LEVELS OF POLYCHLORINATED BIPHENYLS AND CHLORINATED INSECTICIDES IN THE LIVER AND MUSCLE TISSUES OF THE EUROPEAN CHUB (LEUCISCUS CEPHALUS) FROM THE SAVA RIVER (CROATIA)

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Introduction

The Sava River (945 km) is the biggest tributary to the Danube River. The 95551 km² large catchment is extended over Slovenia, Croatia, Bosnia and Herzegovina and Serbia and Montenegro. Although the methodological bases for data collection have been reasonably unified, data on the ecological character of the river basin, inventory of pollution sources, dangerous substances, socio-economic parameters, cost and benefit implications are still lacking due to the insufficient financing and the recent warfare.

Aquatic organisms can be used as biological indicators to monitor contamination of aquatic ecosystems with persistent organic contaminants (POPs) such as organochlorine pesticides (OCPs) and polychlorinated biphenyls (PCBs). Because of their strong lipophilic properties and their corresponding low solubility in water, it is much easier to determine POPs in the lipid fractions of aquatic organisms. The potential of fish as biomonitors was demonstrated by several monitoring studies.^{1,2,3,4} It has to be stressed out that relatively small number of the papers report about the exposure of the chubs to chlorinated hydrocarbons in the European rivers. This short paper is among the first reporting about the systematic tracking of these pollutants in European chub in the rivers of the Balkan area.

Materials and Methods

Sampling locations and samples

The sampling was carried out in the section from the Slovenian-Croatian border at Jesenice to the mouth of the Una River at Jasenovac (see stations on Figure 1 - downstream: Otok Samoborski, Jarun, Oborovo, Lukavec Posavski and Jasenovac). The sampling was postponed from February to the end of March 2005, due to cold weather. On both occasions, in March and September, high water flows caused sampling problems. Due to extremely high water level at the location of Jesenice, the fish sampling was not performed during the first field campaign. A sampling of fish with electricity was used as an efficient sampling method. Alive fishes were transported to the laboratory and dissected. It was proved that the fish catchment technique and the transport do not affect the survival of the caught specimens. Altogether 126 chubs were sampled (49 in spring and 77 in autumn of 2005).



Figure 1. Location of the sampling stations of E. chubs from the Sava River

Chemical analysis

Analyses were performed on composite samples, 9 of the muscle tissue and 8 of the liver. Pooled tissue samples were homogenized after addition of Na₂SO₄ anh. and extracted with n-hexane in a high revolution blender. In one third of the extract, extracted organic matter (EOM) was determined gravimetrically. In the other third of the extract, Mirex was added as an internal standard and analysis was performed. Separation of chlorinated insecticides from polychlorinated biphenyls was made on a silica gel column. Quantification of chlorinated hydrocarbons was performed by high resolution EC gas chromatography. After concentration down to 1 cm³, elutes were analyzed by an Agilent Technologies 6890N network GC system equipped with an electron capture detector (ECD). The column used was a high resolution glass capillary HP-5 (cross-linked 5% phenyl methyl siloxane), 30 m × 320 μ m × 0.25 μ m film thickness. Detailed descriptions of the methods used are presented in numerous published papers.⁶⁻⁸

Results and Discussion

The basic biometric data on fishes in analysed composite samples are presented in Table 1.

COLLECTION AREA		SAMPLING DATE	NO. OF FISH	TOTAL LENGTH	TOTAL MASS	SEX	% OF FAT	ON W.W.
				(cm)	(g)	(F/M)	MUSCLE	LIVER
1st SAMPLING	OTOK SAMOBORSKI	3/29/2005	15	21.1 ± 2.4	93.4 ± 35.4	2 M 13 F	0.8	2.4
	OBOROVO	3/30/2005	9	14.1 ± 1.8	25.2 ± 9.7	10 F	2.5	-
	LUKAVEC POSAVSKI	3/31/2005	10	13.2 ± 2.2	25.0 ± 16.2	10 F	1.6	10.9
	JARUN	4/1/2005	15	16.0 ± 3.2	45.1 ± 36.7	15 F	0.8	3.3
2nd SAMPLING	OTOK SAMOBORSKI	9/19/2005 and 9/26/2005	22	18.5 ± 3.8	67.6 ± 43.8	8 M 14 F	0.5	2.4
	OBOROVO	9/20/2005	15	21.0 ± 2.1	96.1 ± 29.1	5M 10 F	0.7	4.3
	LUKAVEC POSAVSKI	9/21/2005 and 9/28/2005	15	16.3 ± 1.3	39.9 ± 10.4	5 M 10 F	0.4	3.2
	JARUN	9/22/2005	15	19.5 ± 1.4	72.7 ± 17.4	7 M 8 F	0.7	3.1
	JASENOVAC	9/23/2005	10	19.9 ± 4.3	91.2 ± 66.9	5 M 5 F	0.3	3.5

Table 1. A basic biometric data of the analysed European chubs caught in the Sava River in 2005

Lengths and weights of the caught European chubs are relatively uniform but larger differences were found comparing amounts of the extracted lipid materials from the muscle and liver tissues of the samples.

Mass partition of organic pollutants is expressed on wet tissue mass (w.w.) and respectively on lipid mass (l.w.). Levels on the lipid mass are presented in mgkg⁻¹ and on the wet sample mass in μ gkg⁻¹.

The following ranges of PCB concentrations on the wet sample mass are determined: the sum of the seven key PCB congeners, Σ PCB7 (IUPAC: PCB 28, PCB 52, PCB 101, PCB 118, PCB 138, PCB 153 and PCB 180) in the muscle tissue 4.9-19.8 µgkg⁻¹ and in the liver 1.7-44.9 µgkg⁻¹; the sum of the DDTs (Σ DDT) in the muscle tissue 0.3-1.3 µgkg⁻¹ and in the liver 5.0-139.4 µgkg⁻¹. Determined ranges of PCB concentrations on lipid mass are: Σ PCB7 in muscle tissue 1.6-4.1 mgkg⁻¹ and in liver 0.05-1.2 mgkg⁻¹; Σ DDT in the muscle tissue 0.05-0.23 mgkg⁻¹ and in the liver 0.02-0.17 mgkg⁻¹ (Figures 2 and 3).

A smaller variability of the levels of $\Sigma PCB7$ is observed in the muscle tissue of the analysed fish samples (especially when the results are expressed on the lipid mass) compared to the liver. For both, the muscle and liver tissues, the levels of the ΣDDT are much lower compared to the level of PCBs. When expressed on the wet mass, levels of $\Sigma PCB7$ are decreased in the muscle tissues of the chubs caught during the second sampling campaign. On the contrary, when the results of the second campaign are presented on the lipid mass, the levels of $\Sigma PCB7$ are increasing in the muscle tissue downstream the Sava River. The increase of the levels of $\Sigma PCB7$ (except for Lukavec Posavski station) is observed in the liver tissues of second campaign samples expressed both, on the wet and lipid mass. Comparing the available data about the levels of PCBs in the muscle tissue of chubs in the European rivers, with the levels of these pollutants determined in our monitoring, we were able to conclude that chubs from the Sava River are moderately polluted with PCBs.²⁻⁴ There is no data about the levels of PCBs in the liver of the European chubs in the available literature. For this reason determination of the levels of PCBs in the liver of the chubs from the Sava River is especially valuable. This is the first paper reporting about monitoring of levels of PCBs and OCPs in the liver tissue of the European chubs.



Figure 2. a) Levels of the sum of the seven PCB congeners (IUPAC No: PCB 28, PCB 52, PCB 101, PCB 118, PCB 138, PCB 153 and PCB 180) and b) levels of the sum of the DDTs (sum of p,p' DDT, DDD and DDE) in the muscle tissue (w.w.-wet weight, l.w.-lipid weight) of the European chubs from the Sava River collected in March/April and September 2005



Figure 3. a) Levels of the sum of the seven PCB congeners (IUPAC No: PCB 28, PCB 52, PCB 101, PCB 118, PCB 138, PCB 153 and PCB 180) and b) levels of the sum of the DDTs (sum of p,p' DDT, DDD and DDE) in the liver tissue (w.w.-wet weight, l.w.-lipid weight) of the European chubs from the Sava River collected in March/April and September 2005

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References

- 1. De Boer J, Brinkman UAT. Trends Anal Chem 1994; 13:397-404.
- 2. Jurajda P and Bernardova I. Folia Zool 1996; 45(1): 77-86.
- 3. Flammarion P, Devaux A, Nehls S, Migeon B, Noury P. and Garric J. *Ecotox Environ Safe* 2002; 51: 145-153.
- 4. Winter MJ, Verweij F, Garofalo E, Ceradini S, McKenzie DJ, Williams MA, Taylor EW, Butler PJ, van der Oost R, Chipman JK. *Aquat Toxicol* 2005; 73: 394–405.
- 5. Picer M and Ahel M. J Chromatogr 1978; 150: 119-127.
- 6. Picer M, Perkov S and Picer N. Water Air Soil Poll 1995; 69: 559-581.
- 7. Picer M. Croat Chem Acta 2000; 73(1): 123-186.
- 8. Picer M, Picer N, Kovač T and Hodak Kobasić V. 9th professional meeting of laboratory authorized for water investigations 2005, Vinkovci, Croatia, Proceedings: 91-94.