ASCORBATE EFFECT ON PROTEIN CONTENT DURING NICKEL INTOXICATION IN THE FRESHWATER BIVALVE, *LAMELLIDENS CORRIANUS*

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ABSTRACT

The effect of L-ascorbic acid on protein content during nickel intoxication of the freshwater bivalve, *Lamellidens corrianus* has been studied. These bivalves were exposed to chronic (0.3981 ppm) dose of Nickel chloride with and without ascorbic acid. Protein content from gill, gonad, digestive gland, mantle, foot of control and experimental bivalves from different group were estimated after 15 days and 30 days. After 30 days Nickel chloride treated bivalves were allow to recover in normal water with and without ascorbic acid. During recovery protein content was estimated after 5 days and 10 days. Protein content was decreasedless in nickelwith ascorbic acid as compare to exposure to Nickel chloride without ascorbic acid. Bivalves show faster recovery with ascorbic acid as compare to normal water recovery.

Key words: Ascorbate, Protein, Nickel, bivalve

INTRODUCTION

Biochemical composition of aquatic organisms and their different biochemical processes are useful in determining the mechanism of toxicity and severity of various toxicants. Naturally, there is protective mechanism of the body to resist and combat the toxic effect of the pollutant like heavy metals. Besides, it is observed that some biochemical alterations occurring in the body gives the alarming first indication of stress condition. Heavy metals such as lead and arsenic are the most common forms of anthropogenic pollutants in aquatic environment. Prolonged exposure to organisms produces cellular alteration in variety of body tissues. Some trace metals have a variety of biochemical function and play an important role in the growth and development of living organisms, but their excess adversely affect the organisms. The trace metals are known to be non bio-degradable and highly toxic to most organisms (Kaoud and Dahshan, 2010).

The heavy metal had a suppressing effect on the biochemical constituents such as protein, carbohydrate and lipid content in the crustaceans. Krishnamoorthy and Subramanian (1995) observed decreased protein content in muscle, gill and hepatopancreas of *Macrobrachium lamarreilamarrei* when exposed to copper. Decrease in the biochemical component was observed commonly in various crustaceans in response to heavy metals (Sarojini *et al.,* 1990; Reddy and Bhagyalakshmi, 1984).

Nickel is a silver white metal with siderophilic properties that facilitate the formation of nickel-iron alloys. In contrast to the soluble nickel salts. Metallic nickel, nickel sulfides and nickel oxides are poorly water soluble. Nickel carbonyl is a volatile liquid at room temperature that decomposes rapidly in to carbon monoxide and nickel. Drinking water and food are the main sources of exposure for general population with the overage American diet containing about 300 µg Ni/d. Nickel is highly mobile in soil, particularly in acid soils. There is little evidence that nickel compounds accumulate in the food chain. Nickel is not a cumulative toxin in animals or in humans. The initial effects involve irritation of the respiratory tract and non specific symptoms. Patients with severe poisoning develop intense pulmonary and cerebral edema are the main cause of death (Barceloux, 1999).

Proteins are long chains of amino acids forming three dimensional structures. Proteins do play both structural and functional role of cellular level. Being an integral part of the cell membrane, intracellular and extra cellular passages are linked through it.

Any sort of cellular metabolism occurring in body involves one or many different proteins. The proteins are among the most abundant biological macromolecules and are extremely versatile in their function and interaction during metabolism of proteins, amino acids, enzymes and co-enzymes (Harper *et al.*, 1978). Deshmukh and Lomte (1998) studied the biochemical content of protein in mantle, foot, gill, digestive gland and whole body of fresh water bivalve, *Parreysia corrugata* after acute and chronic exposure to copper sulphate. Decrease in protein content was found due to increased proteolytic enzymes under stress (Jallauddin, 1987).The biochemical variations in marine bivalve, *Mytilus edulis* was studied by William (1969). Rao and Mane (1987) studied the biochemical composition of Indian freshwater bivalves.

MATERIALS AND METHOD

The freshwater bivalves, *Lamellidens corrianus* were collected from the Nathsagar dam at Paithan Tq. Paithan. Dist. Aurangabad (M.S.). After collection, bivalves were acclimatized in the laboratory condition at room temperature for 2-3 days. The healthy and active acclimatized bivalves of approximately same size were selected for experiment. These bivalves were divided in to five groups and were treated as follows.

First group was maintained as Control. In second group theBivalves were exposed separately to chronic doses (LC50/10) of Nickel chloride (0.3981 ppm) Third group bivalves were exposed separately to chronic doses (LC50/10) ofNickel chloride along with ascorbic acid (50mg/l).

After 30 days exposure to nickel chloride, bivalves from group 2 were divided in to two groups for recovery studies. The bivalves pre exposed to chronic dose (LC50/10) of nickel chloride were treated as. Bivalves pre-exposed to chronic doses (LC50/10) of nickel chloride were allowed for self cure in normal water. Bivalves pre-exposed to chronic doses (LC50/10) of nickel chloride were exposed to ascorbic acid (50mg/l).

The experimental bivalves from 1 to 3 groups were dissected after 15 days and 30 days and from each recovery group (4 to 5) after 5 days and 10 days. Gills, gonad, digestive glands,mantle and foot and from all experimental and recovery group were dried at 80 ^{0}C in an oven until constant weight was obtained. The dried powders of these different tissues of control, experimental and recovery group animals were used for estimation of their protein contents.

Total protein was estimated by Lowry's method (Lowry *et al.*, 1951) using bovine serum albumin as standard from each powder. The average results of three repeats are presented in the table No. 1 and are expressed as percentage of dry weight. Percent variations, standard deviation, and "t" test of significance were calculated and are expressed in respective tables

RESULTS AND DISCUSSION

Biochemical estimation of protein contents were estimated from the different tissues i.e. gills, gonads, digestive glands, foot and mantle of experimental model, the freshwater bivalve *Lamellidens corrianus* from control and experimental groups are presented in table no. 1

Protein profile:

Table No.1 indicate changes in protein levels of gills, gonads, digestive glands, mantle and foot of *Lamellidens corrianus* on chronic exposure of nickel (0.3981 ppm) with combinations of ascorbic acid and during recovery. The experimental control bivalves in ascorbic acid showed slight non significant alterations in the protein levels in all tissues. It is noticed that protein contents were significantly reduced after nickel exposure in all tissues of the bivalves as compared to control. Bivalves exposed to nickel with ascorbic acid showed fewer alterations in the protein contents showing the protective role of the ascorbic acid. When ascorbic acid was simultaneously given with the nickel the alterations in the protein contents were still minimized.

When the bivalves exposed for 30 days to nickel was allowed to recover, protein recovery was at a very slow rate in naturally curing bivalves and in most cases was non-significant. Protein contents recovered faster during ten days in all tissues in ascorbic acid. rate of recovery was better in ascorbic acid than in normal water recovery. Sometimes due to the photo induced effect, some of the substances have complex toxicological effects. For example, polycyclic aromatic hydrocarbons (PAHs) are of much environmental concern they are hydrophobic and bio-accumulate

Table 1: Protein content in selected tissues of *Lamellidens corrianus* after chronic exposure to $NiCl_2$ without and with Ascorbic acid and during recovery.(Values represent percentage in dry weight)

Treatment		*Tissue*	15days	30days	Recovery	
					5days	10days
Control		Gill	59.22±8.767	57.37±8.226		
		Gonad	50.29±6.321	46.63±5.434		
		Dig. Glands	63.92±10.214	61.22±13.491		
		Mantle	51.22±6.558	49.11±2.955		
		Foot	69.52±7.942	64.33± 6.788		
$NiCl_2$		Gill	43.26±4.678❖❖ (-26.95)	39.25±3.850❖ ❖(-31.58)		
		Gonad	39.62±3.924❖❖ (21.21)	38.57±3.718❖ (-17.28)		
		Dig. Glands	50.33±6.332❖ 21.26	48.16±5.798❖ (-21.33)		
		Mantle	36.29±3.684❖❖ (-29.14)	34.19±2.921❖ ❖(-30.38)		
		Foot	55.92±7.817❖ (-19.56)	50.33±4.849❖ ❖(-21.76)		
$NiCl_2$ + Ascorbic acid		Gill	47.98±5.755❖ (-18.98)	42.59±4.533❖ ❖(-25.76)		
		Gonad	41.09±4.219❖ (-18.29)	39.16±3.833❖ (-16.01)		
		Dig. Glands	51.63±6.500❖ (-19.22)	49.62±6.155❖ (-18.94)		
		Mantle	37.19±3.456❖❖ (-27.39)	35.13±3.084❖ ❖(-28.46)		
		Foot	58.11±8.440❖❖ (-16.41)	51.36±6.594❖ (-20.16)		
After 30 days Exposure to $NiCl_2$ & $NiCl_2$+Ascorbic acid	Normal Water (D)	Gill			42.33±4.478■ ■[+26.21]	43.97±4.832■■ [+23.35]
		Gonad			40.73±4.145NS[+12.65]	41.63±4.331NS [+10.72]
		Dig. Glands			49.52±6.130■ [+19.11]	50.03±6.257■ [+18.27]
		Mantle			35.11±3.080■ ■[+28.50]	35.98±3.236■■ [+26.73]
		Foot			51.22±6.558■ [+20.37]	52.62±6.923■ [+18.20]
	Normal Water + A. A.	Gill			45.06±5.076■ ■[+21.45]	47.22±5.566■ [+17.69]
		Gonad			41.12±4.227NS[+11.81]	42.01±4.411NS [+9.90]
		Dig. Glands			50.16±6.290■ ■[+18.06]	51.13±6.534■ [+16.48]
		Mantle			35.92±3.225■ ■[+26.85]	37.12±3.444■■ [+24.41]
		Foot			52.69±6.939■ [+18.09]	53.97±7.280■ [+16.10]

Values in the ( ) brackets indicate percent change over control

N.S. - Non Significant ❖ - Compared with respective (A)
❖/■ - P < 0.005 ■ - Compared with respective 96hrs of (B)
❖❖/■■ - P < 0.01
❖❖❖/■■■ -P < 0.001

to form residues which may lead to carcinogenic effect. The possibility of PAH carcinogenicity being induced by ultra violet rays, has been recorded suggesting that photo activation of ground state PAHs to excited state may be the key in predicting environmental effects (Morgan *et al.,* 1997). Out of the present, thousands of chemical entering into societal use a very few have the empirical properties needed for risk assessment, ecotoxicological and environmental fate data. In such a situation the development and application of quantitative structure – activity relationship has expanded the general research activities significantly. This includes development of models as a complementary tool in management of toxicological risk assessment. This is becoming popular as available alternative tool for predictive toxicology.

Scanning of pertinent literature reveals that biochemical studies under nickel intoxication in freshwater teleosts are rather mearge Gill and Tant(1981). Chaudhary and Nath (1985), Since considerable quantities of nickel are sometimes present in the ambient environment of fish Rehwoldt *et al.,* (1973) reported the manner in which the metal interferes with the physiology of the fish has not been fully understood, it was thought worth while to identify some of the metabolic dysfunctions in the fish, *Anabas testudineus,* intoxicated with a chronic (30days) sublethal concentration (140.70 mg/l) of mercuric chloride. The variable monitored are glycogen, total protein and lipids in the liver and gonads of the fish.

Andhale and Zambare (2011), studied the nickel induced biochemical alterations in freshwater bivalve, *Lammellidens marginalis* and reported that the protein contents were decreased in treated animals than the control. In the present study ascorbic acid recovered the total protein content and it play important role as detoxication of nickel which recovered the protein content.

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