STATUS OF MERCURY CONTAMINATION IN MUSCLE OF WALLAGO CATFISH FROM AYEYARWADY RIVER SEGMENT BETWEEN SINGU AND SAGAING TOWNSHIPS, MANDALAY REGION, MYANMAR

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Abstract

The Impact of mercury pollution has been a critical issue worldwide. The present study focused on assessment of mercury contamination status in Ayeyarwady River using adult Wallago catfish as bio-indicator during June 2017 to December 2019. Mercury concentration in the muscle of fish was analysed by mercury analyser (MA-3 Solo NIC). Mean mercury concentration in fish muscle was recorded as 0.62 ± 0.23 ppm (dry weight basis), and 0.14 ± 0.05 ppm (wet weight basis). Average mercury concentration in the fish muscle in wet weight basis showed lower than the WHO (1990) standard ($<0.5\mu g/g$), while higher than this standard in dry weight basis. Mercury concentration in fish muscle of the present study was about five times lower than last five years record in Ayeyarwady River. Illegal gold mining is the major source of mercury contamination to the aquatic ecosystem of the Ayeyarwady River, so as education program to local people and regular monitoring about the hazard of mercury are effective tools for the conservation of vulnerable aquatic organisms and health safety of the people. This finding would be provided the information for assessing the impact of the mercury pollution on biodiversity, especially Irrawaddy Dolphin in the Ayeyarwady River of Myanmar.

Keywords: Mercury contamination, Wallago, bio-indicator, Ayeyarwady River.

Introduction

Nowadays, the hazard of mercury pollution, especially in aquatic environment, has been a critical issue in environmental management, conservation of wildlife and public health around the world. An estimation of the level of contamination in a particular environment can be revealed by the assessment of the status of aquatic organisms such as algae, macrophyte, zooplankton, bivalve mollusks, seabirds and fish (Manickavasagam *et al.*, 2019). Many researchers recommended the fish as a bio-indicator to detect the mercury contamination in the environment because of highly bio-accumulation in its body (Olaifa *et al.* 2004).

Methyl mercury toxicity can cause a neurological disorder called Minamata disease in humans. This mercury-related disease occurred in Japan round about 1950-1960 when mercury pollution occurred in Minamata Bay due to the wastewater discharges of the chemical industry (Kyaw Myint Oo, 2010). People ingested the fish and shellfish contaminated with mercury developing neurological symptoms as loss of consciousness and sometimes death. This disaster pointed out the importance of mercury management and alarmed to developed countries. Recently, mercury pollution has become a serious problem not only in developed countries but also in developing countries, since excessive use of mercury in artisanal and small-scale gold mining has increased in developing countries (Harada, 1995).

In Myanmar, some research works on mercury contamination in fishes from some segments of Ayeyarwady River were carried out by Wildlife Conservation Society and Whale and Dolphin Conservation Society (Smith, *et al.* 2003), Khin Myint Mar (2011), Soe Soe Aye and Khin Ni Ni Win (2015). However, regular and localized studies of mercury contamination in fish muscles are still needed to conduct for assessing the mercury pollution along the Ayeyarwady River. Therefore,

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the present study was focused on mercury contamination status in Ayeyarwady River using Wallago catfish as bio-indicator with following objectives:

- to detect the mercury concentration in muscle of Wallago catfish collected from the study area in relation to well-being of fish
- to evaluate the safety consumption to Wallago catfish in the study area using WHO (1990) standard.
- to assess comparatively the status of mercury contamination in the Ayeyarwady River using the recorded data of present and previous studies.

Materials and Methods

Study Area and Study Period

Study area is the Ayeyarwady River segment between Singu and Sagaing townships, Mandalay Region, Myanmar, and located between $22^{\circ}33^{\circ}N$, $95^{\circ}58^{\circ}E$ and $21^{\circ}52^{\circ}N$, $95^{\circ}59^{\circ}E$ (Figure 1). It is included the portion of Irrawaddy Dolphin protected area (from Kyauk Myaung to Mingon) where a biologically unique human-dolphin cooperative fishery is famous. Study period lasted from June 2017 to December 2019.

Studied Fish Species

Fish species for analysing mercury contamination was chosen *Wallago attu* (Bloch & Schneider, 1801) of Family Siluridae. It is carnivore, bottom dweller and potamodromous fish. *Wallago attu* was chosen as target fish species because it is sensitive indicator of environment according to the finding of Singh and Tandon (2009).



Figure 1 Location map of the study area

Specimen Collection

Adult fish specimens were purchased from local fishermen in the study area. Specimen collection was conducted as three specimens per month. A total of 30 specimens including 9 samples in the rainy season, 12 samples in the cold season, and 9 samples in the dry season were collected during the study period. Fish specimens were not available in June and July when the fishing activities were legally prohibited due to the spawning season. Collected specimens were

kept in the ice box and bring to the laboratory of Zoology Department, University of Mandalay for preparing mercury analysis.

Preparation for Mercury Analysis

Collected fish specimens were washed with distil water, skinned and cut out approximately 50g of the axial muscles. Then, fish muscle was cut into slices for dry rapidly and weighted in wet condition. Consequently, flesh slices were dry in drying oven at 60°C until reaching the constant weight. Each dry specimen was weighted again and kept in separate polyethylene bag and stored in the refrigerator at 20°C before mercury analysis. Code number of each specimen, collection date, wet weight and dry weight were labelled on the respective specimen bag.

Method of Mercury Analysis

Each specimen was homogenized by using electric blander before conducting mercury analysis. And then, 5g each of three specimens collected in the same month were composed into one sample and mix thoroughly for mercury analysis.

Digestion procedure was conducted followed after Hajeb *et al.* (2009). Firstly, 0.1g of dry powder of fish muscle was weighted using analytical balance and put into 100ml digestion tube, then, 5ml of analytical grade nitric acid (HNO₃) was added. After that the mixture was digested at 40-90°C in water bath for 3hs till getting clear solution. The digested sample was then cooled at room temperature about 30mins and subsequently diluted 40ml volume with deionized water. Blank solutions were prepared at the same time. Consequently, mercury concentrations in samples were detected duplicating for each sample using mercury analyser (MA-3 Solo NIC) at Nanova Laboratory, Yangon.

The unit of mercury concentration was expressed as microgram per gram ($\mu g/g$).

The mercury concentration in fresh fish was calculated according to the method of Sanders, *et al.* (2008).

Moisture on dry weight basis = $\frac{\text{Concentration dry weight}}{1 + \text{moisture on dry weight basis}}$

Hg concentration (wet weight) = $\frac{\text{Wet weight} - \text{Dry weight}}{\text{Dry weight}}$

Condition Factor

Condition factor (K) was calculated according to Bagenal (1978) as follows:

$$K = 100 \text{ W/L}^3$$

Where W is the total body weight in grams and L the standard length in centimeters, the factor 100 is used to bring K close to a value of one. The number 1 indicates a "normal" fish in good condition.

Statistical Analysis

Recorded data were statistically analysed using Microsoft Excel 2010 and IBM SPSS Statistics Version 22. Variation of the mercury concentration among the seasons was analysed by "t" test. Relationship between mercury concentration in the fish muscle and condition factor of fish was analysed by Pearson's correlation coefficient and regression tests.

Mercury Level Limit for Human Consumption

Safety guideline consumption of fish by WHO (1990) was $<0.5 \mu g/g$ of mercury.

Results

Analysis of mercury concentration in the muscle of Wallago Catfish based on 30 specimens in the study area indicated that mercury concentration in fish muscle varied seasonally (Table 1). Mean mercury concentration in fish muscle was found to be significantly highest in the rainy season ($0.85\pm0.01 \ \mu g/g dry$ weight basis and $0.19\pm0.03 \ \mu g/g$ wet weight basis) (t=9.798, p<0.001), decreased in the cold season ($0.60\pm0.13 \ \mu g/g dry$ weight basis and $0.13\pm0.03 \ \mu g/g$ wet weight basis), and slightly increased again in the dry season ($0.66\pm0.25 \ \mu g/g dry$ weight basis and $0.16\pm0.06 \ \mu g/g$ wet weight basis).

Season	No. of	Mercury concentration (µg/g)				
		Dry Weight Basis		Wet Weight Basis		
		Mean±SD	Range	Mean±SD	Range	
Rainy	9	0.85 ± 0.01	0.84-0.85	0.19±0.03	0.18-0.22	
Cold	12	0.60 ± 0.13	0.44-0.75	0.13 ± 0.03	0.10-0.17	
Dry	9	0.66 ± 0.25	0.37-0.85	0.16 ± 0.06	0.09-0.21	

Table 1 Mercury concentration in	the muscle of	f Wallago catfi	sh in	different seasons
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The condition factor was analyzed to detect the well-being of fish in the study area during the study period (Figure 2). Monthly variation of condition factor value was observed, the lowest value (K=0.33) was recorded in August 2017 while the highest value (K= 0.96) was recorded in May 2018. In all studied fish samples, the condition factor values were found to be less than one indicating poor condition of fish in all seasons. During the study period, condition factor values of studied fish samples were significantly negative correlation with mercury concentrations in their muscle (r=-0.65, p<0.05) (Figure 2).



Figure 2 Relation of monthly condition factor value and mercury concentration in the muscle of Wallago catfish in the study area

WHO mercury level limit for human consumption is noted as <0.5 μ g/g. Mercury concentrations (wet weight basis) of all studied fish samples were detected to be lower than the permissible limit of safety consumption (<0.5 μ g/g) in all seasons. However, mercury concentrations (dry weight basis) in most of samples were found to be exceeding the permissible limit of safety consumption (>0.5 μ g/g) in all seasons as 100% of fish in the rainy season, 75% of fish in the cold season and 67% of fish in the dry season (Figure 3).



Figure 3 Percentage of fish exceeding WHO Hg level limit in different seasons

The comparison of the present and previous studies analyzed on mercury concentration in the muscle of Wallago Catfish indicated that the mercury concentration increased significantly in different segments of the Ayeyarwady River from 2003 to 2015, while mercury concentration decreased remarkably at the present (Table 2).

Location		Mercury			
Region	Ayeyarwady River Segment	Concentratio n (µg/g)	Reference	Remarks	
Mandalay	Bamaw and Kyauk Myaung	0.317	Smith <i>et al.</i> (2003)	<who standard<="" td=""></who>	
Mandalay	Mandalay	0.674	Khin Myint Mar (2011)	> WHO Standard	
Magway	Magway	0.978	Khin Myint Mar (2011)	> WHO Standard	
Magway	Pakokku	0.671	Soe Soe Aye & Khin Ni Ni Win (2015)	> WHO Standard	
Mandalay	Singu and Sagaing	0.142	Present Study (2019)	< WHO Standard	

Table 2	Comparison of	mercury concentr	ration (µg/g	wet weight)	in muscle	of Wallago
	catfish between	present study and	previous stud	lies of Myanı	mar	

Mercury concentration in the present study was about five times lower than the data of last five years ago at Pakokku Segment (Soe Soe Aye and Khin Ni Ni Win, 2015) and the data of last nine years ago at Mandalay Segment (Khin Myint Mar, 2011). The highest mercury concentration (0.978 μ g/g) in the muscle of Wallago catfish was noted in Magway Segment (Khin Myint Mar, 2011), it is nearly seven times higher than the Hg concentration of the present study. These previous records were higher than the WHO permissible limit of human consumption (>0.5 μ g/g).

Discussion

Ayeyarwady River, one of the largest free-flowing rivers in Southeast Asia, is not only unique and special, but also the lifeline of Myanmar and majority of the people is dependent on the river for their daily life. The water quality of Ayeyarwaddy River has been in decline for many years especially due to mining operations, deforestation, and lack of soil protection and other human activities (Bowles, 2013). The ecosystem of the Ayeyarwady River is vulnerable especially due to the artisanal and small-scale gold mining by using mercury (Yousafzai *et al.*, 2010).

In the present study, Wallago catfish was used as bio-indicator to detect the mercury contamination in the Ayeyarwady River, since it is carnivorous, bottom dweller and potamodromous. They have more chances to contact both water and sediment ran off from gold mining and mercury used from artisanal and small-scale gold mining. In the rainy season, high and torrent water drags mercury contaminated sediment from upstream to the downstream. The more mercury-contaminated sediment deposited the more chance to contact the fish, so as mercury concentration in fish muscle was detected to be higher in the rainy season. This finding is similar to the finding of Soe Soe Aye and Khin Ni Ni Win (2015) who studied the Ayeyarwady River Segment in Pakokku Township, Magway Region. Therefore, mercury contamination can be predicted not only in the study area but also in the upstream of the river by analysing of Wallago catfish. Gupta (2015) also documented Wallago catfish as a good bio-indicator for heavy metal pollution in the river.

The previous authors stated that very low-levels of pollution may have no apparent impact on the fish itself, which would show no obvious signs of illness. However, fecundity of fish population may decrease gradually, and fish population may decline in long term leading to extinction of these important natural resources (Dupuy *et al.*, 2014). In the present study, condition factor values of studied fish samples were detected to be less than one indicating poor condition of these fishes. Besides, these values were negatively correlated with the concentration of mercury in their muscles. Although these fishes have not showed serious symptoms of mercury toxicity at the moment, they may suffer gradually weakness of their health by mercury toxicity in their lifetime and this impact may lead to their next generations as stated by previous authors. It is the warning for conservation of the endemic and endangered species inhabiting in the study area especially Irrawaddy Dolphin since they can also suffer long term impact of mercury toxicity the same as the case of the studied Wallago catfish.

Methyl mercury is an intensely toxic developing neurological symptom and can enter into the human body by various ways. One of the ways to enter mercury into the human body is ingesting such polluted fish resulting in mercury toxicity (Harada, 1995). Kyaw Myint Oo (2013) suggested that it is a good choice to eat fish once or twice a week for getting cancer fighting fats, but beware the hazard of mercury which is a contaminant accumulated in many fish species. In the present study, average mercury concentration in the muscle of fish samples as wet weight basis was lower than the permissible limit of WHO standard $(0.5\mu g/g)$ in all seasons, while in the muscle of fish as dry weight basis was higher than this standard. Therefore, it should be caution that consuming a large amount of dried and salted fish made in the study area can be dangerous to regular fish consumers in all seasons.

The data of the present study and previous studies (Smith *et al.*, 2003; Khin Myint Mar, 2011; Soe Soe Aye and Khin Ni Ni Win, 2015) indicated the effect of the gold mining operations as the source of mercury contamination in the Ayeyarwady River by using Wallago catfish as bioindicator. In 2002, mercury concentration in fish muscle was low ($0.317\mu g/g$) and within the WHO standard, when gold mining operations were few in the Ayeyarwady River (Smith *et al.*, 2003). After 2005, although the government has tried to reduce the gold mining operations, these operations have been gradually growing (Smith and Mya Than Tun, 2006), and Ayeyarwady River is more and more polluted. It is clearly seen in the data of Khin Myint Mar (2011) in Mandalay Segment (0.674 µg/g), Khin Myint Mar (2011) in Magway Segment (0.978 µg/g), and Soe Soe Aye and Khin Ni Ni Win (2015) in Pakokku Segment (0.671 µg/g). These mercury concentrations are exceeding the WHO standard for human consumption. In 2015, the government initiating the action plan to control the gold mining operations (Kawakami *et al.*, 2019), as a result, the numbers of mining operations were relatively decreased (Bates *et al*, 2015). In the present study, mercury concentration in fish muscle of wet weight basis (0.142 μ g/g) was nearly five times lower than the last five years report of Soe Soe Aye and Khin Ni Ni Win (2015), and becoming within the WHO standard of human consumption. Therefore, it is assumed that illegal gold mining is the major source of mercury contamination to the aquatic ecosystem of the Ayeyarwady River.

Conclusion

The Ayeyarwady River plays an important role as a niche to a large diversity of fish and aquatic animals, including Irrawaddy Dolphin which is the endemic of Myanmar and endangered species in IUCN Red List. Recently, the health of the Ayeyarwady River become the importance issue, since mercury is a globally threaten contaminant in both wildlife management and public health, so as sustainable awareness programs to mining companies and local workers should be conducted to safely handle mercury and eliminate or reduce its toxic effects. So far, Ayeyarwady River seems to be recovered from mercury toxicity according to the comparative data of the present and previous studies. Therefore, sustainable monitoring is recommended to assess the toxic pollution for the conservation of biodiversity, especially Irrawaddy dolphin and health safety of people who depend on the Ayeyarwady River in Myanmar.

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