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Investigation of heavy metals (Cadmium, Lead) in *Chironomidae* and *Gammarus pulex* Namrood River – Tehran Province

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Abstract: Marine ecosystem pollution is one of the important problems of today environment. In this study the existence of heavy metal in the Namrood River, situated in Firoozkooh in Tehran province, Iran has been investigated. The Namrood River is located near Firoozkooh route, and is affected by pollutant from tourist centers, entertainment, gas stations, nearby villages' sewage systems, farming effluent, and hatchery farms. In some areas, its water is heavily polluted possibly by heavy metals. After selecting two stations in upstream and downstream of the river, they were sampled three times in both cold and hot seasons of year (mid-March, and June) for *Chironomidae,* and *Gammarus plux* sediments. The measured heavy metals were cadmium and lead. The results showed that the concentration of cadmium in measured samples varied from 0.010-0.2033 ppm. The concentration of lead in samples varied from 0.11-2.16 ppm. The results also indicated that sediments of samples taken from the upper station in the cold season had a higher proportion of cadmium and a higher concentration of lead than sediments in the lower station during the hot season.

Keywords: cadmium, lead, *Chironomidae*, *Gammarus pulex*, Namrood River

INTRODUCTION

Heavy elements are part of the earth's mantle and a group of elements with atomic weights of 63.546-200.59; their specific gravity is higher than 6 g/cm^3 when surfaced by natural factors or human beings. Heavy metals have entered the biosphere and food chain systems in this way (Esmaeely Sari, 2000).

Selecting biological characteristics is one of the most important biological studies applicable in connection with determining the water quality of rivers. Biological characters are various creatures that on the basis of their specific biology have the ability to live and survive in a particular environmental condition; otherwise they cease to exist and lose from the area.

Microbentos are thought to determine the rate of pollution in the marine ecosystem, for they are motionless and because of their superiority to others of their kind. In comparison with swimming fishes, microbentos sampling is relatively simple. Microbentos also have a longer life span, variance and change in biomass, and their form of species is less than plankton; therefore, microbentos reflect reactions to incoming loads, variety of running water, and sewage. Even bimonthly sampling (once every two months) or semiannually is sufficient enough to show major changes (Ghafoori, 1995).

The Namrood River is located in Ghezghanchai village, Firoozkooh county, Damavand city, Iran. This river is one of the rivers flowing year-round in the area, which flows to Hablehrood in the Dasht-e-Kavir. The length of The Namrood River is 51 kilometers. Its altitude at source is 3000 meters. The river falls 1720 meters over these 51 km and its relative grade is 2.5%. Its general path is to the east, and its annual volume is 160 million cubic meters.

The Namrood River originates from north of the Gharedagh Mountains, 20 km to the northeast of Damavand Mountain in a village called Ghezghanchai. It passes through a canyon between Pashoore and Rangrokh Dogan in the north, and three mountains Gharedagh, Asbgiroon and Zarin Kooh in the south. After watering the villages of Najafdar, Behan, and Asoor, and then combining with a current in the northwest, two kilometers north of where Vashtan villag joins Farahrood, and subsequent passing through Selebon village, the Namrood River crosses the Tehran-Firoozkooh road. After watering Harland and Hamze villages, 14 km southwest of Firoozkooh, the Namrood joins the Firoozkooh River to form the Hablehrood River (Atef yekta, 2005).

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The population of Arjmand city is 1701. The population of the surrounding county and villages is 8822. There are 9 fish hatchery centers in the region, and the area of hatchery farming is 750.10 m^2.

In graduate thesis entitled, "Inspection and measuring heavy metals in water and sediments of the Sepidrood River," the occurrence of metals such as chromium, mercury, cadmium, arsenic, and lead was investigated. Except for arsenic, the river was found to be polluted with all of these elements (Soleymani Mokhtari, 1996).

The study, "Comparison measurement of concentration of lead and zinc in sea fish, and hatchery fish," found that the zinc concentration in south sea fish is higher than Caspian sea fish , and that the concentration of lead in hatchery fish muscle is relatively high (Asgari Sari, 2002).

Similar issues have been uncovered in Portugal. In Soares " Sediments as monitors of heavy metal contamination in the Ave River basin (Portugal), " the accumulation of heavy metals Cd, Cr, Cu, Ni, Pb, and Zn in sediments was determined. By analysis and characterization of heavy metals pollution, a proper relation among all heavy metals at the sampling stations was specified (Soares et al., 1999).

The Danube River flows through highly industrialized nations. " Analysis and assessment of heavy metal pollution in suspended and sediments of the river Danube " addresses the rates at which some heavy metals such as Al, As, Cd, Cr, Cu, Fe, Pb, Mn, Hg, Ni, and Zn are found in suspended materials and sediments of the Danube River and its branches. Results showed a high concentration of cadmium in the Danube (Woitke et al., 2003).

Fetlen et al. (2007) reported in " Physiological and behavioral responses of *Gammarus pulex* (Crustacea, Amphiphoda) exposed to cadmium " about the effect of cadmium on *Gammarus pulex*'s physiological, and behavioral responses. In this study, cadmium LC_{50} was evaluated at different times, and then physiological and behavioral responses of *Gammarus pulex* were studied. *Gammarus pulex* exposed to cadmium under laboratory conditions was investigated as well.

In "Persistence of *Chironomids* in metal polluted environment," accumulation form of *Chironomidae* stations in Belgium's Dommel River were compared with cadmium pollution in individual larva. The Dommel River has been adversely affected by farming livestock, and industrial pollution. It has been shown that *Chironomidae* have great ability in governing cadmium-polluted environments, while the rest of the toxins may be omitted in such an environment (Boonstra et al., 2009).

MATERIALS

Sampling stations

Station 1 (the upstream station near Arjmand village) was chosen for being near a village in which there is the possibility of being affected by habitat sewage, i.e., farming, running water polluted by fertilizers and pesticides. This station was situated at 35° 47' N, 52° 30' E, at an altitude of 2101 meters.

Station 2 (the downstream station) was selected for inspection of the river's water quality because there the river's flow mixes with fish hatcheries' waste water. The station lies at 35° 43' N, 52° 39' E, at an altitude of 1819 meters.

Sampling method

After being evaluated, two stations in upstream and downstream of the river in the cold and hot seasons of the year (mid- March and June) were selected for, *Chironomidae* and *Gammarus pulex* sediments sampling. For macrobentos sampling, then the most current available method was used in accordance to EPA (2006) instructions. After researchers were situated at each stations, samples were taken off the riverbed by 0_1m^2 grab. Each sample was washed in the river's current, using a 500 micron sieve. The washed samples were stored in polyethylene containers, or plastic bags, and were maintained in an icebox containing ice. They were transferred to the laboratory after recording the specifications. The rocky riverbed stations were sampled 3 times via the Surber sampling device (Burkle GmbH, Bad Bellingen, Germany).

Sample preparation

In the preparation of tissue samples, the MOOPAM 1999 instructions were used. For sediment samples, the ROPME, 1989 instructions were adhered to.

Tissue sample preparation

The tissue samples were taken out from the freezer and allowed to thaw under laboratory conditions. Each station's samples were identified by Maccafferty, 1981 and Pennak, 1978 keys, dried in a 60 $^{\circ}C$ oven for 24 h, then made into a uniform powder by agate mortar. One gram of powdered sample was precisely weighed and added to an Arlene Mayer 8 ml flask of 65%nitric acid, to which 4 ml of 70% perchlorate acid was added. The samples were maintained in the hood overnight and heated the next day to 120$^{\circ}C$. After cooling down, they were filtered through Whatman paper filter, and reached a volume of 200 ml. The samples then were determined through atomic flame absorption spectrophotometer.

Sediment sample preparation

The sediment samples were taken out from the freezer, and allowed to thaw under laboratory conditions. A specific amount of sediment was placed in a 60 $^\circ C$ oven for 24 h to dry out. Then particles below 63 microns were sieved by using a special sieve. The sieved sediment was made into a uniform powder in an agate mortar. Two grams of the powdered sediment sample was precisely weighed and added to an Arlene Mayer 8 ml flask 65% nitric acid, and 4 ml 70% perchloride acid was slowly added, and was digested at 120 $^\circ C$ for 3 hours. The samples were filtered by paper filter and in a volumetric flask, by adding twice-distilled ion less distilled water, and reached a volume of 200 ml. The samples were ready to be presented for analysis.

Data Analysis

Data analysis was performed with SPSS and Excel software. The one- way analysis of variance test was done by ANOVA, plus Duncan comprehensive tests, LSD, and mean modulus $P<0.05$.

RESULTS AND DISCUSSION

In this section, the average concentration of heavy metals from different stations has been revealed.

The bar graph is a representative of the average of samples which has been shown as a number in the upper part of each bar.

It can be claimed that the upstream in cold season has the highest amount of Cadmium rather than others. The downstream sediments in hot season have the highest amount of lead.

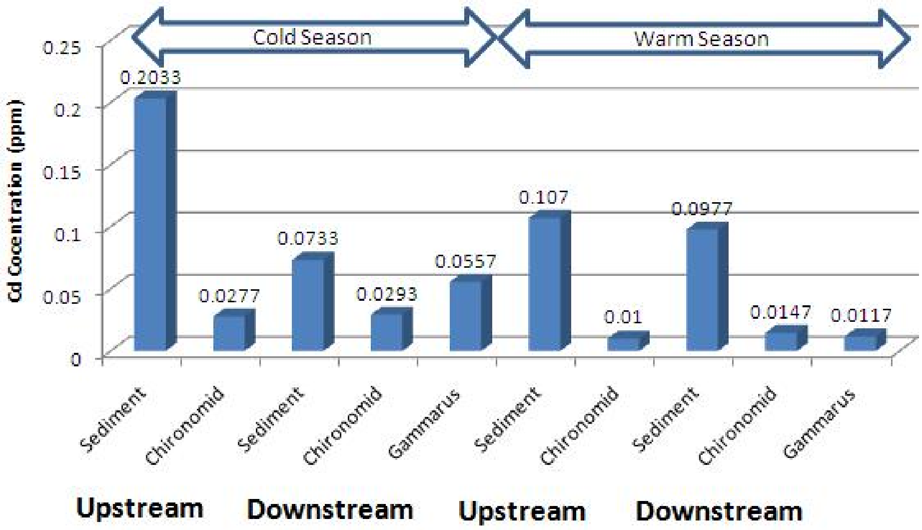

Figure 1: Average concentration of cadmium

In the cold season, existing cadmium in the upstream and downstream sediments displayed a significant difference ($P<0.05$). But the concentration of this metal in *Chironomidae* from the upper and lower stations in hot and cold seasons did not show any significant difference statistically.

In the hot season, both upper and lower stations sediment concentration in *Gammarus* in cold seasons has been greater than in hot seasons ($P<0.05$). In every one of the inspected stations, also the cadmium concentration has always been less in concentration of this metal in sediments and macorbento.

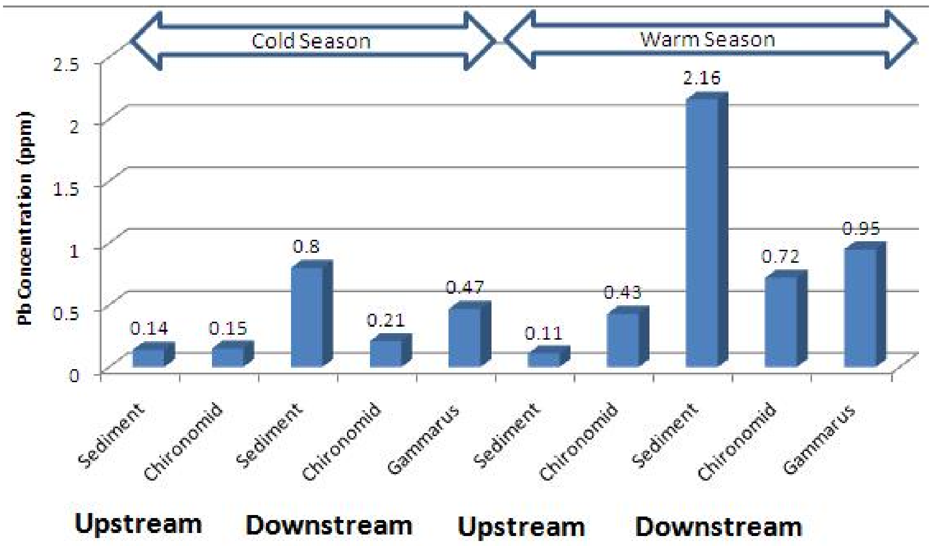

Figure 2: Average concentration of lead

In cold season, no significant difference was seen in concentration of this metal in sediment, and *Chironomidae* at the upstream. But, at downstream in cold season, the concentration of lead in sediments was four times more than in *Chironomidae* ($P<0.05$). So the lead in sediments at the upper station was lower than other samples. In other words, the concentration of this metal in sediments at downstream was measured as twenty times higher than at the upstream ($P<0.05$). In addition,
the accumulated lead in *Chironomidae* at the lower station was more than at the upstream ($P<0.05$). The accumulated amount of lead in *Gammarus* found in the hot season was more than in the cold season ($P<0.05$). The amount of lead in sediments has had significant difference only at the lower station in the cold season relative to other sediments ($p<0.05$).

DISCUSSION

As shown in obtained results, the sediments at the upstream in mid-March have the highest concentration of cadmium among the samples (0.2033 ppm). The lowest amount of cadmium belongs to *Chironomidae* at the upper station in mid-March (0.0377 ppm), *Chironomidae* at the lower station in mid-March (0.0293 ppm), *Chironomidae* at the upper station in June (0.010 ppm), *Chironomidae* at the downstream in June (0.0147 ppm), and *Gammarus pulex* at the lower station in June (0.0117 ppm). Perhaps we witnessed a high average concentration of cadmium in sediments at the upper station compared with other samples because of pollution by habitat sewage in nearby Arjmand village, and through the use of organic fertilizers, chemical fertilizers and pesticides at farms around the river.

However, lead, not cadmium, has the highest average concentration in sediments at the lower station in June (2.16ppm). The lowest average concentration was observed from sediments at the upper station in mid-March (0.14 ppm), and *Chironomidae* at the upper station in mid-March (0.11ppm). High lead concentration at the lower station, seen in June, is caused by a higher rate of traffic in the summer, and using the riversides for entertainment. We should also consider that the Firoozkooh Department of Transportation is located near the lower station, so that lead created by fuel combustion and oil changes would be mixed with river water by rain water in the rainy season, or is disposed in the river directly, and would accumulate in sediments.

Table 1: heavy metals in water and sediments of the Sepidrood River

Metal	Water(ppm)	Sediment(ppm)
Pb	0.18	42.6
Cd	0.06	0.33

Most pollution of cadmium is caused by farming toxins, chemical fertilizers and habitat sewage. The main source of pollutant in lead-polluted river is fuel combustion; it can be added by rain and snow which bring polluted air to the ground and surface water (Solaymani Mokhtari, 1996).
In this study, also the high cadmium concentration in sediments at the upper station with respect to other samples may be caused by habitat sewage and farming waste water. And also high lead concentration in sediments at the lower station probably is caused by exhaustion of automotive fuel combustion, because of being near the road. The upper station, distant from the road is not affected by the autos' fuel combustion.

Table 2: the lead concentration in sea fish and hatchery fish (ppm)

Sciaenidae	0.86
Mugilidae	0.56
hatchery fish	11.5

This table reveals that pollution in most marine creatures at the bottom levels is higher than in the upper levels of water. Furthermore, lead in muscles of bottom fed-fish such as *Mugilidae* and *Sciaenidae* is higher than in other fish. The reason for high lead concentration in hatchery fish muscle such as trout is possibly because the hatchery is near the road in which the used water has a high lead concentration (AsgariSari, 2002).

In this study, macorbentos are used as a biomonitor because of their characteristics. One of these characteristics is being bottom-fed creatures. Regarding lead average concentration, the highest average concentration belongs to sediments at the downstream in the summer, and this is because the downsream is close to the road, where cars stop to rest and use the riverside as a recreation area in summer.

Table 3: heavy metals concentration in sediment

Metal	greater than 63 μm(gkg^{-1})	smaller than 63 μm(gkg^{-1})
Pb	0.86	0.51
Cd	0.115	0.160

The results of this study show that sediments greater than 63μm could be a good indicator for determining pollution of heavy metals under study (Soares et al., 1999).

Table 4: heavy metal pollution in suspended and sediments of the river Danube

Metal	Sediments(μgg^{-1})	Suspended(μgg^{-1})
Pb	2.7	1.4
Cd	48.6	17.8

The results show that the heavy metals concentration pattern in suspended solid particles and sediments are very similar (Woitke et al., 2003). In the present study, sediments are used as one of the indicators that could express the river's pollution status as well.

Chironomidae has higher ability with respect to other toxins in governing cadmium-polluted environment. The mechanism which allows them to be present in such polluted environment is not completely known yet, and physiological changes might be responsible for this matter. Perhaps, when *Chironomidae* is exposed to cadmium distribution, a metallothionein-like protein is involved. As a result, *Chironomidae* could be used as an indicator of an environment which most creatures could not tolerate (Boonstra et al., 2009).

In the present study, *Chironomidae* was present in all sections of river path which being monitored.

CONCLUSION

The stable sediments and animals are more appropriate indices than water in order to do the ecological assessments of water pollution. If the sediment has the smaller grain size, it can be less affected by the sudden changes of water. Also, the water physic-chemical factors and environmental situation can influence the river and its aquatic animals.

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