# Heavy Metal Contamination of the Loučka River Water Ecosystem

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#### Abstract

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Heavy metal contamination of the Loučka River water ecosystem was assessed in July 2005. We analyzed concentrations of T-Hg (total mercury), Cd, Pb, Cr, Cu, Zn, and Ni in water, sediments, zoobenthos, and in the brown trout (Salmo trutta m. fario) muscle and liver tissues (a total of 28 individuals) at four sampling sites. The highest Pb and Ni concentrations (4.634 - 12.050 and 0.689 - 24.980 mg kg<sup>-1</sup>) were found in sediments. The zoobenthos was most contaminated by Zn and Cu (0.556 - 1.505 and 2.925 - 74.300 mg·kg<sup>-1</sup>). The heavy metal contamination of river water was highest in Ni and Cr (0.1 - 6.8 and 0.5 - 10.0 mg·l<sup>-1</sup>). Concentrations of heavy metals in the brown trout muscle were following (in mg·kg<sup>-1</sup>): Pb  $0.108 \pm 0.073 - 1.010 \pm 0.506$ , Cd  $0.003 \pm 0.002$  - $0.026 \pm 0.022$ , Zn  $3.956 \pm 0.371 - 5.801 \pm 1.718$ , Ni  $0.058 \pm 0.018 - 0.102 \pm 0.046$ , Cr  $0.028 \pm 0.005$  $-0.073 \pm 0.039$ , Cu  $0.329 \pm 0.079 - 0.437 \pm 0.064$  and Hg  $0.065 \pm 0.008 - 0.106 \pm 0.047$ . Statistical differences (P < 0.05) in the brown trout muscle were in Pb and Zn. Cd, Cu and Zn were markedly accumulated in the brown trout liver (concentrations  $0.107 \pm 0.066 - 0.223 \pm 0.078$ , 59.973  $\pm 38.951 - 145.800 \pm 48.286$  and  $30.671 \pm 3.574 - 34.274 \pm 7.226$  mg·kg<sup>-1</sup>). Humans of 60 kg body mass may consume 1.5 kg of brown trout muscle from the Loučka River weekly without any risk. Adverse influence of the Uniglas distillery on the Loučka River environment contamination by heavy metals was not confirmed.

Czech Republic, stream ecosystems, brown trout, mercury, lead

The contamination of the water ecosystems is a worldwide problem of great importance. In the Czech Republic we can consider the situation of water pollution as improving from year to year, but it is not optimal yet. Heavy metals do not have primary lethal effect on hydrobionts but they have long-term negative influences on them.

Heavy metals have negative impacts on ecological stability of water ecosystems because of long-term environmental chronic stress. They cause fish growth disturbances, reproduction failure, immunosupression (Stave and Roberson 1985), histopathological changes in fish skin, gills, liver and kidneys and skeleton deformations (Sloof 1982; Hinton and Laurén 1990). Exposure of fish to heavy metals can cause metallothionein synthesis in the liver, kidneys and gills, and these metals can thus infiltrate the fish body directly by this non-alimentary way (Hamilton and Mehrle 1986). In addition, copper and mercury slow down fish metabolism by gill damage and enormous mucus secretion (Rice 1990).

Over the last 15 years, the heavy metals monitoring in stream ecosystems and distribution of heavy metals in fish tissues of different fishes has been carried out in many countries (Olsvik et al. 2001 - Norway, brown trout; Falandysz et al. 2000 - different species, Vistula River, Poland; Albeke et al. 2001 - Colorado, brown trout; Bervoets and Blust 2003 - Belgium, gudgeon; Linde et al. 2004 - Spain, brown trout and eel; Allengyl and Martynov 1995 - 9 species, Pechora River, Russia). Spurný et al. (2002 - Jihlava River, chub) and Svobodová et al. (2004 - Tichá Orlice River and tributaries, brown trout) monitored the situation in Czech and Moravian rivers. Beside the serious negative influence of heavy metal contamination on ichthyocenosis diversity and productivity, there is a possible health risk connected with regular consumption of heavy metal-contaminated fish in the long run, especially in sport fishermen and their families.

The Loučka River is divided into four fishing yards with salmonid stocking, which are of the ten most important in South Moravia. Beside low summer flow, ichthyocenosis prosperity is affected by water pollution. Over the last ten years there were several serious accidents resulting in mass fish death, that were mainly associated with Uniglas Company distillery industry near the Radešín village. Therefore field research was conducted, evaluating heavy metal contamination of the river water ecosystem.

#### **Materials and Methods**

The Loučka River is an important right-side tributary of the Svratka River with about 60 km of length and average annual flow of 2.0 m<sup>3</sup>·s<sup>-1</sup> at the mouth. Samples for laboratory analyses were taken from four sampling sites of the upper and middle course of the river (Loučka 4 and Loučka 2 fishing yards, Plate XIV, Fig. 1). Sampling sites were localized exactly by GPS Garmin iQue 3600 locator. Sampling sites 1 and 2 were situated upstream, whereas localities 3 and 4 downstream of the Uniglas Company distillery as the main supposed source of pollution. Due to the stream zonation, the localities 1, 2 and 3 corresponded to the brown trout zone; locality 4 is a typical grayling zone. Following ichtyological parameters were used to characterize fish community: total abundance, total biomass, diversity index (according to Shannon and Weaver 1963) and equitability index (according to Sheldon 1969). The brown trout (Salmo trutta m. fario) was used as an ichthyoindicator. Seven individuals of this species from each sampling site were taken in July 2005. River water, riverbed sediment and macrozoobenthos were also sampled for further laboratory manipulation. Fish for heavy metal analyses were brought to the laboratory immediately after returning from the field researches. Dorsal muscle samples (in the amount of about 2 g, without skin) and whole liver from each of the brown trout individual were taken carefully for individual assessment of heavy metal concentrations. These tissue samples were kept in a freezer at a temperature of -18 °C together with the water, sediment and zoobenthos samples until further analyses. To determine the age of the brown trout, 5 - 8 scales were sampled from each fish.

The concentration of total mercury (T-Hg), cadmium (Cd), lead (Pb), chromium (Cr), copper (Cu), zinc (Zn), and nickel (Ni) was analyzed after dry mineralizing using the Czech mineralizing equipment APION. Methods are described precisely by Spurný et al. (2002). Mercury was analyzed directly on the AMA 254 apparatus. Remaining heavy metals were detected by the AAS method on the SPECTR AA-30 apparatus with electro-thermal atomizer GTA-96 (in the case of Cd, Pb, Cr and Ni). Concentration of Cu and Zn was assessed using the method of flame AAS on the AA3000 apparatus. For verification standards KS 1014 and KS 1015 were used from UKZUZ Brno.

The results were statistically compared using software Unistat 5.1 (ANOVA, test of Scheffe).

## **Results and Discussion**

Heavy metal concentration in water, sediments and in macrozoobenthos of the Loučka River is given in Table 1. The highest concentration of lead, cadmium, zinc and mercury in sediments was detected in locality 2 (12.050, 0.303, 75.100 and 0.092 mg kg<sup>-1</sup>). Markedly higher nickel and chromium concentrations were found in sediments of locality 4 (24.980 and  $36.196 \text{ mg} \cdot \text{kg}^{-1}$ ) and the highest copper concentration was detected in locality 1 (8.200 mg \cdot \text{kg}^{-1}). Spurný et al. (2002) found in sediments of the Jihlava River below Třebíč higher concentrations of cadmium and zinc (0.638 and 102.840 mg·kg<sup>-1</sup>), similar concentrations of mercury, chromium and copper  $(0.108, 15.438 \text{ and } 12.974 \text{ mg} \cdot \text{kg}^{-1})$  and lower concentrations of lead and nickel (18.013 and 6.390 mg·kg<sup>-1</sup>) compared to the Loučka River sediments. In zoobenthos samples the highest concentration of zinc and nickel was in locality 2 (67.300 and  $1.505 \text{ mg}\cdot\text{kg}^{-1}$ , locality 1 showed the highest chromium, copper and mercury levels (3.832, 74.300, 0.030 mg·kg<sup>-1</sup>), the highest contamination by lead was found in locality 3 (0.865 mg kg<sup>-1</sup>) and by cadmium in locality 4 (0.255 mg kg<sup>-1</sup>). Heavy metal contamination of zoobenthos in the Jihlava River (Spurný et al. 2002) is similar (maximal concentrations in mg·kg<sup>-1</sup> are for Hg 0.054, for Cd 0.126, for Cr 1.595, for Cu 5.198, for Zn 34.120 and for Ni 0.942) except for lead (in the Jihlava River concentration of 1.920 mg·kg<sup>-1</sup> was detected, which is about two times higher than the maximum in the Loučka River). Heavy metal

Specimen	Site	Pb	Cd	Zn	Ni	Cr	Cu	Hg
Sediments	1	11.8900	0.2450	15.6090	0.6890	9.3840	80.2000	0.0448
	2	12.0500	0.3030	75.1000	13.1470	14.0000	19.9400	0.0921
	3	9.1400	0.1650	44.0000	16.3640	19.2400	6.1620	0.0411
	4	4.6340	0.0760	0.2078	24.9800	36.1960	11.2600	0.2078
Benthos	1	0.8160	0.0340	2.0240	0.5560	3.8320	74.3000	0.0303
	2	0.3270	0.1000	67.3000	1.5050	0.5950	4.8000	0.0237
	3	0.8650	0.0320	43.0000	0.6540	0.1760	2.9250	0.0157
	4	0.2360	0.2550	0.0197	1.0610	0.5270	4.5000	0.0197
Water	1	0.0016	0.0001	0.0110	0.0068	0.0100	0.0200	0.0004
	2	0.0025	0.0002	0.0100	0.0005	0.0006	0.0110	0.0002
	3	0.0016	0.0008	0.0050	0.0002	0.0004	0.0090	0.0001
	4	0.0020	0.0002	0.0007	0.0001	0.0005	0.0020	0.0007

Table 1. The Loučka River ecosystem contamination by heavy metals at four sampling sites in July 2005 (mg·kg<sup>-1</sup>)

contamination of the Loučka River water was similar at all the sampling sites, except for a higher concentration of nickel and chromium in locality 2 (6.8 and 10.0 mg·l<sup>-1</sup>). Jihlava River is about a five times bigger stream than Loučka and flows through big towns (such as Jihlava with about 50 000 and Třebíč with 35 000 inhabitants); therefore contamination of the Loučka River water ecosystem by heavy metals can be considered as rather high.

Table 2. Basic ichthyologic parameters of the Loučka River fish community at four sampling sites in July 2005

Sampling site	Abundance (indiv·ha <sup>-1</sup> )	Biomass (kg·ha <sup>-1</sup> )	Diversity index (H')	Equitability index (E)
1	3241	231.58	1.612	0.694
2	635	109.73	1.325	0.836
3	387	28.00	1.087	0.686
4	1635	74.07	2.841	0.896

Main characteristics of the fish community in the analyzed sector of the Loučka River are given in Table 2. The highest fish concentration and biomass was detected in locality 1 (3 241 fish ha<sup>-1</sup> and 231.58 kg·ha<sup>-1</sup>). It is the highest upstream locality, and the proximity of a trout-rearing brook can result in higher values. The highest diversity and equitability was in the furthest downstream situated locality 4 (H' = 2.841 and E = 0.896).

The results of heavy metal concentration analyzed in the brown trout muscle and liver are presented in Tables 3 and 4. The concentration of copper in the brown trout liver in the

Table 3. Heavy metal concentration (mean ± SD, n=7) in the brown trout (*S. trutta* m. *fario*) muscle from the Loučka River (in mg·kg<sup>-1</sup> of wet matter)

Sampling site	1 age 2+-3+ average 3+	2 age 2+-3+ average 3+	3 age 2+-3+ average 2+	4 age 2+-3+ average 2+
Pb	$0.390 \pm 0.311^{a}$	$0.108 \pm 0.073^{a}$	$0.281 \pm 0.322^{a}$	$1.010 \pm 0.506^{b}$
Cd	$0.023 \pm 0.028$	$0.007 \pm 0.004$	$0.201 \pm 0.022$ $0.003 \pm 0.002$	$0.026 \pm 0.022$
Zn	$5.531 \pm 0.530$	$4.631 \pm 0.639$	$5.801 \pm 1.718^{a}$	$3.956 \pm 0.371^{b}$
Ni	$0.058 \pm 0.018$	$0.080 \pm 0.043$	$0.079 \pm 0.020$	$0.102 \pm 0.046$
Cr	$0.041 \pm 0.023$	$0.059 \pm 0.030$	$0.073 \pm 0.039$	$0.028 \pm 0.005$
Cu	$0.399 \pm 0.163$	$0.437 \pm 0.064$	$0.329 \pm 0.079$	$0.386 \pm 0.043$
Hg	$0.065 \pm 0.008$	$0.089 \pm 0.017$	$0.088 \pm 0.013$	$0.106 \pm 0.047$

Statistical differences are indexed by letters, same letters for homogenous groups

Sampling site	1	2	3	4
Pb	$0.347 \pm 0.184^{a}$	$0.073 \pm 0.030 a$	$0.374 \pm 0.176^{a}$	$1.721 \pm 1.238^{b}$
Cd	$0.107\pm0.066$	$0.126\pm0.066$	$0.202\pm0.064$	$0.223\pm0.078$
Zn	$30.671 \pm 3.574$	$31.131 \pm 6.538$	$34.274 \pm 7.226$	$32.533\pm2.825$
Ni	$0.060 \pm 0.017^{\rm a}$	$0.079 \pm 0.074^{\rm a}$	$0.099 \pm 0.068^{\rm a}$	$0.328 \pm 0.117^{b}$
Cr	$0.068\pm0.024$	$0.035\pm0.021$	$0.060\pm0.040$	$0.067\pm0.047$
Cu	$59.973 \pm 38.951^{\rm a}$	$51.709 \pm 24.749^{\rm a}$	$92.510 \pm 54.454$	$145.800 \pm 48.286^{\text{b}}$
Hg	$0.143\pm0.024$	$0.147\pm0.038$	$0.123 \pm 0.019$	$0.159\pm0.056$

Table 4. Heavy metal concentration (mean  $\pm$  SD, n=7) in the brown trout (*S. trutta* m. *fario*) liver Loučka from the River (in mg·kg<sup>-1</sup> of wet matter)

Statistical differences are indexed by letters, same letters for homogenous groups

Loučka River is about five times higher compared to Piguena River in northern Spain (Linde et al. 1999). As Svobodová et al. (2004) report, in the upper reaches of the Tichá Orlice River brown trout muscle contamination by cadmium was lower (maximum 0.0073 as opposed to 0.026 mg·kg<sup>-1</sup>) and higher by nickel (maximum 0.146 against 0.102 mg·kg<sup>-1</sup>), than in the Loučka River. In the case of copper, zinc and chromium the values were similar (0.343, 4.53 and 0.081 as opposed to 0.437, 5.801 and 0.073 mg·kg<sup>-1</sup>).

Presently, in the Czech Republic the FAO/WHO limits are important (Table 5). The maximum tolerable daily or weekly human intakes of brown trout muscle from the Loučka River are included in Table 5 (values for a human consumer of 60 kg body mass). The maximum concentration of all the localities for each heavy metal was used there. Apparently, the most limiting heavy metal is lead. People can eat 1.5 kg of brown trout muscle weekly without any risk. Although the concentration of lead in the zoobenthos in the Jihlava River is higher, the chub muscle contamination by lead is lower (mean value below Třebíč 0.636 mg·kg<sup>-1</sup>; S purný et al. 2002). Other heavy metals are of no concern for human consumption. As Houserová et al. (2006) report, concentration of total mercury in the chub muscle at the lower reach of the Loučka River was also lower than in our research.

metal	Pb	Cd	Zn	Ni	Cr	Cu	Hg
Limit	25	7	1000			500	5
(µg·kg <sup>-1</sup> )	PTWI	PTWI	PMTDI	-	-	PMTDI	PTWI
intake	1.5	16.2	10.3	-	-	68.6	2.8

Table 5. Maximal brown trout muscle intake from Loučka River according to FAO/WHO limits (in kg·60 kg<sup>-1</sup> body mass of human consumer)

PTWI-provisional tolerated weekly intake, PMTDI-provisional maximum tolerable daily intake

When we compare heavy metal concentrations in the brown trout muscle to its liver, a strong increase of cadmium, zinc and copper is obvious. These metals are bioaccumulated in the liver for detoxication. There is a risk of metallothionein synthesis in the fish body, which can cause another increase of these metals concentration in fish body as Hogstrand et al. (1991) describe in perch, Olsvik et al. (2001) in brown trout in Norwegian rivers and Linde et al. (1999 and 2001) in brown trout in Spain. As our ichthyological research in the Loučka River shows, the occurrence of individuals older than 3 years is rare, and the analyzed fish were purely in the age of 2+ and 3+ (Table 3).

Comparing heavy metal contamination of the brown trout muscle from four sampling sites of the Loučka River, statistically significant differences were found between locality 4 and the other ones in lead concentration, when the value in locality 4 was markedly higher. Significantly lower concentration of zinc in locality 4 compared to locality 3 was also found (Table 3).

In the case of brown trout liver contamination by heavy metals, the statistical comparison also showed several differences. At locality 4, the concentration of lead (corresponding to concentration in the muscle) and the concentration of nickel significantly exceeds the remaining sampling sites. Another significant difference was in copper at locality 4 (a higher value) compared to localities 1 and 2 (Table 4). Because no statistical differences were found between localities 2 and 3 in the heavy metal concentration of brown trout tissues, we can exclude the adverse influence of the Uniglas distillery on the Loučka River environment in relation to contamination by heavy metals.

If we consider brown trout as an indicator of heavy metal contamination in the water environment (Linde et al. 1998), the water, sediment and zoobenthos in locality 4 should be the most contaminated by lead, but the reality is different. It may be due to feeding migrations or a consequence of brown trout stocking (this species is stocked yearly at the age of 1+ and 2+). The analyzed fish thus may not belong to the original population, originating from a different stream, or they may have moved upstream from probably more contaminated lower parts of this river.

# Zatížení vodního ekosystému řeky Loučky těžkými kovy

V červenci 2005 bylo provedeno sledování zatížení vodního ekosystému řeky Loučky těžkými kovy. Analyzovali jsme koncentraci T-Hg (celková rtuť), Cd, Pb, Cr, Cu, Zn, a Ni ve vodě, sedimentu a bentosu a dále ve svalové a jaterní tkáni pstruha obecného f. potoční (Salmo trutta m. fario; celkem 28 jedinců), a to na čtyřech lokalitách. Nejvyšší koncentrace v sedimentu byla zjištěna u Pb a Ni (4,634 - 12,050 a 0,689 - 24, 980 mg·kg<sup>-1</sup>). Zoobentos byl nejvíce kontaminován Zn a Cu (0, 556 - 1,505 a 2,925 - 74,300 mg·kg<sup>-1</sup>). Zatížení říční vody těžkými kovy bylo nejvyšší u Ni a Cr  $(0, 1 - 6, 8 a 0, 5 - 10, 0 \text{ mg} \cdot 1^{-1})$ . Koncentrace těžkých kovů ve svalovině pstruha potočního byly následující (v mg.kg<sup>-1</sup>): Pb  $0,108 \pm 0.073 - 1,010$  $\pm 0,506$ ; Cd 0,003  $\pm 0,002 - 0,026 \pm 0,022$ ; Zn 3,956  $\pm 0,371 - 5,801 \pm 1,718$ ; Ni 0,058  $\pm 0,018$  $-0.102 \pm 0.046$ ; Cr 0, 028  $\pm 0.005 - 0.073 \pm 0.039$ ; Cu 0.329  $\pm 0.079 - 0.437 \pm 0.064$  a Hg  $0.065 \pm 0.008 - 0.106 \pm 0.047$ . Byly zjištěny statistické rozdíly (P < 0.05) v obsahu Pb a Zn ve svalovině pstruha obecného. Čd, Ču a Žn jsou výrazně akumulovány v játrech pstruha obecného (koncentrace  $0,107 \pm 0,066 - 0,223 \pm 0,078; 59,973 \pm 38,951 - 145,800 \pm 48,286$  $a 30,671 \pm 3,574 - 34,274 \pm 7,226 \text{ mg} \cdot \text{kg}^{-1}$ ). Člověk vážící 60 kg může bez jakéhokoliv rizika zkonzumovat 1,5 kg svaloviny pstruha obecného týdně. Negativní vliv lihovaru firmy Uniglas na zatížení ekosystému řeky Loučky těžkými kovy prokázán nebyl.

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Fig. 1. Location of the Loučka River sampling area