# MUSSELS (Mytilus galloprovincialis) AS BIOINDICATORS OF PRESENCE OF HEAVY METALS IN BOKA KOTORSKA BAY

Andraš Š. Štajn<sup>1</sup>, Radoslav V. Žikić<sup>1</sup>, Ranđel P. Mihajlović<sup>1</sup>, Sreten Mandić<sup>3</sup>, Branka I. Ognjanović<sup>1</sup>, Slađan Z. Pavlović<sup>2</sup>, Zorica S. Saičić<sup>2</sup>, Vojislav M. Petrović<sup>2</sup>

<sup>1</sup> Faculty of Sciences, University of Kragujevac, Kragujevac <sup>2</sup> Institute for Biological Research "Siniša Stanković", Belgrade <sup>3</sup>Institute of Marine Biology, Kotor E-mail: Stajn@knez.uis.kg.ac.yu

#### **ABSTRACT**

Rapid urbanization and eutrophization, disturbing normal ecological equilibrium of water ecosystems and directly endangering living organisms is the main problem in Boka Kotorska bay. Mediterranean mussel (Mytilus galloprovincialis, Bivalvia, Mollusca) is a species which naturally inhabits the waters of Boka Kotorska bay, but it is artificially bred and increasingly used in human diet. We established that the presence of heavy metals in sea water even in allowed concentrations caused increased accumulation in tissues of mussel (Cu, Mn, Cr, Ni, Al, Mg, Zn, Cd, Pb) pointing to a numerous negative consequences not only in those organisms but in humans, potential consumers of mussels as well.

Key words: Boka Kotorska, mussels, metals.

## DAGNJE (Mytilus galloprovincialis) KAO BIOINDIKATORI PRISUSTVA TESKIH METALA U BOKOKOTORSKOM ZALIVU

#### **REZIME**

Osnovni problem u Bokokotorskom zalivu predstavlja ubrzana urbanizacija i eutrofizacija, zbog čega se bitno remeti normalna ekološka ravnoteža vodenih ekosistema i direktno ugrožava živi svet. Mediteranska dagnja (*Mytilus galloprovincialis, Bivalvia, Mollusca*) predstavlja vrstu koja prirodno naseljava vode Bokokotorskog zaliva, ali se i veštački uzgaja i sve više koristi u ljudskoj ishrani. Ustanovili smo da prisustvo teških metala u morskoj vodi, i u dozvoljenim koncentracijama, izaziva povećanu akumulaciju u tkivima dagnje (Cu, Mn, Cr, Ni, Al, Mg, Zn, Cd, Pb) i ukazuju na niz negativnih posledica, kako u tim organizmima, tako i kod ljudi kao potencijalnih konzumenata školjki.

Ključne riječi: Boka Kotorska, dagnje, metali

#### INTRODUCTION

Geographically and oceanographically, Boka Kotorska Bay (Figure 1.) represents a closed basin with specific hydrographic and dynamic characteristics. In comparison to open part of Montenegro coast, this narrow aquatic area shows a large number of specific differences in some climatological, hydrological and hydrographical elements. The basic problem in Boka Kotorska Bay is the rapid urbanization and eutrophization which led to disturbed ecological equilibrium of aquatic ecosystems and directly endangers living world, (Doklestić, 1996).

In addition to rainfalls, waters from river basin area and freshwater of springs, about 5 · 10<sup>6</sup> m³ of waste water are added per year in this area, which amounts about 0,2 % from total water mass of Boka Kotorska Bay.

Increased pollution of sea water by metals occurs near industrial plants, shipyards and other light industrial area.

In the bay there are 62 sewerage system outputs, zone for reloading and storage of gasoline in Lipci, as well as numerous city harbours (Zelenika, Kotor, Herceg-Novi, Risan, Bonići, Meljine, Tivat, etc). All these places represent a potential danger for plants and animals in the bay. The greatest dangers are the shipyard in Bijela and Maintinance Institute in Tivat, where periodically relase of waste substances in sea water occurs due to damage (e.g. crude oil, dyes, etc.), (Doklestić, 1996). In waste substances released in water of Boka Kotorska Bay some heavy metals can be found which may cause different negative consequences for living world. Heavy metals are accumulated in tissues causing disturbances of oxidative status. Moreover, metals induce the forming of proteins with low molecular mass called metalothioneins, (Gould & Karolus, 1974; Hoghstrand *et al.*, 1996; Žikić *et al.*, 1997).

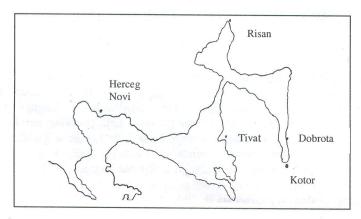


Figure 1. The map of Boka Kotorska Bay

Mediterranean mussel (*Mytilus galloprovincialis*, *Bivalvia*, *Mollusca*) is the species which naturally inhabits waters of Boka Kotorska Bay. This species is edible and is bred artificially. Mussels are fed by filtration of water, so their gills are structurally modified both to feeding and respiration. In order to enhance feeding, gills are supplied with numerous cilia which direct the food particles toward digestive tract.

In the process of feeding and respiration, mussels take metals into the organism via gills. Metals are than transported to tissues where there are accumulated, particularly in gills, hepatopancreas and muscle. Due to these and accumulation of other waste substances, mussels may cause harmful consequences if they are used in human nutrition. The content of heavy metals in mussels depends on the amount of metals in surrounding water, salinity, (Struck *et al.*, 1997), as well as on seasonal conditions with maximum accumulation in winter and minimum in summer period, (Regoli, 1998). Therefore, mussels are suitable bioindicators of pollution with heavy metals and recently they have been used as test-organisms, (Ostapczuk, 1997; Szefer, 1997).

Special attention in scientific investigation should be focused on biology and ecology of fish and other marine organisms which are potentially important in human nutrition, that is on production of healthy food through organized mariculture. It is also necessary to monitor the quality of sea water and prevent its pollution.

The aim of this investigation was to determine the content of some metals (Cu, Cr, Fe, Zn, Ni, Mn, Pb, Cd, Al, Ca, Mg) in different tissues of mussels (whole shell, gills, foot muscle and rest of shell) and to compare obtained results with the results of other investigators.

## MATERIAL AND METHODS

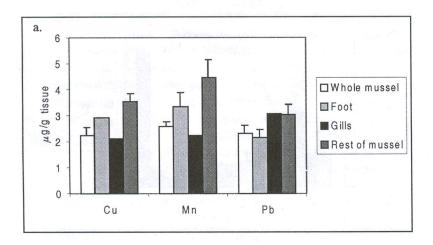
Mussels were collected at three localities (Figure 1) in the vicinity of Institute of Marine Biology in Kotor. Immediately after collecting, mussels are determined, dissected and the tissues where frozen in saline. Following tissues were selected for analysis: whole mussel, gills, foot muscle and the rest of soft tissue of mussel. Material was destroyed in the mixture of nitric and perchloric acid (in 17:3 ratio) with heating in sand bath, (Shirley *et al.*, 1949). Concentrations of metals in tissues were determined by using atomic-absorption spectrophotometer (Perkin Elmer, model 3300).

## RESULTS AND DISCUSSION

Concentrations of metals in different tissues and in the rest of mussels are shown in Figures 2a, b and 3a, b.

The highest concentrations of copper and manganese were detected in rest of mussels as well as in foot. The obtained values are significantly higher than the concentrations of copper in blood cells and other tissues of sole (Pleuronectes platesa) determined by Harvey (1972). Concentrations of manganese are also slightly higher than concentrations obtained in tissues of carp in natural freshwaters, (Žikić et al., 1988). Distribution of lead and cadmium shows that these metals are maast present in gills and rest of mussels. Results obtained in this work are in accordance with results obtained by Ashworth & Farthing (1981) in the area of Port Philip (USA). Results of Pagenkopf et al. (1972) concerning the muscle of fish, show much lower values of lead concentration in comparison to values obtained in this work. Due to the specific way of life and feeding, mussels are susceptible to accumulation of metals in organism. The increased content of some metals (Cd, Pb) was found by other investigators, (Szefer, 1997), who studied mussels in high industrialized area of Kyushu Island in Japan. On the other hand concentrations of nickel and chromium are significantly decreased when compared with results obtained in shells in natural waters of other regions, (Moor & Ramamurti, 1987).

Concentration of zinc in analyzed mussels is also decreased in comparison to values obtained by Goldberg *et al.* (1978) in species *Mytilus edulis* collected from coastal waters of USA. Results obtained in this work show that the concentration of aluminium is the highest in the rest of soft tissue of mussels, which is in accordance with the results obtained for iron as well. Concentration of calcium obtained in this work is almost equailized in all examined tissues. The content of magnesium is the highest in the rest of tissue of mussels, whereas in foot and gills it is at the same level for whole shell.



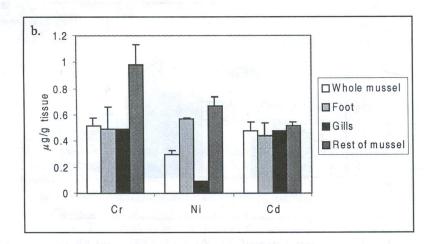
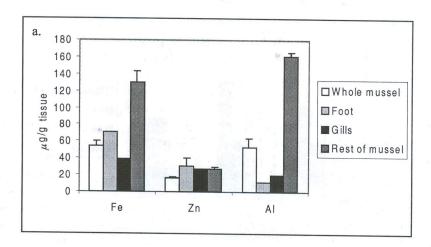


Figure 2a,b: The concentrations of Cu, Mn, and Pb (a) and Cr, Ni and Co (b) in investigated tissues of mussels



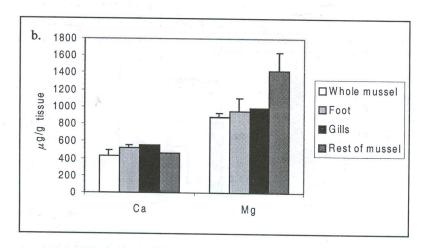


Figure 3a,b: The concentrations of Fe, Zn, and Al (a) and Ca and Mg (b) in investigated tissues of mussels

#### **CONCLUSION**

On the base of obtained results it can be concluded that the presence of heavy metals in sea water, even in allowed concentrations causes increased accumulation in tissues of mussel (Cu, Mn, Cr, Ni, Al, Mg, Zn, Cd, Pb) and points to numerous negative consequences in those organism and in humans as potential consumers of shells as well. Concentration of some metals (Ni, Cr, Zn) in tissues of mussels is lower in comparison to values obtained by other investigators, which depends on geographic area where the shells were collected. On the other hand, concentrations of some metals (Cu, Mn, Pb) are higher in tissues of shells from Boka Kotorska bay than concentrations obtained by other investigators. The differences in concentrations of some metals in tissues of mussels can be explained by seasonal variations and by differences in salinity as well. The increased content of metals in tissues of mussels may point to increased contamination of water by metals which is the consequence of rapid industrialization. Heavy metals induce numerous biochemical and physiological disturbances including the disturbance between reactive oxygen species and antioxidative defense system. In that way the quality of food is diminished, which is proved by presence of antioxidants.

#### REFERENCES

- Ashworth, M.J. and R.H. Farthing (1981): International Journal Analitical Chemistry, 10: 35. *In*: Thomas R. Crompton. *Ed.* Analysis of solids in Natural Waters, Springer-Verlag, Berlin, 1996.
- Chisela, F., D. Gawlik and P. Bratter (1986): Analyst (London) 111: 405. In: Thomas R. Crompton. Ed. Analysis of solids in Natural Waters, Springer-Verlag, Berlin, 1996.
- Doklestić, O. (1996): Akcidentne situacije morskog (obalnog) pojasa od Ulcinja do Igala. Zaštita voda: 32-37.
- Goldberg, E.D., V. T. Bowen, J. W. Farrington, G. Harvey, J. H. Martin, P. L. Parker, R. W. Risebrough, W. Robertson, E. Schneider and E. Gamble (1978): The Mussel Wach Environ Conserv. 5: 101. *In*: Thomas R. Crompton. *Ed*. Analysis of solids in Natural Waters, Springer-Verlag, Berlin, 1996.
- Gould, E. and J. J. Karolus (1974): Physiological response of the cunner, *Tautogolabrus dispersus*, to cadmium. *NOAA Tech Rep*, NMFS, SSRF., **681**: 21-25.
- Harvey, B.R. (1972): Analytical Chemistry **50**: 1866. *In*: Thomas R. Crompton. *Ed*. Analysis of solids in Natural Waters, Springer-Verlag, Berlin, 1996.
- Hoghstrand, C., F. Galvez and C. M. Wood (1996): Toxicity, Silver Accumulation and Metallothionein Induction in Fresh-Water Rainbow Trout During Exposure to Different Silver Salts. Environ. Toxicol. Chem., 15 (7): 1102-1108.
- Moor, D.V. and S. Ramamurti (1987): Heavy metals in natural waters. Moskva Mir, 1-285.

- Ostapczuk, P., J. D. Schladot, H. Emons, K. Oxynos, K. W. Schramm, G. Grimmer and J. Jakob (1997): Environmental monitoring and banking of marine pollutants by using common mussels. Chemosphere., **34** (9-10): 2143-2151.
- Pagenkopf, G.K., D. R. Neuman and R. Woodruff (1972): Analytical Chemistry, 44: 2248. *In*: Thomas R. Crompton. *Ed.* Analysis of solids in Natural Waters, Springer-Verlag, Berlin, 1996.
- Regoli, F. (1998): Trace metals and antioxidant enzymes in gills and digestive gland of the Mediterranean mussel *Mytilus galloprovincialis*. Arch. Environ. Contam. Toxicol., **34** (1): 48-63.
- Shirley, R.L., W. J. Beune and E. J. Miller (1949): Cadmium in biological materials and food. Anal Chem., 21: 300-303.
- Struck, B.D., R. Pelzer, P. Ostraoczuk, H. Emons and C. Mohl, (1997): Statistical evaluation of ecosystem properties influencing the uptake of As, Cd, Co, Cu, Hg, Mn, Ni, Pb and Zn in seaweed (*Eucus vesiculosus*) and common mussel (*Mytilus edulis*). Sci. Total. Environ., **14: 207** (1): 29-42.
- Szefer, P., K. Ikuta, S. Kushiyama, K. Szefer, K. Frelek and J. Geldon (1997): Distribution and Association on Trace Metals in Soft Tissue and Byssusof Mytilus edulis from the East Coast of Kyushu Island, Japan. Arch. Environ. Contam. Toxicol., 32 (2): 184-190.
- Žikić, R.V., A. Štajn and R. Živković (1988): The activity of superoxide dismutase and catalase in the tissues and erythrocytes of the carp (*Cyprinus carpio* L) exposed to manganese. Iugoslav Physiol Pharmacol Acta., **24** (3): 273-281.
- Žikić, R.V., A. Š. Štajn, B. L. Ognjanović, S. Z. Pavlović, and Z. S. Saičić, (1997): Activities of Superoxide Dismutase and Catalase in Erythrocytes and Transaminases in the Plasma of Carps (*Cyprinus carpio L.*) Exposed to Cadmium. Physiol Res., **46**: 391-396.