

## EXTRACT FROM CHILLIS, REMEDY FOR HEAVY METAL TOXICOSIS INDUCED ALTERATIONS IN THE LIPID CONTENT IN VARIOUS ORGANS OF FRESH WATER MUSSELS, *L. CONSOBRINUS*

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### ABSTRACT

The effect of sub lethal concentration of chromium trioxide on the lipid content in the various organs of fresh water mussel, *Lamellidens consobrinus* like gills, whole body & digestive gland is studied. Among the control set lipid content is  $5.58 \pm 0.0020$  in gills,  $8.74 \pm 0.0024$  in digestive gland and  $7.70 \pm 0.0030$  in whole body respectively. In set B, on exposure to heavy metal salt chromium trioxide (Lc50/10 of 96 hours i.e. 1.65 ppm), the lipid content is  $5.02 \pm 0.0028$  in gills,  $7.70 \pm 0.0036$  in digestive gland &  $7.14 \pm 0.0020$  in whole body respectively. In set C when animals are treated with 1.65 ppm of CrO<sub>3</sub> + 5ml/litre extract of capsicum annum, the lipid content is maximum in the gills i.e  $5.26 \pm 0.0030$ ,  $8.08 \pm 0.0032$  in digestive gland &  $7.38 \pm 0.0026$  respectively after 7 days exposure of treatment. The chronic treatment is given upto 42 days. The lipid level significantly decreased on exposure to heavy metal salt chromium trioxide, while increase in presence of chilli extract. During recovery the lipid content recovered and rate of recovery was faster in chilli extract exposed fresh water muscels as compare to those recovered in normal water. The probable role of antioxidants from chilli *Capsicum annum* linn. on heavy metal salt of chromium trioxide on the lipid contents on fresh water mussels *Lamellidens consobrinus* studied in this paper.

**Keywords:** Chromium Trioxide, Lipid Content, *Lamellidens consobrinus*, Extract of *Capsicum annum* Linn.

### INTRODUCTION

Heavy metals play a crucial role in various biological functioning of aquatic organisms and remain present in trace amount in the body but increase in the concentration lead to high level of toxicity in different organs. The rapid industrialization in India has resulted into substantial increase in the liquid effluents, which traditionally discharged into open land or nearby natural water, causing a number of problems like threat to aquatic flora and fauna and surface water logging and salinizing of land (Ramana *et al.*, 2001). Effluent of electroplating industry containing heavy metals, is an industry which experts an impact on aquatic organisms (Kokila *et al.*, 2005; Kaur and Kaur, 2006).

Fresh water mussel *Lamellidens marginalis* is a common bivalve occurring in freshwater bodies in ponds, rivers, dams, ditches, canals etc. They are cilliary filter feeder feeding mainly on planktons (algae protozoa and bacteria) as well as riverine sediments. To obtain their food they circulate large quantity of water through their body cavity from which the food substances are filtered out with the help of gill filaments. In gills, they are known to accumulate toxicants like heavy metals (Santosh and Diniz, 2007). The mussels are used as food by tribal people in India. Their flesh is rich in protein carbohydrate and lipid, it is soft, tasty and easily cooked and digestible. Most recently it is observed that the body extract of fresh water mussel has therapeutic use in remedy of arthritis (Chakraborty *et al.*, 2010).

In aquatic environment, Cr is one of the biochemically active transition metal. Weathering of earth crust is the primary and natural source of chromium in the surface water through an essential trace nutrients and a vital component for the glucose tolerance factor, chromium toxicity damages the liver, lungs and causes organ hemorrhages (Goyer and Clerian, 1995) chromium compounds are used as pigments,

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mordant and dyes in the textiles and as the tanning agent in leather. Anthropogenic sources of emission of Cr in the surface waters are from municipal wastes, laundry chemicals, paints, leather industries, road run off due to tire wear, corrosion of bushings, brakes wires, radiators (Dixit, 2008). The hexavalent chromium when present in excess amount, induces toxic effects in the cells like genotoxicity and oxidative damage primarily occurs through production of reactive oxygen species (ROS) and can damage lipids, proteins and DNA contributing to loss of activity and structural integrity of enzymes. Lipids are the molecules that contain hydrocarbon and make up the building blocks of the structure and function of living cell. Lipid is the most efficient organic reserve of most of the bivalve and other animals. They are the major structural component of body tissues and is therefore essential to study the effect of variables on the lipid content.

Green chilli contains a wide array of phytochemicals such as neutral acidic phenolic compounds, which are important nutritional antioxidants which may reduce the risk of degenerative diseases (Howard *et al.*, 2000).

Carotenoids, which are fat soluble antioxidants found in peppers/chilli, have received considerable interest by researchers due to their antioxidant properties (Rao and Rao, 2007) now a day, chilli is an important vegetables crop due to colour, flavour and aroma to the food. In peppers, there is phytochemical property which include antioxidants, anti-inflammatory, anti allergic and anticarcinogenic activities (Lee *et al.*, 2005). It is also has been proven that, ripe red peppers can reduces the risk of cancers (Nishino *et al.*, 2009). So, in present study, the probable role of antioxidants from green chilli *Capsicum annum* on chromium induced toxicosis on lipid content in fresh water bivalve *Lamellidens consobrinus* is discussed.

### MATERIALS AND METHODS

The fresh water bivalve mollusc, *Lamellidens consobrinus* were collected from Hartala lake near Muktainagar. The bivalves which were healthy, well developed were selected for laboratory experiment. After bringing to Laboratory, shells of bivalve were cleaned and washed with fresh and clean water to remove algal biomass and other waste material; the healthy bivalves were acclimatized in the laboratory for the further study.

The healthy and acclimatized bivalves of approximately same size were selected for experiment. These bivalves were divided into five groups and were treated as follows: first group was maintained as control. In second group, the bivalves were exposed separately to chronic doses ( $LC_{50}$  values of 96 hrs/10) of chromium trioxide (1.65ppm) upto 21 days.

Third group of bivalves were exposed separately to chronic concentration of chromium along with chilli extract (extract of *capsicum annum*), (5ml/lit) upto 21 days. After 21 days' exposure to chromium trioxide, the bivalves from group B were divided into two sub groups D and E for recovery studies. Bivalve pre exposed to chronic doses of chromium trioxide were allowed for self cure in normal water while the bivalve of group E are exposed to 5 ml/lit. chilli extract upto 21 days. The experimental bivalves from A to C groups were dissected after 7 days, 14 days and 21 days and from each recovery group (D and E) were collected after 28, 35 and 42 days.

The gills, digestive gland and whole body from all experimental and recovery group were dried at 80°C in oven until constant weight is obtained. The dried powder of these tissues of control, experimental and recovery group animals were used for estimation of their lipid contents the total lipids from dried tissues were estimated by vanilline reagent method as given by Barnes and Blackstoke (1973) using cholesterol as standard. 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 test of significance were calculated and are expressed in respective tables.

For lipid analysis, animals were dissected and soft body tissues like whole body, gills and digestive glands were removed. The animal's body part was dried and powder was prepared. Lipid was determined by the method proposed by Barnes and Black stoke in 1973. The results were expressed in milligram

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**Table 1: Alteration of lipid content in carious tissues of *Lamellidens consobrinus* after chronic exposure to heavy metal salt CrO<sub>3</sub> and CrO<sub>3</sub> with 5ml/lit extract of capsicum annum.**

| Day/Set   | Tissue | 7 days     | 14 days     | 21 days     | 28 days | 35 days | 42 days |
|---|--------|------------|-------------|-------------|---------|---------|---------|
| A<br>Control set  | Gills  | 5.58       | 5.40        | 5.12        | --      | --      | --      |
|   |        | ± 0.0020   | ±0.0017     | ±0.0024     |         |         |         |
|   |        |            |             |             |         |         |         |
|   | D.G.   | 8.75       | 8.42        | 7.94        | --      | --      | --      |
|   |        | ±0.0024    | ± 0.0022    | ±0.0026     |         |         |         |
|   | W.B.   | 7.70       | 7.16        | 6.88        | --      | --      | --      |
| B<br>Treated with<br>LC <sub>50/10</sub> at 96 hrs<br>of CrO <sub>3</sub><br>(1.65ppm)                          | Gills  | ± 0.0030   | ±0.0028     | ±0.0032     |         |         |         |
|   |        | 5.02       | 4.88        | 4.54        | --      | --      | --      |
|   |        | ±0.0028    | ±0.022      | ±0.0035     |         |         |         |
|   | D.G.   | P=-4.1404  | P= -6.3192  | P=-3.5013   |         |         |         |
|   |        | (-10.04%)  | (-9.63%)    | (-11.33%)   |         |         |         |
|   |        | 7.70       | 7.72        | 6.88        | --      | --      | --      |
|   | W.B.   | ±0.0036    | ± 0.0030    | ±0.0020     |         |         |         |
|   |        | P=-5.00101 | P=-9.6213   | P=10.0542   |         |         |         |
|   |        | (-11.90%)  | (-14.25%)   | (-13.35%)   |         |         |         |
|   |        | 7.14       | 6.80        | 6.20        | --      | --      | --      |
|   |        | ± 3.4300   | ±0.0017     | ±0.0028     |         |         |         |
|   |        | (-7.27%)   | P=2.0112    | P=12.4756   |         |         |         |
| C<br>Treated with 1.65<br>ppm of CrO <sub>3</sub> +<br>5 ml/lit extract<br>from <i>Capsicum</i><br><i>annum</i> | Gills  | (-5.03%)   | (-9.88%)    |             |         |         |         |
|   |        | 5.26       | 5.12        | 4.78        | --      | --      | --      |
|   |        | ±0.0030    | ±0.0024     | ±0.0026     |         |         |         |
|   | D.G.   | P=-1.9600  | P=-2.3940   | P= -10.6108 |         |         |         |
|   |        | (-5.73%)   | (-5.19%)    | (-6.84%)    |         |         |         |
|   |        | 8.08       | 7.72        | 7.34        | --      | --      | --      |
|   | W.B.   | ±0.0032    | ±0.0026     | ± 0.0037    |         |         |         |
|   |        | P=-5.6714  | P= -10.4654 | P=-3.3534   |         |         |         |
|   |        | (-7.55%)   | (-8.31%)    | (-7.56%)    |         |         |         |
|   |        | 7.38       | 6.96        | 6.50        | --      | --      | --      |
|   |        | ±0.0026    | ±0.0024     | ±0.0028     |         |         |         |
|   |        | P=5.5328   | P=3.232     | P=6.9717    |         |         |         |
|   |        | (-4.16%)   | (-2.79%)    | (-5.52%)    |         |         |         |

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**Table 2: The variation of lipid contents of preexposed *Lamellidens consobrinus* to  $\text{CrO}_3$  for 21 days during recovery**

| Day/Set   | Tissue | 7 days | 14 days | 21 days | 28 days                 | 35 days                | 42 days               |
|---|--------|--------|---------|---------|-------------------------|------------------------|-----------------------|
| D<br>Normal water<br>cure   | Gills  | --     | --      | --      | 4.84                    | 5.06                   | 5.30                  |
|   |        |        |         |         | $\pm 0.0028$            | $\pm 0.0014$           | $\pm 0.0020$          |
|   |        |        |         |         | P=-5.4721<br>(13.26%)   | P=0.5521<br>(-6.30%)   | P=-2.4528<br>(+3.52%) |
|   | D.G.   | --     | --      | --      | 7.36                    | 7.62                   | 7.80                  |
|   |        |        |         |         | $\pm 0.0017$            | $\pm 0.0028$           | $\pm 0.0035$          |
|   |        |        |         |         | P=11.7815<br>(-15.79%)  | P=-8.2720<br>(-9.50%)  | P=-0.5988<br>(-1.017) |
| E<br>Cure with 5.00<br>ml/lit extract of<br><i>Capsicum annum</i> | W.B.   | --     | --      | --      | 6.04                    | 6.86                   | 7.12                  |
|   |        |        |         |         | $\pm 0.0022$            | $\pm 0.0020$           | $\pm 0.0030$          |
|   |        |        |         |         | P=8.4988<br>(-13.77%)   | P=2.2181<br>(-4.19%)   | P=-9.585<br>(3.49%)   |
|   | Gills  | --     | --      | --      | 4.88                    | 5.24                   | 5.54                  |
|   |        |        |         |         | $\pm 0.0030$            | $\pm 0.0028$           | $\pm 0.0035$          |
|   |        |        |         |         | P=-4.2875<br>(-12.54%)  | P= -0.8939<br>(-2.96%) | P=-2.5355<br>(8.20%)  |
|   | D.G.   | --     | --      | --      | 7.48                    | 7.92                   | 8.28                  |
|   |        |        |         |         | $\pm 0.0036$            | $\pm 0.0028$           | $\pm 0.0017$          |
|   |        |        |         |         | P=-6.6751<br>(-14.42%)  | P=-5.1700<br>(-5.94%)  | P=-2.2792<br>(4.28%)  |
|   | W.B.   | --     | --      | --      | 6.76                    | 7.22                   | 7.62                  |
|   |        |        |         |         | $\pm 0.00036$           | $\pm 0.0024$           | $\pm 0.0017$          |
|   |        |        |         |         | P=-9.0519<br>(-12.21 %) | P=-0.9698<br>(0.84%)   | P=-3.1691<br>(10.76%) |

Figures in bracket indicate percent variation in lipid content

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content per 100mg wet tissue. Triplicate values of each biochemical constituent were subjected for statistically confirmation by p value standard deviation was calculated during every step of experiment.

### RESULTS AND DISCUSSION

The biochemical estimation of lipid contents was estimated from the different tissues like gills, whole body and digestive gland are presented in the respective tables. Thus from the result obtained indicates that there is severe alteration in the lipid metabolism in fresh water bivalve *Lamellidens consobrinus* after exposure to chromium trioxide in the present study, significant decrease in the lipid content was observed in set C of *Lamellidens consobrinus* compared to those of control bivalves.

There was increase in lipid content in heavy metal salt with extract of *Capsicum annum* exposed bivalve compared to those exposed to only Chromium trioxide. The bivalve shows fast recovery in the tissue lipid level in presence of *Capsicum annum* extract than those allowed to cure naturally. When the bivalve exposed for 21 days to chromium trioxide was allowed to recover, the lipid recovery was at a slow rate in naturally curing bivalves. Lipid contents recovered faster during 7 days in all tissues in *Capsicum annum* extract. The rate of recovery was better in *Capsicum annum* extract than in the normal water recovery.

The decrease in the lipid content in fresh water bivalve, after chromium trioxide treatment may be due to reduced synthesis of lipid or increased activity of lipase involved in oxidation of lipids. The overall decrease in lipid content of tissue indicates the pronounced lipolysis and its utilization during heavy metal exposure (Shivprasad *et al.*, 1979). Jalaluddin (1987) observed decrease in lipid content in muscles of fish after exposure to heavy metal stress, might be due to compensate the energy requirement in animals. Mahajan (2005) observed decrease in lipid content in fresh water gastropod snail *Bellamya bengalensis* after heavy metal salt HgCl<sub>2</sub>, PbNO<sub>3</sub> treatment may be due to reduced synthesis of lipids and increased activity of lipase involved in oxidation of lipids. In present study, antioxidants from *Capsicum annum* recovered the total lipid content and it play important role as detoxification of chromium which recovered the lipid content.

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