Arsenic, zinc and copper accumulation in cultured milkfish from ponds using groundwater near the urban area in southwest Taiwan

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Abstract: Bioaccumulation of arsenic (As), zinc (Zn) and copper (Cu) in milkfish (Chanos chanos) from the culture ponds using groundwater near the urban area in southwest Taiwan was investigated. Samples of juvenile milkfish and ambient water were obtained from 8 culture ponds from the suburban area. The bioconcentration factor (BCF) of milkfish was calculated to indicate the accumulation ability of milkfish. The resulting data showed that the As, Zn and Cu concentrations in pond water were 63.92 ± 57.71 µg L⁻¹, 69.36 ± 27.81 µg L⁻¹ and 11.09 ± 15.22 µg L⁻¹, respectively, while in milkfish these concentrations were 0.94 ± 1.34 µg g⁻¹, 2.01 ± 0.96 µg g⁻¹ and 40.31 ± 17.25 µg g⁻¹, respectively. The As, Zn and Cu levels in milkfish showed significant positive relations to the As, Zn and Cu concentrations in pond water. The high BCF values of As, Zn and Cu accumulated in fish (12.60 ± 4.68, 32.31 ± 12.53 and 4029.04 ± 1623.96, respectively) show that those aquacultural milkfish from the suburban area are contaminated by the ambient water and have a high tolerance against these pollutants. Since those fish will be sold to the markets in cities nearby, ingestion of in this way contaminated milkfish could result in overexposure of As, Zn and/or Cu in inhabitants and lead to adverse health effects. Besides the risk on human health, using groundwater for aquaculture may also have a negative effect on the land above the aquifer because it may sink and cause damage on property and fields in the suburban and/or urban areas.

Key-Words: Arsenic, zinc, copper, bioaccumulation, groundwater, milkfish (Chanos chanos)

1 Introduction

Trace elements, such as arsenic (As), zinc (Zn) and copper (Cu), are introduced into the environment by a wide spectrum of natural and anthropogenic sources. These elements are non-biodegradable, and once they enter organisms from the environment, bioconcentration may occur in tissue by means of metabolic and biosorption processes [1, 6, 23]. Many studies have shown that trace elements can be accumulated in fish [22]. Uptake of sublethal concentrations of these elements may lead to altered physiological processes, which reduce the normal functioning of the organism. From an environmental point of view, a study on bioconcentration is important because elements usually occur in low concentrations and subtle physiological effects go unnoticed until gross chronic reactions (e.g. changes in population structure, altered reproduction, etc.) become apparent.

Among trace elements, As is well known as a toxin [20]. Arsenic has been classified as a carcinogen, based on human epidemiological data; it is known to increase the risk of producing or inciting cancer [16]. Since As is mainly transported by water, it can easily be accumulated by aquatic organisms [12, 16, 18]. Several studies have been conducted and demonstrated that an overexposure of As could result in accumulation in fish and lead to adverse health effects [5, 8, 10]. Consumption of As-polluted fish might cause an overexposure of As and pose a cancer risk to human health via the food chain [9]. It has also been well documented that As is the major risk factor for blackfoot disease (BFD), which is a peripheral vascular disease that ends with dry gangrene and spontaneous amputation of affected extremities [2]. The increase in internal organ and skin
cancers as well as BFD disease was significantly associated with the use of high-As groundwater [16]. Although Zn and Cu are essential nutrients for normal metabolic functioning [13], they may become toxic to aquatic organisms, particularly fish, when ambient concentrations exceed physiological thresholds [21, 24]. These elements can be incorporated into the food chain and concentrated by aquatic organisms to a level that affects their physiological state [7]. Ultimately they pose a health hazard to humans. Many studies have demonstrated a modifying influence of water quality on Zn and Cu toxicity to fish [15]. For example, pH affects Zn and Cu speciation, which in turn affects bioavailability [3]; calcium (Ca²⁺) associated with water hardness tends to reduce toxicity by competitively inhibiting Cu binding to fish gills [17]; and increasing concentrations of dissolved organic matter sequester waterborne Cu from biological uptake [4, 14, 19].

Milkfish (Chanos chanos) is common seafood in Taiwan. Most of the milkfish culture ponds are located in the coastal region of southwest Taiwan, which is subjected to groundwater polluted with As [11]. A high amount (38,000-49,000 ton ha⁻¹) of freshwater is needed for milkfish culture. Several studies have been conducted to demonstrate that to use the groundwater for aquaculture may cause an overexposure of As [8, 9, 10]. In addition, milkfish from the aquacultural ponds in this area are also contaminated by Zn and Cu from the ambient water. The purpose of this study is to evaluate the bioaccumulation of As, Zn and Cu in milkfish. The bioconcentration factor of milkfish was estimated to analyze to potential risk to human health.

2 Materials and Methods

2.1 Sampling
Samples of juvenile milkfish (range 4.0-6.0 cm in length and 0.41-1.41 g in weight) and ambient water were obtained from 8 culture ponds near the urban area. Three fish and three 500 ml water samples per pond were collected. The milkfish were placed on ice immediately, and kept at 4°C during transfer to the laboratory. The water samples were fixed by adding 5 ml 1N HNO₃.

2.2 Chemical Analysis
The frozen flesh of milkfish was dehydrated in a dryer (40) for 96 h, and then grounded into powder. Aliquots of dry flesh powder weighing 0.5 g were placed into a 250 ml beaker. Nitric acid (65%, 10 ml) was added for an overnight digestion. The beaker with flesh solution, after the digestion, was heated with a water bath at 70-80°C for 2-4 h until the total volume is reduced to 1-2 ml. The solution was transferred to a volumetric flask (50 ml), and then filled with 0.01N of HNO₃ to make a 50 ml of final solution. After filtration, this 50 ml solution was transferred to test tubes for As, Zn and Cu analysis using ICP-MS (Agilent 7500a). Analytical quality control was achieved by digesting and analyzing identical amounts of rehydrated (90% H₂O) standard reference materials (DORM-2, Dogfish Liver-2-organic matrix, NRC-CNRC, Canada). Recovery rates were ranged from 95% to 97%.

2.3 Calculation of Bioconcentration factor (BCF)
When steady-state chemical concentrations of tissue are attained, the equilibrium bioconcentration factor (BCF) of the milkfish can be calculated from the ratio of the chemical concentration in fish to that in water. The BCF can also be calculated from the ratio of the uptake rate constant to the depuration rate constant as,

\[
BCF = \frac{C_b}{C_w}
\]

where \(C_b (\mu g g^{-1})\) is the chemical concentration in biota; \(C_w (\mu g ml^{-1})\) is the chemical concentration in water.

3 Results and Discussion
The resulting data of this study showed that the As, Cu, Zn concentrations in pond water were
63.92 ± 57.71 µg L⁻¹, 69.36 ± 27.81 µg L⁻¹ and 11.09 ± 15.22 µg L⁻¹, respectively (Table 1), while in fish were 0.94 ± 1.34 µg g⁻¹, 2.01 ± 0.96 µg g⁻¹ and 40.31 ± 17.25 µg g⁻¹, respectively (Table 2).

The As, Zn and Cu levels in milkfish showed significant positive relations to the As, Zn and Cu concentrations in pond water: $C_b = -0.27 + 0.02 C_w$, $C_b = 25.43 + 1.02 C_w$ and $C_b = 1.22 + 0.01 C_w$, respectively (Fig. 1), where $C_b$ is the chemical concentration in biota (µg g⁻¹); $C_w$ is the chemical concentration in water (µg ml⁻¹). It shows that milkfish can accumulate As, Zn and Cu from the ambient water. The high BCF values of As, Cu and Zn accumulated in fish (12.60 ± 4.68, 32.31 ± 12.53 and 4029.04 ± 1623.96, respectively) show that those cultured milkfish from are contaminated by the ambient water and have a high tolerance against the pollutants (Table 3). The accumulation of these chemicals may contribute to chronic toxicity in fish [1, 4]. Since those fish will be sold to the markets in cities nearby, ingestion those contaminated milkfish could result in overexposure of As, Zn and/or Cu in inhabitants and lead to adverse health effects. Besides the risk on human health, using groundwater for aquaculture may also have a negative effect on the land above the aquifer because it may sink and cause damage on property and fields in the suburban and/or urban areas.

Lin et al. [9, 11] demonstrated that consumption of cultured milkfish from the As-contaminated ponds may pose a cancer risk to human health. Thus far no adverse effect on health of the people due to exposure to As, Zn and Cu contaminated milkfish is evidenced. A wider study involving As, Zn and Cu analyses of milkfish from unpolluted areas, as well as other aquacultural products should be investigated to assess the extent of chemical contamination in seafood. The interactions among the elements and the subsequent uptake and depuration rates associated with the individual elements are needed to be analyzed [4]. Further more, studies concerning the effects of the individual elements in the mixture are needed to be undertaken [23].

4 Conclusion
Milkfish can accumulate remarkably high levels of As, Zn and Cu from the ambient water. The As, Zn and Cu levels in milkfish showed significant positive relations to the As, Zn and Cu concentrations in pond water, respectively. The high BCF values of As, Zn and Cu accumulated in fish show that those cultured milkfish are contaminated by the ambient water and have a high tolerance against the pollutants. Since those fish will be sold to the markets in cities nearby, ingestion those contaminated milkfish could result in overexposure of As, Zn and/or Cu in inhabitants and lead to adverse health effects. Besides the risk on human health, using groundwater for aquaculture may also have a negative effect on the land above the aquifer because it may sink and cause damage on property and fields in the suburban and/or urban areas.

Reference:


Table 1. Arsenic (As), Zinc (Zn) and Copper (Cu) concentrations in water of milkfish culture ponds (µg L\(^{-1}\)).

<table>
<thead>
<tr>
<th>Location</th>
<th>As Mean ± SE</th>
<th>Zn Mean ± SE</th>
<th>Cu Mean ± SE</th>
</tr>
</thead>
<tbody>
<tr>
<td>Putai</td>
<td>88.10 ± 65.63*</td>
<td>21.65 ± 11.06</td>
<td>85.70 ± 8.34*</td>
</tr>
<tr>
<td>Yichu</td>
<td>102.57 ± 63.62*</td>
<td>5.97 ± 1.91</td>
<td>60.01 ± 2.44*</td>
</tr>
<tr>
<td>Hsuehchia</td>
<td>30.44 ± 6.82</td>
<td>7.45 ± 0.23</td>
<td>40.78 ± 15.92*</td>
</tr>
<tr>
<td>Peimen</td>
<td>34.58 ± 3.76</td>
<td>8.49 ± 0.23</td>
<td>121.88 ± 7.18*</td>
</tr>
<tr>
<td>Average</td>
<td>63.92 ± 57.71</td>
<td>11.90 ± 15.22</td>
<td>69.36 ± 27.81</td>
</tr>
</tbody>
</table>

* As > 50 µg L\(^{-1}\); Zn > 500 µg L\(^{-1}\); Cu > 30 µg L\(^{-1}\)

Table 2. Arsenic (As), Zinc (Zn) and Copper (Cu) levels in milkfish (µg L\(^{-1}\)) from culture ponds.

<table>
<thead>
<tr>
<th>Location</th>
<th>As Mean ± SE</th>
<th>Zn Mean ± SE</th>
<th>Cu Mean ± SE</th>
</tr>
</thead>
<tbody>
<tr>
<td>Putai</td>
<td>1.89 ± 1.66</td>
<td>46.33 ± 13.75</td>
<td>2.11 ± 0.03</td>
</tr>
<tr>
<td>Yichu</td>
<td>1.29 ± 0.93</td>
<td>33.10 ± 4.61</td>
<td>1.84 ± 0.11</td>
</tr>
<tr>
<td>Hsuehchia</td>
<td>0.32 ± 0.00</td>
<td>39.92 ± 2.63</td>
<td>1.76 ± 0.31</td>
</tr>
<tr>
<td>Peimen</td>
<td>0.40 ± 0.11</td>
<td>32.57 ± 2.65</td>
<td>2.63 ± 0.45</td>
</tr>
<tr>
<td>Average</td>
<td>0.94 ± 1.34</td>
<td>40.31 ± 17.25</td>
<td>2.01 ± 0.96</td>
</tr>
</tbody>
</table>

Table 3. Bioconcentration factors of Arsenic (As), Zinc (Zn) and Copper (Cu) of milkfish from culture ponds.

<table>
<thead>
<tr>
<th>Location</th>
<th>BCF</th>
<th>As Mean ± SE</th>
<th>Zn Mean ± SE</th>
<th>Cu Mean ± SE</th>
</tr>
</thead>
<tbody>
<tr>
<td>Putai</td>
<td></td>
<td>16.85 ± 9.72</td>
<td>3191.7 ± 2404.89</td>
<td>24.84 ± 2.92</td>
</tr>
<tr>
<td>Yichu</td>
<td></td>
<td>9.01 ± 6.64</td>
<td>5541.02 ± 1469.51</td>
<td>30.78 ± 3.81</td>
</tr>
<tr>
<td>Hsuehchia</td>
<td></td>
<td>10.94 ± 3.55</td>
<td>5357.59 ± 2125.76</td>
<td>47.43 ± 15.61</td>
</tr>
<tr>
<td>Peimen</td>
<td></td>
<td>10.35 ± 4.17</td>
<td>3919.48 ± 593.07</td>
<td>21.62 ± 7.44</td>
</tr>
<tr>
<td>Average</td>
<td></td>
<td>12.51 ± 4.70</td>
<td>4029.04 ± 1623.96</td>
<td>32.31 ± 12.53</td>
</tr>
</tbody>
</table>
Figure 1. Plot of the As, Zn and Cu levels in cultured milkfish and the chemical concentrations in pond water.