Sargassum, Padina and Turbinaria as bioindicators of cadmium in Bais Bay, Negros Oriental

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ABSTRACT

Marine pollution is becoming one of the global environmental problems around the world. Heavy metal concentrations in the environment is 100-1,000 folds higher than those in the Earth's crust due to anthropogenic sources. The use of algae as bioindicators of metal pollution and the effective removal of toxic metal ions has received increasing attention. Brown algae is thought to be excellent in the sequestration of metal. In this study, cadmium (Cd) concentration in the thalli of *Sargassum* sp., *Padina* sp. and *Turbinaria* sp. from North Bais Bay, Negros Oriental was determined using Atomic Absorption Spectrophotometer (AAS). The concentration of cadmium in the sediment of North Bais Bay was also determined because environmental pollutants could also accumulate in the sediment. Results of this study showed the cadmium contamination in Bais Bay, wherein *Sargassum* sp., *Padina* sp., and *Turbinaria* sp. absorbed these cadmium ions. *Sargassum* sp. had the highest concentration of cadmium which ranged from 2.14 to 4.45 mg kg^{-1}. The concentration of cadmium in *Padina* sp. and *Turbinaria* sp. ranged from 2.2 to 3.4 mg kg^{-1} and 2.36 to 2.76 mg kg^{-1}, respectively. The concentration of cadmium in the sediment ranged from 3.72 to 5.53 mg kg^{-1} dry weight. This indicates that these brown algal species could be utilized as bioindicators of cadmium contamination in marine waters and possible phytoremediation of cadmium in wastewater.

Keywords: brown algae, phytoremediation, pollution, biomonitoring, cadmium

INTRODUCTION

In recent years, heavy metal pollution has become a persistent problem in coastal and estuarine ecosystems around the world (Khan et al. 2017). Accumulation of heavy metals in the aquatic environment has been associated with agricultural and urban run-off, boating activities, quarrying activities, and discharges from mining, industrial and municipal wastes (El-Serehy et al. 2012; Borja et al. 2015). These pollutants can enter and contaminate estuarine and marine waters from feeder rivers. Unlike organic contaminants, heavy metals are not biodegradable and tend to accumulate in the sediment and living organisms to reach the toxic concentration causing ecological damage and potential danger to human health (El-Serehy et al. 2012; Kaparapu et al. 2015). Example of heavy metal which is considered highly toxic and poses a threat to the development of flora and fauna as well as human health is cadmium (Rzetala 2016). It is easily absorbed and accumulated in tissues, which may lead to damage of kidneys, liver, testes and prostate when exposed to cadmium for long term (Olmedo et al. 2013).

The use of marine algae as bioindicators to trace metal pollution and monitor the extent of contamination in the marine environment has received increasing attention (Torres et al. 2008). Algae are of special interest in search for the development of new biosorbent materials due to their high sorption capacity and readily available in practically unlimited quantities in seas and oceans (Rincon et al. 2005). Among the algal species, brown algae have proven to be the most promising substrate for bioremediation of toxic heavy metals (Torres et al. 2008). Its basic biochemical cell wall constituents are chiefly responsible for heavy metal biosorption (Davis et al. 2003).

Bais Bay is known for its rich fishery resources in Negros Oriental. However, wastes, sewages and untreated effluents from domestic, agricultural and industrial sources may be discharged into the bay resulting to poor water quality. In 2015, fish kills were experienced in Bais Bay which could be attributed to poor water quality. An investigation conducted by the Environmental Management Bureau-Department of Environment and Natural Resources (EMB-DENR) showed that dissolved oxygen in Bais Bay dropped significantly due to untreated wastewater (Matus 2015). Though water quality monitoring was conducted by DENR and Bureau of Fisheries and Resources (BFAR), biomonitoring of heavy metals needs to be intensified. Thus, this study was conducted to determine the concentration of cadmium in brown algae *(Sargassum, Padina* and *Turbinaria* spp.), and sediment in North Bais Bay, Negros Oriental, Philippines. Results of this study can be used as a reference for cadmium contamination in Bais Bay, and utilization of *Sargassum, Padina* and *Turbinaria* species for possible application in wastewater treatment.

METHODS

Description of the Study Area

This study was conducted in North Bais Bay (9°39'4.4208"N, 123°8'41.2908"E) located on the eastern side of Negros Island (Figure 1). There were human settlements areas, industrial plants, resorts and two rivers draining water into the bay.

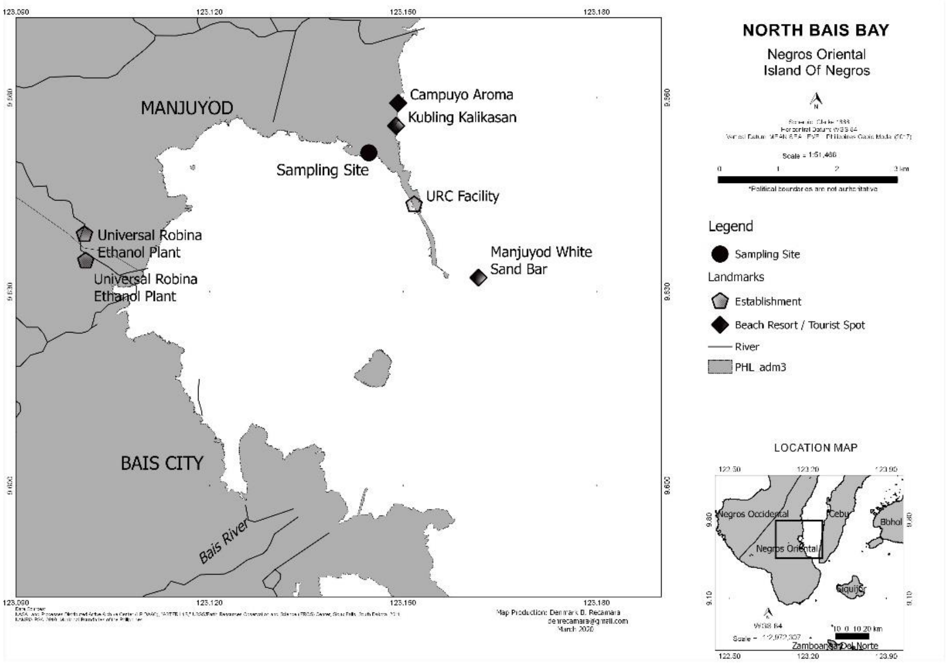

Figure 1. Map of North Bais Bay showing the sampling site (black circle), industries, resorts and Bais River.

Collection and Treatment of Samples

From August to October 2018, six sampling events were conducted to collect samples of algae belonging to the following genera: *Sargassum, Padina,* and *Turbinaria*. These months are within the southwest monsoon (Habagat) which may bring frequent heavy rains and surface runoff. The samples were then washed with filtered seawater and brought to NORSU Biology Laboratory in a polyethylene bags with ice. Upon arrival at the laboratory, the algal samples were thoroughly cleaned from epiphytes and sediment using distilled water and air dried on an absorbent paper for at least 1 week. After which, the dried samples were oven dried at 100°C for 1.5 h to remove the remaining moisture in the samples. The samples were then pulverized in an electric grinder for more efficient digestion and brought to University of San Carlos Water Laboratory, Cebu City for heavy metal analysis.

Since bioaccumulative substances are also incorporated into the sediment (Ansari et al. 2004), concentration of cadmium in the sediment of North Bais Bay was also determined. Two hundred grams (200 g) of sediment samples were collected from the same sites where the algal samples were collected three times a month from August to October 2018 using a polyethylene core borer (5 cm diameter x 50 cm length) at a depth of 5 cm. The collected sediment was then placed inside a plastic bag and brought to NORSU Biology Laboratory for air drying. The dried sediment samples were then submitted to University of San Carlos Water Laboratory, Cebu City for heavy metal analysis.

Analytical Method for Heavy Metal Determination in Brown Algae and Sediment

To determine the heavy metal in *Sargassum, Padina* and *Turbinaria* species, 1 g each of dried sample (dry weight) was digested with 20 ml of nitric acid (HNO_3) and 5 ml of hydrogen peroxide (H_2O_2). The samples were stirred gently to homogenize with the acids. The solution was then heated to 95°C and concentrated for analysis.

Heavy metal in the sediment samples was determined using the method from US EPA (1996). The sediment samples were mixed thoroughly to achieve homogeneity and filtered using a USS #10. After homogenization, 1 g of dry sample was digested in a digestion vessel using 10 ml of HNO_3. It was then heated to 95°C and refluxed for 10 to 15 min without boiling. Five (5) ml of concentrated HNO_3 was added and refluxed for 30 min until production of brown fume has ceased. The solution was allowed to evaporate to approximately 5 ml using a ribbed watch glass or vapor recovery system. If the sample has cooled, 2 ml of water and 3 ml of 30% H_2O_2 was added. Acid-peroxide digestate was again heated until the volume has been reduced to approximately 5 ml. Acid-peroxide digestate was allowed to cool then diluted to 100 ml with water. Particulates in the digestate should then be removed by filtration, by centrifugation, or by allowing the sample to settle. The sample is now ready for analysis.

The concentration of cadmium in brown algae and sediment was analyzed using Shimadzu Atomic Absorption Spectrophotometer (AAS) at the Water Laboratory of University of San Carlos, Cebu City following AAS Flame technique. All analyses were carried out in triplicate. For each run, three "blanks" were analyzed to check the purity of reagents and any possible contamination.

Statistical Analysis

The data represent the mean and standard deviation (mean±SD) of triplicate samples. Monthly cadmium concentration in the sediment of Bais Bay were compared using Kruskal Wallis test, while two-way ANOVA was used for the three brown algal species among sampling months at 95% confidence interval.

RESULTS

Among the three species, *Sargassum* sp. had the highest concentration of cadmium which ranged from 2.14 to 4.45 mg kg^{-1} dry weight. The concentration of cadmium in *Turbinaria* sp. ranged from 2.36 to 2.76 mg kg^{-1} dry weight while *Padina* sp. had cadmium concentration which ranged from 2.23 to 3.4 mg kg^{-1} dry weight. The highest concentration of cadmium across the three species was observed in September (Figure 2). However, there is no significant difference between algal species across sampling months (*P*=0.3; CI=95%). The concentration of cadmium in the sediment ranged from 3.72 to 5.53 mg kg^{-1} dry weight. Highest cadmium concentration was recorded in October while lowest in August (Figure 3) but the difference was not statistically significant (*P*=0.4; CI=95%).

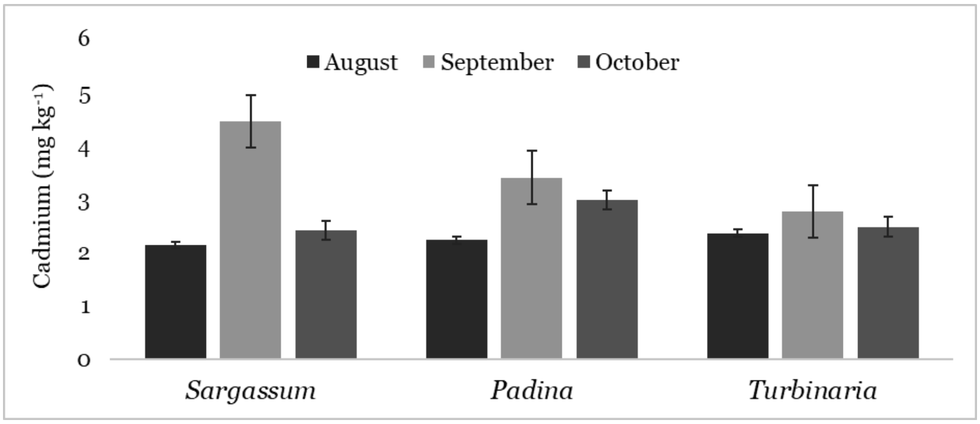

Figure 2. Concentration of cadmium (Cd) in 3 species of brown algae from August to October 2018. (*P*>0.05; error bar=±SD).

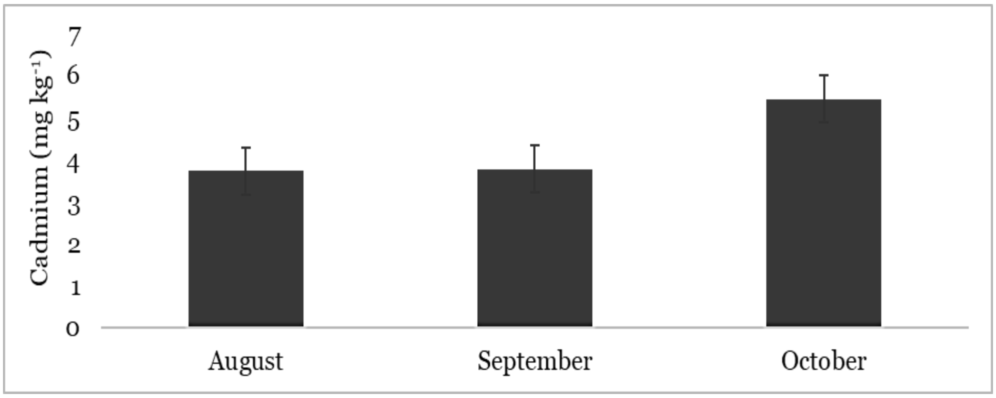

Figure 3. Concentration of Cadmium (Cd) in the sediment from August to October 2018. (P>0.05; error bar=±SD).

DISCUSSION

Cadmium concentration in *Sargassum* sp., *Padina* sp. and *Turbinaria* sp. provided an estimate on the bioavailability of cadmium in Bais Bay. Comparison of Cd concentration in the algal samples collected from Bais Bay were higher than other areas in the Philippines such as the coastal waters of Batangas, Philippines with 0.50 mg kg^{-1} (Borja et al. 2015). Likewise, cadmium level in Bais Bay during this study was higher by several orders of magnitude than the natural range from 0.15 to 0.20 mg kg^{-1} of concentration for this element (Kabata-Pendias and Pendias 1979). This implies input of cadmium in Bais Bay from various sources within the bay.

Electroplating, smelting, paint pigments, batteries, fertilizers, mining and alloy industries are considered as the major anthropogenic sources of cadmium in the environment (Iqbal and Edyvean 2005). In Bais, agricultural, domestic and industrial wastes are the most important sources of pollution. In 2001, waste generation of the domestic, commercial and institutional sectors in Bais City was estimated at 7,300 tons yr^{-1} but will most likely increase to at least 10,000 tons yr^{-1} in 2020. The domestic sector generated an average waste of 0.20 kg capita^{-1} day^{-1} while the two sugar mills generated approximately 85,000 tons of mudpress, lime-mud and ash every year as main process residues. Widespread and regular application of chemical fertilizer and pesticides on more than 19,000 ha for agricultural production every year and using the river system for scattered waste disposal provided further waste sources (Paul 2002). In 2015, release of industrial untreated wastewater into the bay was reported (Matus 2015). Though the chemical composition of untreated industrial effluents and urban runoff has not been characterized, accumulation of environmental pollutants may occur in Bais

Bay due to its enclosed condition with limited water exchange as observed in the Gulf of Thailand (Wattankorn 2006).

The presence of cadmium in the thalli of *Sargassum*, *Padina* and *Turbinaria* showed that these species absorbed cadmium from Bais Bay. Thus, these species could be most effective for biomonitoring and phytoremediation of toxic heavy metals because their thalli directly interact with the water column. It has been observed that *Sargassum* and *Padina* had the highest potential as biosorbents for the removal of heavy metals such as Pb^{2+}, Cu^{2+}, Cd^{2+}, Zn^{2+}, and Ni^{2+} from aqueous solutions. The general affinity sequence for *Sargassum* was Pb>Zn>Cd>Cu>Ni while that of *Padina* was Pb>Cu>Cd>Zn>Ni (Sheng et al. 2004). Due to abundance of alginate in the cell wall of brown algae especially order Fucales and Laminariales, metal ions such as Pb^{2+}, Cu^{2+}, Cd^{2+}, Zn^{2+}, Ca^{2+} are sequestered from the aqueous solution through the formation of network junctions by the cations of homopolymeric guluronic acid blocks in alginic acid (Davis et al. 2003). When guluronic acid content increased, affinity of alginates for divalent cations also increased (Haug 1961). The higher specificity of polyguluronic acid residues for divalent metals is explained by its "zigzag" structure which can accommodate the Ca^{2+} (and other divalent cations) ion more easily (Haug et al. 1967). The alginates are thought to adopt an ordered solution network, through inter-chain dimerization of the polyguluronic sequences in the presence of calcium or other divalent cations of similar size (Rees 1981). Fourier-transformed infrared (FTIR) spectral analyses have shown that cadmium biosorption to *Sargassum* arises from bridging or bidentate complex formation with the carboxylate groups of the alginate (Fourest and Volesky 1996). It has been also shown that ion-exchange takes place between metals when binding to alginate (Myklestad 1968). Untreated biomass generally contains light metal ions such as K^{+}, Na^{+}, Ca^{2+}, and Mg^{2+}. These are originally bound to the acid functional groups of the alga. During exposure of macroalgae to heavy metal solutions, metal ions replace for some of the cations such as K^{+}, Na^{+}, Ca^{2+}, and Mg^{2+} that were initially bound to the acid functional groups and make stronger cross-linking (Saravanan et al. 2011). Aside from alginate, sulphonate groups of fucoidan also contribute to heavy metal biosorption. But its role could become prominent if the binding of the metal occurs at a low pH (Fourest and Volesky 1996).

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