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Preliminary Studies on Mercury Bioaccumulation within Various Fish Tissues as Heavy Metal Stressor in Aquatic Ecosystems

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This study was carried out to determine bioaccumulation levels of total mercury on various fish tissues in two streams (Banseok and Hasin Stream) and one lake (Yeongsan Lake) during April-May 2007. We also determined natural background levels of total mercury bioaccumulation to evaluate relative individual impacts of fish and compared the levels with reference sites as a preliminary study to evaluate heavy metal stressor using fish. For the study, we collected fishes in the sampling sites and analyzed the concentrations of total mercury in the liver, kidney, gill, vertebral column, and muscle tissues using Direct Mercury Analyzer (DMA-80, US EPA Method 7473). The levels varied depending on the types of waterbody and tissues used. Concentrations of total mercury ranged between 5.1 μg kg^{-1} and 108.6 μg kg^{-1} in the streams and between 5.3 μg kg^{-1} and 87.3 μg kg^{-1} in the reservoir, and the values were highest in the muscle tissues. Levels of natural background levels of total mercury, even though the sampling number was few, averaged 23.6 μg kg^{-1} in the study sites. The individual and mean values in each system was not so high in terms of US EPA criteria of fish health and human health, indicating that the impact was minor in the study site. Further studies should be done for the determination of mercury levels in the systems

Key words : total mercury, Hg, bioaccumulation, fish, tissue, aquatic ecosystem

INTRODUCTION

Numerous researches (Lee, 1985; Eum *et al.*, 1987; Sohn *et al.*, 1992; Hwang *et al.*, 1992; Bang and Choi, 1993; Hwang *et al.*, 1998; Cho *et al.*, 2004) have been reported that mercury is toxic on aquatic biota such as fishes in the aquatic environments and has bioaccumulation effects of heavy metal, especially fish liver and fat tissues. Lee *et al.* (2001) pointed out that mercury along with lead and cadmium caused deleterious effects on various tissues of biological organisms in Korea. The mercury inputs in aquatic ecosystems are known as to be frequently caused by effluents from smelting industries, wastewater disposal plants, and rice paddy by agricultural pesticides (Lee *et al.*, 2007). Mercury in aquatic ecosystems is accumulated in the water column and then deposited in habitat substrate. These sources in stream ecosystems can be uptaken by various trophic levels of aquatic biota through a periphyton as a primary producer, macroinvertebrate as a secondary consumer, and fish as a secondary or tertiary consumer, resulting in degradations of aquatic ecosystem health and human health problems by fish consumption.

US EPA (1994, 2000, 2005) alerted that metal mercury can cause human health problems such as decreases of eye sight and hearing abilities

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and the nerve defect when human consume regularly fish contaminated by mercury. Thus, US EPA (1994, 2000, 2005) established fish advisory criteria on mercury contamination for public health, but these studies have been mainly focused on seawater fish rather than freshwater fish. For this reason, little is known about mercury contamination and bioconcentration in freshwater fish, in Korea as well as other developed countries. Recently, just a few studies on mercury contamination of freshwater fish have been done in aquatic ecosystems in Korea (Kim and Park, 1981; Kang *et al.*, 1986; Sohn *et al.*, 1992; Bang and Choi, 1993).

Previous few studies of heavy metal on fish tissue suggested degradations of water quality in various streams and rivers. Reseach Institute of Pubulic Health and Environment conducted fish survey in 2000 and Korean Federation for Environmental Movement conducted in 2003. These studies found that in 2000, total mercury concentration was 40 $\mu g\ kg^{-1}$ in the tissues of *Carassius auratus*, while it was 154 $\mu g\ kg^{-1}$ in 2003. This founding implied the rapid bioaccumulation of mercury during the short period. Also, mercury levels in Cyprinidae fish were surveyed in various watersheds such as Han River, Junrang Stream, Tan Stram and Chungpyung Reservoir (Reseach Institute of Pubulic Health and Environment, 2006).

The objectives of this study were to determine mercury levels in various fish tissues such as liver, kidney, gill, vertebral column, and muscle tissues in aquatic ecosystems, and to compare the spatial levels among the sampling sites within aquatic ecosystem. These preliminary outcomes may provide basic data of mercury levels depending on various fish tissues through the field sampling.

MATERIALS AND METHODS

1. Sampling sites and periods

This study was conducted at three sites of Banseok Stream, which is 1st order stream and is located in the military base, and one site of a upstream reach of Hasin Stream during April-May 2007. We collected fish samples twice from both streams to analyze the mercury contents in the various fish tissues. Also, we took fish samples from five sites in the Yeongsan Lake during May 2007. The purpose of the sampling form the lake was to compare the mercury level of the lake, which is known as polluted by various chemicals including nutrients, heavy metals, and pesticides (Lee *et al.*, 2007). The sampling sites and sampling dates are as follows (Fig. 1):

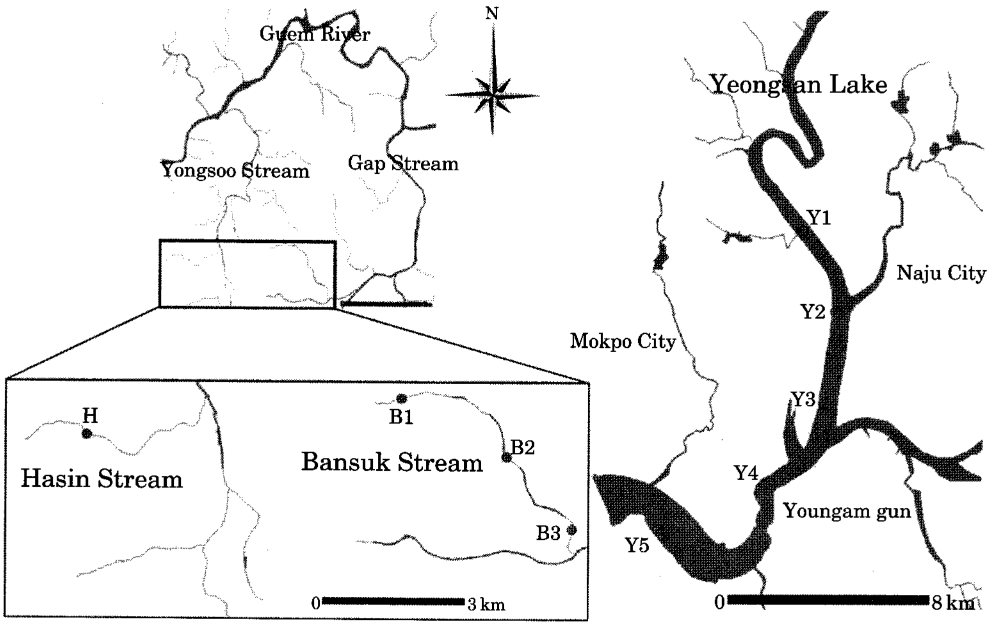

Fig. 1. Sampling sites in Hasin Stream (H), Banseok Stream (B), and Yeongsan Lake (Y).

Hasin Stream (H)
- H: Hasin-ri, Banpo-myeon, Gongju city, Korea

Banseok Stream (B)
- B1: Military base, Banseok-dong, Yuseong-gu, Daejeon city, Korea
- B2: Jyukdong bridge, Hagi-dong, Yuseong-gu, Daejeon city, Korea
- B3: Gungdong 2^{nd} bridge, Jangdae-dong, Yuseong-gu, Daejeon city, Korea

Yeongsan Lake (Y)
- Y1: Dangho-ri, Mongtan-myeon, Muan-gun, Jeonnam Province, Korea
- Y2: Bokryong-ri, Ilro-eup, Muan-gun, Jeonnam Province, Korea
- Y3: Euisan-ri, Ilro-eup Muan-gun, Jeonnam Province, Korea
- Y4: Chungho-ri, Ilro-eup Muan-gun, Jeonnam Province, Korea
- Y5: Nabul-ri, Samho-myeon, Toungam-gun, Jeonnam Province, Korea

2. Sampling methods

We took fish samples using the sampling gears such as casting net (5 × 5 mm) and kick net (4 × 4 mm). The fish population for the mercury analysis in the upstream reach of Bansuk Stream was *Rhynchocypris oxycephalus*, which was a sole species sampled in the sampling sites, while the population sampled in the downstream was *C. auratus*, which was dominant species. Fish population of *R. oxycephalus* sampled in the Hansin Stream, which was considered as a reference site, was acclimated in the laboratory during one month. In addition to the stream samples, two populations were sampled in the Yeongsan Lake; one population was *C. auratus*, which is known as an omnivore species and feed mainly bottom sediments and the other was *Micropterus salmoides*, which is known as piscivore species and is top exotic carnivore species. We disected the fish and then took samples of various fish tissues such as liver, kidney, gill, vertebrae, and skin muscles.

3. Methods analyzing total mercury

Concentrations of total mercury (total [Hg]) in the various fish tissues were analyzed by Direct Mercury Analyzer (DMA-80, US EPA Method 7473; Fig. 2) The method 7473 of US EPA (2000) was known as a spectroscopic analytical method which have no any pre-treatments of samples and ignite at high temperatures. Samples of fish tissues were analyzed by the DMA-80, based on thermal decomposition, catalytic reduction, and amalgamation desorption. The samples of liver, kidney, gill, vertebral column, and muscle tissues were initially dried in the oxygen stream passing through a quartz tube located inside a controlled heating coil under the condition of oxygen supplies as a carrier gas to each cylinder. The combustion gases were further decomposed on a catalytic column at 750°C. Mercury vapor was collected on gold amalgamation traps and subsequently desorbed for a quantification. Thus, the mercury content was determined using a single

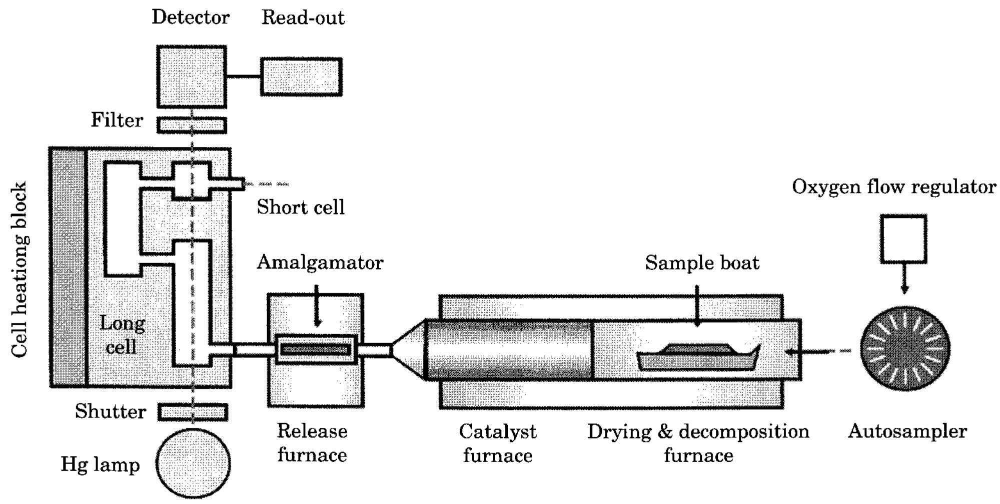

Fig. 2. System structure of Direct Mercury Analyzer (DMA-80).

Table 1. Total [Hg] concentrations in Hasin Stream (H) and Banseok Stream (B).

Sites	Species	Types of tissues	1^{st} survey (μg kg^{-1})	2^{nd} survey (μg kg^{-1})	Mean (μg kg^{-1})
		Liver	21.1	31.5	26.3
		Kidney	20.3	34.7	27.5
H	*Rhynchocypris oxycephalus*	Gill	32.8	21.9	27.4
		Vertebral column	22.3	36.2	29.3
		Muscle	44.8	69.7	57.3
		Liver	62.6	34.7	48.7
		Kidney	72.3	34.9	53.6
B1	*Rhynchocypris oxycephalus*	Gill	64.3	18.6	41.5
		Vertebral column	127.6	62.5	95.1
		Muscle	88.7	108.6	98.7
		Liver	5.1	15.0	10.1
		Kidney	4.9	18.0	11.5
B2	*Carassius auratus*	Gill	7.8	8.1	8.0
		Vertebral column	14.5	18.6	16.6
		Muscle	16.8	31.3	24.1
		Liver	12.2	13.3	12.8
		Kidney	22.5	14.5	18.5
B3	*Carassius auratus*	Gill	6.5	7.1	6.8
		Vertebral column	18.3	18.2	18.3
		Muscle	36.3	33.9	35.1

beam spectrophotometer with two sequential, flow-through measurement cells under the condition of 254 nm. The light source for the spectrophotometer was a low pressure mercury vapor lamp and the silicon UV photo-detector was used for the analysis.

RESULTS AND DISCUSSION

Total mercury [Hg] concentrations in Hasin and Banseok streams varied from 4.9 μg kg^{-1} to 17.6 μg kg^{-1} depending on the types of fish tissues, fish species, and the survey period (Table 1). Various tissue analyses of *R. oxycephalus* showed that the bioaccumulation levels were muscle (78 μg kg^{-1}), vertebral column and kideny (41 μg kg^{-1}), respectively, liver (78 μg kg^{-1}), and gill (35 μg kg^{-1}) in the order of magnitude in Hasin and Banseok streams. The species of *R. oxycephalus* was dominant and characteristic species in the B1 and H sites. In contrast, gold fish of *C. auratus*, which is known as omnivore and tolerant species, was a dominant species in the B2 and B3 sites. Concentrations of total mercury in the species were highest in the muscle (30 μg kg^{-1}) and then in the vertebral column (17 μg kg^{-1}). The levels of total Hg in the *C. auratus* showed a

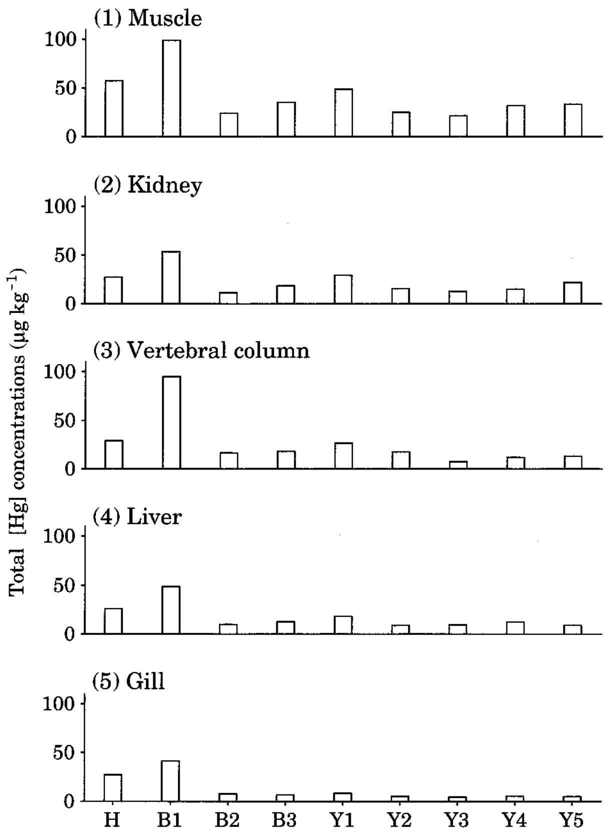

Fig. 3. Total [Hg] concentration of each tissues in sampling sites.

Table 2. Total [Hg] concentrations in Yeongsan Lake (Y).

Species	Types of tissues	Sites				
		Y1	Y2	Y3	Y4	Y5
Carassius auratus (μg kg^{-1})	Liver	11.8	8.6	6.6	5.9	8.1
	Kidney	18.1	11.9	10.3	7.8	14.8
	Gill	8.5	6.2	8.1	19.1	5.3
	Vertebral column	28.7	21.9	7.2	14.8	16.3
	Muscle	43.2	29.3	14.8	22.1	31.4
Micropterus salmoides (μg kg^{-1})	Liver	32.7	23.8	16.7	21.3	13.5
	Kidney	33.6	22.1	6.0	6.7	16.2
	Gill	16.5	12.0	5.8	7.1	5.8
	Vertebral column	36.0	27.4	9.0	13.7	10.5
	Muscle	87.3	59.2	40.8	43.5	34.1

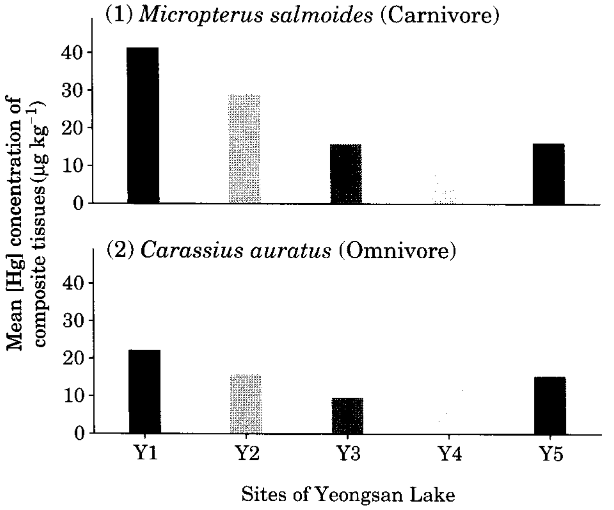

Fig. 4. Mean [Hg] concentration of composite tissues in Hasin Stream (H) and Banseok Stream (B).

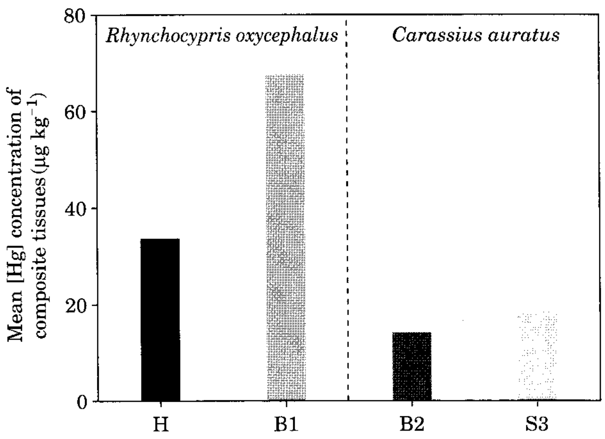

Fig. 5. Mean [Hg] concentration of composite tissues in Yeongsan Lake (Y).

similar pattern with those in the *R. oxycephalus* (Fig. 3).

In addition to the stream samples, we collected fish samples of *M. salmoides* and *C. auratus* from a lentic waterbody of Yeongsan Lake (Y). We selected the *M. salmoides* which is a top carnivore and dominant species in the aquatic ecosystem. In all sites (Y1-Y5) of the reservoir, mean concentrations of *M. salmoides* were 53 μg kg^{-1} in muscle, 22 μg kg^{-1} in liver, and 19 μg kg^{-1} in vertebral column (Table 2). In the mean time, mean levels of *C. auratus* in muscle were 28 μg kg^{-1}, which was highest values among the various tissues and the mean in the liver tissues was 18 μg kg^{-1} (Table 2). Thus, the mercury level in muscle tissues was higher than any other types of tissues in Yeongsan Reservoir (Fig. 3). Total mercury concentration in *M. salmoides* as top carnivore, based composite sample tissues in the dataset of Y1-Y5 sites, was 1.5 times higher than that of *C. auratus*. Also, the content in the *M. salmoides* in the muscle and liver was 1.9 and 2.6 times, respectively greater than those of *C. auratus* (Fig. 4). This result suggests that the bioaccumulation level is greater in the piscivorous fish than in the omnivorous fish as shown in numerous researches in other countries. For this reason, it is necessary to establish a database system of many fish species collected from various types of watersheds in the future studies.

Mean level of total mercury in the *R. oxycephalus* was greater two fold in the B1 site (67.5 μg kg^{-1}) than the level in the H site (33.6 μg kg^{-1}, Fig. 5). Especially, mercury level in the vertebral column was almost 3.2 times higher in the B1 site (95 μg kg^{-1}) than that of site H (29 μg kg^{-1}, Fig. 3). This phenomenon may be associated with mercury inputs from the nearby military base, but this guess should be further studied in the future. All tissues in *C. auratus* did not show large differences between B2 site and B3 site (Figs. 3, 5). In this study, we expected that mercury contents of tolerant species were greater than the sensitive species, but the result was opposite. Mercury level of *R. oxycephalus* (50.5 μg kg^{-1}) common sensitive species habitating living in clean upstream reach, was higher than that of tolerant *C. auratus* (16.1 μg kg^{-1}). This result indicates that the mercury bioaccumulation varied depending on the species used, and previous study of Eum *et al.* (1987) support our results.

Our results in mercury levels showed that muscle and vertebral column had greater than any other types of fish tissues. This finding agreed with previous researches of Kim and Park (1981) and Kang *et al.* (1986). In contrast, this finding was contrasted with previous research (Lee, 1985; Hwang *et al.*, 1998), in which liver and kidney had greater bioaccumulation than any other types of fish tissues. Hereafter, further researches to analyze bioaccumulation of the mercury at each tissues is necessary for demonstrating the bioaccumulation impacts on mercury in aquatic ecosystem.

In this study, we found that there were some differences in total mercury concentration depending on the types of waterbody and types of fish tissues used. However, the mercury impacts on bioaccumulation in the fish tissues was minor, based on the US EPA criteria of total mercury (US EPA, 1994, 2005), in this study. However, still it will be harmful to aquatic ecosystems when the mercury has continuous bioaccumulation in the fish tissues without any control action. This study is a preliminary study to evaluate heavy metal stressor in the various fish tissues and waterbodies. We suggest that measurements of the mercury level throughout the extensive fish sampling from various watersheds are conducted for effective watershed managements and conservations.

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