

## Long-Term Monitoring of Mercury Content in Fish From the Želivka Reservoir-Syndrom of Newly Filled Reservoir

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A systematic investigation of bioaccumulation of mercury in fish from the manmade reservoir Želivka (Czech Republic) was performed from 1974 to 2011. Changes of mercury concentrations in muscle and liver of four predatory fish species in this location are summarized and discussed. Tissue mercury concentrations in *Esox lucius*, *Aspius aspius*, *Perca fluviatilis*, and *Abramis brama* are related to fish age and time of sampling in the thirty-seven year period after flooding. Monitoring data were divided into five time periods from 1974 to 2011. Although no local or point mercury sources in the Želivka reservoir are known, high mercury content in all examined fish species were found in the years shortly after impoundment. Mercury concentration in muscle and liver showed significant differences among time periods for all species. The highest mercury content was found in the first period after impoundment (1985 and earlier) whereas the lowest content was determined to be from 1990 to 1995 (15-20 years after flooding). A significant decrease during the monitored years was observed both for muscle and liver of fish and is presently stabilized at around 0.2 mg kg<sup>-1</sup>.

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**Keywords:** predator, mercury methylation, bioaccumulation, flooded reservoir

### 1. INTRODUCTION

Mercury contamination of the environment has been studied since mercury poisoning in Minamata, Japan in the mid-twentieth century. Mercury poisoning has been caused by the consumption of contaminated fish; thus, mercury contamination is a critical issue in the aquatic environment and the focus of scientific attention. Both human activities and atmospheric deposition

are sources of elevated mercury levels. Consequently, the consumption of fish may pose a serious environmental health risk to humans.

In summer 1970, gradual flooding of a valley on the river Želivka (Czech Republic) to create a reservoir of potable water was begun. Flooding of the area of 14 km<sup>2</sup> containing about 2.5x10<sup>8</sup> m<sup>3</sup> of water was completed in 1975, and began to be used to supply drinking water for Prague. Immediately after impoundment in 1974, elevated Hg levels in fish were reported [1]. This unexpected event initiated long-term systematic investigation of bioaccumulation of Hg in fish in the reservoir. To our knowledge, neither Želivka River itself (a small tributary of the Vltava river basin) nor the flooded area of the reservoir was contaminated with mercury prior to this time [2].

Elevated content of Hg in fish from newly constructed manmade reservoirs has been reported several times since then [3-6]. It is now assumed that elevated Hg levels in such reservoirs are connected with biochemical decomposition processes in flooded soil and vegetation at the reservoir bottom. Mercury, especially as the methylmercury organic form [7] originates in bacterial activity and accumulates in the nutrient base of the food chain of aquatic fauna. The strong bond of mercury to sulphur-containing groups is well known [8]. Extraordinarily strong bonding to protein SH-groups leads to accumulation of Hg in the food chain beginning with simple organisms to fish, acting as integrator of Hg. Hence, high values of Hg in older fish, especially predatory species, are not surprising.

The completion of the decomposition of flooded organic materials leads to diminishing of the amount of methylmercury in benthic fauna and the zooplankton. Changes in the environment are also reflected in the occurrence of different fish species in the locality and their amount of Hg. Over the course of time, we observed, in accord with the assumption, changes in numbers of fish species caught in the dam. From 1974 to 2011 a total of 15 fish species were sampled. For the purpose of this study we selected from the data four relatively frequent species. Predatory fishes were chosen, since mercury bioaccumulation in predators is highest, due to their position in the food chain [9].

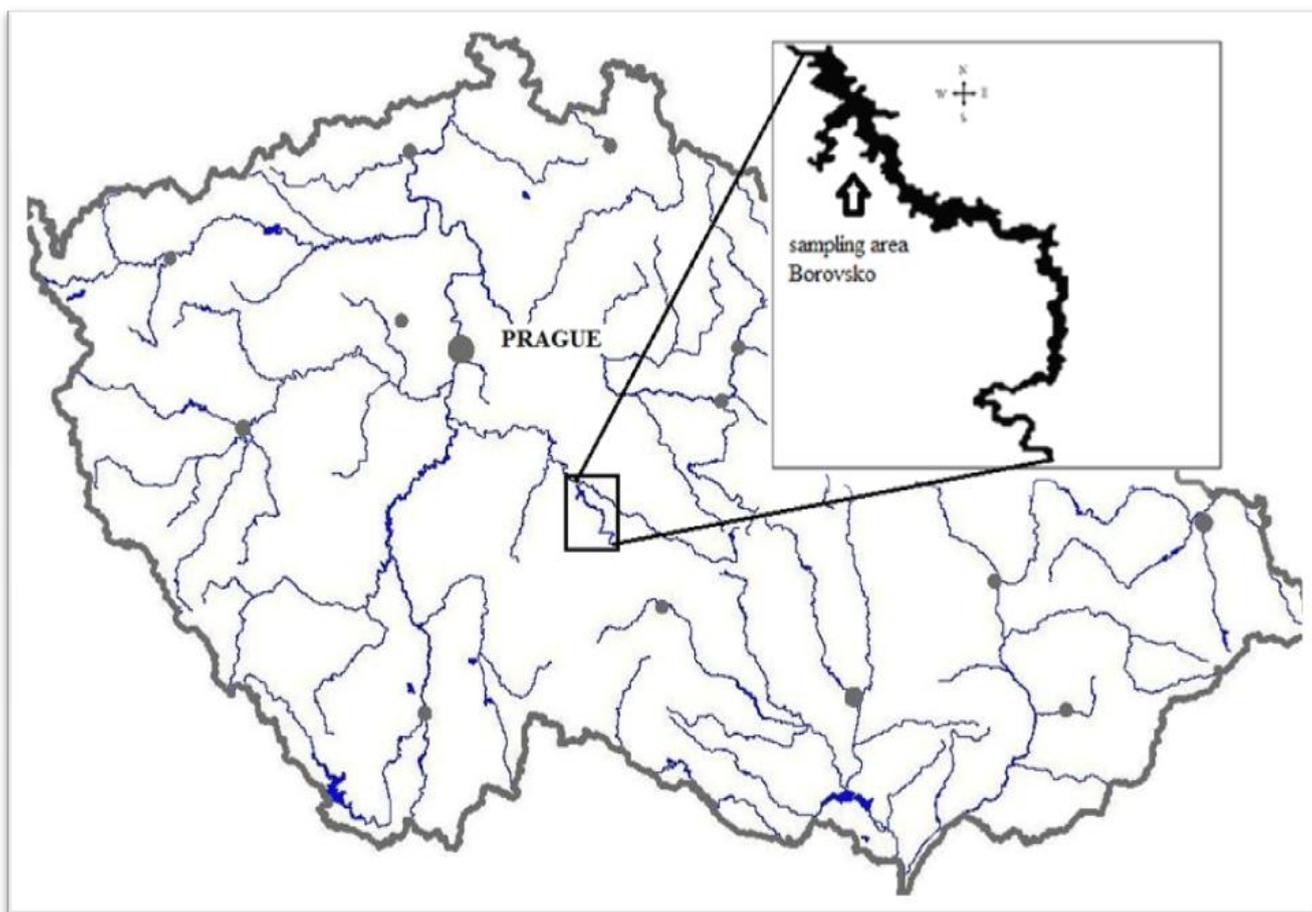
The weight of sampled individuals of the same species and age changed in time, in some cases considerably. We presume that these changes in growth were influenced partially by temporary conditions created by the decomposition processes in the flooded area. Under such circumstances the most useful quantitative information about changes in Hg levels was assumed to be gained from the relationship between the Hg concentration and age of the fish relative to time of initial flooding.

## 2. MATERIALS AND METHODS

### 2.1. Study area

The Želivka Reservoir is located 4 km upstream of the confluence with the Sázava River, in the central Czech Republic, southeast of the capital city of Prague. A valley on the Želivka River in Bohemia was gradually flooded from 1970 to 1974 to a current area of about 1400 ha. About 15% of agricultural areas and 85% of forest stamp in the Moravian and Bohemian highlands were flooded. After initial flooding, the water surface was continually increased until 1996. In 1975, the removal of

water for purification was begun, and this reservoir is the main water supply for Prague with an output of  $7000 \text{ l s}^{-1}$ . The Czech Republic has a temperate continental climate, summers are relatively hot and winters are cold, cloudy, and snowy. The temperature difference between summer and winter is relatively high, due to the landlocked position. On average, summer temperatures are about  $20 \text{ }^\circ\text{C}$  higher than in winter. Especially in the past decade, temperatures in summer above  $30 \text{ }^\circ\text{C}$  ( $86 \text{ }^\circ\text{F}$ ) are not unusual. Summer is also characterized by rain and storms.



**Figure 1.** Map of the Želivka Reservoir

## 2.2. Data collection

For the purpose of this study only results from Borovsko, were considered. The first sampling was performed in 1974 and the most recent in 2011. The predatory fish species were taken during the whole year. By means of electrofishing in spring time at near shore mainly (reproduction time for pike, pikeperch, asp), in summer time in the night by beach seining nets, and from deeper parts of reservoir by gill nets (in autumn and winter time). A total of 525 predatory fish individuals taken from this site 1974 to 2011 were selected for the present study. Table 1 presents the number of individuals of selected species that were examined.

### 2.3. Predatory fish

The predatory fish were captured and immediately weighed. Scales were collected for age determination. Samples of muscle (the cranial area dorsal to the lateral line) and liver were taken from, placed in polyethylene bags, labelled, and stored at -18 °C.

The dataset consisted of 525 specimens of four fish species. The fish were sampled from 1974 to 2011 and five time periods (I: 1985 and earlier; II: 1986–1990; III: 1990–1995; IV: 1995–2000; V: 2001 and later) were defined prior to comparative analyses.

Perch – *Perca fluviatilis*: perch is the common name for fish of the genus *Perca*, freshwater gamefish belonging to the family *Percidae*. Total of 298 perch were sampled, more than 56 % of all predatory fish analysed in this study.

Pike – *Esox lucius*: pike is a species of carnivorous fish of the genus *Esox*. Pike is typical of brackish and freshwaters of the northern hemisphere. With sufficient nutrition, a rapid elevation of weight, beginning at relatively low age, is usual. A total of 49 pike were sampled and analyzed.

Asp – *Aspius aspius*: the asp is a European freshwater fish of the family Cyprinidae. A total of 86 asp were sampled and analyzed in this study.

Pikeperch – *Sander lucioperca* : a total of 92 pikeperch were captured over the 37 year period. It is 9.3% of all fish. Young pikeperch live on plankton, molluscs, and other invertebrates. Adult pikeperch consumes mainly small fish.

### 2.4. Total mercury analysis

The total mercury (THg) concentration (organic and inorganic) in fish muscle and liver was determined by CV-AAS after acid oxidizing and digestion of samples. Digestion of the fish samples and liver in the way and the total mercury contents estimation by means of the flameless atomic absorption were carried out as previously describes. [10].

About 5 g of samples was used because of a better digestion procedure (in case of measuring till 1995). Mineralization was carried out by means of a mixture of nitric acid and sulphuric acid in the ratio 10:1 in closed glass device with acid inlet and effective reflux water coller, so that the loss of mercury by vaporization might be thoroughly prevented.

This principle was utilized to construct the AMA 254 analyzer (Altec, Czech Republic), which has been used for mercury determination since 2009. This device is based on the unique concept of in situ dry decomposition of liquid or solid sample (maximally 300 mg) in a stream of oxygen and passage of the combustion gases through the catalytic column, followed by trapping Hg on the gold amalgamator.

Heating the amalgamator rapidly evaporates preconcentrated mercury which is then transported into the system of measuring cells, and its atomic absorption is measured. The whole analysis is

performed automatically without the need of preliminary dissolution and/or decomposition of the sample.

The method is direct and does not require sample preparation. For mercury analysis after 1995 about 50 mg of muscle/liver samples was used. The limit of detection of this method is  $0.1 \mu\text{g kg}^{-1}$ . The accuracy for THg value was validated using standard reference material BCR-CRM 464 (Tuna Fish, IRMM, Belgium). The total mercury concentrations in fish muscle and liver are given in  $\text{mg kg}^{-1}$  wet weight.

### 2.5. Statistical analysis

Standard descriptive statistics were used to summarize primary data: absolute and relative frequencies for categorical variables, arithmetic means in cases of normal distribution, and geometric means in cases of log-normally distributed continuous variables. A 95% confidence interval was applied to mean estimates.

Mercury concentration values proved to be log-normally distributed. Therefore, logarithmic transformation ( $X_{\text{tr}} = \text{Ln}[X]$ ) of the primary values was applied prior to parametric statistical testing. The transformation was proven effective in normalization of the distribution (Kolmogorov–Smirnov test) and also helped to reach homogeneity of variance among compared variants (Levene test). Statistical significance of differences among time periods was tested using analysis of variance (one-way ANOVA model) followed by Tukey post-hoc test for continuous variables.

In addition to processing of primary values, mercury content in muscle and liver was adjusted for fish age prior to further analysis, using a linear regression model on log-transformed data. Non-standardized residuals of regression models based on age as a predictor variable were added to the mean of primary data to obtain age-adjusted concentration values, with adjustment was computed separately for each species.

Both primary and age-adjusted mercury content values were employed in principal comparisons of sampling time periods. The analysis was computed using SPSS 19.0.1 (IBM Corporation, 2010).

## 3. RESULTS AND DISCUSSION

The main characteristics of the sampled fish are presented in Table 1.

As expected, the mercury concentration in muscle and liver significantly depended on the age of fish, with this variable explaining from 11 to 61% of the variation in concentration levels (primary data in Figure 1a, b).

This finding, together with the age structure of samples obtained in different sampling campaigns was used to compute age-adjusted mercury concentrations in muscle and liver. The adjusted values were used for statistical comparisons within fish species.

In case of pike is depending slightest both for muscle and liver, whereas high mercury value

was found in relatively young pike (see Figure 1a).

This is probably related to feeding habitat.

Pike can grow very quickly at a relatively young age when appropriate nutrition is available. This means enhanced intake of feed and higher bioaccumulation of mercury in young fish.

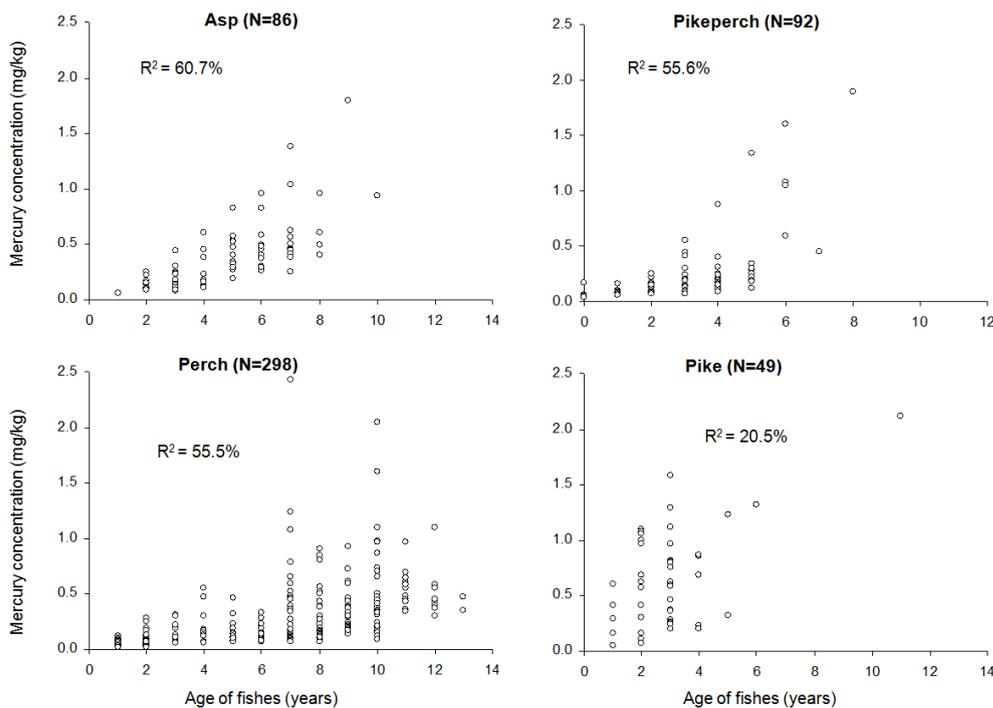
**Table 1.** The main characteristics of sampled fishes in each period <sup>1</sup>

<sup>1</sup> Characteristics of sampled fish specimen: sample size in given time period (N), age in years and weight in kg (arithmetic mean ± standard error), absolute and relative frequency of males as sex category (13.6% of specimens were juveniles of undetermined sex)

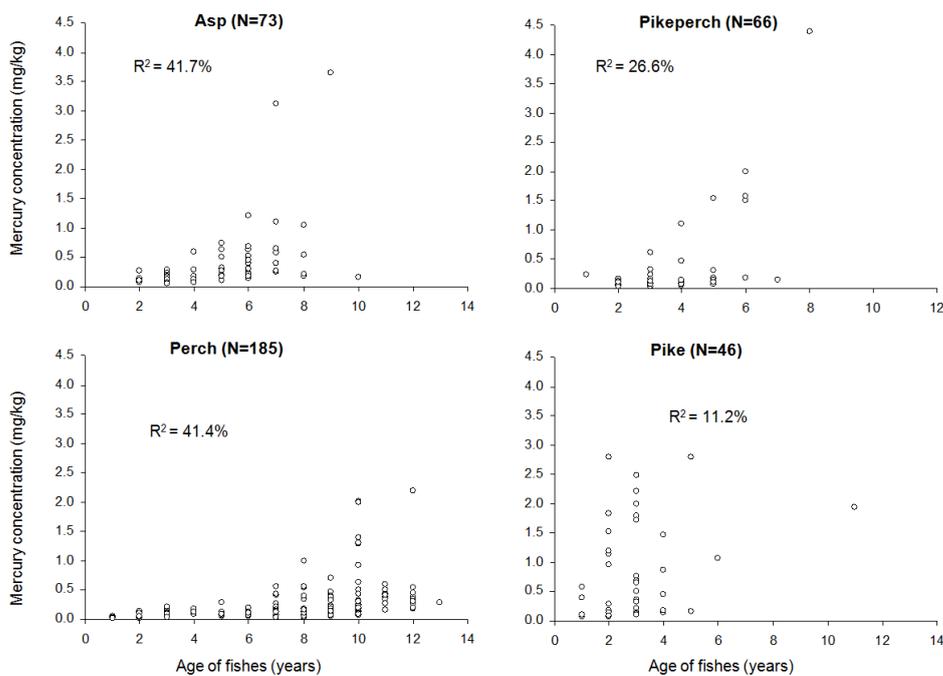
	Asp (N=86)	Pikeperch (N=92)	Perch (N=298)	Pike (N=49)
1985 and earlier	N=11	N=9	N=39	N=30
age	6.1 (0.4)	5.0 (0.6)	7.9 (0.3)	3.2 (0.3)
weight	1.54 (0.31)	2.04 (0.45)	0.40 (0.03)	1.49 (0.30)
males	5 (45.5%)	6 (66.7%)	1 (2.6%)	19 (63.3%)
1986-1990	N=10	N=7	N=50	N=6
age	5.5 (0.3)	3.7 (0.5)	8.9 (0.4)	2.7 (0.8)
weight	1.44 (0.06)	0.94 (0.15)	0.39 (0.02)	1.29 (0.53)
males	3 (30.0%)	5 (71.4%)	6 (12.0%)	4 (66.7%)
1990-1995	N=24	N=58	N=124	N=3
age	5.1 (0.4)	2.7 (0.2)	6.0 (0.3)	1.7 (0.3)
weight	1.45 (0.10)	0.69 (0.07)	0.22 (0.02)	0.39 (0.15)
males	6 (25.0%)	32 (55.2%)	31 (25.0%)	2 (66.7%)
1995-2000	N=17	N=12	N=53	N=3
age	2.9 (0.3)	2.2 (0.3)	4.1 (0.5)	3.0 (1.0)
weight	0.77 (0.14)	0.57 (0.14)	0.16 (0.02)	0.80 (0.28)
males	7 (41.2%)	2 (16.7%)	16 (30.2%)	1 (33.3%)
2001 and later	N=24	N=6	N=32	N=7
age	4.5 (0.4)	4.3 (0.8)	1.8 (0.2)	3.0 (0.4)
weight	1.25 (0.24)	1.94 (0.36)	0.10 (0.03)	0.82 (0.22)
males	8 (33.3%)	2 (33.3%)	1 (3.1%)	4 (57.1%)
Total	N=86	N=92	N=298	N=49
age	4.7 (0.2)	3.1 (0.2)	6.0 (0.2)	3.0 (0.2)
weight	1.27 (0.09)	0.90 (0.09)	0.25 (0.01)	1.26 (0.20)
males	29 (33.7%)	47 (51.1%)	55 (18.5%)	30 (61.2%)

The mercury concentrations in muscle and liver samples according to fish species and time period are presented in Tables 2 and 3, both as primary and age-adjusted data.

Mercury concentrations showed significant differences among time periods for all species in both muscle and liver. Results of detailed post-hoc analysis of age-adjusted values are shown in Tables 2 and 3.



**Figure 1a.** Relationship between mercury concentration in muscle and age of fish.  $R^2$  - mercury concentration variability explained by age in linear regression model for age and log transformed concentration value



**Figure 1b.** Relationship between mercury concentration in liver and age of fish.  $R^2$  - mercury concentration variability explained by age in linear regression model for age and log transformed concentration value

**Table 2.** Mercury concentration in muscle of predatory fish

<sup>1</sup> geometric mean and its 95% confidence interval

<sup>2</sup> statistical significance evaluated by one-way ANOVA based on log transformed data

a, b, c, d the same lettermarks time periods which are not statistically significantly different (Tukey post hoc test)

	Asp (N=86)	Pikeperch (N=92)	Perch (N=298)	Pike (N=49)
Non-adjusted mercury concentration (mg/kg) <sup>1</sup>				
1985 and earlier	0.57 (0.41; 0.81) <sup>a</sup>	0.90 (0.62; 1.30) <sup>a</sup>	0.52 (0.43; 0.64) <sup>a</sup>	0.73 (0.61; 0.89) <sup>a</sup>
1986-1990	0.42 (0.37; 0.48) <sup>ab</sup>	0.19 (0.14; 0.26) <sup>b</sup>	0.39 (0.31; 0.48) <sup>a</sup>	0.71 (0.38; 1.33) <sup>a</sup>
1990-1995	0.23 (0.17; 0.30) <sup>b</sup>	0.11 (0.10; 0.13) <sup>b</sup>	0.10 (0.09; 0.11) <sup>b</sup>	0.07 (0.05; 0.12) <sup>b</sup>
1995-2000	0.23 (0.19; 0.28) <sup>b</sup>	0.18 (0.16; 0.19) <sup>b</sup>	0.12 (0.11; 0.14) <sup>b</sup>	0.20 (0.16; 0.24) <sup>c</sup>
2001 and later	0.26 (0.18; 0.36) <sup>b</sup>	0.37 (0.29; 0.48) <sup>c</sup>	0.06 (0.04; 0.08) <sup>c</sup>	0.23 (0.19; 0.29) <sup>c</sup>
Total	0.29 (0.25; 0.33)	0.17 (0.14; 0.20)	0.15 (0.14; 0.17)	0.50 (0.39; 0.63)
p (ANOVA) <sup>2</sup>	0.001	<0.001	<0.001	<0.001
Mercury concentration adjusted for age by linear regression (mg/kg) <sup>1</sup>				
1985 and earlier	0.39 (0.29; 0.52) <sup>a</sup>	0.46 (0.38; 0.56) <sup>a</sup>	0.35 (0.30; 0.40) <sup>a</sup>	0.70 (0.59; 0.84) <sup>a</sup>
1986-1990	0.33 (0.26; 0.42) <sup>a</sup>	0.15 (0.13; 0.18) <sup>b</sup>	0.21 (0.17; 0.25) <sup>b</sup>	0.77 (0.54; 1.09) <sup>a</sup>
1990-1995	0.21 (0.18; 0.23) <sup>b</sup>	0.13 (0.12; 0.14) <sup>b</sup>	0.10 (0.10; 0.11) <sup>c</sup>	0.10 (0.07; 0.14) <sup>b</sup>
1995-2000	0.37 (0.32; 0.43) <sup>a</sup>	0.24 (0.20; 0.29) <sup>c</sup>	0.18 (0.16; 0.20) <sup>b</sup>	0.19 (0.15; 0.26) <sup>bc</sup>
2001 and later	0.27 (0.23; 0.32) <sup>ab</sup>	0.24 (0.16; 0.36) <sup>c</sup>	0.14 (0.10; 0.19) <sup>d</sup>	0.23 (0.21; 0.26) <sup>c</sup>
Total	0.29 (0.26; 0.31)	0.17 (0.15; 0.19)	0.15 (0.14; 0.17)	0.50 (0.40; 0.62)
p (ANOVA) <sup>2</sup>	<0.001	<0.001	<0.001	<0.001

**Table 3.** Mercury concentration in liver of predatory fish

<sup>1</sup> geometric mean and its 95% confidence interval

<sup>2</sup> statistical significance evaluated by one-way ANOVA based on log transformed data

a, b, c, d the same lettermarks time periods which are not statistically significantly different (Tukey post-hoc test)

	Asp (N=73)	Pikeperch (N=66)	Perch (N=185)	Pike (N=46)
Nonadjusted mercury concentration (mg/kg) <sup>1</sup>				
1985 and earlier	0.82 (0.37; 1.85) <sup>a</sup>	1.14 (0.68; 1.93) <sup>a</sup>	0.41 (0.28; 0.61) <sup>a</sup>	0.92 (0.67; 1.24) <sup>a</sup>
1986-1990	0.41 (0.28; 0.59) <sup>ab</sup>	0.13 (0.08; 0.20) <sup>b</sup>	0.33 (0.29; 0.37) <sup>a</sup>	0.67 (0.25; 1.75) <sup>a</sup>
1990-1995	0.15 (0.11; 0.22) <sup>c</sup>	0.08 (0.07; 0.09) <sup>b</sup>	0.10 (0.09; 0.11) <sup>b</sup>	0.07 (0.07; 0.08) <sup>b</sup>
1995-2000	0.19 (0.15; 0.23) <sup>bc</sup>	0.13 (0.11; 0.16) <sup>b</sup>	0.14 (0.12; 0.16) <sup>c</sup>	0.14 (0.10; 0.19) <sup>b</sup>
2001 and later	0.18 (0.13; 0.26) <sup>bc</sup>	0.15 (0.12; 0.19) <sup>b</sup>	0.05 (0.04; 0.07) <sup>d</sup>	0.13 (0.11; 0.15) <sup>b</sup>
Total	0.21 (0.17; 0.26)	0.14 (0.11; 0.18)	0.14 (0.13; 0.16)	0.49 (0.35; 0.69)
p (ANOVA) <sup>2</sup>	<0.001	<0.001	<0.001	<0.001
Mercury concentration adjusted for age by linear regression (mg/kg) <sup>1</sup>				
1985 and earlier	0.57 (0.35; 0.93) <sup>a</sup>	0.71 (0.58; 0.88) <sup>a</sup>	0.35 (0.26; 0.47) <sup>a</sup>	0.86 (0.62; 1.19) <sup>a</sup>
1986-1990	0.30 (0.18; 0.51) <sup>b</sup>	0.11 (0.08; 0.15) <sup>b</sup>	0.18 (0.15; 0.21) <sup>b</sup>	0.73 (0.38; 1.43) <sup>a</sup>
1990-1995	0.13 (0.11; 0.16) <sup>c</sup>	0.09 (0.08; 0.10) <sup>b</sup>	0.09 (0.08; 0.10) <sup>c</sup>	0.10 (0.08; 0.12) <sup>b</sup>
1995-2000	0.30 (0.25; 0.35) <sup>b</sup>	0.22 (0.16; 0.30) <sup>c</sup>	0.17 (0.15; 0.19) <sup>b</sup>	0.14 (0.11; 0.18) <sup>b</sup>
2001 and later	0.18 (0.14; 0.24) <sup>bc</sup>	0.12 (0.07; 0.22) <sup>b</sup>	0.14 (0.11; 0.17) <sup>b</sup>	0.13 (0.12; 0.15) <sup>b</sup>
Total	0.21 (0.18; 0.25)	0.14 (0.11; 0.17)	0.14 (0.13; 0.16)	0.49 (0.35; 0.68)
p (ANOVA) <sup>2</sup>	<0.001	<0.001	<0.001	<0.001

Significantly elevated values of mercury were found in Period I (1985 and earlier) for all species and tissue in comparison with other periods. Impoundment of agriculture area made worse hydrochemical quality and thus oxygen concentration in 8-9 m upper of the bottom was less than 2 mg/l (1971) and gradually was decreased till anaerobic condition on bottom sediment was created (1973 and later). It caused intensification of bacterial methylating process and easier conversion of Hg to the food chain. For some species elevated values remained in Period II (1986–1990), especially in pike. The Period III (1991–1995) revealed generally the lowest mercury concentration. After this period a slight, but significant, increase in mercury concentration was found in all species and tissue with the exception of pike liver, where concentrations remain low and stable. In Period V (2001 and later), a stabilization in mercury concentration (asp, pikeperch, and pike muscle; asp, perch, and pike livers) or its decrease (perch muscle, pikeperch liver) was found.

Adjusted values of mercury concentration in fish muscle and liver of all analyzed species are presented in Figure 2.

#### Asp – *Aspius aspius*

Mercury content in asp muscle and liver was highest in Period I in comparison to other periods with the mean value of mercury in liver and muscle twice that in Period V (37 years after impoundment). In the first period mercury predominates in the liver of fish than in muscle. The statistically lowest mercury content in asp muscle and liver was found from 1990 to 1995 fifteen years after flooding at approximately 0.23 and 0.15 mg kg<sup>-1</sup>. In Period V the Hg levels remained below 0.3 mg kg<sup>-1</sup>.

#### Perch – *Perca fluviatilis*

The greatest amount of data was obtained for perch. The highest mercury content in muscle and liver were observed in 1981 (0.79 ± 0.47 and 1.31 ± 0.69 mg kg<sup>-1</sup> respectively), seven years after flooding. Year 1981 is in the first period, in which significantly higher mercury levels were found compared with the other periods. Samples obtained after 1981 showed lower amount, and the results from Period V show values stabilized at about 0.2 mg kg<sup>-1</sup>. The lowest mean concentration was found in Period III, 15-20 years after flooding. Similar results were obtained for perch liver. The trend in mercury bioaccumulation in muscle and liver were analogous.

Proportion between mercury in muscle and liver during the monitoring period was interesting. While in the first period the mercury level in liver was greater than that in muscle, in Period II the mercury level were higher in muscle than in liver. Havelková (2008) found that, in heavily contaminated locations, mercury content of liver was significantly greater than in muscle. Mercury is preferentially deposited in the liver if the location is highly contaminated. Our results are in agreement with this. Higher mercury levels in fish liver demonstrates that more mercury was released into the food chain, probably via anaerobic microorganisms from the flooded land [11].

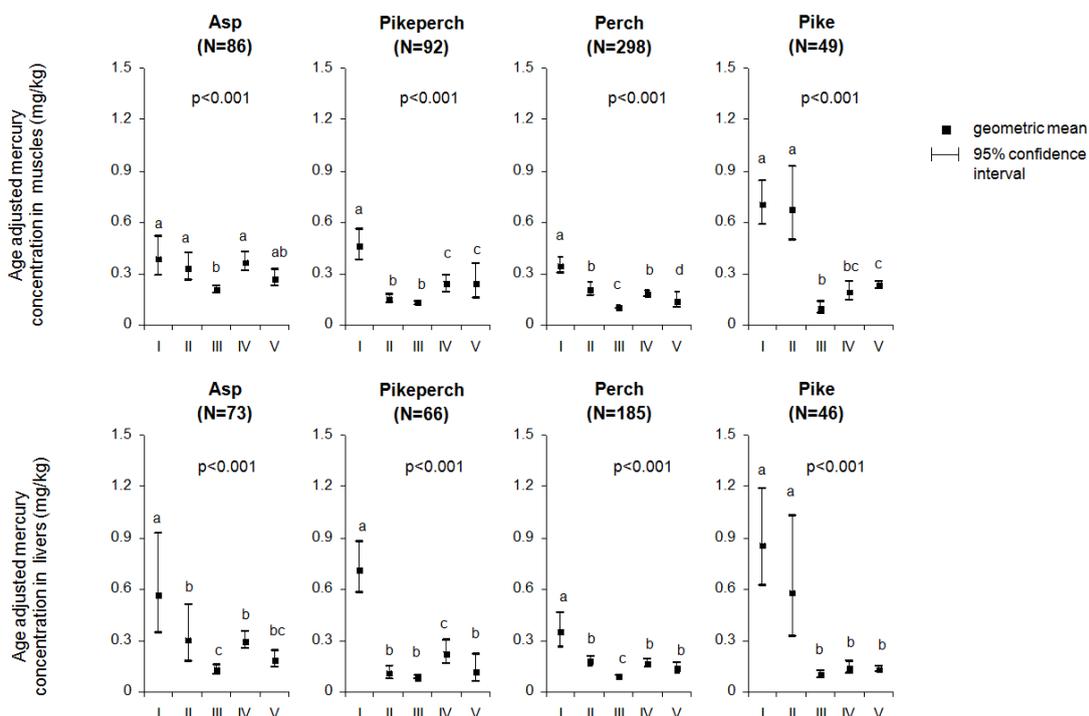
#### Pike – *Esox lucius*

Ten percent of all analyzed fish were the indicator species pike. Mean mercury values in muscle and liver are shown in Figure 2. Most pike were sampled during the Period I (N = 30). In the

ensuing years few fish were caught, reflecting collocation of individual fish species in the flooded reservoir. Therefore it is difficult to qualify mercury accumulation in pike, although the trend is similar to that in other fish species. The highest mean mercury content was observed during the fifteen years after flooding in Period I and reached 0.73 mg kg<sup>-1</sup>. In individual samples the highest amount was 2.12 and thus exceeds health safety limits (1 mg kg<sup>-1</sup>) for predatory species set by Commission Regulation 1881/2006 [12]. Statistically, higher mercury values in Periods I and II are in agreement with many authors [13, 4, 6]. Subsequently, mercury in pike decreased and stabilized at about 0.2 mg kg<sup>-1</sup>, which is common in non-contaminated localities in the Czech Republic [14]. As in other species, mercury in liver was higher than in muscle in the first decades of the monitored period.

Pikeperch – *Sander lucioperca*

Data for Pikeperch are show in Figure 2. The highest mercury levels were found in Period I comparison. Mercury geometric mean reached 0.90 mg kg<sup>-1</sup> in liver and 1.14 mg kg<sup>-1</sup> in muscle in the first period after flooding. The lowest value was found in both liver and muscle in Period III. Values have been stabilized at under 0.4 mg kg<sup>-1</sup> since 1990, approximately fifteen years after impoundment. This is in agreement with reports of Bodaly [15]. As in other analyzed fish species, mercury predominated in liver of pikeperch at the beginning of the monitoring period.



**Figure 2.** Mercury concentration adjusted for age of fish. Time periods: I - 1985 and earlier; II - 1986-1990; III - 1990-1995; IV - 1995-2000; V - 2001 and later. Statistical significance evaluated by one-way ANOVA based on log transformed data. <sup>a, b, c, d</sup> the same letter marks time periods which are not statistically significantly different (comparison within given species; Tukey post-hoc test)

Results of thirty-seven years monitoring were confirmed findings of many authors [6, 9,13]. Mercury level in fish was highest in the first 15-20 years following flooding. It is assumed that elevated mercury concentration is caused by the bacterial methylation of Hg that occurs naturally in the environment along with long range atmospheric deposition in combination with transformations and mobilizations from catchments and sediments [13]. Mercury concentration in fish in flooded reservoir has also been reported to correlate with the rate of flooding [16]. Climate, pH of water, soil and vegetation type, water hardness, and dissolved organic matter [17-19] could also be important factors in mercury accumulation in the food chain. More than 85% forest stamp were flooded in 1974 which caused sufficiency of dissolved organic matter. Forested catchments are an important part of terrestrial Hg cycle and a link between the atmospheric and aquatic environment. Mercury forms strong complexes with dissolved organic matter. Therefore the export of mercury from soils to lakes is largely controlled by the export of dissolved organic matter [20]. Pelcova [21] found positive correlations between C, N, and S content in sediment and mercury adsorption have been found. Finally, sediment composition also plays an important role in mercury adsorption [21].

To establish the possible source of contamination of the aquatic environment at the bottom of the reservoir sediment samples from several sites in the reservoir were analyzed in years 1984 to 1987 and in 1995 [14]. At Borovsko, the approximate mean value, 0.13 mg Hg in 1 kg of dry sediment (or 1.4 mg Hg in 1 kg of organic mass of sediment), is in accord with the data for similar localities in the country, e. g. Podhora reservoir at Mariánské Lázně [14] and is consistent with non-elevated mercury levels.

The mercury content in the fish muscle from the monitored reservoir exceeded the safe limits of mercury in fish for human consumption of 0.5 mg kg<sup>-1</sup> and, for the target predatory fish, 1 mg kg<sup>-1</sup>. This limit was exceeded mainly in the first years after impoundment (until 1988). From 1974 to 1988 approximately 50-70% of analyzed fish exceeded 0.5 mg kg<sup>-1</sup> each year, and up to 40% were higher than 1 mg kg<sup>-1</sup>. No source of mercury in the Želivka Reservoir is known. From 1990 to 1997 no value exceeding 0.5 mg kg<sup>-1</sup> was found. In the most recent years about 10% of analyzed fish have exhibited mercury content over 0.5 mg kg<sup>-1</sup>. This is typical for older individuals of predator fish.

At present, mercury contamination of fish is decreasing and stabilizing to levels found out in non-contaminated locations. This finding is in agreement with some authors [9, 14].

#### 4. CONCLUSION

Although the Želivka reservoir is not considered a mercury contaminated location, unnaturally high mercury levels in fish were obtained after flooding. Higher mercury levels in newly flooded reservoirs in comparison with natural lakes have been observed. Although no sources of mercury contamination are known in the Želivka reservoir, mercury in fish muscle and liver in the first two periods was significantly higher than currently. Increased mercury levels in fish should be taken into account in environmental assessment of proposed reservoir projects.

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