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Deposition of the Mn in the Tissues of Crucian Carp in the Coal Gangue Polluted Water and Natural Water

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Abstract

Coal gangue is the solid waste material in the process of coal mining and coal washing, which contains a lot of heavy metal: Manganese (Mn), which poses a serious threat to food security and human health. In order to study the present pollution situation of Mn and the deposition of Mn in the tissues of fish from gangue polluted water of Yongcheng, Shangqiu, Mn was determined by using the flame atomic absorption method. The results show that the concentration of Mn in the heart, hepatopancreas, gills, muscles of crucian carp from pollution-free waters in Tianmu Lake were 138.96.±35.85 (μg/g), 10.20±4.92 (μg/g), 30.02±7.62 (μg/g), 14.64±4.06(μg/g); whereas they were 316.48±23.41 (μg/g), 25.84±6.03 (μg/g), 70.18±14.67 (μg/g), 30.09±10.44(μg/g) in the crucian carp from the coal mining subsidence area. The Mn deposits were all significantly higher than natural pollution-free waters. The sedimentary rule of Mn in the crucian carp is: heart > gill > hepatopancreas and muscles. The concentration of Mn in heart is significantly higher than in the other tissue. So, we draw a conclusion that coal gangue pollution aggravates the deposition of Mn in the tissues of crucian carp and it is more likely to deposit in the heart of the crucian carp. Meat from fishes of coal gangue cannot be eaten exceeding 333g per day.

Keywords: Crucian carp (*Carassius auratus*); Heavy metal; Flame atomic absorption spectrometry (FAAS).

Abbreviations: FAAS: Flame Atomic Absorption Spectrometry; SOD: Superoxide Dismutase

Introduction

Coal gangue is the solid waste, which is produced in the process of exploiting coal. Every year, the emission of coal gangue is about 10%-20% of coal yield [1,2]. Coal gangue contains about 20 kinds of elements, such as Ca, Mg, Fe, S, Si, As, Cr, Pb, Hg, Mn, Se, Mn, Ni, Cu, Zn, Sb, Co, Mo, Be, V, Ba, Ti, Th, U, Ag [3]. Among these, Hg, Mn, Cr, Pb, Cu, As, Zn are harmful to human health [4,5].These heavy metal elements affect the growth and immune function of aquatic animals and also get deposited in their muscles [6-12]. When people consume the meat from these aquatic animals, the heavy metals get absorbed, and affect the human health [13]. Even though Mn is required by the human body as a trace element [14-16], Law reported that intake of too much Mn would affect the Central Nervous System [17], damaging the mechanism of switch off enzyme, thus acting as a great threat to human health [18].

In China, mine subsidence area often comprises of lakes, and some people feed on the fish from these mine subsidence area. This becomes a threat to food safety. But, there have been no reports about effects of heavy metal elements deposited in fish tissues by coal gangue pollution. Hence, this study on the heavy metal elements deposition in the tissue of fish is aimed to help us know the pattern of heavy metal deposition, and evaluate the water pollution. In this experiment, the deposition of Mn in crucian carp in coal gangue polluted lake and pollution-free lake is determined, by detecting the content of Mn in fish tissues. The results will help us to know the pattern of Mn deposition and evaluate the safety of the fish meat from the coal gangue polluted lake.

Materials

Fish

40 Fish were sampled from 2 Lakes. The control group fish (20 fish) were collected from a native lake named Tianmu Lake, which is situated on Yellow River old path in Shangqiu (34° 35´N, 115° 41´E). It is far from factory and coal, and hence not polluted. The tested fish (20 fish) were collected from a lake which is a mine subsidence area near the coal, situated on XiaoXu village in Yongcheng Shangqiu (33°57´N, 116°22´E). The body weight of

the fish was 86.8±5.8g.

Reagents and Instruments

Mn Standard Substance, Scissors, Tweezers, Electronic analytical balance, Porcelain crucible, electric furnace, Muffle furnace, Flask (50mL), 65%HNO3, 3mol/L HNO3, Deionized water, AFS3300 flame atomic absorption spectrophotometer, Electric oven thermostat.

Methods

Determination was referred to the method as reported by Li Hua [19].

Sample preparation

- After weighing the fish, the hepatopancreas, hearts, gills, muscles were removed from the fishes, and put into porcelain crucible.
- The porcelain crucible containing samples was put into the electric oven thermostat at 150°C for 3 h. After cooling, the samples were weighed, and the data was recorded.
- 5 mL 65% HNO_3 was added into the samples. The samples were allowed to stand 0.5h, and then put onto electric furnace for heating to have no smoke.
- The samples were placed into muffle furnace for ashing for 3h. (If the sample still have black particles, repeat step (3)and (4) to all samples till they were completed ashed.)
- After cooling, samples were dissolved into 4 mL 3mol/L HNO_3, poured into 50mL flask, and deionized water was added to 50 mL.

Preparation of standard solutions

Formulated 1% HNO_3 as a standard solution. Used 1% HNO_3 for standard blank solutions, and weighed Mn standards, then dissolved by 1% HNO_3, diluted to 0.10, 0.20, 0.30, 0.40, 0.50 μg/mL standard solutions.

Sample Determination

Sample was determined by flame atomic absorption spectrometry. The parameters were wavelength 228.8nm, slit 0.7nm, lamp current 3.0 mA, the air flow 13500 mL / min, and the burner height 13.5 nm. Every sample was determined 5 times.

Data processing

A standard curve was drawn according to the standard solutions measured data, then the content of each sample was calculated according to the standard curve.

All data were analyzed by SPSS. Mn contents in every kind of tissue were compared by Duncan multiple comparisons. The differences between the fish from polluted water and natural clean water were compared by T-test.

Results

In our results, the least content was 1.65μg/g, and the highest was 74.31μg/g. The contents of Mn in the tissues of fish from unpolluted lake(Tianmu Lake) was 1.65-19.77 μg/g. The contents of Mn in the tissues of fish from polluted water(Xiaoxu village) was 4.58-74.31 μg/g.

The results, comparing the contents in different tissues has been shown in Table 1. In the unpolluted native waters, the content of Mn in heart was significant higher than hepatopancreas, gills and muscles. In coal gangue polluted water, the content of Mn in heart was extremely higher than the hepatopancreas, gills and muscles.

The contents of Mn in fish tissues compared between unpolluted native water and coal gangue polluted water has been shown in Figure 1. The content of Mn in hepatopancreas, gills, hearts and muscles of fish in polluted water were all extremely higher than native water.

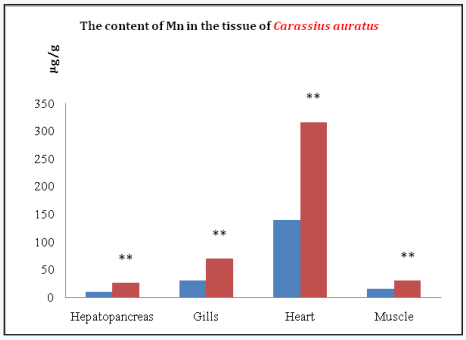

Figure 1: The comparison of the concentration of Mn in the tissues of crucian carp in the two different kind waters (Dry mater).

Table 1: The concentration of Mn in the tissues of crucian carp.

Liver	Gills	Heart	Muscle	
Native waters (Dry mater)	10.20±4.92^{Aa}	30.02±7.62^{Aa}	138.96±35.85^{Bb}	14.64±4.06^{Aa}
Polluted waters (Dry mater)	25.84±6.03^{Aa}	70.18±14.68^{Aa}	316.48±23.41^{Bb}	30.09±10.44^{Aa}
Native waters	2.21±0.98^{Aa}	5.80±2.04^{Aa}	14.93±4.54^{Bb}	2.42±0.57^{Aa}
Polluted waters	6.22±2.00^{Aa}	13.94±1.86^{Aa}	54.90±13.14^{Bb}	6.10±1.68^{Aa}

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Discussion

The contents of Mn in the tissues of fish from unpolluted lake (Tianmu Lake) were 1.65-19.77 μg/g. The contents of Mn in the tissues of fish from polluted water (Xiaoxu village) was 4.58-74.31 μg/g. Ozgur Dogan Uluozlu analyzed Mn in 9 kinds of fish from Black Sea and the Aegean Sea, the results showed that: the content of Mn in these fishes was 1.28-7.40 mg/kg [20]. Li also detected Mn in muscles of Atlantic salmon (*Salmo salar*), the result being 1.6 mg/kg [19]. It was lower than our result. This could have come from the difference of fish types and waters. Yllmaz analyzed metal elements of grey mullet (*Leuciseus eephalus*) and Lepomis cyanellus (Lepomis gibbosus) in the same sea area and found that, the content of Mn was: 0.112-24.230 mg/kg and -12.434 mg/kg [21]. The result from Wang detected that the metal elements in fishes of Huma River showed that the content of Mn in Crucian carp was 0.095 mg/kg, whereas in Burbot (*Lirmatus*) in the same water was 0.606 mg/kg [22]. Wang compared the content of Mn in Northern pike (*Esox lucius*) in different section of the Ussuri river. The results show that in different sections of the river, the content of Mn is different, such as: 2.192 mg/kg in Stellar Section of the river, 1.386 mg/kg in Haiqing Section of the river, 1.069 mg/kg in Fuyuan Section of the river [22]. In our research, the fishes come from different water, especially one of them from coal gangue. Mn deposits are found higher in the tissues of fish from polluted water than native unpolluted water. Coal gangue has heavy metal elements, including Mn. So the high Mn content in the fish from coal gangue polluted water is quite possible, in line with our speculation. We thus draw a conclusion that Mn deposition is more in coal gangue water.

As seen from the results above, in both polluted water and native unpolluted water, the contents of Mn in heart were highest in these 4 kinds of tissues. The deposit pattern was: hearts > gills > hepatopancreas > muscles. Wang analyzed 8 kinds of fishes in Jialing river, and reported that the contents of Mn in hepatopancreas were higher than in the muscles [23], which is same as our result. Liu also reported that Mn in gills of *Procambarus clarkii* was higher than in the muscles. She also reported that Mn in visceral was higher than gills, but she did not separate heart and hepatopancreas [24]. There was no report about comparison of Mn in heart and gills of fishes or shrimps. Even of these, we can have a conclusion that the deposit pattern of Mn is: hearts > gills > hepatopancreas >muscles.

Mn is a trace element required by the body. Mn2+ is the cofactor of Arginase, Ribonucleotide reductase [14]. In animal body, there was Mn-SOD, which contains Mn [16]. Mn is also as an activator of Polysaccharide polymerase, and Galactosyl transferase, which are 2 kinds of enzyme required for forming the organic matrix of bone. So Mn is very important element for formation of bone. Mn is also an essential element for catalyzing cholesterol synthesis by Dicarboxylic acid kinase. Cholesterol acts as a precursor of sex hormone. Lack of Mn would affect reproductive function [15]. So, intake of Mn at a suitable level is helpful for maintaining normal physiological functions. While Law thought that too much Mn can introduce damaging startup mechanism of enzyme and identify obstacles [17], Zhang also reported that intake of too much Mn can introduce poisoning and affect Central Nervous System [18]. So, we can see that intake of Mn at a suitable dose is helpful, but too much is harmful to human health. According to China Intakes Dietary scale, an adult can intake 10 mg of Mn per day [25]. In this research, We detected that Mn in fish muscle was 14.64 μg/g; 30.09 μg/g (Dry mater). So, an adult can eat these two kinds of fish meat about 714.29 g, 332.34 g (Dry mater) one day in a safe scale. For wet mater, we detected 1.79-3.31μg/g in native unpolluted water, and 4.34-7.60 μg/g in coal gangue polluted water. For safety measures, consumption of fish from polluted water should not exceed 1316 g (Wet weight) per one day. Not only for Mn, Yan also reported that coal gangue polluted water have high dose Cd in fish meat [26]. Eating fish from coal gangue polluted water would increase cancer risk by Cd intake [26,27]. As a conclusion, we cannot eat fish from coal gangue polluted water.

Conclusion

In summary, we can draw conclusions as follows: Mn was deposited more in Coal gangue polluted water. The deposit law is: hearts > gills > hepatopancreas > muscles. Seen from food safety, fish in coal gangue polluted water should not be consumed exceeding 333 g (Dry mater) per day.

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