A MODEL STUDY TO USE DUCKWEED, *LEMNA MINOR* AS INHIBITOR OF HEAVY METALS (CU & ZN) STRESS GENES IN NILE TILAPIA *ORIOCHROMIS NILOTECUS*

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Abstract
Aquatic plants have been known for metals bioaccumulation and sequestration. The ability of Duckweed (*Lemna minor*) to regulate heavy metals uptake and to reduce their impacts on the activity and gene expression of main enzymatic antioxidants was evaluated. *L. minor* was exposed to high levels of the most dominant heavy metals in the Egyptian environment; copper and zinc. Nile tilapia within different treatment groups were exposed to water contaminated with copper and zinc for 4 weeks to assess the damage in DNA and alteration in expression of the stress related genes as GST, CAT, SOD and GPx as well as its protective mechanism on DNA structure against damage in response to the exposure to low and high doses of Cu and Zn.

Keywords: Biotechnologies, Mediterranean Sea

Introduction
Cu naturally occurs in the aquatic environment in low concentrations. Cu is one of the most toxic elements to aquatic species, at levels just above that needed for growth and reproduction it can accumulate and cause irreversible harm to some species. Copper is an essential trace metal necessary for growth and metabolism of all living organisms Cu is acutely toxic (lethal) to freshwater fish via their gills in soft water at concentrations ranging from 10 – 20 ppb.
Zn is an essential trace element for organisms, but in excessive amount is toxic for organisms. The present wide industrial use of zinc makes it one of the most common pollutants in natural waters. Excessive zinc could inhibit physiological activities of aquatic organisms and even be lethal. Zinc can be accumulated in organisms and transmitted by the aquatic food chain and finally harm human health.

Duckweed (*Lemna minor* L.) is used in water quality studies to monitor heavy metals and other aquatic pollutants, because duckweed, like other water plants, may selectively accumulate certain chemicals and may serve as biological monitors.

Material & methods
Ten fish from each treatment were sampled after 4 weeks of treatment. At the end of the experimental period, all fish were sampled and dissected. Gills and liver samples from tilapia within different treatment groups were collected in liquid nitrogen and stored at -80ºC until used for RNA extraction. Several techniques including enzyme activity determination, comet assay, alteration in gene expression, and Quantitative Real Time-Polymerase Chain Reaction were applied.

Results and discussion
Treatment of Cu or Zn contaminated water with Duckweed decreased the DNA damage by 36 and 37% respectively. On the other hand duckweed increased the activity levels of GST, SOD, CAT and GPx (Figure 1). Expression levels of GST, and GPx genes were significantly increased in contaminated water fishes treated with duckweed.

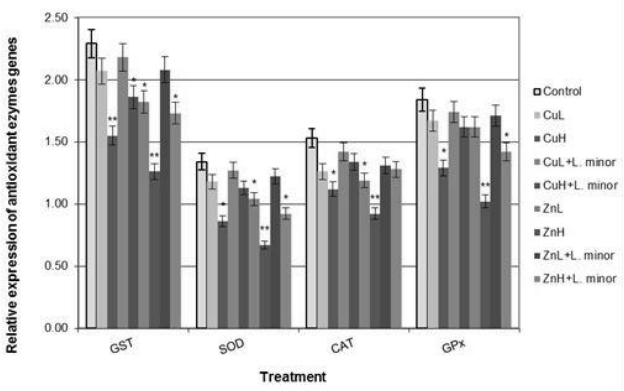

Fig. 1. The relative expression of antioxidant enzyme genes (GST, SOD, CAT, GPs) in liver of Nile tilapia exposed to heavy metals (Cu and Zn) in water with or without duckweed (*L. minor*) (L: low dose, H: high dose). *P < 0.05 and ** P < 0.01 for the treated groups compared with control group.

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