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Heavy Metal Bioaccumulation in Soil Arthropods at Malaysian Sanitary Landfill

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ABSTRACT

Orthomorpha coarctata and *Trigoniulus corallines*, which are common soil arthropods found in Southeast Asia, were found in abundance at a Malaysian sanitary landfill. This indicates that both these species were able to tolerate heavy metal contaminated soils. A study was conducted to assess the bioaccumulation of heavy metals in these soil arthropods, to determine the possibility of these heavy metals contaminating our food chain and also the possibility of developing natural absorbents of heavy metals. This study was conducted at Air Hitam Sanitary Landfill, one of the largest landfills in Malaysia. Chemical analyses were conducted to determine the concentration of heavy metal in the top soil (10.0 cm), washed and unwashed *Orthomorpha coarctata* and *Trigoniulus corallines* samples. Comparison between the heavy metal concentrations in the washed arthropods samples, unwashed arthropods samples and the top soil were conducted using one way analysis of variance (ANOVA) at significance difference of $P \leq 0.05$, followed by Tukey HSD test, to determine if the soil arthropods were bioaccumulators for any of the heavy metals. Only Cd was found to be significantly higher in both the soil arthropods when compared to the top soil content, meaning that both *Orthomorpha coarctata* and *Trigoniulus corallines* were soil arthropods that were bioaccumulators of Cd. More research should be conducted to determine the exact part the Cd is stored in these arthropods and also the possibility of Cd entering our food chain via these soil arthropods.

1. Introduction

Soil arthropods are organisms that get their name from their jointed (arthros) legs (podos), living in top soils to a depth of 10 cm [1]. A common soil arthropod is *Orthomorpha coarctata*, a millipede that is endemic in tropical regions all over the world [2]. This black and yellow millipede breeds and lives in grassy soils, but can also be found in residential areas [2]. Another common soil arthropod is the *Trigoniulus corallines*, a millipede species native to South East Asia but can also be found in North America as an introduced species [3]. This rusty coloured millipede normally inhabits moist areas such as rotten wood or top soils with organic matter [3].

Heavy metal pollution is becoming increasingly prevalent in Southeast Asia, especially in areas with heavy industries, mining and landfills [4]. These metal deposits remain predominantly in the top few centimeters where soil micro arthropods live, which can cause the decrement in soil arthropod species abundance and richness [1,5]. Some soil arthropods have been found to be able to tolerate these heavy metals, sometimes accumulating it in their bodies [1, 5, 6]. This bioaccumulation of heavy metals can cause heavy metals to slip into our food chain, usually via grazing animals consuming the grass alongside the contaminated arthropods [1, 7]. Besides that, some researchers have found that chitin, the exoskeleton of arthropods, can be converted into chitosan to be used as a natural absorbent of heavy metals [6]. Therefore, studies on the bioaccumulation of heavy metals in soil arthropods can lead to better understanding of the possibility of these heavy metals contaminating our food chain and also the possibility of developing natural absorbents of heavy metals. This study was conducted to determine the bioaccumulation of heavy metals in two soil arthropods, *Orthomorpha coarctata* and *Trigoniulus corallines*, at a Malaysian sanitary landfill.

2. Experimental Methods

2.1 Study Site

This study was conducted at Air Hitam Sanitary Landfill (AHSL). AHSL is located near the Air Hitam Forest Reserve in Mukim Petaling, Daerah Petaling, Puchong, Selangor and is located at longitude 101° 39' 55" E and latitude 03° 0' 10" N (Fig. 1). The Selangor State Government Council approved Worldwide Sita Environmental Management to develop this sanitary landfill, on 22nd March 1995. ASHL was built in 1995 and was the first engineered sanitary landfill site in Malaysia, covering a total of 42 hectares. During the 11 years ASHL operated, it received approximately 6.2 million tons of domestic waste. AHSL was officially closed on 31 December 2006 and the 5-year Landfill Closure and Post Closure Maintenance Plan (LCPCMP) were in place (2007-2011). ASHL is surrounded by residential housing and schools. One of the main reasons this site was selected, was because AHSL is one of the largest landfills in Malaysia. Despite having undergone closure, researchers have found high heavy metal concentrations in AHSL landfill leachate [8], which is another reason why this site was selected.



Fig. 1 Location of study site, Air Hitam Sanitary Landfill

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2.2 Soil and Arthropod Sampling

Six subplots (20 m x 20 m) was randomly established (completely randomized design). Soil composite samples were obtained using an auger from each subplot at 0 cm–20 cm. All soil samples were kept in a standard plastic container and air-dried before chemical analyses. Samples of the soil were air dried for three days, pounded with a mortar and pestle, and then sieved through a 2 mm mesh. This is done to produce a homogenous mixture for analyses.

Arthropods samples were collected using the same soil sampling plots. Arthropods were collected using appropriate pitfall methods at these sampling plots. The average density of the arthropods was also calculated using the formula:

$$\bar{x} = \frac{\sum_{i=1}^n x_i}{n}$$

Where x_i = each sample observation, n = sample size

The arthropods samples that were collected were *Orthomorpha coarctata* and *Trigoniulus corallines*. These species were selected for this study as they were found in abundance at AHSL. All arthropods collected was killed in a deep freezer and dried in oven at 60 °C for 10 hours and then stored at room temperature in plastic tubes. Before being analyzed, the arthropod samples were dried overnight at 105 °C. The arthropod samples were then divided into two, unwashed and washed samples. Unwashed samples underwent acid digestion without any prior preparation while the washed samples were washed with distilled water prior to being analyzed. This was done to determine if particles (soil) sticking to the samples had any effect on the heavy metal concentrations of the samples [9].

2.3 Chemical Analyses

Acid digestion using aqua regia was conducted to determine the concentration of heavy metal in all the samples collected. After digestion, the total concentrations of Cd, Fe, Zn, Cu, and Pb were determined using the Atomic Absorption Spectrometer (AAS) [10]. These metals were selected as other heavy metals such as Cr and As had non-detectable concentrations.

2.4 Statistical Analyses

Statistical analyses were conducted using the SPSS program (Version 17). Comparison between the heavy metal concentrations in the washed arthropods samples, unwashed arthropods samples and the top soil were conducted using one way analysis of variance (ANOVA) at significance difference of $P \leq 0.05$, followed by Tukey HSD test.

3. Results and Discussion

The average density of *Orthomorpha coarctata* was $35.17 \pm 7.7 / 400 \text{ m}^2$, while the average density for *Trigoniulus corallines* was $14.33 \pm 5.6 / 400 \text{ m}^2$.

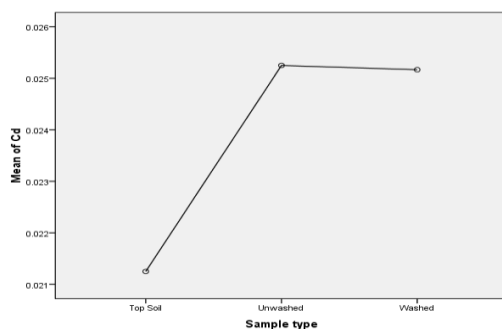


Fig. 2 Mean concentration of Cd according to top soil and *Orthomorpha coarctata* (washed and unwashed)

3.1 Comparison Between Cd Concentrations in Top Soil and *Orthomorpha coarctata*

There was a significant difference between top soil and *Orthomorpha coarctata* (washed and unwashed) Cd concentrations [$F(2, 33) = 13.081, p = 0.000$]. A Tukey post-hoc test revealed that the top soil had statistically significant lower concentration of Cd ($0.021 \pm 0.003 \text{ ppm}$) than both the *Orthomorpha coarctata* samples, that is the unwashed samples ($0.025 \pm 0.001 \text{ ppm}, p = 0.0$) and washed samples ($0.025 \pm 0.001 \text{ ppm}, p = 0.0$). There were no statistical differences in Cd concentrations between the

unwashed and washed *Orthomorpha coarctata* samples. This indicates that *Orthomorpha coarctata* is an arthropod species that accumulates Cd (Fig. 2).

3.2 Comparison Between Cu Concentrations in Top Soil and *Orthomorpha coarctata*

There was a significant difference between top soil and *Orthomorpha coarctata* (washed and unwashed) Cu concentrations [$F(2, 33) = 3169.315, p = 0.000$]. A Tukey post-hoc test revealed that the top soil had statistically significant higher concentration of Cu ($28.82 \pm 1.53 \text{ ppm}$) than both the *Orthomorpha coarctata* samples, that is the unwashed samples ($3.6 \pm 0.18 \text{ ppm}, p = 0.0$) and washed samples ($3.6 \pm 0.18 \text{ ppm}, p = 0.0$). There were no statistical differences in Cu concentrations between the unwashed and washed *Orthomorpha coarctata* samples. This indicates that *Orthomorpha coarctata* is not an arthropod species that accumulates Cu (Fig. 3).

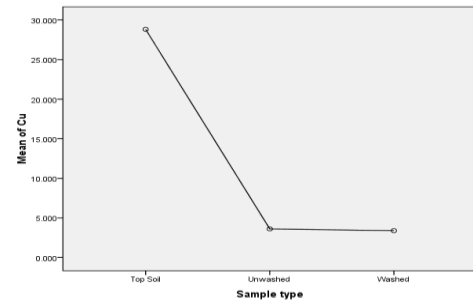


Fig. 3 Mean concentration of Cu according to top soil and *Orthomorpha coarctata* (washed and unwashed)

3.3 Comparison Between Fe Concentrations in Top Soil and *Orthomorpha coarctata*

There was a significant difference between top soil and *Orthomorpha coarctata* (washed and unwashed) Fe concentrations [$F(2, 33) = 2751.191, p = 0.000$]. A Tukey post-hoc test revealed that the top soil had statistically significant higher concentration of Fe ($20.56 \pm 0.83 \text{ ppm}$) than both the *Orthomorpha coarctata* samples, that is the unwashed samples ($7.48 \pm 0.17 \text{ ppm}, p = 0.0$) and washed samples ($7.41 \pm 0.14 \text{ ppm}, p = 0.0$). There were no statistical differences in Fe concentrations between the unwashed and washed *Orthomorpha coarctata* samples. This shows that *Orthomorpha coarctata* is not an arthropod species that accumulates Fe (Fig. 4).

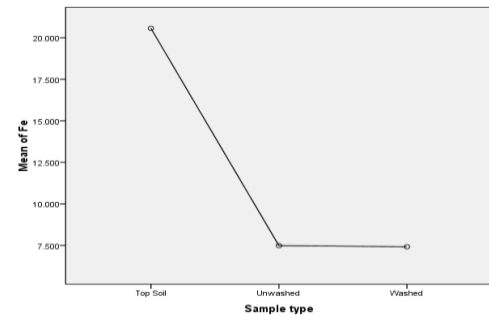


Fig. 4 Mean concentration of Fe according to top soil and *Orthomorpha coarctata* (washed and unwashed)

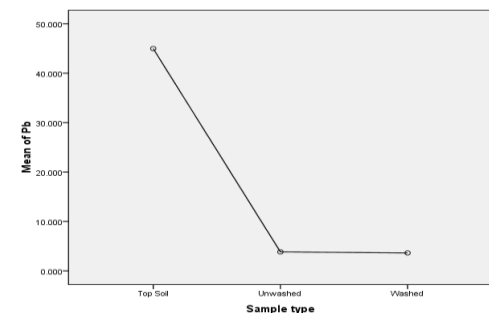


Fig. 5 Mean concentration of Pb according to top soil and *Orthomorpha coarctata* (washed and unwashed)

3.4 Comparison Between Pb Concentrations in Top Soil and *Orthomorpha coarctata*

There was a significant difference between top soil and *Orthomorpha coarctata* (washed and unwashed) Pb concentrations [$F(2, 33) =$

2647.852, $p = 0.000$]. A Tukey post-hoc test revealed that the top soil had statistically significant higher concentration of Pb (44.99 ± 2.76 ppm) than both the *Orthomorpha coarctata* samples, that is the unwashed samples (3.82 ± 0.08 ppm, $p = 0.0$) and washed samples (3.61 ± 0.21 ppm, $p = 0.0$). There were no statistical differences in Pb concentrations between the unwashed and washed *Orthomorpha coarctata* samples. Therefore, similar to Cu and Fe, *Orthomorpha coarctata* is not an arthropod species that accumulates Pb (Fig. 5).

3.5 Comparison Between Zn Concentrations in Top Soil and *Orthomorpha coarctata*

There was a significant difference between top soil and *Orthomorpha coarctata* (washed and unwashed) Zn concentrations [$F(2, 33) = 1471.065$, $p = 0.000$]. A Tukey post-hoc test revealed that the top soil had statistically significant higher concentration of Zn (50.16 ± 4.03 ppm) than both the *Orthomorpha coarctata* samples, that is the unwashed samples (5.5 ± 0.11 ppm, $p = 0.0$) and washed samples (5.36 ± 0.171 ppm, $p = 0.0$). There were no statistical differences in Zn concentrations between the unwashed and washed *Orthomorpha coarctata* samples. This indicates that *Orthomorpha coarctata* is not an arthropod species that accumulates Zn (Fig. 6).

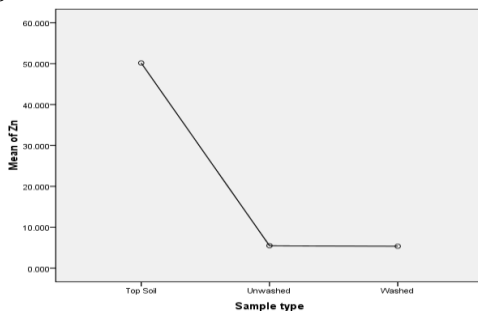


Fig. 6 Mean concentration of Zn according to top soil and *Orthomorpha coarctata* (washed and unwashed)

3.6 Comparison Between Cd Concentrations in Top Soil and *Trigoniulus corallinus*

There was a significant difference between top soil and *Trigoniulus corallinus* (washed and unwashed) Cd concentrations [$F(2, 33) = 4.149$, $p = 0.000$]. A Tukey post-hoc test revealed that the top soil had statistically significant lower concentration of Cd (0.021 ± 0.003 ppm) than both the *Trigoniulus corallinus* samples, that is the unwashed samples (0.023 ± 0.0008 ppm, $p = 0.0$) and washed samples (0.023 ± 0.0009 ppm, $p = 0.0$). There were no statistical differences in Cd concentrations between the unwashed and washed *Trigoniulus corallinus* samples. This indicates that *Trigoniulus corallinus* is an arthropod species that accumulates Cd (Fig. 7).

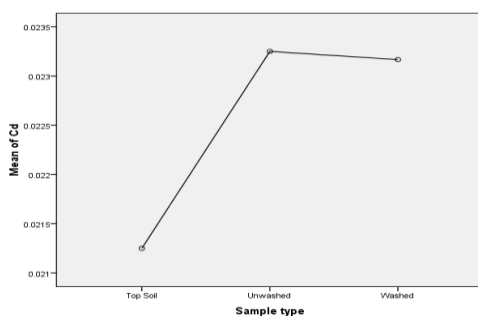


Fig. 7 Mean concentration of Cd according to top soil and *Trigoniulus corallinus* (washed and unwashed)

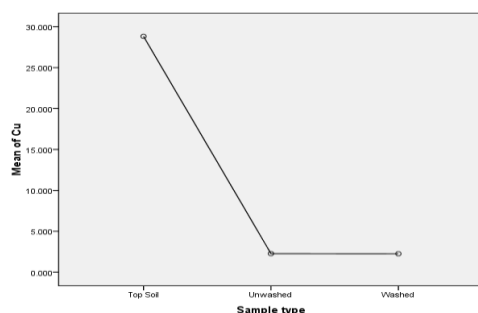


Fig. 8 Mean concentration of Cu according to top soil and *Trigoniulus corallinus* (washed and unwashed)

3.7 Comparison Between Cu Concentrations in Top Soil and *Trigoniulus corallinus*

There was a significant difference between top soil and *Trigoniulus corallinus* (washed and unwashed) Cu concentrations [$F(2, 33) = 3502.350$, $p = 0.000$]. A Tukey post-hoc test revealed that the top soil had statistically significant higher concentration of Cu (28.82 ± 1.53 ppm) than both the *Trigoniulus corallinus* samples, that is the unwashed samples (2.27 ± 0.02 ppm, $p = 0.0$) and washed samples (2.25 ± 0.02 ppm, $p = 0.0$). There were no statistical differences in Cu concentrations between the unwashed and washed *Trigoniulus corallinus* samples. This shows that *Trigoniulus corallinus* is not an arthropod species that accumulates Cu (Fig. 8).

3.8 Comparison Between Fe Concentrations in Top Soil and *Trigoniulus corallinus*

There was a significant difference between top soil and *Trigoniulus corallinus* (washed and unwashed) Fe concentrations [$F(2, 33) = 3496.523$, $p = 0.000$]. A Tukey post-hoc test revealed that the top soil had statistically significant higher concentration of Fe (20.56 ± 0.83 ppm) than both the *Trigoniulus corallinus* samples, that is the unwashed samples (6.21 ± 0.69 ppm, $p = 0.0$) and washed samples (6.2 ± 0.63 ppm, $p = 0.0$). There were no statistical differences in Fe concentrations between the unwashed and washed *Trigoniulus corallinus* samples. This shows that *Trigoniulus corallinus* is not an arthropod species that accumulates Fe (Fig. 9).

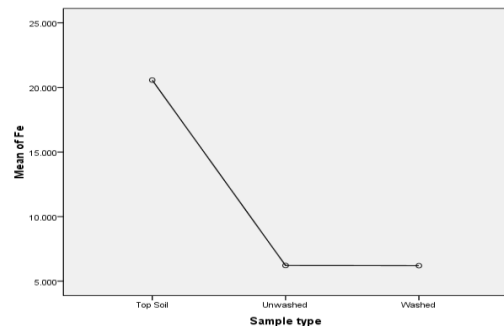


Fig. 9 Mean concentration of Fe according to top soil and *Trigoniulus corallinus* (washed and unwashed)

3.9 Comparison Between Pb Concentrations in Top Soil and *Trigoniulus corallinus*

There was a significant difference between top soil and *Trigoniulus corallinus* (washed and unwashed) Pb concentrations [$F(2, 33) = 2708.415$, $p = 0.000$]. A Tukey post-hoc test revealed that the top soil had statistically significant higher concentration of Pb (44.99 ± 2.76 ppm) than both the *Trigoniulus corallinus* samples, that is the unwashed samples (3.39 ± 0.45 ppm, $p = 0.0$) and washed samples (3.36 ± 0.07 ppm, $p = 0.0$). There were no statistical differences in Pb concentrations between the unwashed and washed *Trigoniulus corallinus* samples. These findings indicate that *Trigoniulus corallinus* is not an arthropod species that accumulates Pb (Fig. 10).

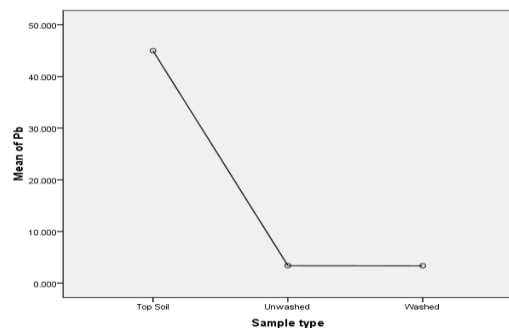


Fig. 10 Mean concentration of Pb according to top soil and *Trigoniulus corallinus* (washed and unwashed)

3.10 Comparison Between Zn Concentrations in Top Soil and *Trigoniulus corallinus*

There was a significant difference between top soil and *Trigoniulus corallinus* (washed and unwashed) Zn concentrations [$F(2, 33) = 8143.904$, $p = 0.000$]. A Tukey post-hoc test revealed that the top soil had statistically significant higher concentration of Zn (50.16 ± 4.03 ppm) than

both the *Trigoniulus corallinus* samples, that is the unwashed samples (5.04 ± 0.02 ppm, $p = 0.0$) and washed samples (5.04 ± 0.01 ppm, $p = 0.0$). There were no statistical differences in Zn concentrations between the unwashed and washed *Trigoniulus corallinus* samples. Similar to Cu, Fe and Pb, these findings show that *Trigoniulus corallinus* is not an arthropod species that accumulates Zn (Fig. 11).

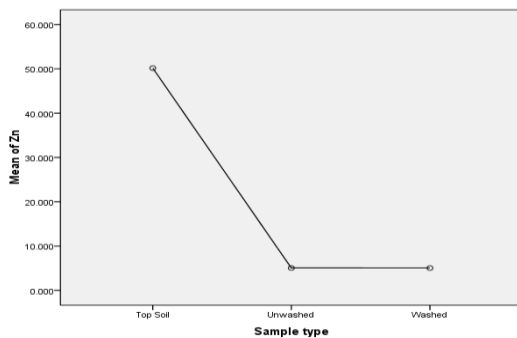


Fig. 11 Mean concentration of Zn according to top soil and *Trigoniulus corallinus* (washed and unwashed)

3.11 Comparison of Heavy Metal Concentrations in Top Soil with Soil Arthropods

Only Cd was found to be significantly higher in both species of arthropods, for both the unwashed and washed samples. This is an indication that both *Orthomorpha coarctata* and *Trigoniulus corallinus* are potential bioaccumulators of Cd. This is a characteristic that has been found to be prevalent in most soil arthropods; most researchers citing that the outer skeleton of these arthropods, the chitin, is the primary part that is found to accumulate Cd [6, 11, 12]. As some researchers have established, this chitin can be converted into a natural absorbent of heavy metals, which makes these two soil arthropod species possible candidates for the development of such technologies in the future [6]. Besides that, these species can also be used as a bioindicator for Cd contamination, to study the extent of Cd contamination in an ecosystem [12]. There is the possibility of these soil arthropod species contaminating the food chain via dietary cadmium, which has been found to be quite prevalent in Cd contaminated ecosystems [13].

Other heavy metals, Cu, Pb, Fe and Zn didn't show any indication of bioaccumulation in both soil arthropod species. Although some soil arthropods have shown bioaccumulation of other heavy metals, Cd remains to be the most common heavy metal accumulated by these organisms [6-13].

4. Conclusion

There is a possibility that the heavy metal content in the soil had caused the reduction in the abundance and richness of the soil arthropods at AHSL. Hence, the two soil arthropods in this study, *Orthomorpha coarctata* and *Trigoniulus corallinus*, is able to tolerate soils contaminated with heavy metals as they were found in abundance at AHSL. Cu, Pb, Fe, and Zn did not accumulate in both these soil arthropods, indicating that these species did not bioaccumulate these metals. However, Cd was significantly higher in both the soil arthropods when compared to the top soil content, meaning that both *Orthomorpha coarctata* and *Trigoniulus corallinus* were soil arthropods were bioaccumulators of Cd.

More research should be conducted on the exact part the Cd which is stored in these arthropods, as it can be used as a platform to develop natural absorbents of Cd. Besides that studies should also be conducted to determine the possibility of these species causing Cd to slip into our food chain.

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