Effect Of Sublethal Chromium On Biochemical Constituents And Bio Accumulation In The Post Larvae Of *Penaeus Monodon*.

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Abstract:
Heavy metals occur naturally in the ecosystem with large variations in concentration. Living organisms require varying amounts of some heavy metals. Small amounts of these elements are common in our environment and diet and are actually necessary for good health, but large amounts of any one of them may cause acute or chronic toxicity. For some heavy metals, toxic levels the background concentrations naturally found in nature. Therefore, it is important to elucidate their impact on organisms and take protective measures against excessive exposure. As the coastal waters are frequently used by aqua farms coastal districts, it is proposed to undertake the present investigation on postlarvae of *Penaeus monodon* exposed to chromium. The objective of this study was to determine the effect of sublethal chromium concentrations on biochemical constituents and metal accumulation when subjected to sublethal chromium i.e. 1/5 th of LC_{50}[1.2056ppm]. The present investigation also reveals the marginal levels of safety for chromium i.e., 60.28 g/L using the PL of *P. monodon* and this is very much essential for future monitoring studies of metal contamination.

Keywords: Chromium, biochemical constituents, bio-accumulation, post larvae, *Penaeus monodon.*

Introduction:
The ocean, being so vast is constantly being used as a dumping ground which can neutralize some of these chemical wastes. Estuaries, marine and other aquatic bodies serve as an important habitat for many organisms including crustaceans, fish, shellfish, various reptiles and birds. Some of these marine organisms absorb the toxins, and these toxins become more concentrated in animals higher in the food chain. This adversely affects the animals at the top of the food chain and may cause a decline in their populations. Other toxins may kill the organisms that come in contact with them. Heavy metals occur naturally in the ecosystem with large variations in concentration. Living organisms require varying amounts of some heavy metals. Small amounts of these elements are common in our environment and diet, and are actually necessary for good health, but large amounts of any one of them may cause acute or chronic toxicity. For some heavy metals, toxic levels can be just above the background concentrations naturally found in nature. Therefore, it is important to elucidate their impact on aquatic organisms and take protective measures against excessive exposure.

The coastal waters are constantly being affected by these industrial and domestic effluents. Most of these effluents contain several toxic materials and among all of them, the heavy metals are considered to be more important. Chromium is one of the heavy metals reported in the industrial effluents. The major water polluting industries are tanneries or leather industries, these industries are responsible for release of very toxic metals like Chromium, Lead and Arsenic. Pulp and Paper industries, the bleaching agent used in these industries as well as other chemicals result in release of chemicals like Chromium.

Materials and methods:
Postlarvae (PL) of *P. monodon* were brought from a local hatchery. The PL were transported to the laboratory in plastic bags as explained earlier. These PL were acclimatized to laboratory conditions for 48hrs in plastic troughs containing seawater. The salinity of the seawater was maintained at 10ppt throughout the experiment and sufficient aeration was provided using aerator. The pH and temperature were also kept constant at 8 and 29 ± 1°C respectively. The larvae were maintained in the troughs without any crowding.

Uniform sized PL (0.9 – 1.0cm) were chosen for the experiment and the PL were fed with commercial diet (Highashi 3000 started B, Highashimanu Co. Ltd., Japan) twice a day. Excess feed and excretory wastes were removed every day by siphoning. Plastic troughs of 20L capacity were used and 15L of 10ppt seawater was taken in each trough. During the experimentation, the PL were exposed to 1.2056ppm of chromium which represents 1/5th of 96hr LC_{50} value, for a period of 30 days. A control was maintained simultaneously without the metal toxicant. The toxic solution was prepared by dissolving potassium chromate (AR) in distilled water (1% stock solution). Appropriate amounts of stock solution were added to 15L of seawater to get the final desired concentration. Samples were taken from both the control and exposed at

intervals of 24hrs, 48hrs, 96hrs, 10days, 20days and 30days. At the end of each interval, 30 PL were collected both from the control and exposed tanks for the estimation of biochemical constituents viz. carbohydrates, proteins and lipids by following the methods detailed below:

Biochemical and bioaccumulation analysis:
At each of the above six different intervals, 30 PL were isolated from both control and exposed tanks separately and were blot dried to remove adhered water. These PL were then dried in an oven at 60ºC for 48hrs, powdered using mortar and pestle and preserved in glass vials, in a desiccator until further experimentation. The preserved dry tissue powder of both the control and exposed PL was used for estimating the biochemical constituents, such as total lipids total proteins and total carbohydrates and metal accumulation.

Estimation of Total Carbohydrates:
Total carbohydrates were estimated by following the Anthrone method [5]. A known amount of dry tissue powder was homogenized in 5ml of 10% trichloroacetic acid (TCA) and centrifuged at 2500rpm for 15mins. The supernatant was used as the sample source. 1.0ml of sample source was taken in a test tube and 5ml of Anthrone reagent was added and thoroughly vortexed. These test tubes were incubated in boiling water bath for 15mins and cooled to room temperature under running tap water. The colour was read in a spectrophotometer at 620nm against the blank. The amount of total carbohydrate was calculated against glucose as standard and they were presented as µg/mg dry weight of tissue powder.

Estimation of Total Proteins:
Total proteins were estimated by Lowry's method [22]. A known amount of dry tissue powder was homogenized in 5ml of 1N NaOH and centrifuged at 2500rpm for 15mins. The supernatant was used as the sample source. 0.1ml of sample source was taken in a test tube and was made up to 1ml using distilled water. To this 4ml of Lowry C was added, thoroughly vortexed and incubated at room temperature for 20mins. Then, 0.5ml of Folin-phenol reagent (1:1, Folin-Phenol: distilled water) was added to the test tubes, vortexed thoroughly and incubated at room temperature for 30mins. The developed colour was measured at 700nm in a spectrophotometer (Chemito, 2000). The values were calculated against Bovine Serum Albumin (BSA) standard and they were presented as µg/mg dry weight of tissue powder.

Estimation of Total Lipids:
Total lipids were estimated by following the sulpho-phosphovanillin method [2]. A known amount of dry tissue powder was homogenized in 5ml of chloroform: methanol (2:1) solvent and centrifuged at 2500rpm for 15mins. The supernatant was used as the sample source. 0.5ml of sample source was taken in a test tube and was made up to 1ml using the solvent. The solvent was made to evaporate in an oven for 20mins. To the residue, 0.5ml of concentrated sulphuric acid was added and the mixture was stirred well and kept in boiling water bath for five minutes. After cooling to the room temperature, 2.5ml of the phosphoric acid-vanillin reagent was added, thoroughly vortexed and the test tubes were stoppered with the cotton wool. They were then incubated for 30mins at room temperature. The optical density was measured at 520nm in a spectrophotometer (Chemito, 2000). The values were calculated against cholesterol standard and they were presented as µg/mg dry weight of tissue powder.

Estimation of Total energy levels:
Total energy levels were estimated by following the conversion formula enunciated by [12], taking in account the total amounts of carbohydrates, proteins and lipid. The conversion factors used were 4.1, 4.3 and 9.5cal/mg for carbohydrates, proteins and lipids respectively.

Metal accumulation:
Metal analysis was then carried out with the dried tissue samples of both the control and exposed PL for all the sixtime intervals following dry-ash method [47]. A known quantity of dried tissue powder was dry-ashed in a muffle furnace at 800ºC for 6hrs. The dry ash obtained from the above process was dissolved in a known quantity of 2N HNO_3. The final clear and colourless solution was then used for chromium analysis using graphite furnace atomic absorption spectrophotometer (Model: Perkin Elmer No. 3110). Each sample was analysed in triplicate.

4.2.3 Statistics:
The above experiment was repeated five times. Total carbohydrates, proteins, lipids (n=15) as well as total energy levels (n=15) and metal accumulation were then calculated for all the exposure periods. The mean values and standard deviations (S.D) were calculated for each interval using standard methods [36]. The exposed values were compared with their respective controls by using Student's t-test [36] and significant differences were calculated at $P < 0.05$.

Results:
The results on variations in biochemical constituents such as total carbohydrates, proteins and lipids in *Penaeus monodon* PL on exposure to sublethal chromium along with energy levels are presented in Figures 1.1 to 1.4.
The data presented in Figure 1.1 show variations in total carbohydrates between control and exposed PL between control and exposed PL at different exposure periods for sublethal chromium. A significant decrease was noticed in total

carbohydrates of the exposed PL from 96hrs onwards compared to their respective controls. However, the percentage decrease was14.32, 11.23, 27.04, 34.66, 38.03 and 49.12 for 24hrs, 48hrs, 96hrs, 10days, 20days and 30days respectively. A significant ($P < 0.05$) maximum (49.12%) and minimum (11.23%) decrease in total carbohydrates was observed in the exposed PL with respect to the controls at 30days and 96hrs respectively.

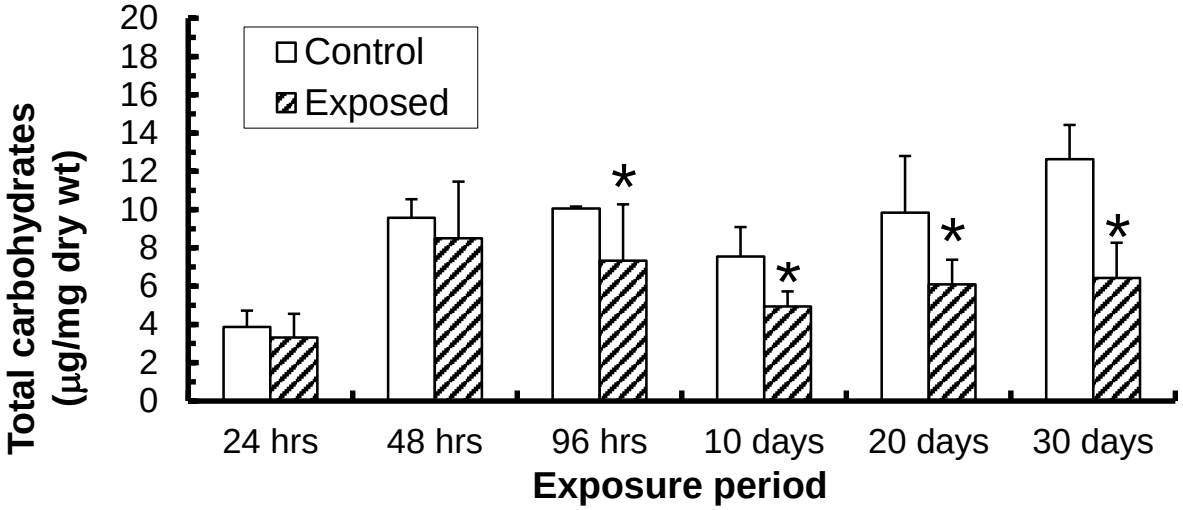

Fig. 1.1 Total carbohydrates in *P. monodon* PL exposed to sublethal chromium
Vertical lines represent standard deviation. *Significantly different from their respective controls at $P < 0.05$

The total protein content of the control and exposed PL of *Penaeus monodon* at different intervals from 24hrs to 30days is presented in Figure 1.2. The results clearly indicate a significant decrease in the total protein content of the exposed PL with reference to its control from 10days till 30days of exposure. The control PL showed a gradual increase in the total protein content from 468.5µg/mg dry weight at 24hrs to 515.0µg/mg dry weight after 30days. Though the control PL showed an increase in total protein content, it was significantly low in exposed PL particularly from 10days onwards. The percent decrease in total proteins of the exposed PL observed at different exposure periods were 0.28, 7.38, 4.82, 8.01, 14.78 and 15.39 for 24hrs, 48hrs, 96hrs, 10days, 20days and 30days respectively. A significant ($P<0.05$) maximum (15.39%) and minimum (8.01%) decrease in total proteins was observed in the exposed PL with respect to the controls on 30days and 96hrs respectively.

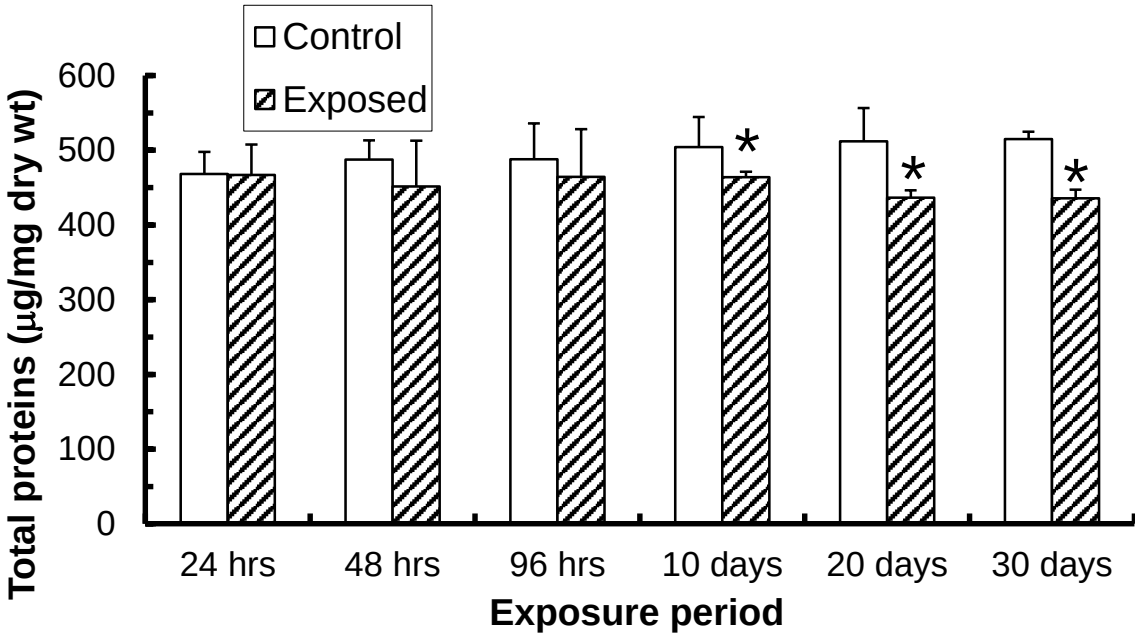

Fig. 1.2 Total protein in *P. monodon* PL exposed to sublethal chromium.
Vertical lines represent standard deviation. *Significantly different from their respective controls at $P < 0.05$

Figure 1.3 show the total lipid content of control and exposed *Penaeus monodon* PL.
The total lipid content in the exposed PL when compared to its respective controls showed a decreasing trend with increasing time from 5.20% (24hrs) to 19.88% (30days). However, a significant (P<0.05) decrease was noticed at 20 and 30 days. The percent decrease in total lipids of the exposed PL observed at different exposure periods was 5.20, 4.35, 2.92, 8.39, 9.91 and 19.88 for 24hrs, 48hrs, 96hrs, 10days, 20days and 30days respectively.

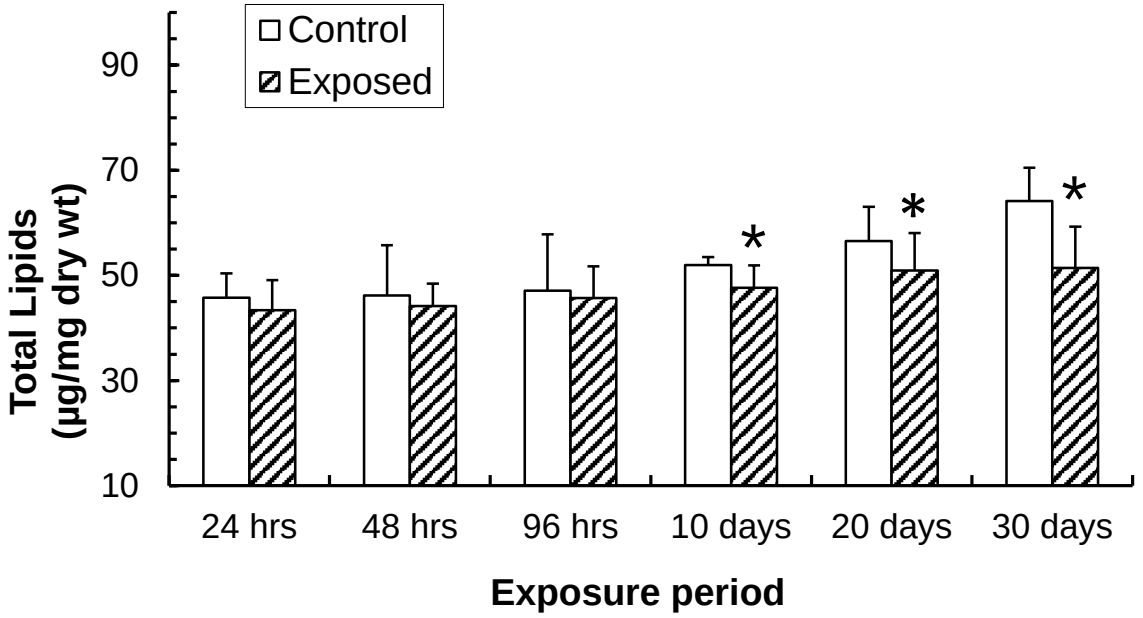

Fig. 1.3 Total lipids in *P. monodon* PL exposed to sublethal chromium.
Vertical lines represent standard deviation. *Significantly different from their respective controls at P < 0.05

The total energy levels of control and exposed PL were presented in Figure 1.4. There was an increase in the total energy level of the control PL from 2347.43cal/mg dry weight at 24hrs to 2874.6cal/mg dry weight after 30days. However, a significant (P<.05) decrease in the energy levels of the exposed PL over their respective controls was noticed from 10days onwards. The percent decrease in total energy levels of the exposed PL observed at different exposure periods was 3.07, 8.37, 5.46, 9.02, 10.40 and 17.36 for 24hrs, 48hrs, 96hrs, 10days, 20days and 30days respectively.

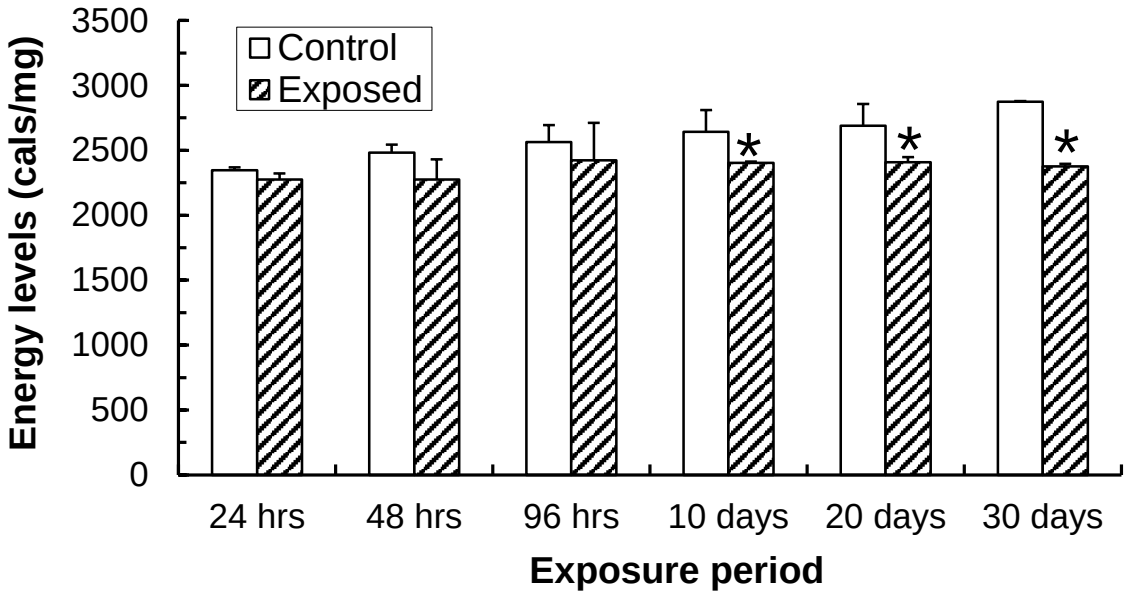

Fig. 1.4 Total energy levels in *P. monodon* PL exposed to sublethal chromium
Vertical lines represent standard deviation. *Significantly different from their respective controls at P < 0.05

A gradual and time-dependent increase in the metal accumulation was noticed in the exposed PL ranging from 80.44% to 194.68%. The percent increase in the metal content of the exposed PL compared to their respective controls for different time intervals was 80.44% for 24hrs,194.68% for 48hrs, 32.60% for 96hrs, 167.80% for 10days and 145.86% for 20days and 100.32% for 30days of sublethal chromium exposure. The increase in the metal content in the exposed PL was from 27.066µg/gm dry weight at 24hrs, 47.150µg/gm dry weight at 48hrs, 33.350µg/gm dry weight at 96hrs, 43.250µg/gm dry weight at 10days, 43.150µg/gm dry weight at 20days and 40.667µg/gm dry weight at 30days of exposure. However, the metal content in the control PL was almost the same for all intervals i.e. 15.0, 16.0, 25.15, 16.15, 17.55 and 20.30µg/gm dry weight for 24hrs, 48hrs, 96hrs, 10days, 20days and 30days respectively.
The results obtained from the studies on the biochemical constituents such as total proteins, total carbohydrates, total lipids and total energy levels indicate that all the values of above biochemical constituents in the soft tissues of *Penaeus monodon* PL were found to decrease at different intervals. The depletion was more in total carbohydrates, followed by total lipids and total proteins. Maximum depletion of total carbohydrates, total lipids and total proteins was observed at 30days of sublethal chromium exposure. Similarly, the total energy levels also showed a maximum decrease from 24hrs to 30days exposure period to sublethal chromium concentration. However, a significant decrease in all these values was observed from 10days of sublethal chromium exposure.

The results of metal accumulation in the PL of *Penaeus monodon* exposed to sublethal chromium at six different time-intervals are presented in Figure 2.1. The data clearly showed a significant ($P<0.05$) accumulation of chromium in the exposed PL compared to their respective controls from 24hrs onwards till 30days of exposure. However, the metal content in the control PL for all the six exposure periods remained almost the same.

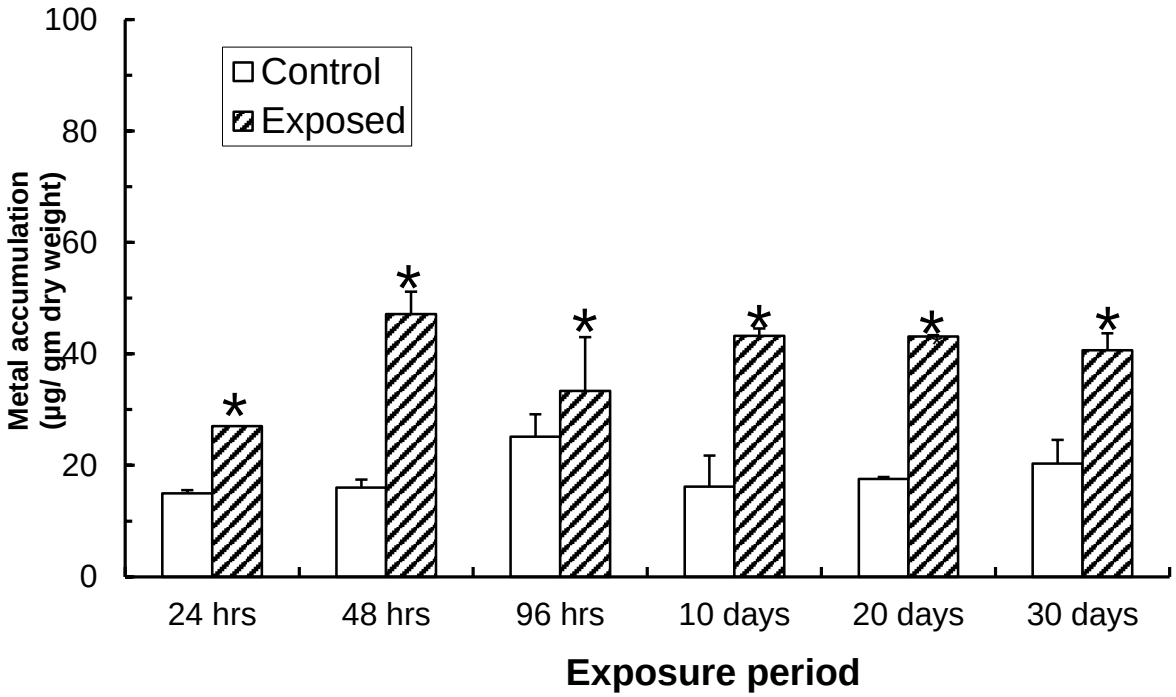

Fig. 2.1 Metal accumulation in *P. monodon PL* exposed to sublethal chromium.
Vertical lines represent standard deviation. *Significantly different from their respective controls at $P < 0.05$

DISCUSSION

Overall decrease in the total biochemical constituents as well as energy levels in the exposed PL when compared with their respective controls indicate the effect of sublethal chromium (1.2056ppm) on chronic exposure in *Penaeus monodon* PL. The results of the present study showed a maximum decrease on 30days of exposure in total carbohydrates (49.12%) followed by total lipids (19.87%) and total proteins (15.38%) in the exposed PL of *Penaeus monodon* with reference to their controls. The total energy levels also decreased significantly ($P < 0.05$) in the exposed PL from 9.02% at 10days to 17.36% after 30days of exposure when compared with the controls.

A significant decrease in the total carbohydrates of the exposed PL from 96hrs onwards corroborated well with similar reports on metal-induced variations in carbohydrate levels of other crustaceans. [37] reported that sublethal long-term (30, 60 and 90days) exposure of chromium on carbohydrate metabolism resulted in depletion of glycogen and glucose levels and induced alteration in metabolite levels and enzyme activities in the tissues of freshwater field crab, *Barytelphusa guerini*. While conducting experiments on carbohydrate metabolism in the tissues of marine prawn, *Penaeus indius* exposed to phosphamidon, a reduction in carbohydrate content in the midgut gland of *Penaeus indicus* was reported by [15]. An exposure of cadmium chloride to crayfish *Procambarus clarkii* resulted in decrease in the amylase activity in the

digestive juice as reported by [30]. While reporting the effects of synthetic detergent sodium lauryl sulphate on *Macrobrachium rosenbergii,* [18] reported that the level of glycogen decreased significantly in the hepatopancreas and a maximum decrease in protein concentration was noticed in the hepatopancreas followed by gills and muscles.

The present experiment also exhibited a decrease in total proteins of the exposed PL when compared to its control. Similar results were obtained by several investigators in other aquatic animals. The impact of endosulfan on protein metabolism of the freshwater crab *Oziotelphusa senex senex* was studied by [40]. The *in vitro* addition of organochlorine insecticide, Lindane was reported to cause significant changes in protein and amino-transferase system of *Penaeus indicus* [31]. [44] reported that lead altered the haemolymph metabolite levels in *Penaeus monodon*. Tannery effluents were also reported to significantly reduce the hemolymph protein content of the freshwater prawn *Macrobrachium idella* [39]. However, [45] reported an increase in the concentration of protein after 48hrs exposure to copper in an estuarine teleost and suggested that the increased protein might be due to the transport and storage of these metal ions as an effort to detoxify the metal toxicant.[21] observed that *P. monodon* following 24hrs exposure to ambient copper as low as 4.76mg/L, shows a reduction of oxyhemocyanin, protein, and develop metabolic acidosis in the hemolymph. [3] reported endosulfan toxicity in juveniles of *Macrobrachium malcolmsonii* and observed the breakdown of various polypeptides in tissues of test prawn, which resulted in protein denaturation. [10] observed reductions in protein and DNA of shrimp, *Litopenaeus stylirostris* larvae exposed to pesticides DDT, azinphosmethyl, permethrine, parathion, chlorpyrifos, malathion, endosulfan, and carbaryl, and reported that reduction in protein indicates a decrease in larvae growth rate.

Finally a decrease in total lipids along with other biochemical constituents upon chromium exposure in the exposed PL corroborated well with the other reports. [42] observed a decrease in the lipid content of hepatopancreas, glycogen of the muscles and protein content of gills hepatopancreas and the muscles of red crayfish, *Procambarus clarkii* over the 96hr period exposure to 0.25mg/hg/L mercury. Glycogen/lipid and glycogen/protein ratios increased after 48 and 96hr of mercury exposure. [43] studied the effect of sublethal exposure of mercury on the freshwater crayfish, *Procambarus clarkii* and noticed a decrease in the levels of total proteins, total lipids and glycogen concentrations in hepatopancreas. Decrease in total proteins, lipids and carbohydrates were observed by [35] in *Penaeus indicus* post larvae exposed to sublethal lead. [11] reported that *M. malcolmsonii* exposed to dichlorvos exhibited a major decline in biochemical constituents, such as glycogen, protein and lipid indicating the operation of mechanism to overcome the required energy demand due to dichlorvos toxicity. At sublethal concentrations of cadmium, [11] observed that low salinities caused increase in levels of protein concentration whereas the amount of lipids decreases in estuarine hermit crab, *Clibanarius infraspinatus*. [4] noticed the utilization of major biochemical constituents, such as total carbohydrate, glycogen, protein and lipid to generate required energy as an attempt to withstand the carbaryl toxicity in *Macrobrachium malcolmsonii* and further suggested that the carbaryl toxicity resulted in severe energy crisis in the juvenile stages of prawns.

Based on the above mentioned reports on the effect of metals and other pollutants on the biochemical constituents in crustaceans, it can be concluded that the decrease in total carbohydrates, proteins and lipids on chromium exposure in the exposed PL in the present study might be attributed to the effect of metal on oxidative metabolism. Since maximum decrease was noticed in total carbohydrates, it can be concluded that utilization of carbohydrates was more than other biochemical constituents to generate required energy as an attempt to withstand chromium toxicity, similar to carbaryl toxicity as suggested by [4]. The role of lipids in generating the required energy demand in the exposed PL undergoing stress can be supported by the fact that a maximum of 19.87% decrease in the total lipids was noticed in the exposed PL compared to its control. Though proteins (15.3%) were also utilized to combat the energy crisis by the exposed PL, its utilization was much less compared to carbohydrates and lipids. Cu, like Zn probably induced synthesis of specific metal-binding proteins (metallothionin) causing an increase in the synthesis of proteins. Lower levels of percent decrease in total protein content (8.01% at 10days to 15.38% at 30days) of the exposed PL compared to its control can thus be attributed to the fact that, heavy metals unlike other metals might be involved in synthesis of metallothionin proteins as reported by[26], [45] and [19].

[24] reported that both copper and cadmium resulted in overall metabolic depression, decreasing energy allocation to both maintenance and production in adult grass shrimp *Palaemonetes pugio* upon exposure to either aqueous copper (ranging from 7.54 to 41.29µg Cu^{2+}/L) or cadmium (2.48- 6.55µg Cd^{2+}/L) for 14days. In the present experiment, the total energy levels in the control PL was 4314.28cal/mg dry weight whereas the exposed PL showed 3097.36 cal/mg dry weight after 30days of experimentation. Therefore, it can be concluded that chromium toxicity, even at sublethal concentration of 164.1µg/L can significantly induce energy crisis in the PL of *Penaeus monodon* from 10days exposure onwards indicated by gradual decrease in energy levels from 9.29% to 17.37% over 30days of chromium exposure. This demand was met, by maximum utilization of carbohydrates followed by lipids and proteins in the exposed PL. Finally, the decrease in the biochemical constituents of *Penaeus monodon* PL upon chromium exposure might be either due to increased utilization or reduced synthesis of biochemical constituents because of inhibition of different metabolic pathways by the metal.

The present investigation revealed a gradual accumulation of chromium in the tissues of the exposed PL with increasing exposure period. However, the metal content in the control PL almost remained same at all the time intervals. During ecdysis, shrimps intake of water increases owing to increase in their size, resulting in hydration of their tissues and

favouring metal accumulation in PL [1]. Further, the dissolved metals are considered more toxic since they are more easily absorbed by aquatic organisms than the particulate fraction [9]. This may be the probable reason for metal accumulation in the tissues of the PL where ecdysis occurs frequently.

In crustacean species, it is also thought that there is a relationship between metal permeability, regulation and accumulation. If the metal uptake rate in organisms is higher than the excretion rate, then metal accumulation occurs. An ideal bioindicator should be a net accumulator of the trace metal [14]. The effect of body weight on trace metal concentrations of Cd, Cu, Pb, Ni and Cr in the muscle and gill of marine crustaceans of the Bohai Sea was studied by [46] and they observed that the muscle of the smaller individual accumulated more metal per gram wet weight than that of the larger ones.

While studying the nitrite accumulation in *Penaeus monodon*, [6] reported that the concentration of nitrite in the tissue increased directly with ambient nitrite and exposure time (except for muscle) following 48hr exposure to 0.36ppm nitrite. Nitrite concentration progressively increased from the muscle (0.40µmol/g), hepatopancreas (1.24µmol/g), gill (1.82µmol/g), foregut (2.03µmol/g), hemolymph (0.39µmol/ml), heart (2.43µmol/g), eyestalk (3.07µmol/g), and to the midgut (4.14µmol/g), which is 1.1, 3.4, 5.0, 5.6, 6.6, 6.8, 8.5 and 11.4 times the ambient nitrite concentration, respectively. [13] reported the metal (Cd, Zn, Pb, Fe) accumulation in the shrimp *Penaeus monodon* muscles. Concentration of the metals in shrimp muscle ranged as follows: Cd 0.11 – 3.2; Zn 7.3 – 4809.5; Pb 22.9 – 42.1; Fe 5.0 – 495.0µg/gm. Except for lead, the metals in shrimp muscle varied significantly among the localities. [7] reported that the organ-specific increase in the nitrate concentration of *Penaeus monodon* was progressive from muscle, hepatopancreas, heart, foregut, gill, haemolymph and eyestalk to the midgut which is 0.057, 0.092, 0.148, 0.177, 0.260, 0.270, 0.340, and 0.428 times more than the ambient nitrate concentration. [33] observed a differential tissue cadmium accumulation in the snow crab *Chionoecetes opilio* in the order hepatopancreas > gut > antennal glands > hemaolymph > gonads > gills > muscle > eye. [25] noticed that the accumulation of lead in the postlarvae of *Penaeus monodon* reached a stable state in 10days with an accumulation factor of 139. [8] observed a time-dependent increase in the metal concentration of *Peneaus indicus* postlarvae exposed to sublethal lead. While investigating trace metals from Antarctic copepods, [17] reported high cadmium concentrations (10µg Cd/gm) in the *Metridia* species whereas zinc concentration in *M. gerlachei* and *R. gigas* were much higher i.e., 518µg Zn/mg and 43µg Zn/gm whereas cobalt and lead concentrations were much lower (<0.1µg Co/gm and <1µg Pb/gm). [41] studied the percolation of heavy metals into cellular level through the cell membrane and interact with cellular macromolecules to inhibit the essential cellular metabolism in *Penaeus monodon* and *Metapenaeus moyebi* from Ennore brackish water ecosystem. [28] and [29] reported cadmium uptake and accumulation in the juveniles of *Penaeus indicus* over the exposure concentration range of 1.8-31.5µg/L, the hepatopancrease and gills showing the highest concentration of cadmium. [29] reported cadmium, zinc and calcium uptake by two crabs, *Carcinus maenus* and *Eriocher sinensis*. A significant ($P<0.05$) time-dependent and concentration-dependent increase in copper accumulation in giant freshwater prawns, *Macrobrachium rosebergii* was reported by [20] in juveniles, and [32] in juveniles and postlarvae, and [38] in postlarvae of *M. rosenbergii*. Cadmium accumulation in the postlarvae of *Penaeus monodon* on exposure to sublethal cadmium for a period of 30days was reported by [27]. [16] reported the suitability of brown shrimp *Crangon crangon* as a bioindicator of trace metals cadmium and lead from German Wadden Sea. [23] reported zinc accumulation with increase in exposure time in *Penaeus monodon* postlarvae. [34] observed a gradual increase in accumulation of copper in postlarvae of *Penaeus indicus.*

Thus, the results of our investigation are well corroborated with the above literature on crustaceans. Our present findings therefore, suggest the possible use of this species as a good indicator of chromium pollution.

Conclusion:

In conclusion the PL of *P. monodon* are sensitive to sublethal concentration of chromium indicating a reduction of biochemical constituents with the increase in their accumulation levels on exposure to sublethal chromium and these can be considered as indicators of chromium toxicity. The present investigation also reveals the marginal levels of safety for chromium i.e., 60.28µg/L using the PL of *P. monodon* and this is very much essential for future monitoring studies of metal contamination.

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