Egyptian Journal of Aquatic Research 44 (2018) 83–87

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Contents lists available at ScienceDirect

Egyptian Journal of Aquatic Research

journal homepage: www.sciencedirect.com/locate/ejar

Acute toxicity of some heavy metals to the fresh water snail, *Theodoxus niloticus* (Reeve, 1856)

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ARTICLE INFO

Article history:
Received 28 April 2018
Revised 7 June 2018
Accepted 7 June 2018
Available online 4 July 2018

Keywords:
Toxicity
LC50
Theodoxus niloticus
Zn
Fe
Pb

ABSTRACT

Three toxicity experiments were carried out separately to study the toxicity effects of Zn, Fe and Pb on adult freshwater snail *Theodoxus niloticus* (Gastropod, Prosobranchia, Nertidae). The LC50 values for the 96 hr exposures of Zn, Fe and Pb on *Theodoxus niloticus* were 12.199, 8.6 and 18 mg/l respectively. These values increased by decreasing the time of exposure. Fe was more toxic followed by Zn and Pb to *Theodoxus niloticus*. The rate of mortality increased by increasing the exposure period and concentration of metals in solution. Moreover, bioconcentration of Zn, Fe and Pb in soft parts and shell increased gradually by increasing the concentrations of each metal in solutions. After 96 hr of exposure, the bioaccumulation factors for each metal were decreased from highest values in the control to the lowest one with the highest concentration of the metal. It is difficult to compare LC50 values for metals for this species with those for other gastropod species due to different abilities of closely related taxa or the species belonging to the same genus, which live in the same habitats, to accumulate metals.

Introduction

Cupper (Cu), manganese (Mn), and zinc (Zn) iron (Fe), selenium (Se) and others are essential nutrient metals that are required for living organisms. Low levels of these elements lead to essential metal deficiencies. Nevertheless, they are toxic for all forms of life at excessive levels (USEPA, 1985). Copper is essential for the normal function and components of some enzymes in all living organisms, iron is an important component of hemoglobin in red blood cells and zinc is required as necessary component for more than 150 enzymes (Walker et al., 2006). Other metals like cadmium, lead are non-essential metals. Their toxic effect may be relatively high in comparison to other more essential metals (Zhiyou et al., 2016). It is important to know that heavy metals are natural substances but they are always considered contaminants or pollutants. In aquatic environments, contamination with heavy metals increases and becomes a critical problem that causes environmental risks (Nriagu et al., 1998). Unlike other organic pollutants such as pesticides that break down to less harmful components, managing metal contaminants demand an understanding of the concentration dependent toxicity. They do not degrade in general; therefore, they accumulate throughout the trophic chain (Gupta and Singh, 2011). Dose-response relations give the base for the assessment of risks caused by the chemicals (Shuhaimi-Othman et al., 2012). When the body accumulates heavy metals in the tissues and do not metabolize them, they become toxic. Toxicity tests is an important tool to assess the effect and fate of toxicants in the aquatic environment and they can be measured by many ways. Death is the widely way used to measure the toxicity because death is the acceptable sign of acute toxicity in living organisms (Adams and Rowland, 2003; Luoma and Rainbow, 2008).

Freshwater molluscs are among the aquatic biota which are advantageous organisms for biomonitoring purposes (Andral et al., 2004; Viarengo et al., 2007). These organisms, especially Gastropoda, can accumulate high levels of heavy metals in their bodies and have the ability as potential bioindicator (Abdel Kader et al., 2016). Fresh water snail *Theodoxus niloticus* (Prosobranchia, Nertidae) are used in the toxicity assay during the present study. It is abundant and widely distributed along the River Nile and its tributaries from Lake Nasser to Lower Egypt. It occurs in colonies on and under rocky limestone near the shoreline, associated with many aquatic plant species. *Theodoxus* needs high levels of oxygen to thrive (Orabi and Osman, 2015). Its average size is about 9 mm high and 8 mm width.

This research aimed to study the toxicity effects of some heavy metals Zn, Fe and Pb separately on adult fresh water snail *Theodoxus niloticus* to determine the mortality rate for each metal exposure then estimating LC50, the bioaccumulation and fate of Zn, Fe and Pb in the tissues of snail after 96 hr exposure to different concentrations of each metal.

Peer review under responsibility of National Institute of Oceanography and Fisheries.
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https://doi.org/10.1016/j.ejar.2018.06.004
1687-4285/

Materials and methods

Samples collection

Snail samples were collected from the Nile River at Mansoura City in front of the Company Sale Manufactures beside Talkha Bridge where there is no enough pollution. Samples were transported to the laboratory in glass tanks (30 × 30 × 30 cm) with water from the same habitat and suitable aeration. Snail were acclimatized to laboratory conditions using dechlorinated tap water which was exchanged every 12 hr interval for four days.

Preparation of stock solution

Stock solutions (1gm/l) of Zn^{2+} and $Fe^{3+}$and Pb^{2+} (from Zn $SO_4{\cdot}7H_2O$, $Fecl_3$ and Pb $(No_3)_2$ were obtained from Merck, Germany) were prepared separately. Tests concentrations were prepared from the stock solutions using appropriate calibrated pipettes and graduated cylinders. Zinc concentrations used in this study were control, 1.0, 4.0, 8.0, 12.0 and 20.0 mg/l, zinc concentration in the control was measured and added to previous concentrations. In Fe experiment, the used concentrations were control, 1.0, 4.0, 7.0, 10.0 and 15.0 mg/l, concentration of Fe in the control was added to them. Lead was used at concentrations of control, 1.00, 2.0, 5.0, 20.0 and 40.0 mg/l, lead concentration was measured in control and added to previous concentrations. These ranges of each metal were chosen and based on results of preliminary experiments.

Acute toxicity assay

Three toxicity experiments were carried out in the laboratory for toxicity assay of Zn, Fe and Pb on snail *Theodoxus niloticus*. Acclimatized snails were transferred to (20 × 20 × 40 cm) test glass aquaria. Each container contains 750 ml of a definite concentration, air pumps and individual stone diffusers were provided for well aeration. Twenty snails were put per container and two replicates per treatment and control were carried out. The toxicity experiments were performed at room temperature of 25– 28°C with fluorescent lights 12 hr light: 12 hr darkness. Adult snails used in each experiment were nearly of the same size and were exposed to the same level of aeration. No food was supplied during the experiments. Test solutions were changed by another new one of the same concentration every 24 hr interval (APHA et al., 1999). Dead animals were counted every 12 hr and removed from each solution and mortalities were recorded at the end of the experiment (96 hr exposure), the median lethal concentration (LC50) was estimated for each metal by the Probit transformed concentration - response curves (USEPA, 2002).

The remaining alive snails were removed from each concentration solution and from control. Metal detection in all the soft tissues and shells were determined by using the method described by Siraj et al. (2016), Khan et al. (2018) and the detected metals were measured by Atomic Absorption Reader (Savant AAS with GF 5000 Graphite furnace). Data were expressed in mg/kg dry weight and bioaccumulation of Pb, Zn and Fe in the soft tissue and the shell of *Theodoxus niloticus* compared with those in solutions to measure the steady-state bioaccumulation factors (BAF).

Table 1
Percentages of mortality of *Theodoxus niloticus* in different Zn concentrations (mg/l) for 96 hr exposure.

Hours	mg/l					
	C (0.199)	1.199	4.199	8.199	12.199	20.199
24 hr	0	0	0	5	10	15
48 hr	0	10	15	15	20	30
72 hr	0	15	25	25	35	50
96 hr	0	20	25	30	50	85

Table 2
Percentages of mortality of *Theodoxus niloticus* in different Fe concentrations (mg/l) for 96 hr exposure.

Hours	mg/l					
	C (0.235)	1.235	4.235	7.235	10.235	15.235
24 hr	0	0	0	0	5	5
48 hr	0	0	5	5	10	30
72 hr	0	25	35	40	45	60
96 hr	0	30	35	40	55	80

Table 3
Percentages of mortality of *Theodoxus niloticus* in different Pb concentrations (mg/l) for 96 hr exposure.

Hours	mg/l					
	C (0.004)	1.004	2.004	5.004	20.004	40.004
24 hr	0	0	0	0	5	0
48 hr	0	5	10	5	15	20
72 hr	0	15	20	15	20	50
96 hr	0	35	40	40	55	75

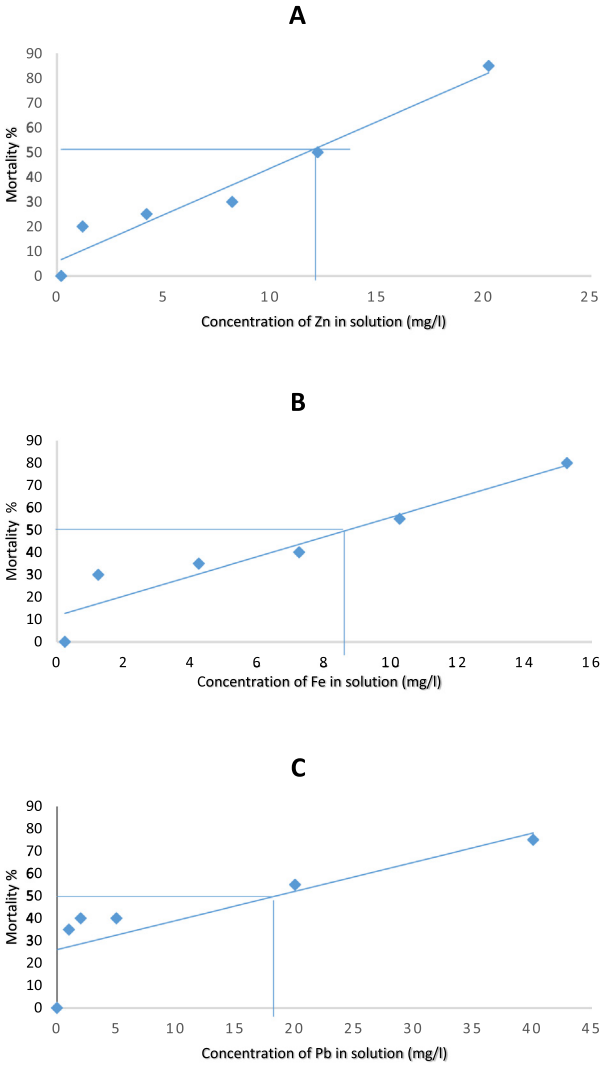

Fig. 1. Probit line graph of Zn (A), Fe (B), Pb (C) toxicity on gastropod, *Theodoxus niloticus* in different concentrations after 4 days exposure.