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BIOCHEMICAL CHANGES IN HAEMOLYMPH OF FRESH WATER CRAB *PARATELPHUSA JACQUEMONTII* (RATHBUN) EXPOSED TO COPPER.

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Abstract: *Paratelphusa jacquemontii*, a fresh water crab is abundantly found in the Akola district of Indian state Maharashtra. They were brought to the laboratory from their natural habitat, and first subjected to acclimatization to the laboratory conditions. They were subsequently exposed to sublethal concentrations of copper and the haemolymph total protein, free sugar and lipid level of the animal were estimated to study the stress caused by this heavy metal toxicant. In the present study, total protein, free sugar and lipid level of the animal showed a gradual declining trend in experimental animals exposed for 96 hours for 0 to 15 µg/l copper concentrations.

Keywords: Biochemical, Copper, Fresh water crab, Haemolymph

Introduction:

Environmental poisoning of water by heavy metals has increased in recent years due to extensive use of heavy metals in agriculture, and chemical and industrial processes, posing a serious threat to living organisms. Among the heavy metals, copper, chromium, and iron are being major pollutants from industrial effluents and agricultural waste in aquatic environment, cause maximum effects on non-target aquatic organisms resulting in imbalance of the ecosystem. The effect of heavy metals on aquatic organisms is currently attracting widespread attention, particularly in studies related to industrial and agriculture pollution. Today heavy metals in aquatic ecosystems often show levels above the accepted levels. Extensive use of various chemical contaminants is known to adversely affect the growth of various organisms. The toxicity of such chemical constituents in invertebrates influences other physiological, biochemical and metabolic processes.

In biological systems, the biocatalysts play a vital role in the metabolic pathways. Animal exposed to stress conditions alter their physiological status with the help of enzymes. Toxicants like pesticides are known to secrete hyper or hypo level of enzymes. When any aquatic animal is exposed to polluted medium, a sudden stress is developed for which the animals should meet more energy demand to overcome the toxic stress (Mortimer, 2000). In crustacean Copper may acts as a toxicant (Maharajan *et al.,* 2012). Hence, the present investigation was conducted on the fresh water crab, *Paratelphusa jacquemontii* after exposure to copper in various concentrations.

Material and methods:

Fresh water crab, *Paratelphusa jacquemontii* of carapace size ranging from 4-5cm and weight 50-70g were selected for experiment. They were acclimatized in well aerated aquarium for one week. Before introducing into the tank, the fishes were screened for any visible pathological symptoms and were treated with 0.1% of KmNO4. The water quality during experiment was maintained as per APHA (1985).

For sublethal toxicity tests, the crabs were grouped into four batches. Each batch had 10 animals and had 3 replicates.

Group I: Crabs were maintained in normal Fresh water and served as control.

Group II: Crabs were exposed to the sublethal concentration of 5 µg/l

Group III: Crabs were exposed to the sublethal concentration of 10 µg/l

Group IV: Crabs were exposed to the sublethal concentration of 15 µg/l

The media were renewed every alternate day. Crabs were fed daily with artificial feed. Two specimens each from all the groups were sacrificed after 96 hours of the experiment.

For biochemical analysis, the haemolymph was extracted from the thigh

region of the crab with the help of a syringe, and the biochemical constituents (proteins, carbohydrates and lipids) were estimated by following standard procedures. The total protein (TP) and the total carbohydrate (TC) concentrations in haemolymph were determined according to the methods of Lowry *et al.* (1951) and Roe (1955). The total lipid (TL) content was estimated by the method of Barnes and Blackstock (1973).

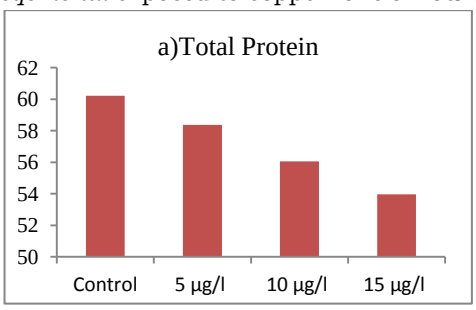
Observations and results:

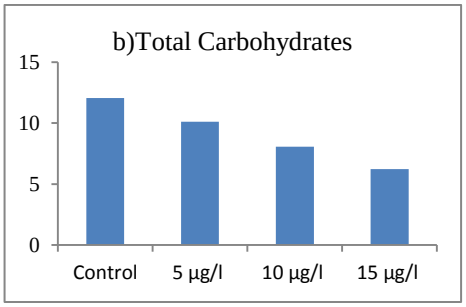
In the present study, the fresh water crab, *Paratelphusa jacqemontii* was exposed to different concentrations of copper for 96 hours. The total protein, free sugar and lipid level of the animal showed a gradual declining trend in experimental animals with increase copper concentrations (Table 1, Figure 1).

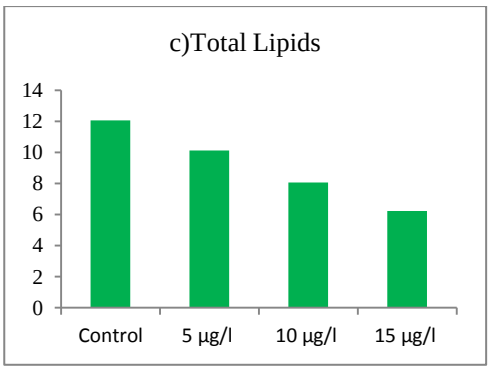
Table-1: Biochemical constituents of *P. jacqemontii* exposed to copper for 96 Hours.

Conce.	Total protein	Total carbohydrates	Total lipids
Control	60.22 ± 0.57	12.05± 0.27	50.19 ±0.95
5 µg/l	58.37 ± 0.48	10.12 ± 0.30	46.71 ± 0.55
10 µg/l	56.06 ± 0. 13	8.06 ± 0.41	40.37 ± 1.15
15 µg/l	53.97 ± 0.39	6.23 ± 0.05	37.94 ± 0.72

Fig.-1:Biochemical constituents of *P. jacqemontii* exposed to copper for 96 Hours.







Discussion:

In recent years, there is a growing concern worldwide on the environmental pollution due to indiscriminate use of pesticides due to their persistence, toxicity at low concentrations and bioaccumulation by biota. Biochemical changes induced by heavy metal stress is due to disturbed metabolism manifested by inhibition of enzymes, retardation of growth and reduction in the fecundity and longevity of the organism. Most of the heavy metals act as metabolic depressors and generally affect the activity of biologically active molecules such as proteins, carbohydrates and lipids (Agrahari and Gopal, 2009). The exposure of aquatic organisms to even very low levels of pesticides causing alterations in the nutritional value of finfish and shellfish as well as their biochemical constituents, physiological and histological functions has been widely documented by Bhavann and Geraldine (2001 and 2002),

Oxygen consumption is one of the most important physiological phenomenon, which controls all metabolic activities. It is the most important indicator of metabolic rate and status of the stress condition of exposed animals (Newell 1973). Since cellular and sub-cellular functions form the basis of all disorders, the toxic effects of xenobiotics mainly influence the cellular responses. The injury caused to cells by foreign compounds may be direct or indirect. Direct cell injury occurs when a toxicant interacts with one or more cell components. In indirect cell injury, the effect is due to a disturbance in the microenvironment of the cell. If tissues have insufficient supply of oxygen during hypoxia or anoxia, the energy metabolism is

disturbed leading to damage to the cellular metabolism. The increased glycol tic activity during oxygen deficiency cannot meet the energy requirements of the cell. As a result, the energy requiring processes such as protein synthesis, and glycol tic activity during oxygen deficiency cannot meet he energy requirements of the cell. As a result, the energy requiring processes such as protein synthesis, phospholipids metabolism, and membrane transport recesses are inhibited. Many investigators have demonstrated harmful effects of heavy metals on histological structure of gills of crustaceans (Ramanna. and Ramamurthy, 1996; Bhise, 2011).

Similarly, hen the crabs were exposed to sub lethal concentration of the same toxicant for 0, , 10 and 15 days showed an elevation in the haemolymph sugar level with a maximum decrease at 15 days. Physiological processes are mostly coordinated by hormones, and changes in hormone levels are expected to occur soon after exposure to environmental stress, such as pollutants, eventually acting as endocrine disruptors. Hyperglycemia is a common stress response of many aquatic animals. In crustaceans it occurs following the involvement of the hyperglycemic hormone produced in the eyestalk; it mainly regulates glucose homeostasis. It belongs to the neuropeptide family synthesized in the eyestalk by medulla terminal is x-organ, and is accumulated by and released from the sinus gland. We are in agreement with the views of earlier researchers that an elevation in the haemolymph sugar level of the fresh water crab, *Paratelphusa jacqemontii* can occur due to copper, which may act on the neurotransmitter acting on hyperglycemic hormone, a haemolymph sugar level regulating hormone **(**Varghese *et al.,* 1992; Maharajan *et al.,* 2012)

In conclusion, the total protein, free sugar and lipid level of the *Paratelphusa jacqemontii* showed a gradual declining trend in experimental animals exposed for 96 hours for 0 to 15 µg/l copper concentrations hence copper is proved to be toxic.

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