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# Availability, Accumulation and Elimination of Cadmium by *Artemia urmiana* in Different Salinities

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## ABSTRACT

**The effect of salinity on cadmium uptake and elimination by brine shrimp *Artemia urmiana* which lives only in Urumieh Lake, Iran, was studied for the first time. Brine Shrimp were acclimated to different salinities and exposed to the metal in solution at differing salinities and composition. Experimental salinities ranged from 50 to 125 $gL^{-1}$ and the concentrations for cadmium added to the solution were 20 and 5 mg/L. At first, bioaccumulations of Cd (5, 10, 15 and 20 mg/L Cd) were measured on day 1, 5, 11 and 17 of life. The results showed that, accumulation of cadmium in *A. urmiana*, depends upon the concentration of cadmium in the environment and increased with time. It also increased with the concentration of Cd in the environment. Maximum accumulation of Cd (0.748 mg kg -1) was observed with 20 mg/L Cd and on the 11th day of life of *Artemia*. Within each acclimation group, the cadmium uptake decreased with increasing exposure to salinity.**
**To study cadmium elimination, samples were taking at 0, 6, 12, 24, 48, and 72 h. Results showed that, *Artemia urmiana* has the ability to accumulate and eliminate cadmium. Accumulation and also elimination of Cd by *Artemia urmiana* in 20 mg/L Cd were higher than in 5 mg/L Cd in all studied salinities. The cadmium accumulation pattern which seems well conserved in *Artemia* is characterized by a fast elimination of the metal.**

**Key Words: *Artemia urmiana*, salinity, bioaccumulation, elimination, Heavy metal**

## INTRODUCTION

The brine shrimp *Artemia* (crustacean, Anostraca) is distributed worldwide with the exception of Antarctica (Medina *et al*. 2007). *Artemia* is a crustacean that is widely used in laboratory toxicity studies due to its small body size and short lifespan together with its availability from dry cysts. *Artemia* has a broad tolerance to environmental factors such as salinity, temperature and dissolved oxygen in the water allowing it to live in hyper saline waters (Sarabia *et al.* 2006) such as Urmieh Lake.

Urmieh Lake with an area of about 5000 $km^2$ is one of the largest permanent water catchments in west Asia. Urmieh Lake is a thalssohaline, sodium chloride lake with oligotrophic characteristics, located at an attitude of 1250 m above sea level (Agh *et al.* 2008). *A. urmiana* was first reported in Urmieh Lake by Gunther in 1899 (Agh 2002).

Cadmium is a ubiquitous non-essential element that possesses high toxicity and is easily accumulated from the environment by aquatic organisms (Del Ramo *et al.* 1993).

Cadmium is a reference toxicant as well as a widespread environmental pollutant, and its mechanism of toxicity is relatively well understood. There have been no reported cases of catastrophic episodes of pollution of natural salt lakes at potentially dangerous levels of cadmium and it is accepted that it is not easily bio-available at the high salinities predominant in these waters. However, the atmosphere is an important and continuous source of cadmium for the waters. A decrease in salinity of these lakes has already occurred as a consequence of climate change (Sarabia *et al.* 2003).

It is accepted that life history traits can be modified as a consequence of long- term changes in the environment, which may be of great relevance to the viability of the population (Sarabia *et al.* 2003). The uptake and accumulation of cadmium by aquatic organisms is a long standing environmental problem (Nriagu 1988 , Nriagu and Pacyna. 1988).

Cadmium has no known biological function and the internal body concentration is not regulated (Rainbow 1985). Uptake and toxicity of trace metals such as cadmium by

aquatic organisms are often affected by salinity whereby toxicity increases at low salinity (Blust *et al. 1992*). The problem of metal speciation at different salinities may be addressed in several ways. Varying the amount of total metal added to solution alters the chemical equilibrium of the solution such that the free ion concentration changes. Alternatively, chelating agents (such as ethlylendiaminetetraacetic acid [EDTA]) may be added to a solution to alter the availability of metal- binding ligands, altering the free ion concentration without altering the total metal concentration. Physiological mechanisms, especially water and ion exchange during osmoregulation, may also play a role in metal uptake. For example, metal uptake may be reduced at an organism's isosmotic point when ion exchange between the organism and the external medium is reduced because of hemolymph osmotic pressure being in equilibrium with the

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external medium. However, comparatively few data are available for examining these possible mechanisms of metal uptake in euryhaline crustaceans (Roast *et al.* 2001).

The chemical speciation of cadmium in saline waters with a composition similar to that of seawater is dominated by the formation of weak complexes with chloride. Only a small fraction of the cadmium exists as the free metal ion while most of the cadmium is found in chloride complexes. The concentration of the free metal ion increases with decreasing salinity (Boyle *et al.* 1976 , Mantoura *et al.* 1978 , Turner *et al.* 1981).

Aquatic organisms control the movement of water and ions across the exchange surfaces by altering the permeability of body surface and / or by actively regulating the influx and efflux of water and ions. Acclimation of an aquatic organism to salinity involves the alteration of the exchange surfaces (e.g. gill and gut epithelium) in order to maintain the composition of the internal environment within certain physiological limits. Generally, the permeability of aquatic organisms is lower in low salinity than in high salinity, environments. Several studies have shown an inverse relationship between the salinity and the uptake or toxicity of cadmium in aquatic organisms. This observation has been explained in different way including: (1) increased availability of cadmium at low salinity caused by the increased free cadmium ion level (Sunda *et al.* 1978 , Engel and Fowler 1979, Del Lisle and Roberts 1988), (2) increased influx of cadmium as a result of the decreased osmolarity of the solution (George *et al. 1978*) and (3) competition of calcium and magnesium with cadmium for similar uptake sites (Part *et al. 1985*).

To understand how these chemical and biological processes influence the availability of cadmium to salt water organisms it is necessary to determine effects of these different salinity components on metal uptake. For this purpose we have studied the accumulation and elimination of cadmium in *Artemia urmiana* for a better understanding of strategies used by *Artemia* to elude cadmium toxicity and to investigate the dependence uptake and elimination on cadmium concentration in water.

## MATERIALS AND METHODS

### *Culture procedure*

This study was conducted on *Artemia* nauplii hatched from dried cysts in laboratory of Faculty of Marine science and Technology, Azad University in 2009. Cysts were hatched in a funnel shaped plastic container filled with synthetic seawater. Newly hatched nuclei were processed following the procedure described by Amat (2005). The larvae hatching from the cyst samples were siphoned into separate beakers and then transferred into separate 30 L aquaria, where they were cultured until adulthood (Agh *et al*. 2008). Adult *Artemia* were all transferred into separate 1.5 L cylindro-conical containers. The animals were cultured at 27±1 ºC under constant aeration. The salinity in each flask was checked twice a day in order to maintain salinities according to the experimental set up. *Artemia* were fed unicellular algae *Dunaliella tertiolecta* and chemically treated yeast. Density of *Artemia* was adjusted to one animal/mL at the beginning of the experiment, but the density was gradually decreased to one animal per 3 mL on day 8 and per 4 mL on 14 of growth.

### *Bioaccumulation experiments in different stages of life in Artemia*

These tests were performed on one-day Nauplii, 5 & 11 and finally, 17 day *Artemia*. Four different concentrations of cadmium (5, 10, 15 & 20 mg/L) were prepared with control group in 3 repetitions. About 2,000 *Artemia* (1,000 for larger *Artemia*) were subjected to the mentioned concentrations for 24 hours and the concentrations were then measured (Hadjispyrou *et al.*, 2001).

### *Bioaccumulation experiments*

To study bioaccumulation adult *Artemia* pre-exposed to cadmium for 3 h were used as a measure of biological availability of the metal. Experiments were carried out under semi-static conditions in 2-L glass flasks containing 1.5L of test solution at two sub lethal concentration of Cd (5, 20 mg/L) and four concentration of salinity (50, 75, 100 and 125 ppt). All experimental flasks were kept under the same conditions, three independent replicates per treatment (three flasks exposed to one of four salinity concentrations and two Cd concentrations and one control) were performed (Sarabia *et al.* 2006). Preliminary assays showed considerable differences in tolerance to chronic Cd exposure in *Artemia* populations studied. *Artemia* population, as a function of its sensitivity, was exposed to at least one of the different Cd concentrations tested in such a way that at least 80% of the population was alive at the end of the experiment to find out the time course of metal uptake and elimination. Adult *Artemia* were cadmium exposed for 14 to 27 days in order to achieve a presumed equilibrium. Afterwards, animals were rinsed

and transferred to clean seawater to start the elimination phase and were maintained there for 3 days during the uptake phase. Ten animals were taken randomly from each experimental flask and the medium was completely renewed every 3 days. After collection, samples were rinsed with distilled water, dried on absorbent paper, weighed and frozen at -20 °C until the analyses were carried out. For the elimination study, samples were taken at 0, 6, 12, 24, 48 and 72 h (Sarabia *et al.* 2006).

***Chemical Analysis***

The separated *Artemia* samples were washed with distilled water. Each repetition was separately transferred to a container which had previously been completely cleaned and washed with distilled water and then kept in freezer at a temperature of –20 ºC up to digestion and analysis phases (Blust *et al*. 1992). The samples were placed inside an oven for digestion at a temperature of 50 ºC for 24 hours to be completely dried. After cooling the samples in a desiccator, the dried samples were transferred to separate beakers and weighted to the nearest 0.0001g. At first, 1 ml nitric acid was added to dry samples and the samples were heated at a temperature of 60 ºC for 10 minutes. Then, 1 ml of hydrochloride acid was added and they were heated for 30 minutes. Then the solutions were reached to a volume of 10 cc and were kept in different jars until instrumental analysis (seebaough *et al*. 2004). Cadmium concentrations in *Artemia* were determined by atomic absorption spectrophotometer (Varian, model 220 spectra) after digestion in nitric acid and hydrochloric acid (Blust *et al. 1992*). This part of experiment was performed in the Atomic Energy Organization of Iran.

***Data Analysis***

Data obtained were analyzed using SPSS software. All sets of data were tested for homogeneity of one way ANOVA and HSD test. The Excel sofware was used to plot graphics.

## RESULTS

The results of cadmium bio-accumulation in cysts and one-day nauplii samples and 5, 11 & 17 day *Artemia* as well as in the control group in different cadmium concentrations (5, 10, 15 & 20 mg/L) are shown in figure 1. Cadmium accumulation level in cysts of *Artemia* until maturity had no significant difference as compared to the control group. In the investigation of cadmium accumulation level in one-day nauplii, significant difference were detected in all cadmium concentrations with control group and a significant difference was also seen in 5, 10, 15 & 20 mg/L cadmium treatments ($p<0.05$) (median bioaccumulation in 5, 10, 15 and 20 mg/L Cd, were 0.276, 0.253, 0.282 and 0.364 mg kg -1 respectively). There were no significant differences between the control group and 5 mg/L cadmium treatment ($p>0.05$) regarding cadmium accumulation in 5-day *Artemia* (median bioaccumulation in 5, 10, 15 and 20 mg/L Cd were 0.775, 0.634, 0.386 and 0.480 mg kg -1 respectively). The results showed that bio-accumulation level in *Artemia* increased with the increase of cadmium concentration in the manner that bio-accumulation level in 11-day *Artemia* and cadmium 5mg/L treatment were significantly less than cadmium concentration in 15 & 20 mg/L treatments ($p<0.05$). Maximum accumulation levels were obtained in 11-day *Artemia* and in cadmium 20 mg/L concentration which was significantly different from all treatments (median bioaccumulation in 5, 10, 15 and 20 mg/L Cd were 0.345, 0.553, 0.719 and 0.748 mg kg -1 respectively). Moreover, bio-accumulation of cadmium of 17-day *Artemia* in a cadmium concentration of 20 mg/L was significantly different from that in the control group ($p<0.05$) (median bioaccumulation in 5, 10, 15 and 20 mg/L Cd, were 0.179, 0.171, 0.223 and 0.327 mg kg -1 respectively).

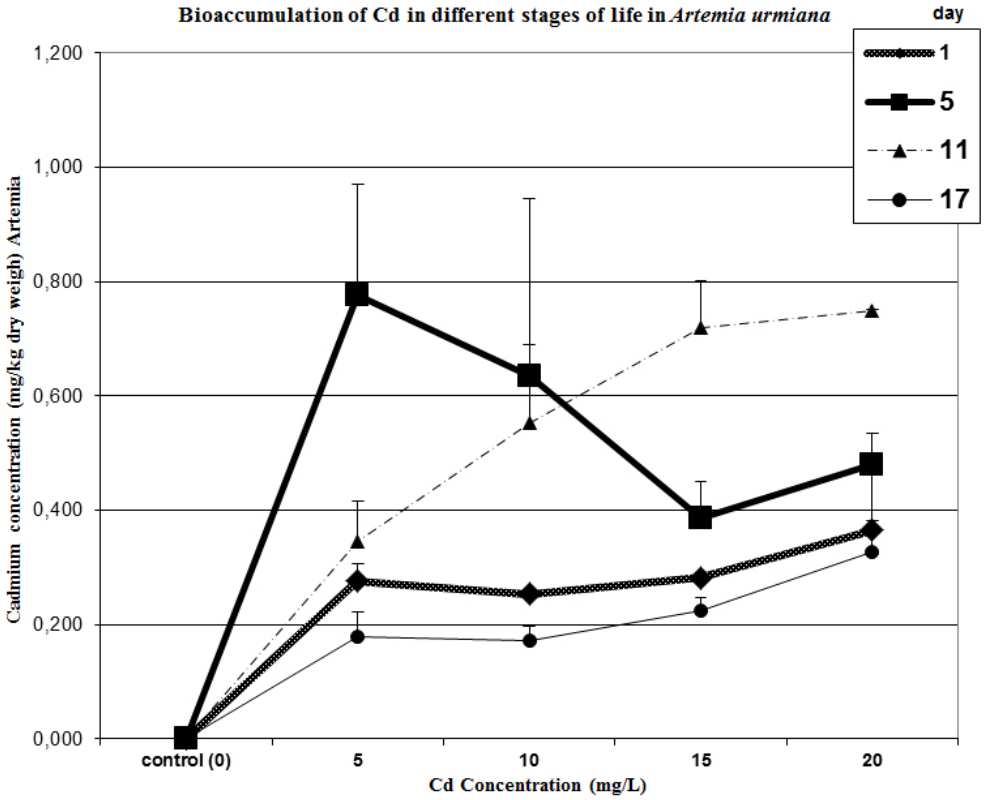


**Figure 1.** Median Cadmium concentration in different days of life of *A. urmiana* exposed to Cd

The accumulation of cadmium by adult *Artemia urmiana* at 2 external cadmium concentrations (20 and 5 mg/L Cd) are shown in fig. 2 and 3. Accumulation of cadmium continues through 3 h exposure under all 4 conditions of salinity (50, 75, 100 and 125 ppt). For each of the 9 different acclimation groups the uptake of cadmium decreased rapidly with increasing salinity of exposure but there were marked differences among the acclimation groups. For the same salinity of exposure uptake of cadmium increased with increasing cadmium concentration. There was no significant difference between levels of median bioaccumulation of cadmium in *A. urimiana* which was exposed to different salinities with dissolved cadmium ($P>0.05$) (fig. 2 and 3). Median bioaccumulation of cadmium in *Artemia*, exposed to 20 mg/L Cd in different salinities (50, 75, 100 and 125 ppt) were 0.258, 1.916, 0.543 and 0.322 mg kg -1, respectively and 0.081, 0.079, 0.084 and 0.045 mg kg -1, respectively when exposed to 5 mg/L Cd. As regards the elimination, the cadmium eliminated by *A. urmiana* at 75 ppt salinity exposed to 20 mg/L Cd, after 6 h in clean water was significantly different from that at 100 and 125 ppt salinities (median concentration of Cd elimination after 6 h following a 3 h pre-exposure to 20 mg/L Cd in 50, 75, 100 and 125 ppt, were 0.075, 0.113, 0.040 and 0.040 mg kg -1 respectively). Maximum accumulation of cadmium was observed at 75 ppt, higher compared to the 100 and 125 ppt. After 12 h, significant differences were found in cadmium elimination at 50 ppt as compared to that at 75 ppt and also between 75 and 125 ppt salinities ($P<0.05$) (median concentrations of Cd elimination, after 12 h, following a 3 h pre-exposure to 20 mg/L Cd, in different salinities 50, 75, 100 and 125 ppt were 0.027, 0.048, 0.032 and 0.020 mg kg -1 respectively).

Cadmium elimination after 24 h was significantly different at 50 ppt as compared with that at 75 ppt. Similarly significant differences were found in cadmium elimination at 75 ppt with that at 125 and 100 ppt (median concentration of Cd elimination after 24 h, following 3h pre-exposure to 20 mg/L Cd, in different salinities of 50, 75, 100 and 125 ppt, were 0.025, 0.045, 0.032 and 0.023 mg kg -1, respectively). After 48 h and 72 h, significant differences were found in cadmium elimination at 50 ppt with that at 75, 100 and 125 ppt salinities ($P<0.05$) (median concentration of Cd elimination after 48 h, after 3h pre-exposure to 20 mg/l Cd, in different salinities 50,75, 100 and 125 ppt, were 0.097, 0.044, 0.039 and 0.022 mg kg -1 respectively) & (median concentration of Cd elimination after 72 h , following a 3h pre-

exposure to 20 mg/L Cd, in different salinities 50, 75, 100 and 125 ppt, were 0.076, 0.020 ,0.035 and 0.019 mg kg -1 respectively).

Results showed that, with pre-accumulation (20 mg/L Cd) cadmium elimination at 50 ppt was significantly different at 6, 12, 24, 48 and 72 h. Also, significant differences were observed in cadmium elimination in *A.urmiana* at 75, 100 and 125 ppt at different hours ($p<0.05$). However as the experiment continued there were no significant differences in the cadmium eliminated from the body of *Artemia* at different salinities studied ($P>0.05$). (median concentration of Cd elimination after 6 h following a 3h pre-exposure to 5 mg/L Cd were 0.047, 0.061, 0.016 and 0.023 mg kg -1 respectively at 50, 75, 100 and 125 ppt) (median concentration of Cd elimination after 48 h following a 3h pre-exposure to 5 mg/L Cd in different salinities of 50, 75, 100 and 125 ppt were 0.030, 0.020, 0.012 and 0.018 mg kg -1, respectively) and (median concentration of Cd elimination after 72 h, after 3h pre-exposure to 5 mg/L Cd in different salinities of 50, 75, 100 and 125 ppt,were 0.022, 0.018, 0.009 and 0.012 mg kg -1, respectively) .

After 12 h, significant differences were found in cadmium between 50 ppt and 75 ppt salinities and between 75 and 100 ppt ($P < 0.05$) salinities. (median concentration of Cd elimination after 12 h following a 3h pre-exposure to 5 mg/L Cd in different salinities 50,75, 100 and 125 ppt, were 0.024, 0.069, 0.016 and 0.036 mg kg -1, respectively ).

However after 24 h no significant differences were found between cadmium elimination at 50 ppt and that eliminated at 100 and 125 ppt ($p<0.05$) (median concentration of Cd elimination after after 3h pre-exposure to 5 mg/L Cd in different salinities of 50, 75, 100 and 125 ppt were 0.037, 0.025, 0.018 and 0.013 mg kg -1, respectively).

On comparing different hours of cadmium elimination at different salinities it was seen that at salinity of 50 ppt, significant differences in cadmium elimination and cadmium uptake were found in Artemia previously pre-exposed to 5 mg/L Cd only after 72 h. However at 75 ppt , significant differences were found after 24, 48 and 72 h. At the same salinity significant differences were also found between cadmium elimination after 12 h and that eliminated after 24, 48 and 72 h. At 100 ppt cadmium elimination was significantly different at different hours (6, 12, 24, 48 and 72 h), while at 125 ppt, no significant differences were noted in cadmium elimination at different hours as compared to cadmium uptake.

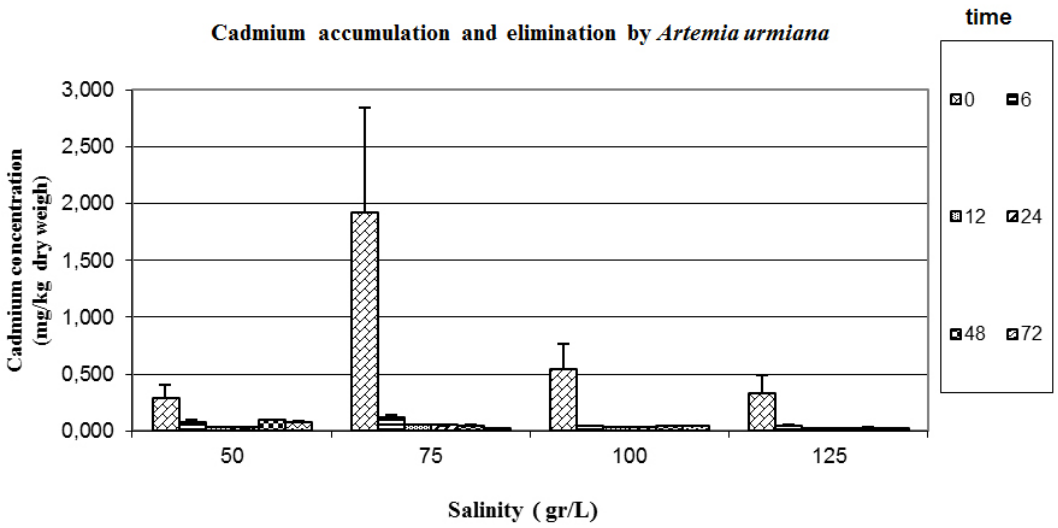


**Figure 2.** Concentrations of cadmium after 3h pre-exposed 20mg/L cd in different salinities and elimination in different times (6-72 h)

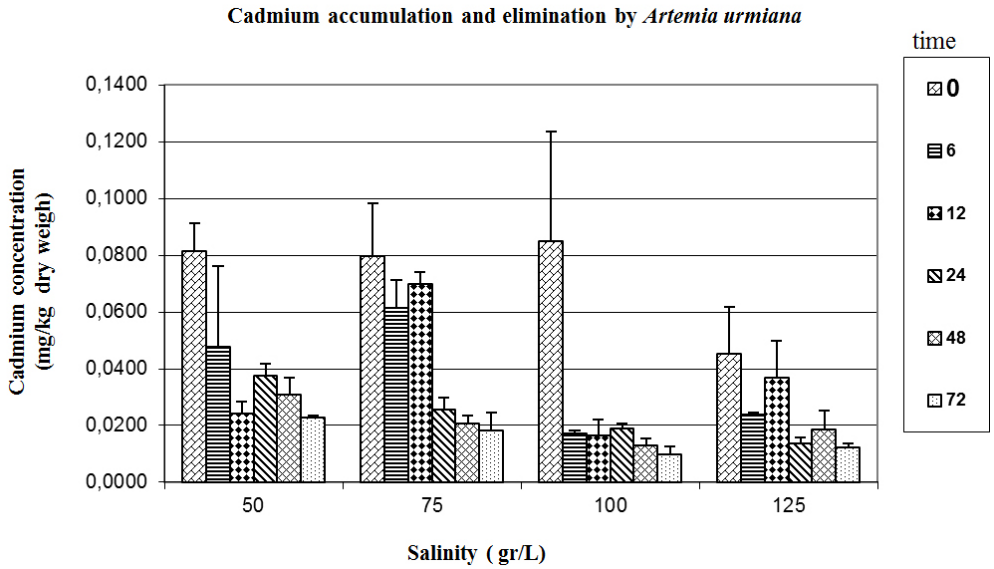


**Figure 3.** Concentrations of cadmium after 3h pre-exposed 5mg/L cd in different salinities and elimination in different times ( 6-72 h )

## DISCUSSION

The results of cadmium bio-accumulation in 1, 11 & 17-days *Artemia* showed that accumulation level increased upon the increase of metal in the solution, but for 5-day *Artemia*, a high mortality occurred compared to all periods of life when they were exposed to Cd. Therefore, in those that remained alive, accumulation level was less by water filtration control mechanisms**.** It seems that the less the cadmium concentration, the less stress will be imposed on *Artemia* and its accumulation level increases. A significant difference in cadmium concentration in 5-day *Artemia* was observed in 5mg/L concentrations compared to control group, but when cadmium concentration increased, the mortality increased, too and the level of accumulation decreased (figure 1). For the rest of their life, a significant difference was observed in cadmium accumulation upon increase in cadmium concentration. But there was no significant difference between different concentrations in 17-day *Artemia*. There was only a significant difference between the 20mg/L cadmium concentration and the control group which indicates larger *Artemia* has more capability to accumulate cadmium compared to their early days of life. There were partial amounts of cadmium in cyst to mature samples of control group. Statistical investigation showed that there was no significant difference in that partial amount. In all cases, no significant differences were observed in one-day nauplli between control group and different cadmium treatments. Investigation and comparison of results show 11-day *Artemia* have a high capability in cadmium accumulation compared to other periods of life (even compared to 17-day period) and accumulation capability will significantly increase with increase in cadmium concentration. This is probably due to increase in feeding capability as well as increase in non-poisoning power and metal removal in larger *Artemia* (17-day). Hence it can be said that even at minimum levels of cadmium, 5-day *Artemia* were able to accumulate the metal through a capillary network on the surface of their body and a higher percentage of them survived, while accumulation levels in larger *Artemia* showed a decrease possibly due to removal and neutralization of metal as well as decrease in surface interchange (figure 1).

This trend was observed in all investigated concentrations and shows lower sensitivity of *A. urmiana* in 11-day samples (instar 8). Studies of other researchers such as (Hadjispyrou *et al.* 2001) proved that accumulation levels of metals such as tin, cadmium and chromiumin *A. franciscacna* is lower compared to fishes, thus showing resistance of this species against heavy metals. They also showed that filter feeding rate decreased severely in the presence of toxins and lethal concentrations and in such conditions the creature makes an effort to survive by minimizing its metabolic activities.

As seen from the results of the present study with 5-day old *Artemia*, bio-accumulation in tested *Artemia* in 5mg/L of cadmium concentration was more than that in other investigated cadmium concentrations. The processes through which different aquatics can regulate the concentrations of different metals in their bodies are quite diverse and complicated. For example, accumulators are

creatures that store the metals on a non-toxic basis in high amounts. These creatures change the metals somehow to a non-toxic form and store them by granulating them and combining them with metallothionein. Metallothioneins are a class of low- molecular weight, cytoplasmic, metal-binding proteins that have a high affinity for various toxic heavy metals, particularly cadmium. Elevated levels of such proteins have been suggested as indicating involvement in uptake, storage, transport, and elimination of toxic metals and in the routine metabolism of metal. Del Ramo *et al* (1995) showed the MT content in *Artemia* increased in a time-dependent fashion. There is a remarkable increase in MT content between 12 and 24 h of cadmium exposure (Del Ramo *et al.* 1995). It was found that MT protects from Cd- induced toxicity by reducing the amount of Cd that is bound to ligands other than MT (Del Ramo *et al.* 1993). Metallothionein synthesis in *Artemia* is very high and one of the reasons of high resistance of this creature to pollutants is attributed to this issue.

It has been proved that none of the known aquatics has the capability of active regulation of cadmium. Whenever cadmium concentration in environment increases, contributing systems in detoxification process are incapable and mortality occurs (Brix and Deforest 2000). But due to relatively high salinity of cultivation and living environment of *Artemia* (a salinity of 35 g/l or more) and high tendency of cadmium to form complex with chloride ion, its bioavailability decreases and so does its toxicity level.

Blust *et al* (1992) and other several studies have showed, the availability of cadmium to the brine shrimp depends on the free cadmium ion level in the solution. Most of these studies have not experimentally considered the functional difference between these 2 related factors. The results of the present study show that the availability of cadmium depends on the activity rather than on the concentration of the free metal ion. However, since activity and concentration are closely related, the functional difference between there 2 factors is only apparent in the low salinity region where changes in activity and concentration are most pronounced. It has been shown that several other factors which vary with salinity such as the osmolarity and composition (i.e. calcium and magnesium concentration) of the solution influence the uptake or toxicity of cadmium in aquatic organisms.

As the results reveal, in higher salinities, the absorption and accumulation of Cd was less than other treatments and after the last stage of elimination, there were a small amount of cadmium in *Artemia's* body. *Artemia* that was previously exposed to 20 mg/L Cd in different times and stages of elimination, showed significant difference of cadmium accumulation in 50 ppt salinity compared to all other treatments ($p<0.05$). This ability to eliminate from the early hours (6 hours) when placed in clean water can be seen and over time (maximum 72 hours) gradual removal of more cadmium takes place (Fig 2).

Similar trends were seen in salinity of 120, 100 and 75 ppt where *Artemia* significantly has the ability to remove (elimination) cadmium. Maximum elimination of cadmium in *Artemia* groups during the 3-hour exposure to 5 mg/L Cd was observed in the salinity of 50, 75 and 100 ppt after 24 hours. On the whole it was seen that *Artemia* exposed to 20 mg/L Cd accumulated more cadmium and therefore eliminated significantly higher concentrations of cadmium as compared to those that were exposed to 5 mg/L Cd. Sarabia *et al* (2006) in comparative toxicokineties of cadmium in *Artemia* observed that parthenogenetic *Artemia* have the ability to absorb and eliminate of cadmium when placed into clean water. In the initial stage of the uptake of cadmium in *Artemia*, the increase in the cadmium levels in this crustacean is linear and directly proportional to the concentration in the water. As the concentration in the animal increased, the net uptake decreased until it reached a state of dynamic equilibrium in which the rates of accumulation and elimination compensated each other. We, therefore, know that the cadmium present in these compartments is not evenly distributed and thus will not be available equally for elimination.

It is generally accepted that organisms may present three different strategies for avoiding the toxic activity of cadmium: (1) reducing the entry of the metal; (2) increasing the excretion; or (3) capturing the metal within the tissues in a way that is non- toxic for the organism (Sarabia *et al.* 2006). According to our results, it seems that the individuals of *Artemia* restrict metal entry.

The concentration of cadmium reached in organisms belonging to populations of the same or related *Artemia* species may vary due to differences in the routes of entry and elimination of the metal, and in the form of storage, in addition to differences in the permeability of their teguments for the metal, the ligands that bind them, and in the metabolic rates. It should not be forgotten that the physiological, anatomical, ecological, and behavioral differences described for the different populations of *Artemia* studied may contribute to this variability. Nevertheless, as has been mentioned before, the accumulation of cadmium in *Artemia* does not present great differences, at least in the population studied. *Artemia* is a hypo-hyperosmotic euryhaline regulator. The adults use the salty medium where they live as drinking water,

which introduces an excess of ions inside of them that they should eliminate through the gill (Sarabia *et al.* 2006). Studies have showed, all populations of *Artemia*, eliminated cadmium significantly on being transferred to clean water, managing to eliminate between 23 % and 64 % of the accumulated cadmium after three days in metal-free water (Sarabia *et al.* 2006).

Nevertheless, the percentages of cadmium eliminated do not reflect the total complexity of the elimination process, since not only is the percentage of cadmium eliminated important, but also the speed with which it occurs. We could think of *Artemia* as an organism capable of regulating, to a certain extent, the content of cadmium accumulated.

Cadmium is considered as a non-essential metal for crustaceans, and therefore, it is not expected to show the accumulation patterns corresponding to essential metals in which body concentration of the metal is maintained to and approximately constant level over a wide range of availabilities in order to meet essential metabolic needs.

The results obtained from this research showed that *Artemia* bio-accumulation capability against cadmium and the accumulation level of this metal depend on environment as well as different living periods of *Artemia* and this accumulation continues during its life. But as it was specified in this research, if they are exposed to cadmium-free water, they will significantly remove the attracted cadmium.

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