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Effects of lead nitrate on oxygen consumption of fresh water prawn, *Macrobrachium dayanum* (Crustacea - Decapoda)

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Abstract

Freshwater prawn, *Macrobrachium dayanum* were subjected to acute concentration, 116.46 mg/l (96 hr LC_{50} value) and sub-acute concentration, 29.12 mg/l (25% of 96 hr LC_{50} value) of lead nitrate to evaluate its effects on oxygen consumption. Initial increase in oxygen consumption was noticed which was followed by gradual decrease up to 96 hr in acute exposure while significant ($F = 20.33$; $P < 0.001$) decrease in oxygen consumption was noticed through out the experiment during sub-acute exposure up to 30 days. Mechanism of lead intoxication and potential of oxygen consumption as bio-marker has been discussed.

Keywords:- *Crustacea, Freshwater, Lead nitrate, Macrobrachium dayanum, Oxygen consumption*

Introduction

Now a day's water pollution is a issue of great concern. Surface water, a limited commodity, is being continuously contaminated by various anthropogenic activities like industrial effluents, sewage, agricultural runoff containing insecticides, pesticides and various other chemicals. The load of contaminants is increasing day by day thereby deteriorating the life sustaining qualities of water bodies as well as adversely affecting aquatic flora and fauna (Jarup, 2003; Sharma and Agrawal, 2005). Among toxicants, heavy metals are lethal because of their long half life period, persistent accumulative and amplificative tendency in the food chain there by increasing the problem to many folds (Burman and Lal, 1994).

Among heavy metals, lead is a nonessential, ubiquitous environmental contaminant and belongs to the group of most toxic heavy metals in the biosphere. It produces cumulative toxic effects if taken in small doses and acute toxicity in higher doses (Sastry and Gupta, 1978). Lead enters into water bodies from industries and smelter discharges or dissolution of old lead plumbin (Moore and Rammamoorthy, 1984; Gupta and Salunke, 1985; De, 1996; Satake *et al*., 1997). Lead is considered as a non-specific poison affecting physiological systems and can cause brain damage, kidney damage, gastrointestinal distress and reproductive disorders (Campana *et al*., 2003; Kutlu and Summer, 1998).

Toxic effect of lead and other heavy metals on opercular beat and oxygen consumption has been mostly investigated in fishes (Hiltbran, 1971; Singh and Singh, 1979; Rao and Ramamurthi, 1987; Gill *et al*., 1988) but crustaceans despite being important member of food chain and having high economic and medicinal value are being documented less in reference to metal toxicity and oxygen consumption (Chinnaya, 1971; Ghate and Mulhelker, 1979; Papathanasiou and King, 1983; Tulasi and Rao, 1989; Reddy and Venugopal, 1993; Chinni *et al*., 2000; Jadhav and Ambore, 2007; Sen *et al*., 2008).

Considering the above facts, present work has been taken into account to evaluate toxic effects of lead nitrate on oxygen consumption of freshwater prawn, *Macrobrachium dayanum* (Crustacea-Decapoda), a potential animal for fresh water aquaculture.

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Materials and Method

Fresh water prawns, *Macrobrachium dayanum* (Henderson) (Sharma *et al.*, 1997) were collected from river Gomti, Lucknow (U.P.), with the help of local fisherman and brought to the laboratory (N- 26° 5' 59" E- 80° 56' 17") in large plastic containers. The animals were maintained in glass aquaria of 20 liter capacity containing 10 liter dechlorinated water having physico-chemical characteristics as:- pH– 7.66 ± 2.67, Temperature- 27.66 ^{0}C ± 0.66, DO- 6.6 mg/l ± 0.74, Total alkalinity- 425.00 mg/l ±11.36, Total hardness- 268.00 mg/l ± 2.67 (Sharma and Shukla, 1990; APHA, 1998). Proper aeration was provided with the help of aerators and air diffusers.

Stock solution of lead (II) nitrate [$Pb(NO_3)_2$, M.W.- 331.21 gm/mole, A.R. Grade, manufactured by E - Merck (India) Ltd. Worli- Mumbai] was prepared by dissolving weighed amount of salt in double distilled water. Lead nitrate was dissolved in water by adding, 0.30 ml/l of concentrated nitric acid.

Adult inter-moult staged *M. dayanum* (Average length- 5.64 cm ± 0.42, weight- 3.261 gm ± 0.68) were used in experiments after 5-7 days acclimation to laboratory conditions. Acute exposure was carried out on 96 hr LC_{50} value (116.46 mg/l) for 24, 48, 72 and 96 hr while sub-acute exposure was carried out on 25% of 96 hr LC_{50} value (29.12 mg/l) for 10, 20 and 30 day respectively. One aquarium containing diluent water and 0.3 ml/l concentrated nitric acid only, served as control for each set. Feeding was suspended 24 hr before during acute exposure and through out experiment while change of exposure medium and food supply was maintained on alternate day during sub-acute exposure. Continuous air supply was provided by air diffusers and aerators in both control as well as experimental aquaria in both the experiments. Experiments were carried out under natural light and dark period. Both acute and sub-acute experiments were carried out according to guideline of APHA (1998). For oxygen consumption, prawns were kept in fully air tight and completely water filled containers of 5 liters capacity provided with stoppered outlets for collecting water samples. The respiratory rate was calculated hourly by monitoring of the falls in the concentration of dissolved oxygen after 24, 48, 72 and 96 hr in acute and 10, 20 and 30 days in sub-acute exposure respectively. For each control as well as exposed animals three replicate were maintained and dissolved oxygen (DO) was determined by using modified Winkler's method as per APHA (1998). The prawns were carefully weighed before being released into the control and toxic media. The oxygen consumption rate was determined as oxygen consumed (mg/l)/gm body wt/ hr. Experiment was replicated thrice and data were subjected to statistical analysis for student "t" test and ANOVA using MINITAB software on PC.

Results and Discussion

The rate of oxygen consumption of freshwater prawn, *M. dayanum* was studied after the acute and sub-acute exposure of lead nitrate. The results of acute exposure are summarized in Table-1 and Fig. 1. The prawns of exposed animals exhibited marked

Table- 1: Effect of acute exposure of Lead nitrate on oxygen consumption of *M. dayanum*

Exposure duration (hr)	Oxygen consumption (mg/l)/gm body weight/hr (Mean±S.E)	
	Controlled	Exposed
24	0.3684±0.0142	0.4686±0.0135 *
48	0.3825±0.0081	0.4275±0.0082*
72	0.3634±0.0079	0.3339±0.0078^{NS}
96	0.3358±0.0078	0.2856±0.0078*

Note: *Denotes difference in means to be significant at P<0.05
NS Denotes difference in means to be insignificant at P<0.05

alterations in oxygen consumption from 24 to 96 hr than control ones. Exposed animals showed initial increase in oxygen consumption than control animals but declining trend in oxygen consumption was observed through out the experiment in exposed animals. A significant ($t = 5.13$; $P < 0.05$) increase in oxygen consumption of exposed animals (0.4686 ± 0.0135) was noticed after 24 hr of exposure than control ones (0.3684 ± 0.0142); thereafter a declining trend in oxygen consumption was observed in experimental

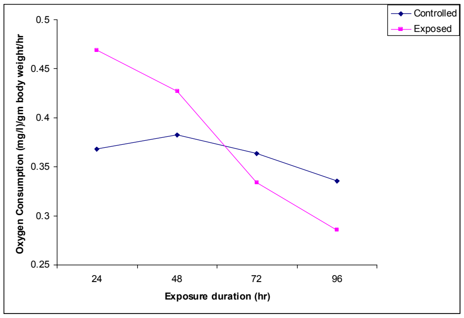

Fig. 1: Effect of acute exposure of Lead nitrate on oxygen consumption of *M. dayanum*

animals (0.4275 ± 0.0082) but it remained higher than control animals (0.3825 ± 0.0081) after 48hr exposure; the declining trend in oxygen consumption was continued in exposed animals (0.3339 ± 0.0078) and observed below than control animals (0.3634 ± 0.0079) after 72 hr exposure. Though the difference between control and exposed animals was found insignificant ($t = 2.66$; $P > 0.05$). Finally a significant ($t = 4.54$; $P < 0.05$) decrease in oxygen consumption was observed in experimental animals (0.2856 ± 0.0078) than controls (0.3358 ± 0.0078) after 96 hr exposure. The overall variations from 24 to 96 hr were found moderately significant in exposed animals ($F = 76.11$; $P < 0.001$) while insignificant in controls ($F = 3.91$; $P > 0.05$).

In sub-acute exposure experimental animals showed marked reduction in oxygen consumption through out the experiment from 10 to 30 days when compared with controls. The results of sub-acute exposure are summarized in Table-2 and Fig. 2. A significant ($t = 3.52$; $P<0.05$) decrease in oxygen consumption were observed in exposed animals (0.2943 ± 0.0077) than control (00.3489± 0.0135) after 10 day exposure. Thereafter a moderately significant ($t = 6.79$; $P < 0.001$) decrease in oxygen consumption was observed in experimental animals (0.2544 ± 0.0133) than control animals (0.3593± 0.0078) after 20 day exposure. Finally after 30 day exposure highly significant ($t = 13.89$; $P < 0.0001$) decrease in oxygen consumption was observed in exposed animals (0.2055 ± 0.0073) than control animals (0.3512 ± 0.0075). The overall variations from 10 to 30 days were found moderately significant in exposed animals ($F = 20.33$; $P < 0.001$) while insignificant in controls ($F = 0.30$; $P > 0.05$).

Table-2: Effect of sub-acute exposure of Lead nitrate on oxygen consumption of *M. dayanum*

Exposure duration (Day)	Oxygen consumption (mg/l)/gm./ body weight/hr (Mean±SE)	
	Control	Exposed
10	0.3489 ± 0.0135	0.2943 ± 0.0077*
20	0.3593 ± 0.0078	0.2544 ± 0.0133**
30	0.3512 ± 0.0075	0.2055 ± 0.0073***

Note: Values are Mean±SE; N=5
***Denotes difference in means to be highly significant at P<0.0001
**Denotes difference in means to be significant at P<0.001
*Denotes difference in means to be significant at P<0.05

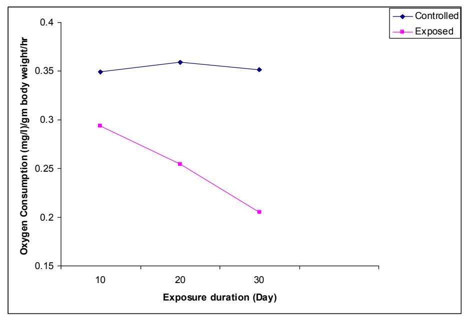

Fig. 2: Effect of sub-acute exposure of Lead nitrate on oxygen consumption of *M. dayanum*

The study revealed that lead caused continuous decrease in oxygen consumption of freshwater prawn, *M. dayanum* after sub-acute exposure while initial increase then continuous decreasing trend during acute exposure.

In crustaceans gills are the main respiratory structure and sites of gaseous exchange, comes in direct contact of surrounding environment hence are more susceptible to damage caused by various toxicants. Toxicants, particularly heavy metals in surrounding medium adversely affect the gills resulting in hypoxia and respiratory failure (Alazemi *et al.,* 1996; Jadhav and Ambore, 2007). Changes in oxygen consumption rate are a good index to measure altered metabolic activity in organism exposed to various toxicants in surrounding medium. Continuous decrease in oxygen consumption, as observed in present study has also been reported in various crustaceans (Chinnaya, 1971; Ghate and Mulhelker, 1979; Tulasi and Rao, 1989; Papathanasiou and King, 1983; Reddy and Venugopal, 1993; Chinni *et al*., 2000; Jadhav and Ambore, 2007; Sen *et al*., 2008). Similar effects have also been reported in fishes (Gill *et al*., 1988; Hiltbran, 1971; Singh and Singh, 1979; Rao and Ramamurthi, 1987). Tasi (1979) reported exposure of metals precipitates gill secretion. Reduced oxygen consumption may be due to precipitated mucous coating on gill surface resulting in asphyxiation. Precipitated gill secretion inhibits the consumption of oxygen by the gill tissues and disrupts osmoregulation as reported in crabs and isopods (Thurberg *et al*., 1973; Jones, 1975; Carrea, 1987). The physiological, histological and ultra-structural studies have shown that metal ion interferes in respiration by disrupting the structure of gill cells by direct cytotoxic effects (Jones, 1975; Gill *et al*., 1988). Cytological damage in crab and shrimp species after heavy metal exposure which results in thickening of bronchial epithelium and profound changes in haemolymph pattern in the gill with concomitant increase in vacuolization and reduced haemolymph spaces causing perfusion stagnation (Spice and Weber, 1991). Hughes and Dutta Munshi (1979) reported a relationship exists between gill area and oxygen consumption. Mitochondrial damage and reduction in ability to synthesize the ATP in gills as reported in *Palaemon serratus* (Papathanasiou and King, 1983) after exposure of cadmium may also be a reason for reduced oxygen consumption in *M. dayanum* after lead exposure. Almost similar findings have been reported by Tulasi and Rao (1989) on oxygen equilibrium curve of the freshwater field crab, *Barytelphusa guerini* after organic and inorganic lead exposure. Recently, Sen *et al.* (2008) reported almost similar findings in *M. dayanum* after cadmium chloride exposure. It is evident from present findings that metallic pollutants, particularly lead exert adverse affects on oxygen consumption of freshwater prawn, *M. dayanum*. Oxygen consumption rate can be used as monitoring tools to asses worsening status of aquatic bodies in reference to metallic pollution, which is of global concern now days.

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