Buletinul Ştiinţific al Universităţii "Politehnica" din Timisoara, ROMÂNIA
Seria CHIMIE ŞI INGINERIA MEDIULUI
Chem. Bull. "POLITEHNICA" Univ. (Timişoara) *Volume 50 (64),1-2,2005*

Pesticides and Heavy Metals Determination in Marine Organisms from Black Sea

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Abstract: This work presents aspects regarding the pollution with pesticides and heavy metals (Cd, Pb, Cu and Zn) from mussels tissue (*Mytilus galloprovincialis*) collected from different zones of the Romanian Black Sea Coast: Mamaia, Tomis Harbour and Agigea. The heavy metals were analysed using atomic absorption spectrometry. For the pesticides analysis a FISONS gas cromatograph was used equipped with an electron capture detector and a capillary cromatograph column filled with mixture of silicone oils on chromosorb WHP.

Keywords: mussels, pesticides, heavy metals

1. Introduction

Pesticide and heavy metals are persistent and non-biodegradable and they can be bioaccumulated through the biologic chains: soil-plant-food and seawater-marine organism-food [1]. So, the presence in high amount of pesticide and heavy metals in environment represents a potential danger for human health and for environment due to their extreme toxicity. For this reason, accurate monitoring of their concentration plays an important role. Population can be contaminated with organic pollutants and heavy metals by ingestion of contaminated or polluted food and water. The gravity of toxic effect depends on nature, concentration, body resistance and presence of other contaminants [2]. The concentration of this elements in food products is varied, depending of their origin, storage conditions and processing technologies.

In literature there are mentioned many methods for heavy metals determination in soils, phosphorus rocks, seawater, plants, biologic materials, steel and cast iron through: inductive coupled plasma - mass spectrometry [3], inductive coupled plasma - atomic emission spectrometry [4], atomic absorption spectrometry with flame or electrothermal atomization [5], electrochemically with ultramicroelectrodes [6], anodic stripping voltammetry [7].

Because of the large scale dilution of organic contaminants in the aquatic matrices, concentrations of many organic pollutants are below the detection limits of standard analytical and sampling methods. Thus, gas chromatography with specific detection methods such as electron capture detector and HPLC has been frequently used for determination of pesticides and polycyclic aromatic hydrocarbons in water samples [8, 9].

Mussels have a high nutritive value as a result of them complex composition, rich in biological active components: essential amino acids, hydrosoluble and liposoluble vitamins, fatty essential acids, microelements, etc.

On the other hand, because of their feeding mode, the mussels are valuable biological indicators since they concentrate microelements from aquatic environment and in consequence in the polluted zones, the contaminant content in mussels in high.

This in the reason we chose the determination of heavy metals and of pesticides in the composition of mussels, in order to locate the zones with high risk of pollution.

This work presents aspects regarding the pollution with pesticides and heavy metals (Cd, Pb, Cu and Zn) of mussels tissue (*Mytilus galloprovincialis*) collected from different zones of the Romanian Black Sea Coast: Mamaia, Tomis Harbour and Agigea. The heavy metals were analysed using atomic absorption spectrometry. For the pesticides analysis a FISONS gas cromatograph equipped with an electron capture detection was used and also, a capillary cromatograph column filled with mixture of silicone oils on chromosorb WHP.

2. Materials and methods

Mussels samples (adult exemplares) and seawater were collected on the Romanian Black Sea Coast at 1. Mamaia, 2. Tomis Harbour and 3. Agigea during August 2003. For determination of heavy metals concentration, the fresh tissue samples were washed, hashed, dried at 105 ^{0}C and mineralized and the shell samples were washed, dried at 105 ^{0}C, pulverized and also mineralized by wet digestion method (HNO_3 - H_2SO_4). About 0.5g of each dried tissue and shells power sample was predigested in 2 mL 65% HNO_3 for 24 hours at room temperature, then 2 mL of 98% H_2SO_4 were added and the mixture was digested in a VELP DK6 heating digester. After cooling, the solution was made up to 25 mL of deionised water. All used reagents were of analytical reagent grade (Merck). The resultant solutions were analysed with an atomic absorption spectrophotometer GBC-AVANTA (air / acetylene flame) in order to determine the heavy metals concentration: Cd (λ = 228.8 nm), Cu (λ = 324.7 nm), Zn (λ = 213.9 nm) and

Pb (λ = 217 nm). Two replicate determinations were done for each solution.

Pesticide residues were extracted from samples (total lipid extract from mussels tissue) with ether of oil and acetone and then were purified on fluorisil column with a layer of anhydrous Na_2SO_4. A total of 10 g fluorisil or aluminium oxide was packed in a glass column with ether of oil. Pesticides were eluted from the column with ethyl ether / ether of oil in the 20 mL fraction. The fraction was concentrated in KUDERNA-DANISH apparatus for concentrating to about 1 mL. For the pesticides analysis was used a FISONS gas cromatograph equipped with an electron capture detection (ECD) and a capillary cromatrograph column filled with mixture of silicone oils (QF-1, OV-11, XE-60) on chromosorb WHP. Conditions: a 1μL aliquot of the extract was injected; column temperature 200 ^{0}C; injector temperature 210 ^{0}C; detector temperature 250 ^{0}C; carrier gas: nitrogen at a flow rate of 4 mL/min.

3. Results and discussions

The heavy metals concentrations in the analysed samples are presented in the figures below:

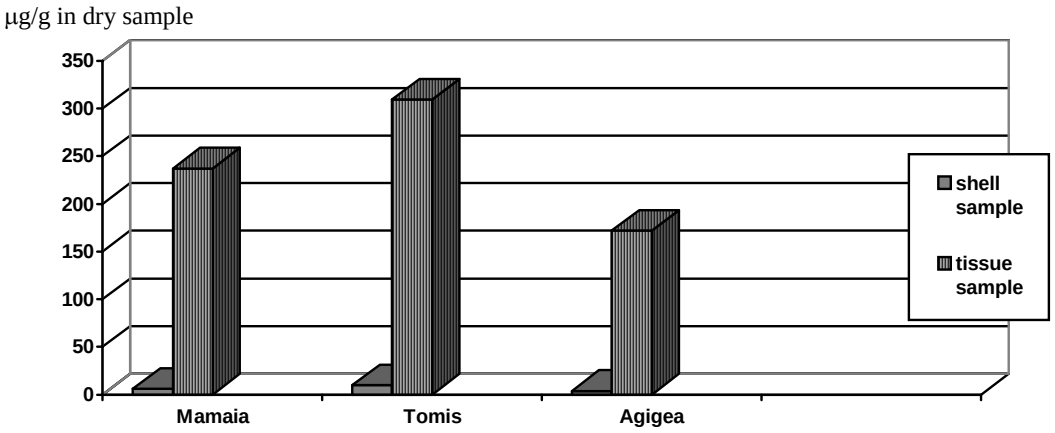

Figure 1. Concentration of Zn in the analysed samples

We remark from figure 1 the high concentrations of Zn in all analysed tissue samples, more over admitted limits [10]: 50 mg/kg.

The maximum admitted limits for Cu, Cd and Pb [10, 11] are: 5 mg/kg (Cu) and 1 mg/kg (Cd and Pb). The concentration of Cu is over admitted limits in samples collected from Mamaia (5.88μg/g) and Tomis Harbour (9.87μg/g). In all analysed tissue samples the concentrations of Cd and Pb are much more over the admitted limits, especially for Pb (fig. 2). The highest concentrations in heavy metals were detected in samples collected from Tomis Harbour: Cu 9.87 μg/g, Cd 1.71 μg/g and Pb 8.92 μg/g. The samples collected from Agigea presented the lowest content in heavy metals.

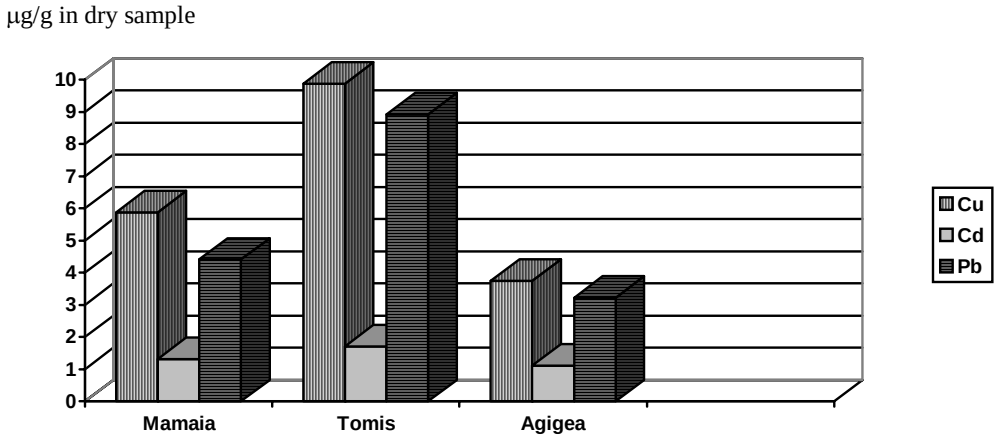

Figure 2. Concentration of heavy metals (Cu, Cd and Pb) in mussel tissue

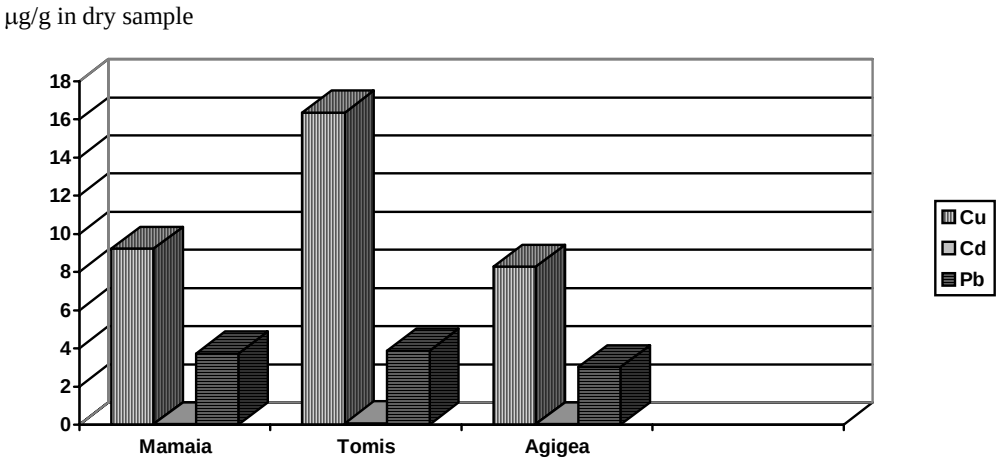

Figure 3. Concentration of heavy metals (Cu, Cd and Pb) in mussel shells

We remark from figure 3 that the shell samples have a low content in Cd (between 0.02 - 0.07 μg/g) but the concentration of Cu is much rise than tissue samples.
In tables 1, 2 and 3 are presented the concentrations of pesticides in mussel tissue:

TABLE 1. Concentrations of organochloride pesticides in mussel tissue from Mamaia

Pollutant	Concentration ppm	Maximal limit admitted ppm	Retention time (min.)	Area (counts)
γ HCH	0.1243	2	1.46	228945
β HCH	0.4820	0.1	1.85	410723
op DDE	0.0759	-	3.10	92467
op DDD	0.0007	-	4.69	1049
pp DDD	0.0023		6.19	3453
pp DDT	0.0010	1	7.34	1020

The data from Table 1 show an increase concentration of β HCH over admitted limit. The others are below maximal limits, but their presence indicates a contamination of marine environmental with such organic pollutants.

TABLE 2. Concentrations of organochloride pesticides in mussel tissue from Tomis

Pollutant	Concentration ppm	Maximal limit admitted ppm	Retention time (min.)	Area (counts)
γ HCH	0.2493	2	1.52	200141
β HCH	2.7449	0.1	1.93	829992
op DDE	0.0120	-	3.37	5761
op DDD	0.0285	-	3.97	56464
pp DDD	0.1342	-	6.25	98716
pp DDT	0.0823	1	7.11	28306

The data from Table 2 show the high concentrations of γ HCH and β HCH over admitted limits, especially at β HCH.

TABLE 3. Concentrations of organochloride pesticides in mussel tissue from Agigea

Pollutant	Concentration ppm	Maximal limit admitted ppm	Retention time (min.)	Area (counts)
γ HCH	0.1000	2	1.52	1204221
β HCH	0.2000	0.1	1.89	907143
op DDE	0.1000	-	3.22	717343
op DDD	0.4000	-	4.66	6838859
pp DDD	0.4000	-	6.24	4413641
pp DDT	0.4000	1	7.09	2064077

HCH = hexachlor cyclohexane; DDE = dichlor-diphenyl-dichlor-ethene
DDD = dichlor-diphenyl-dichlor-ethan; DDT = dichlor-diphenyl-trichlor-ethan

We remark from Table 3 a high concentration of β HCH over the admitted limits.

4. Conclusions

Concentration determination of heavy metals from the Black Sea mussels tissue presents a major importance, because in the sea are discharged the most effluents with high concentrations of pollutants and mussels having bioindicator properties. Mussels are biofilter organisms which retain small particles from sea water, so the presence of some pollutants in mussel tissue indicate a contamination of marine environment.

The concentration values of heavy metals are much over the admitted limits, their presence indicates a high degree of pollution and permits the identification of the principal contamination sources. We have detected a high concentrations of Pb and Cd in tissue sample, over maximal admitted limits which represent an alarming signal for human and environmental health. As concerning the presence of pesticides in mussel tissue, we have detected high concentrations of γ HCH and β HCH over admitted limits, especially at β HCH. The results show a high degree of pollution especially in zone of Tomis Harbour.

References:

1. Shawi A.W.al., Dahl R., *Anal. Chem. Acta*, **1999**, 391, 1, 35
2. Pablos Espada M.C., Garrido Frenich A., Martinez Vidal J. L., Parrilla P., *Analytical Let.*, **2001**, 34 (4), 602
3. Soniassy R., Sandra P., Schlett C., *Water Analysis*, **1995**
4. Liu W., Lee H. K., *Talanta*, **1998**, 45, 632
5. Hagestuen E.D., Campiglia A.D., *Talanta*, **1999**, 49, 548
6. Kalac P., Niznamska M. et al., *Sci. Total Environ.*, **1996**, 177, 251
7. Nusko R., *Fresenius J. Anal. Chem*, **1997**, 357, 1050
8. Liu W., Lee H. K., *Talanta*, **1998**, 45, 632
9. Hagestuen E.D., Campiglia A.D., *Talanta*, **1999**, 49, 548
10. *** *Ordinul MAP 975/98*, MO 268/98
11. *** *Ordinul MAP 352/2001*, MO 812/2001