# THE INFLUENCE OF THE BIOMETRIC PARAMETERS ON METAL AND METALLOTHIONEIN CONTENT IN THE CYTOSOL OF THE WHOLE SOFT TISSUE OF TRANSPLANTED MUSSELS

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### Abstract

The influence of two biometric parameters on metallothionein (MT) level in the cytosol of mussel's whole soft tissue was studied over the period of one year, as well as the correlation between the cytosolic levels of five metals (Cd, Zn, Cu, Mn and Fe) and MTs. MTlevel in the cytosol significantly depends on the mussel's physiological changes, but also positively correlates with Cd and Mn content. The correlation with Cd reflects the known Cd affinity for binding to MTs, while correlation of MTs with Mn is probably the reflection of the strong influence of soft tissue condition index on both parameters.

*Key words: Mytilus galloprovincialis; trace metals; metallothionein; biometric parameters* 

#### Introduction

Metallothioneins (MTs) can function as storage proteins for essential metals, or as chelating agents to bind toxic metals, and essential metals when they are present in excess [1]. Although the synthesis of MTs depends on metal uptake in organism, other factors can influence it, as food availability or the reproductive cycle. Thus, the aim of our study was to define the connection between biometric parameters, MTs and metal content in mussels caged over one year.

# Experimental

In September 1997, mussels Mytilus galloprovincialisof defined length (5.1±0.2 cm) and age (12±1 months) were implanted for one year in Kaštela Bay, and sampled monthly and bimonthly, from October 1997 to September 1998. As biometric parameters, soft tissue condition index (dry mass of whole soft tissue x 100 / shell mass), which varies during the reproductive cycle and with the food abundance [2], and the shell mass which reflects the aging process of mussels [3], were determined. Both MTs and metals were analyzed in the heat treated cytosolic fraction (S30) of the whole soft mussel tissue, and expressed as cytosolic tissue burden (mg and respectively). MTs were analyzed by electrochemical technique [4], and metals by flame atomic absorption spectrometry.

## **Results and discussion**

The cytosolic tissue burdens of metals were ranked as follows: Zn>Fe>Cu>Mn>Cd. MTcytosolic tissue burden was comparable to the level of Cu.

Multiple regression analysis, explaining the influence of biometric parameters on the contents of all analyzed metals and MTs (Table 1), showed that the variability of Fe, Zn and Cd contents could only be slightly explained by biometry, and mostly as a consequence of mussel's age, while the changes of Cu content could not be attributed to the changes of neither of biometric parameters.

The strongest impact of biometric parameters was observed in the case of MTs and Mn. MTcontent could be related both to the aging process of mussels and to the changes of the soft tissue condition index. Although MTis a cellular ligand involved in homeostasis and detoxification of metals, it is also a protein, and it may be assumed that any factor which affects general protein metabolism may also affect MTlevel. For example, the higher food availability in the environment, which enhances somatic growth and, thereby, also the synthesis of total proteins, would be expected to affect the quantity of MTs, too [2]. Unlike MTs, Mn reflects only the changes in condition index of mussel's whole soft tissue. Strong, positive correlation between Mn and condition index (r=0.75; p<0.00001), indicates that this metal precisely follows the changes in tissue mass and makes an excellent indicator of mussel's physiological changes. Thus, it is not surprising that Mn positively, statistically significantly correlates with MTs (Fig. 1). Both parameters probably have important role in the physiological functions of mussel's tissues, which causes them to vary in accordance with the changes of the whole soft tissue mass. Frias-Espericueta et al.[5] suggested that Mn and protein content are influenced by reproductive cycle, and both increase with the gonadal maturation.

The fact that MTcontent increases with mussel's age, too, explains its positive correlation with Cd content (Fig. 1), which also shows increasing trend over the year. Since the accumulation of Cd takes place simultaneously with the enhanced synthesis of MTs, it is possible that Cd accumulation is a consequence of the age-induced MTsynthesis, as it is known that metal uptake can be enhanced by the synthesis of metal-binding proteins, like MTs [6].

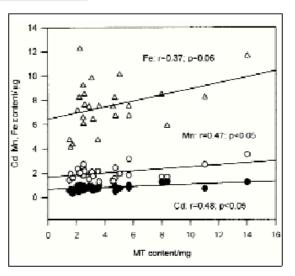


Fig. 1. Linear regression graphs: (MTs vs. Cd, and | MTs vs. Mn.

Table 1. Multiple linear regression analysis explaining the in?uence of biometric parameters on metal and MT content: p values for each biometric parameter and R values for combination of two biometric parameters.

	Mar' PR	Fø/ P8	RK CUV	Za/ PS	Ca/ PR	MT/ #8
Shell muss/g	0.037	0.007	0.029	0.081	0.704	< 0.001
p-value	_					
Soft tissue condition index.%	<0.001	0.103	0.951	0.765	0.861	<0.001
p-value						
R	0.61	0.23	0.14	0.06	0	0.39

#### References

1-Carpene E., 1993. Metallothionein in marine molluscs. Pp. 55-72. In: Dallinger R., and Rainbow P.S. (eds.), Ecotoxicology of metals in invertebrates. Lewis Publishers, Boca Raton.

2-Mourgaud Y., Martinez E., Geffard A., Andral B., Stanisiere J.Y., and Amiard J.C., 2002. Metallothionein concentration in the mussel Mytilus galloprovincialisas a biomarker of response to metal contamination: validation in the field. Biomarkers, 7: 479-90.

3-Fischer H., 1983. Shell weight as an independent variable in relation to cadmium content of molluscs. Mar. Ecol. Prog. Ser., 12: 59-75. 4- Raspor B., Paic M., and Erk M., and Erk M.,2001. Analysis of

4- Raspor B., Paic M., and Erk M., and Erk M.,2001. Analysis of metallothioneins by the modified Brdickaprocedure. Talanta, 55: 109-15. 5-Frias-Espericueta M.G., Osuna-Lopez J.I., and Paez-Osuna F., 1999. Gonadal maturation and trace metals in the mangrove oyster Crassostrea *corteziensis: seasonal variation. Sci. Total Environ.*, 231: 115-23. 6-Langston W.J., Bebianno M.J., and Burt G.R., 1998. Metal handling strategies in molluscs. Pp. 219-284. In: Langston W.J., and Bebianno M.J. (eds.), Metabolism of trace metals in aquatic organisms. Chapman & Hall, New York.

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