhamani, N., 2004. A study on the effect of a pesticide, avaunt on protein changes of the freshwater fish *Cirrhinus mrigala*. Nat. Environ. Poll. Tech. 3(3): 255-259.

Lehninger, A. L. ,2004. In: Biochemistry, Kalyani Publications Ludhiana, New Delhi.

Livingstone, D. R. ,1998. The fate of organic xenobiotics in aquatic ecosystems: quantitative and qualitative differences in biotransformation by invertebrates and fish Comp. Biochem.Physiol Mol. Integ. Physiol. 120 (1): 43-49.

Lowry, O. H., Rosenbrough, N. J., Farr, A. L, Randall, R. J. , 1951. Protein measurement with Folin phenol reagent. J. Biol. Chem. 193:265–275.

Malla Reddy, P. and Bashamohideen, Md. (1995) Alteration in protein metabolism in selected tissue of fish, *Cyprinus carpio*, during sub-lethal concentration of cypermethrin. Environ. Monit. Assess. 36: 183-190.

Martin, D. W., Mayers, P. A. and Rodwell, V. S., 1983. In; Harper's Review of Biochemistry Lange Medical Publications, Meruzen, Asia.

Moore, J.A. ,1964. In: Physiology of Amphibia. Academic Press, New York.

Murty, A. S., 1986. Toxicity of Pesticides to fish. Vol. I and Vol. II Boca Raton CRC Press.

Oluah, N. S., 1998. Effect of sublethal copper (II) ions on the serum transaminase activity in catfish *Clarias albopunctatus*. J. Aquat. Sci. 13: 45–47.

Oluah, N. S., 1999. Plasma aspartate aminotransferase activity in the catfish *Clarias albopunctatus* exposed to sublethal zinc and mercury. Bull. Environ. Contam. Toxicol.63: 343–349.

Palanichamy, S. and Baskaran P.1995. Selected biochemical and physiological responses of the fish Channa striatus as biomonitor to assess heavy metal pollution in freshwater environment. Journal of Ecotoxicology and environmental Monitoring, 5: 131-138.

Prabhakara Rao, K and Radhakrishnaiah, K., 2006. Pesticidal impact on protein metabolism of the freshwater fish, *Cyprinus carpio* (Lin.). Nat. Environ. Poll.

Premdas, F.H. and J.M. Anderson, 1963. The uptake and detoxification of C^{14} labelled DDT in Atlantic Salmon *Salmo salar.* J.Fish. Res.Bd.Can., 30:837. Tech. 5(3): 367-374.

Rajyasree, M. and Neeraja, P. ,1989. Aspartate and alanine aminotransferase activities in fish tissue subcellular fractionation on exposure to ambient urea. Indian J. Fish. 36: 88–91.

Ramakrishnan, R. and Sivakumar, A. A., 1993. Effect of quinalphos on the fish *Oreochromis mossambicus.* J. Ecol. biol. 5: 45-50.

Reitman, S. and Frankel, S.,1957. Calorimetric method for the determination of serum glutamic oxaloacetic and glutamic-piruvic transaminases. Amer. J. Clin. Pathol 28: 65-63.

Saraswathi, R., 2004. A study on the effect of commando on the toxicity of the freshwater fish *Labeo rohita.* M.Phil. Thesis submitted to Bharathiar University, India.

Satyaparameshwar, K., Ravinder Reddy, T. and Vijaya Kumar, N. ,2006. Effect of Chromium on protein metabolism of fresh water mussel, *Lamellidens marginalis*. J. Environ. Biol.27(2): 401-403.

Shivakumar, R.,2005. Endosulfan, induced metabolic alteration in fresh water fish *Catla catla*.Ph.D. Thesis, Karnataka University, Dharwad, Karnataka, India.

Singh, N. N., Das, V. K. and Singh, S. ,1996. Effect of Aldrin on carbohydrate, protein and ionic metabolism of a fresh water catfish *Heteropneustes fossilis*. Bull. Environ. Contam. Toxicol. 57: 204-210.

Singh, A.K. and Banerjee, T.K., 2008. Toxic effects of sodium arsenate (Na2 HASO4 7H20) on the skin epidermis of air – breathing catfish *Clarias batrachus* (L).VeterinarskiArhiv. 78 (1), 73-88.

Tilak, K. S., Veeraiah, K. and Ramana Kumari G. V., 2001. Toxicity and effect of chlorpyriphos to the freshwater fish *Labeo rohita* (Hamilton). Poll. Res. 20(3): 438-445.

Tilak, K. S., Satyavardhan, K. and Thathaji, P. B., 2003a. Biochemical changes induced by fenvalerate in the freshwater fish *Channa punctatus* J. Ecotoxicol. Environ. Monit. 13(4): 261-270.

Tilak, K. S., Veeraiah, K. and Koteswara Rao, D. , 2005. Biochemical changes induced by chlorpyrifos, an organophosphate compound in sublethal concentrations to the freshwater fish *Catla catla, Labeo rohita* and *Cirrhinus mrigala*. J. Environ. Biol. 26 (2 suppl): 341-347.

Vincent, S.; Ambrose T.; Cyril L.; Kumar A. and Selvanaygam M., 1995. Biochemical response of the Indian major carp,. *Catla catla (Ham.)* to chromium toxicity. Indian, J.Environ. Hlth, 37(3): 192-196.

Vutukuru, S.S., 2005. Acute effect of Hexavalent chromium on survival, oxygen consumption, haematological parameter and some biochemical profiles of the Indian Major carp, *Labeo rohita.* Int. J. Environ. Res. Public Health; 2 (3)456-462.

Watts, R. D. and Watts, D. C., 1974. In: Chemical Zoology (ed. Florkin M. and Scheer, B.T.).Vol.VII D. Cuterostomians cyclostomes and Fishes. Academic Press, New York.

Zikic, R.V., Stajn, A. S., Pavlovic, S. Z., Ognjanovic, B. I., Saicic, Z. S. ,2001. Activities of superoxide dismutase and catalase in erythrocytes and plasma transaminase of gold fish (*Carassius auratus gibelio* Bloch.) exposed to cadmium. Physiol. Res. 50, 105–111.