DISTRIBUTION OF MERCURY IN TISSUES OF THE COMMON CARP (*CYPRINUS CARPIO* L.)

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Abstract: The aim of the experiment was to determine the distribution of mercury in ten selected tissues (muscle, skin, fish scales, biliary vesicle, brain, eyes, kidneys, spleen, liver and gills) of common carp (*Cyprinus carpio* L.). Carp fingerlings weighed 47.67±4.61 g. Carps were exposed to increasing concentrations of mercury (0 $\mu g.l^{-1}$ (control), 0.5 $\mu g \cdot l^{-1}$, 1.5 $\mu g \cdot l^{-1}$ and 3.0 $\mu g \cdot l^{-1}$) in fish tanks for 14 days. The concentrations of mercury in fish tanks were continuously monitored and in case of a change they were adjusted to an acceptable value. The fish were not fed during the experiment and mercury got accumulated in fish tissues from fish tank water only. Five fish were collected on the 0^{th}, 4^{th}, 9^{th} and 14^{th} day of experiment from each concentration for the analysis of total mercury content in selected tissues. Total mercury content in water and in selected tissues was determined by the atomic absorption spectrometer AMA 254. The increase of mercury in all tested tissues was not observed in the control group during the 14-day experiment. The time linear increase of mercury content was observed in the muscles, skin, fish scales, biliary vesicle, eyes, kidneys, spleen and gills in all three mercury concentrations under testing. The lowest mercury concentrations were determined in the control group in the range of 0.004–0.052 $mg \cdot kg^{-1}$. Compared to this group, the highest concentration of mercury was found in kidneys (for fish tank with 0.5 $\mu g \cdot l^{-1}$ the mercury concentration was 1.405±0.300 $mg \cdot kg^{-1}$, for fish tank with 1.5 $\mu g.l^{-1}$ the mercury concentration was 5.537±0.027 $mg \cdot kg^{-1}$ and for fish tank with 3.0 $\mu g \cdot l^{-1}$ the mercury concentration was 25.209±2.152 $mg \cdot kg^{-1}$ on day 14 of the experiment).

Key Words: common carp, mercury, atomic absorption spectrometry

INTRODUCTION

As is well known fish are an important constituent of the human diet, but also can represent a dangerous source of certain heavy metals, especially mercury. The monitoring of mercury in the environment is necessary due to the extreme toxicity of its organic forms, its ability of bioaccumulation in aquatic organisms and its long-term persistence in sediments (Ikingura, Akagi 1999, Havelkova et al. 2008).

In many publications the mercury distribution was fish tissues was observed (Houserova et al. 2006, Celechovska et al. 2007, Kruzikova et al. 2013, Cerveny et al. 2014).

These experiments were focused on mercury species accumulation to the fish body through gastrointestinal tract. Only a few publications deal with mercury tissues distribution, if mercury is absorbed to fish tissues only from water environment (Taravati et al. 2012, Kensova et al. 2010).

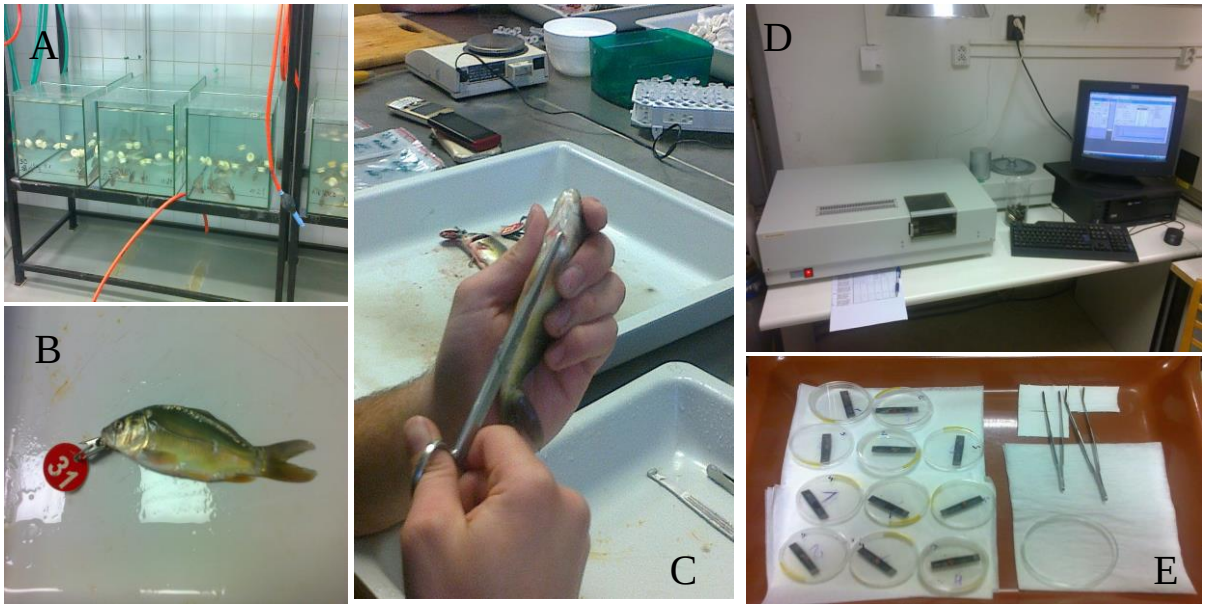
Because common carp (*Cyprinus carpio* L.) is the most consumed fish in the Czech Republic - the observation of accumulation and distribution of mercury in its tissues was the main goal of this study.

MATERIAL AND METHODS

Cyprinus carpio L. (carp fingerlings weighing 47.67±4.61 g) were fed by granules SCREETING F1 PB 40 2.5 mm 10 days before the start of the experiment. This feed contained 0.017 $mg \cdot kg^{-1}$ mercury. Fish were not fed during experiment. Mercury was accumulated to the fish tissues

only from water in glass fish tanks. The glass fish tanks (Figure 1A), which had volume 85 l, were enriched with different concentrations of mercury (control (0 $\mu g.l^{-1}$), 0.5 $\mu g \cdot l^{-1}$, 1.5 $\mu g \cdot l^{-1}$, 3.0 $\mu g \cdot l^{-1}$) for 10 days before experiment. All solutions of mercury were prepared from mercury standard for ICP (c = 1000 $mg.l^{-1}$, Fluka, Canada). During the whole experiment, the concentration of mercury, in the fish tank, were monitored by atomic absorption spectrometer AMA 254 (Altec, Czech Republic) (Figure 1D) and were adjusted in case of change to the appropriate values. The experiment was conducted 14 days. Five fish were collected on the 0^{th}, 4^{th}, 9^{th} and 14^{th} (Figure 1B, 1C). Total 65 fish samples were analyzed. Analyzed tissues of common carp were: muscle, skin, fish scales, biliary vesicle, brain, eyes, kidneys, spleen, liver and gills.

Figure 1 The glass aquariums (A), sampling fish tissues (B, C), atomic absorption spectrometer (D), store the samples before analysis (E)



Determination of total mercury content in *Cyprinus carpio* L. tissues

Atomic absorption spectrometer AMA 254 (Altec, Czech Republic) (Figure 1D, 1E) was used for the determination of total mercury content. Homogenized solid sample of each fish tissues was directly weighted (10± 0.1mg) into pre-cleaned combustion boat, and inserted into the AMA 254 analyser. Samples were dried at 120°C for 60 s and thermally decomposed at 550°C for 150 s under oxygen flow. The selectively trapped mercury was released from the amalgamator by a brief heat-up and finally quantified (measuring cycle, 57 s) as Hg^0 by cold-vapor AAS technique at 253.5 nm. The limit of detection for the determination of mercury was 0.11 $\mu g \cdot kg^{-1}$.

Statistical analyses

Statistical analyses of metal content in tissues were made using one-way analysis of variance (ANOVA) and statistical significance was declared when *p* value was equal to or less than 0.05.

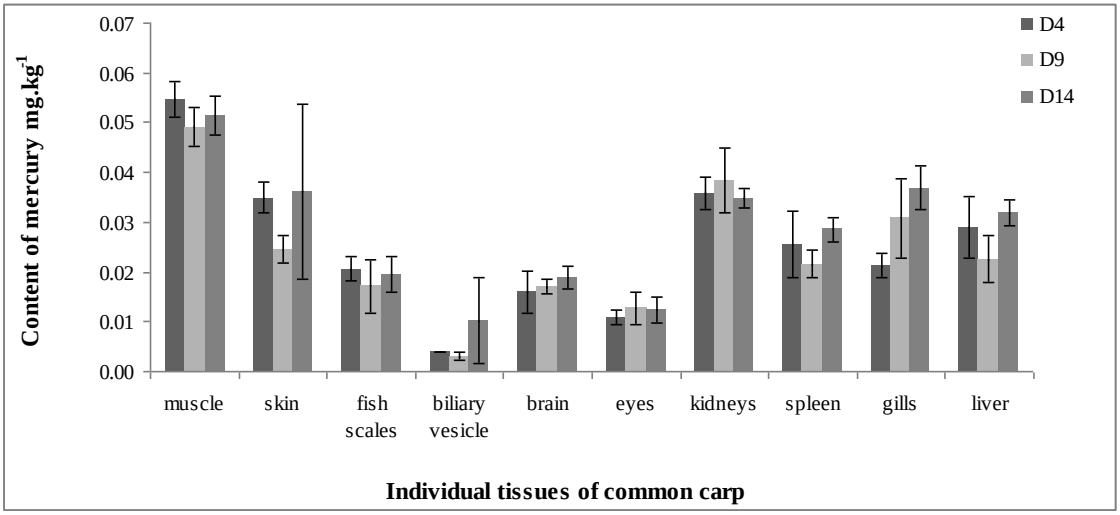
Method validation

The reference material DORM-4 (fish protein Canada, T-Hg: 0.410 ± 0.055 $mg \cdot kg^{-1}$) was used for method validation. Content of mercury in reference material measured by AMA 254 was 0.408±0.009 $mg \cdot kg^{-1}$.

RESULTS AND DISCUSSION

The average contents of mercury in tissues of common carp in control group are shown in Figure 2.

Figure 2 The average contents of mercury in tissues of Cyprinus carpio L. in the control groups (p<0.05)

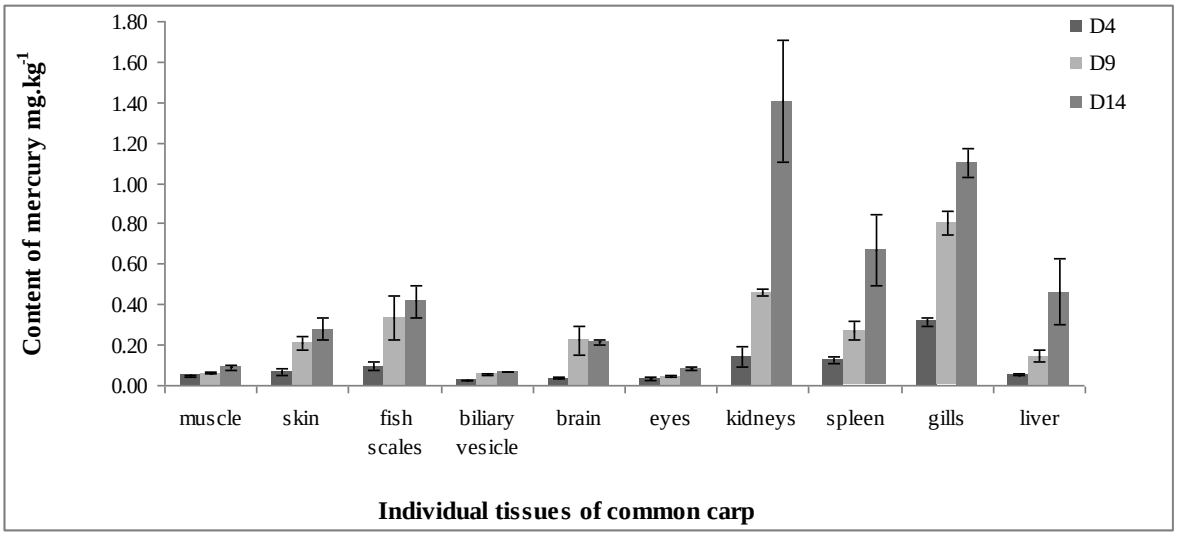


Legend:D4 = 4th day, D9 = 9th day, D14 = 14th day

In comparison with another groups the highest mercury contents were found in control group in muscle (0.052±0.004 mg · kg^{-1}) on day 14 of the experiment, the lowest contents in biliary vesicle (0.010±0.009 mg · kg^{-1}) on day 14 of the experiment. Kruzikova et al. (2013) demonstrated that in non-contaminated locations total mercury concentrations in the muscle are significantly higher compared to liver. Mercury content in water was below the limit of detection (0.11 µg · kg^{-1}). Ten days before start of the experiment fish fed granules, which contained 0.017 mg · kg^{-1} of mercury. Mercury concentrations in the analyzed tissues of the control group probably came from feed. The increase of mercury in all tested tissues was not found in the control group during the 14-day experiment.

The average contents of mercury in carp tissues in the group with concentration 0.5 µg · l^{-1} are shown in Figure 3.

Figure 3 The average contents of mercury in tissues of Cyprinus carpio L. in 0.5 µg · l^{-1} concentration (p<0.05)



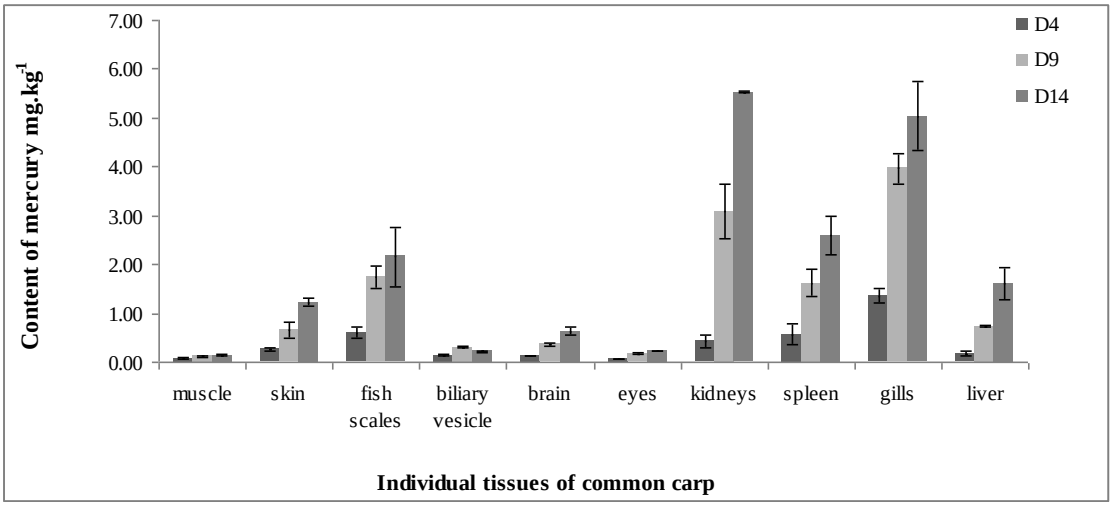
Legend:D4 = 4th day, D9 = 9th day, D14 = 14th day

Total mercury content in selected carp tissues was gradually time increased with the advancing days in muscle, skin, fish scales, biliary vesicle, eyes, kidneys, spleen, gills and liver. The highest concentration of mercury was measured in the kidneys (1.405±0.300 mg · kg^{-1}) on day 14 of the experiment. The lowest concentration of mercury was in the biliary vesicle (0.02±0.003 mg · kg^{-1}) on day 4 of the experiment. The total mercury content in tissues of common carp decreased in order gills = kidneys > spleen > fish scales = liver > skin = brain > muscle = eyes = biliary vesicle.

Statistically significant differences *(p<0.05)* in total mercury contents were observed among gills, spleen, fish scales, liver and brain with time accumulation. Among muscle, eyes and biliary vesicle the differences were not statistically significant *(p<0.05).*

The average contents of mercury in tissues of common carp in the group, where content of mercury was 1.5 μg · l^{-1}, are shown in Figure 4.

Figure 4 The average contents of mercury in tissues of Cyprinus carpio L. in 1.5 μg · l^{-1} concentration (p<0.05)

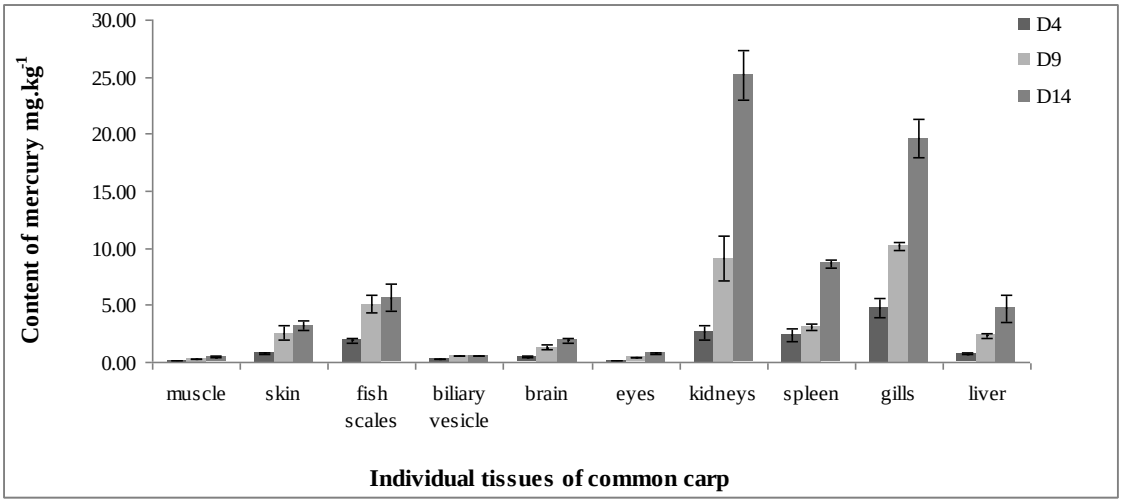


Legend:D4 = 4th day, D9 = 9th day, D14 = 14th day

The total mercury content was gradually increased with the advancing days. The highest concentration of mercury was measured in the kidneys (5.537±0.027 mg · kg^{-1}) on day 14 of the experiment. The lowest concentration of mercury was in the eyes (0.062±0.011 mg · kg^{-1}) on day 4 of the experiment. The total mercury content in tissues of carp decreased in order gills = kidney > spleen = fish scales > liver > skin > brain > biliary vesicle = eyes = muscle. Statistically significant differences *(p<0.05)* in total mercury contents were observed among gills, spleen, liver, skin and brain with time accumulation. Among muscle, eyes and biliary vesicle and between gills and kidney the differences were not statistically significant *(p<0.05).*

The average contents of mercury in tissues of common carp in the groups with mercury concentration 3.0 μg.l^{-1} are shown in Figure 5.

Figure 5 The average contents of mercury in tissues of Cyprinus carpio L. in 3.0 μg · l^{-1} concentration (p<0.05)



Legend:D4 = 4th day, D9 = 9th day, D14 = 14th day

Content of mercury in carp tissues were gradually increased with the advancing days in group with concentration 3.0 μg.l^{-1}. The highest content of mercury was measured in the kidneys

(25.20±2.15 mg · kg^{-1}) on day 14 of the experiment. The lowest content of mercury was in the muscle (0.144±0.020 mg · kg^{-1}) on day 4 of the experiment. The total mercury content in tissues of common carp decreased in order kidney = gills > spleen = fish scales > liver = skin > brain > eyes = biliary vesicle = muscle. Statistically significant differences *($p<0.05$)* in total mercury contents were observed among kidney, spleen, liver, brain and eyes with time accumulation. Among muscle, eyes and biliary vesicle, between spleen and fish scales and between liver and skin the differences were not statistically significant *($p<0.05$)*.

In the control group the contents of mercury in carp tissues on day 14 of the experiment were in the range 0.010–0.052 mg · kg^{-1}. In the fish tanks with concentration 0.5 µg · l^{-1} the contents of mercury in carp tissues on day 14 of the experiment were in the range 0.067–1.405 mg · kg^{-1}. In the fish tanks with concentration 1.5 µ · l^{-1} the contents of mercury in common carp tissues on day 14 of the experiment were in the range 0.224–5.537 mg · kg^{-1} and in the fish tanks with concentration 3.0 µg · l^{-1} the contents of mercury in common carp tissues on day 14 of the experiment were in the range 0.498–25.209 mg · kg^{-1}.

CONCLUSION

The obtained results in this study present the differences in the distribution of mercury among common carp (*Cyprinus carpio* L.) tissues. Overall the highest content of mercury was observed in detoxification organs (kidneys, spleen and liver) and skin, fish scales and gills. The lowest contents of mercury were determined in muscles and biliary vesicles. In the control group contents of mercury were similar to fish caught in the clean aquatic environment. Concentration of mercury in tissues of common carp increased with time accumulation of mercury and with mercury concentration in water environment.

ACKNOWLEDGEMENT

This research was financially supported by the: Innovation of studies programs at Mendel university in Brno for integration between Faculty of Agronomy and Faculty of Horticulture OP VK CZ.1.07/2.2.00/28.0302.

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