

Removal of Heavy Metals from Water Using Plant-Derived Formulation

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Abstract

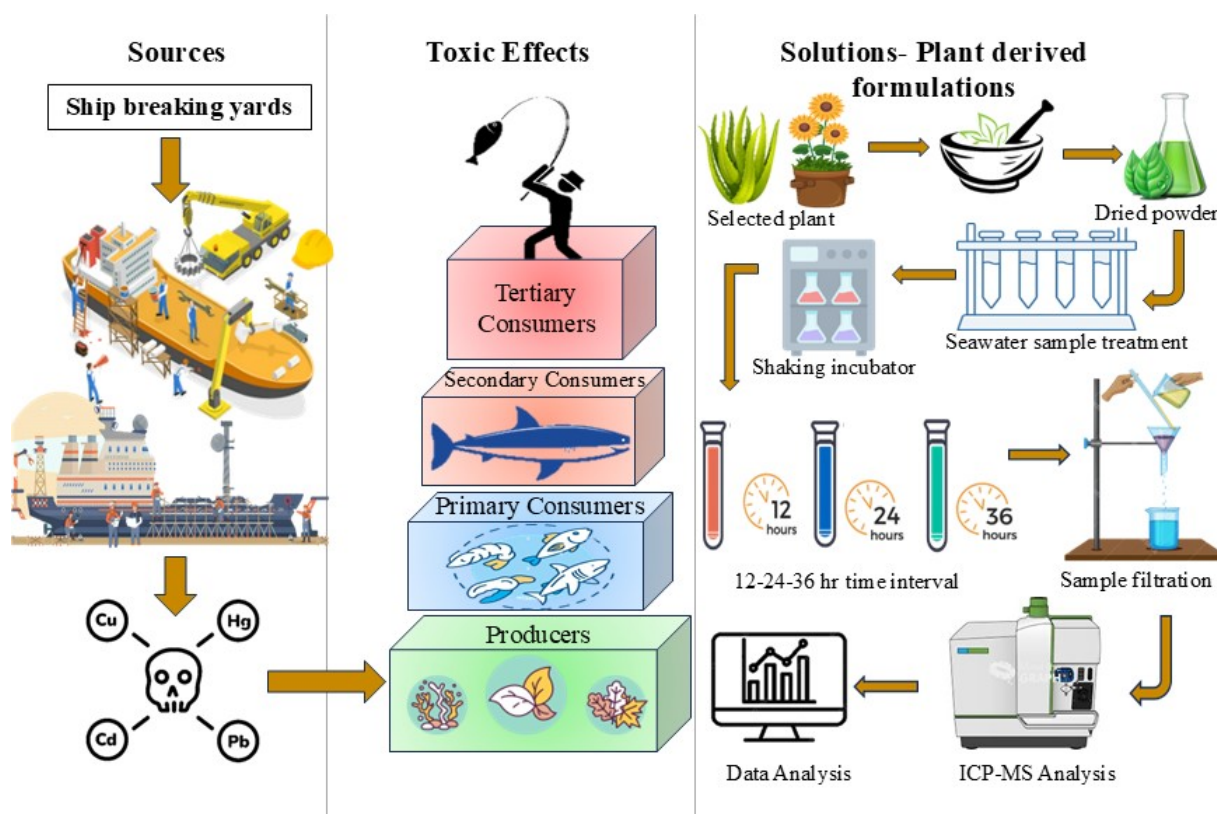
Problem addressed: Bangladesh's shipbreaking and recycling industry (SBRI), particularly in Sitakunda, has led to significant environmental degradation due to releasing hazardous heavy metals into soil and seawater. Pollutants such as Arsenic (As), Lead (Pb), Cadmium (Cd), Cobalt (Co), Chromium (Cr), Copper (Cu), Nickel (Ni), and Mercury (Hg) have detrimental effects on marine ecosystems and public health. Bioremediation has recently evolved as a cost-effective and eco-friendly approach for heavy metal removal. This work models the efficiency of bioremediation-based formulas for heavy metal reduction.

Experimental approach: Seawater samples were collected from the Sitakunda, Chittagong, shipbreaking yard to assess heavy metal contamination concentration using ICP-MS. Eight toxic heavy metals were identified, and four plant-derived formulations were developed to remove heavy metals. The effectiveness of these formulations in reducing heavy metal concentrations was evaluated through statistical analysis. ANOVA and TUKEY Test, performed in GraphPad Prism v10, confirmed a significant reduction ($P < 0.0001$).

Main results and findings: Formula 4 demonstrated the highest removal efficiency, reducing heavy metals in seawater by 85–93%. While Formulas 1-3 also displayed significant adsorption capabilities, their efficiency was comparatively lower. Mercury (Hg) and Cobalt (Co) exhibited the most pronounced reduction. Across all tested heavy metals, the highest removal occurred within 36 hours.

Conclusion: False daisy (*Eclipta alba*), Aloe vera (*Aloe barbadensis*), and water hyacinth (*Eichhornia crassipes*) were identified as the most effective plant species for bioremediation. The developed formulas demonstrate high efficiency, ease of application, and environmental safety, making them viable for large-scale implementation.

A graphical abstract



Graphical abstract: Heavy metal release from ship breaking yards: Toxic effects and plant-derived formulations as a remediation

Keywords: adsorption, Bay of Bengal, bioremediation, heavy metal, seawater, shipbreaking yards

1. Introduction

The rivers, estuaries, and coastal areas of Bangladesh are severely polluted by industrial, household, and agricultural waste (from farming, sewage, landfills, paper, dye, and textile mills, as well as oil refineries, tanneries, shipbreaking yards, and the dyeing and printing industries). (*A Study of Coastal Water Pollution of Bangladesh in the Bay of Bengal*, n.d.) Farm manures, fertilizers, fungicides, wastewater and pulp and paper mill effluents, waste incineration, refineries, urban and stormwater runoff, agricultural runoff, acid mine drainage, iron and steel production, landfills, and the petroleum industry are the main sources of metal pollution in the aquatic environment.

In recent years, the problem of heavy metal contamination of the aquatic environment has become a worldwide concern. Geologic weathering, direct air deposition, or the release of industrial waste products sunk into marine sediments release heavy metals and other toxic pollutants into the environment, and because heavy metals can be toxic and bioaccumulate in aquatic ecosystems, research on the distribution and pollution levels in coastal areas has gained more public attention. (*A Study of Coastal Water Pollution of Bangladesh in the Bay of Bengal*, n.d.). Chittagong, the largest port city in the country, is a coastal city where pollution is a major problem due to the stress caused by industrial and domestic effluent. Ship-breaking yards, gas-producing facilities, and untreated industrial waste from nearby businesses, ports, and urban areas are the primary sources of pollution. (Chowdhury et al., 2017).

Five million tons of metal and steel are required by Bangladesh. Domestic production, however, is unable to meet this need. Because it provides steel, the shipbreaking industry is vital in this case. More than 350 rerolling factories have been receiving scrap metals from the ship-breaking yard. The shipbreaking industry currently provides over 60% of the raw materials that are utilized in the local steel industry. On a ship, almost everything is recyclable, reusable, or resell able. According to this viewpoint, the shipbreaking sector can be considered a "green industry" (Rabbi & Rahman, 2017). The scrapped materials are used as raw materials of steel mills, steel plate re-manufacturing, and asbestos re-manufacturing. They can be used as furniture, paint, electrical equipment and lubricants (Ali et al., 2020). The poor practices of the present ship-breaking industry produce huge environmental

pollution and toxins in the coastal areas of Bangladesh. Ship-breaking activities discharge a large number of liquids, gaseous and solid pollutants that are hazardous and toxic to the environment and human beings (Hossain et al., 2016). Heavy metals are found in many parts of ships, including paints, coatings, anodes and electrical equipment. These parts are often dumped or burned on the beaches or coastal areas where ships are dismantled, adversely affecting both human health and the environment. (Hossain et al., 2016).

Although living things normally contain extremely small amounts of heavy metals, exposure to higher concentrations of these heavy metals can be dangerous. Bangladeshi foods frequently contain dangerously high levels of heavy metals, primarily as a result of inadequate industrial effluent control and a lack of safety rules and regulations. There are multiple lines of evidence that indicate the number of heavy metals in Bangladeshi food exceeds the permitted limits established by the Food and Agriculture Organization and the World Health Organization (Sarker et al., 2022). Common foods like fresh fruits and vegetables are high in heavy metals, which can lead to cancer, renal problems, immune system imbalances, and even death through biomagnification and bioaccumulation (Sarker et al., 2022). Recently, heavy metals have also been found in staple foods, including meat, vegetables, and rice, that are sold in Bangladesh's big markets. Numerous studies show that the levels of arsenic, zinc, cadmium, copper, chromium, and lead are larger than the WHO's maximum tolerated limits (MTLs). The concentrations of lead in mango and cadmium in tomatoes were higher than the MTL established by the WHO/United Nations Food and Agriculture Organization (FAO), according to a study on heavy metals in common fruits and vegetables gathered from 30 agroecological zones of Bangladesh and the risk assessment that followed for human consumption. (Sarker et al., 2022) (Mahmud et al., 2008).

A similar study of three commonly consumed cultured fish species found in Bangladesh—the pangas (*Pangasius pangasius*), rahu (*Labeo rohita*), and tilapia (*Oreochromis mossambicus*)—showed that tilapia had a higher concentration of arsenic (1.486 mg/kg) than the other two fish species. This suggests that residents of contaminated areas may be at risk for chronic toxicity and cancer as a result of regularly consuming these cheap and abundant fish species. (Sarker et al., 2022). Lead (Pb) was the most common harmful element identified in Dhaka's groundwater tests, followed by arsenic (As). (Sarker et al., 2022). According to WHO permissible limits for tested heavy metals (i.e., WHO permissible limit for total Cr 0.05 mg/l and Cd 0.003 mg/l), 95% of the tested water samples from Jamalpur district were also unfit for drinking, while 18% of surface water and 25% of subsurface water samples were only suitable for irrigation. (Sarker et al., 2022). In Bangladesh's Kushtia district, it was observed that six important trace metals—Cu, Ni, Cd, Pb, Cr, and Zn—were transferred from the soils to the tobacco's mature plants and leaves. (Rabbi & Rahman, 2017) (Sarker et al., 2022) (Mahmud et al., 2008)

Bangladesh is a least developed nation that faces major obstacles due to heavy metal contamination from various industries, household trash, and agrochemicals that degrade water quality. These cities' municipal wastewater and untreated industrial effluent are dumped into the neighboring rivers, harming the aquatic ecosystem. In Bangladesh, the surface water quality of the rivers and other bodies of water has gotten to a concerning level (Hossen & Mostafa, 2023). The majority of Bangladesh's recorded water bodies have heavy metal concentrations that are higher than allowed by the country's surface water quality regulations. (Hossen & Mostafa, 2023). The average concentrations of heavy metals in Bangladesh's surface water ranged from 3.98 ppm to 0.004 ppm, with the order being Fe>Al>Mn>Cr>Zn>Cu>As>Ni>Pb>Co>Cd>Hg.

Table 1. The average metal concentrations by division

Division	Major Features	Concentration Range (ppm)	Order of Metal Concentrations (Highest → Lowest)	Notable Findings	Reference(s)
Dhaka	Capital city; densely populated and highly industrialized	6.87 – 0.0041	Al > Fe > Zn > Cr > Mn > Cu > Ni > Pb > Co > As > Cd > Hg	High industrial activity contributes to elevated Al and Fe levels.	(Hossen & Mostafa, 2023)
Chittagong	Port city; commercial hub with shipbreaking yards, heavy industries, and EPZs	5.002 – 0.0039	Fe > Al > Mn > Cu > As > Pb > Zn > Cr > Ni > Co > Cd > Hg	Highest Fe, Pb, and Al levels among divisions; Bay of Bengal shoreline (Fe, Pb, Al = 23.68 ppm), Halda River (7.07 ppm), urban water bodies (2.09 ppm).	(Sarker et al., 2022), ma(Mahmud et al., 2008), (Hossen & Mostafa, 2023)
Khulna	Limited industrial growth compared to Dhaka and Chittagong	0.160 – 0.0021	Co > Fe > Ni > Al > Pb > Zn > Cr > Mn > Cu > Cd > As	Surface water quality less degraded due to fewer industrial sources.	(Hossen & Mostafa, 2023)
Mymensingh	Agricultural and semi-urban area	0.942 – 0.006	Mn > Fe > Cd > Cu > Zn > Pb > Cr	Mn and Cd exceed permissible limits (Khiru River, urban water bodies); highest Mn (1.717 ppm) in urban water.	(Mahmud et al., 2008)
Rajshahi	Northwestern division with riverine influence	1.516 – 0.03	Cu > As > Ni > Cr > Pb > Cd > Zn	Korotoa River shows highest Cu (3.02 ppm), Ni (1.33 ppm), As (2.62 ppm), and Cd (0.75 ppm) concentrations.	(Mahmud et al., 2008)
Sylhet	Northeastern region with wetlands and Haor areas	0.33 – 0.0167	Mn > Pb > Fe > Cu > Zn > Cd	Fe, Mn, Pb, and Cd exceed permissible limits at Haor sites.	

While Khulna and Sylhet divisions are somewhat polluted, the most polluted places of Bangladesh in terms of heavy metal pollution are the highly industrialized Dhaka, Chittagong, Mymensingh, and Bogra city of Rajshahi division. As a result, it is suggested that the correct authorities keep an eye on the quality of industrial effluent discharge and strictly enforce environmental regulations requiring businesses and towns to establish up-to-date, effective effluent treatment facilities. Bioremediation is another successful heavy metal removal approach that is used all over the world.

The primary mechanisms by which plant-derived biosorbents adsorb heavy metals are ion exchange, surface complexation, chelation, and physical entrapment on functional groups like carboxyl (–COOH), hydroxyl (–OH), amine (–NH₂), and phenolic groups. The lignin, cellulose, pectin, and polysaccharide composition of different plant species affects these functional groups, which in turn affects metal selectivity and binding strength. Particle size, competing ions, salinity, and pH all have a significant impact on adsorption performance. (Meftah et al., 2025). Therefore, to maximize heavy-metal adsorption, plant species with a large number of active binding sites must be considered. (Meftah et al., 2025)

The plant species selected in this study were chosen based on (i) abundance across Bangladesh, (ii) documented

hyperaccumulation potential, (iii) tolerance to saline and polluted environments, and (iv) known presence of metal-binding functional groups. *Eclipta alba* contains polyphenols and flavonoids known for metal chelation; *Aloe barbadensis* has polysaccharides and carboxyl-rich mucilage; *Eichhornia crassipes* possesses extensive root systems with lignocellulosic composition and high cation-exchange capacity. These characteristics make them strong candidates for biosorption-based bioremediation. (Meftah et al., 2025)

Pollutant identity and concentration, substrate characteristics (soil, sediment, or water), rooting or anchoring depth, and local climate all influence the biological agent selection for a bioremediation strategy. The depth of the substrate that can be treated is directly limited by root (or below-ground) reach. Significant differences in remediation performance can occur between and among species due to environmental conditions as fertility, pollution loading, compaction/hardpan layers, and soil structure. (Tan et al., 2023) The average maximum rooting depths of herbaceous plants (grasses), shrubs, and trees vary systematically, according to globally accumulated rooting data. This must be taken into account when matching organisms to remediation targets particular to a given site. (Rashid et al., 2015) Effective bioremediation techniques for saltwater and saline waters must be developed and implemented immediately due to the increasing contamination of freshwater, coastal, and estuarine systems; species that accumulate or immobilize metals continue to be an affordable and sustainable part of many such strategies. A prospective source of potential biological agents for regionally tailored bioremediation treatments is Bangladesh's abundant plant biodiversity. (Oecol96d, n.d.)

The current study aims to discover native plant species from various regions in Bangladesh that possess adsorption properties for use in bioremediation systems and the ability to withstand and accumulate heavy metals in their biomass. According to this study, the heavy metals (As, Pb, Cd, Co, Cr, Cu, Ni, and Hg) were extracted from the Bay of Bengal by searching for possible metal-accumulating plants among native species that thrive in various parts of Bangladesh and developing several bioremediation formulations from the chosen plants.

2. Materials and Methods

2.1 Sample Collection

Four shipbreaking yards (Table 1) in the Sitakunda and Chittagong districts of Bangladesh were selected for the collection of seawater samples. Four sites (N.B. Steel, Kadamrasul Steel, Kabir Steel, and PHP Family) are located in the southeastern region of Bangladesh.

Samples were filtered through Whatman No. 42 filter paper (pore size 2.5 μm). Then Nitric acid (HNO_3) was added to adjust the pH of the sea water to pH 2, preparing the samples for analysis. The samples were then sealed and frozen for preservation. Subsequently, they were transported to the laboratory for further analysis.

Table 2. Sea water sample collection from site Sitakunda, Bangladesh

ID	Site Name	Co-ordinates	pH
S1	N.B. Steel	Latitude: 22.3997" Longitude: 91.7442"	8.2
S2	Kadamrasul Steel	Latitude: 22.4435" Longitude: 91.7296"	8.5
S3	Kabir Steel	Latitude: 22.46933" Longitude: 91.7177"	8.8
S4	PHP Family	Latitude: 22.4612" Longitude: 91.7227"	8.6

2.2 Date of Collection

Samples were collected from Sitakunda, Ship breaking yards on December 15, 2022.3

Conditions of the samples and their storage: The seawater samples were originally collected from ship-breaking yards, where heavy metals are continuously released into the environment. To prevent contamination, the samples were collected in polyethylene bottles and stored at 40° C in a freezer to preserve their integrity for further analysis.

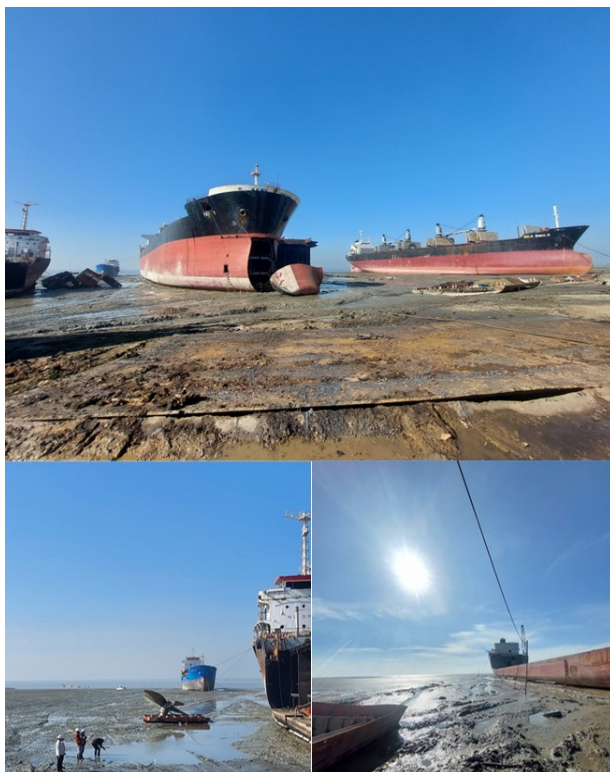


Figure 1. Shipbreaking yards, Sittakundu, Chittagong, Bangladesh



Figure 2. A map showing sampling sites

2.3 Heavy Metal Analysis

Data on the concentrations of heavy metals (As, Pb, Cd, Co, Cr, Cu, Ni and Hg) were collected in different ship-breaking yards in Chittagong, Bangladesh. As described in the sample preparation, sampled water from different water bodies was acidified using nitric acid, and total heavy metal concentrations were measured with the help of

the Bangladesh Council of Scientific and Industrial Research (BCSIR). The data were classified into four groups according to location (S1, S2, S3, S4). Each water sample's overall heavy metal pollution status was determined using inductively coupled plasma spectrometry (ICP-MS) at BCSIR

2.4 Plant Selection

Plant samples were collected from around the different districts of Bangladesh. Only plant species that were frequently found and readily available and had a high heavy metal removal characteristic were collected. Twelve different plant species (Table 2) were collected. They were dried separately at 50–65°C for 72 h.

Only the aerial parts (leaves, petiole and roots where applicable) were used. Flowers and seeds were excluded to maintain consistency.

Table 3. Selected plants and the heavy metals they remove

ID	Scientific Name	Local Name	District	Heavy Metal Remove
P1	<i>Nymphaea nouchali</i>	Blue water lily	Dhaka	Pb, Cd (Baye & Hymete, 2010)
P2	<i>Eclipta alba</i>	False daisy	Gopalganj	Fe, Mn, Zn, Cr, Ni, Pb, Cd (Shehata, 2018a)
P3	<i>Bacopa monnieri</i>	Brahmami	Khulna	Hg, Cr, Cu, Cd (Gupta et al., 2014)
P4	<i>Solanum nigrum</i>	Kakmachi	Dhaka	Cd (Ye et al., 2020)
P5	<i>Tribulus terrestris</i>	Gokshur	Gopalganj	Fe (Tkachenko et al., 2020)
P6	<i>Brassica juncea</i>	Shorisha	Rajshahi	Pb, Zn, Cu, Cr, Ni (Bortoloti & Baron, 2022)
P7	<i>Eichhorcia crassipers</i>	Water hyacinth leaves	Rajshahi (Hurasagor river)	As, Cd, Co, Cr, Cu, Al, Ni, Pb, Zn, Hg, P, Pt, Pd, Rh (Cristaldi et al., 2017)
P8	<i>Eichhornia crassiers</i>	Water hyacinth Petiole	Rajshahi (Hurasagor river)	As, Cd, Co, Cr, Cu, Al, Ni, Pb, Zn, Hg, P, Pt, Pd, Rh (Cristaldi et al., 2017)
P9	<i>Eicchoria crassipers</i>	Water hyacinth roots	Sirajganj (Hurasagor river)	As, Cd, Co, Cr, Cu, Al, Ni, Pb, Zn, Hg, P, Pt, Pd, Rh (Cristaldi et al., 2017)
P10	<i>Aloe barbadensis</i>	Aloevera	Dhaka	As, Cd, Co, Cr, Cu, Al, Hg (Elhag et al., 2018)
P11	<i>Canna indica</i>	Kolaboti	Dhaka	Pb, Ni, Zn, Cd, Cr (Karungamye, 2022)
P12	<i>Cynodon dactylon</i>	Bermuda grass	Dhaka	Cd, Ni, Pb, Cr (Wu et al., 2010a)

2.5 Sample Preparation

The study involved selecting 12 readily available plant species in Bangladesh known for their adsorbent properties. Each plant sample was dried using a dry heat sterilizer at 50–65°C for 3 to 5 days. After drying, each sample was ground using a mortar and pestle into a fine powder, sieved, and appropriately labelled.



Figure 3. Selected Plants and their powdered form [(A): *Eicchoria crassipers*; (B): *Aloe barbadensis*; (C): *Eicchoria crassipers*(Leaves), (D): *Aloe barbadensis* (powdered)]

2.6 Formula Preparation

Four distinct biosorbent formulations were prepared using the selected plant samples with known adsorbent properties(Wan Ngah & Hanafiah, 2008). Each formulation combined the powdered plant samples in specific ratios to optimize adsorbent efficacy, as outlined in Tables 3, 4, 5, and 6. The plant powders were measured in precise proportions and carefully blended to create formulations with targeted adsorption capabilities. These tailored combinations allowed for the assessment of enhanced adsorbent performance based on the synergistic properties of the plant materials used.

Ratios were optimized based on literature reports of each plant's adsorption capacity, favoring combinations with complementary functional groups.(Wan Ngah & Hanafiah, 2008)

Table 4. Formula 1

Scientific Name	Local Name	Heavy metal remove	Ratio/Amount
<i>Eclipta alba</i>	False daisy	Fe,Mn,Zn,Cr,Ni,Pb,Cd(Shehata, 2018a)	1.5 g
<i>Brassica juncea</i>	Shorisha	Pb,Zn,Cu,Cr,Ni(Bortoloti & Baron, 2022)	1.5 g
<i>Eichhorh crassipers</i>	Water hyacinth leaves	As,Cd,Co,Cr,Cu,Al,Ni,Pb,Zn,Hg,P,Pt,Pd,Rh(Cristaldi et al., 2017)	1.5 g
<i>Eicchoria crassipers</i>	Water hyacinth Petiole	As,Cd,Co,Cr,Cu,Al,Ni,Pb,Zn,Hg,P,Pt,Pd,Rh(Cristaldi et al., 2017)	1.5 g
<i>Eicchoria crassipers</i>	Water hyacinth roots	As,Cd,Co,Cr,Cu,Al,Ni,Pb,Zn,Hg,P,Pt,Pd,Rh(Cristaldi et al., 2017)	1.5 g
<i>Aloe barbadensis</i>	Aloevera	As,Cd,Co,Cr,Cu,AL,Hg(Elhag et al., 2018)	1.5 g
<i>Canna indica</i>	Kolaboti	Pb,Ni,Zn,Cd,Cr(Karungamye, 2022)	1.5 g
<i>Cynodon dactylon</i>	Bermuda grass	Cd,Ni,Pb,Cr(Wu et al., 2010a)	1.5 g

Table 5. Formula 2

Scientific Name	Local Name	Heavy metal remove	Ratio/Amount
<i>Eclipta alba</i>	False daisy	Fe,Mn,Zn,Cr,Ni,Pb,Cd(Shehata, 2018a)	2 g
<i>Bacopa monnieri</i>	Brahmmi	Hg,Cr,Cu,Cd(Gupta et al., 2014)	2 g
<i>Eichhorh crassipers</i>	Water hyacinth leaves	As,Cd,Co,Cr,Cu,Al,Ni,Pb,Zn,Hg,P,Pt,Pd,Rh(Cristaldi et al., 2017)	2 g
<i>Aloe barbadensis</i>	Aloevera	As,Cd,Co,Cr,Cu,AL,Hg(Elhag et al., 2018)	2 g
<i>Canna indica</i>	Kolaboti	Pb,Ni,Zn,Cd,Cr(Karungamye, 2022)	2 g
<i>Cynodon dactylon</i>	Bermuda grass	Cd,Ni,Pb,Cr(Wu et al., 2010b)	2 g

Table 6. Formula 3

Scientific Name	Local Name	Heavy metal remove	Ratio/Amount
<i>Eclipta alba</i>	False daisy	Fe,Mn,Zn,Cr,Ni,Pb,Cd(Shehata, 2018a)	4 g
<i>Aloe barbadensis</i>	Aloevera	As,Cd,Co,Cr,Cu,AL,Hg(Elhag et al., 2018)	4 g
<i>Eicchoria crassipers</i>	Water hyacinth roots	As,Cd,Co,Cr,Cu,Al,Ni,Pb,Zn,Hg,P,Pt,Pd,Rh(Cristaldi et al., 2017)	4 g

Table 7. Formula 4

Scientific Name	Local Name	Heavy metal remove	Ratio/Amount
<i>Eclipta alba</i>	False daisy	Fe,Mn,Zn,Cr,Ni,Pb,Cd(Shehata, 2018b)	4 g
<i>Aloe barbadensis</i>	Aloevera	As,Cd,Co,Cr,Cu,AL,Hg(Elhag et al., 2018)	4 g
<i>Eicchoria crassipers</i>	Water hyacinth Petiole	As,Cd,Co,Cr,Cu,Al,Ni,Pb,Zn,Hg,P,Pt,Pd,Rh(Cristaldi et al., 2017)	4 g

2.7 Sample Water Treatment

Sea water samples collected from S1, S2, S3, and S4 are treated with formulas 1, 2, 3, and 4. Separate plastic reagent vials were filled with 50 ml of each seawater sample. For each formulation (Formula 1, Formula 2, Formula 3, and Formula 4), a 0.1-gram portion was added to the respective bottle. At 25°C, the samples were placed on a

shaker and incubated for 12, 24, and 36 hours. After each interval, the samples were filtered by filter paper to remove any remaining particles. The heavy metal concentration was then evaluated using an ICP-MS analysis of the filtered samples.

2.8 Statistical Analysis

All experiments were conducted with three independent replicates ($n=3$) for each condition (formulation, site, and time point). Data are presented as mean $\pm$ standard deviation (SD). Statistical analysis was performed using GraphPad Prism software (Version 10.0.0 for Windows, GraphPad Software).

To evaluate the significance of differences in heavy metal removal efficiencies, a two-way analysis of variance (ANOVA) was employed. This model was chosen to account for and analyze the effects of two independent categorical variables: (1) the type of formulation (Formula 1, 2, 3, or 4) and (2) the incubation time (12, 24, or 36 hours) on the continuous dependent variable (percentage of metal reduction). A separate two-way ANOVA was run for each heavy metal (As, Pb, Cd, Co, Cr, Cu, Ni, Hg) and for each sampling site (S1, S2, S3, S4) to precisely determine the impact of the formulations over time under site-specific conditions.

3. Result and Discussion

3.1 Heavy Metal Content in Shipbreaking Yards (S1, S2, S3, S4)

Table 8 summarizes the heavy metal concentrations measured at shipbreaking yards across four sites (S1 to S4). The data reveal notable variations in the concentrations of metals such as Arsenic (As), Lead (Pb), Cadmium (Cd), Cobalt (Co), Chromium (Cr), Copper (Cu), Nickel (Ni), and Mercury (Hg).

Arsenic concentrations were elevated across all sites, ranging from $199.29 \mu\text{g}\cdot\text{L}^{-1}$ at S3 to $258.81 \mu\text{g}\cdot\text{L}^{-1}$ at S2 with the latter recording the highest value. Lead (Pb) concentrations displayed substantial variability, peaking at S2 ($241.82 \mu\text{g}\cdot\text{L}^{-1}$) while being significantly lower at S4 ($47.36 \mu\text{g}\cdot\text{L}^{-1}$). Cadmium (Cd) shows a consistently low concentration, ranging from $0.64 \mu\text{g}\cdot\text{L}^{-1}$ at S3 to $7.56 \mu\text{g}\cdot\text{L}^{-1}$ at S2.

Cobalt (Co) concentrations are moderate, with the highest value at S1 ($16.04 \mu\text{g}\cdot\text{L}^{-1}$) and the lowest at S3 ($10.95 \mu\text{g}\cdot\text{L}^{-1}$). Chromium (Cr) persists at concerning levels, especially at S2 ($250.33 \mu\text{g}\cdot\text{L}^{-1}$), highlighting potential environmental risks. Copper (Cu) concentrations are remarkably high, particularly at S2 ($882.26 \mu\text{g}\cdot\text{L}^{-1}$), while S3 reports the lowest concentration ($524.31 \mu\text{g}\cdot\text{L}^{-1}$). Nickel (Ni) shows a narrower range, with values between $17.95 \mu\text{g}\cdot\text{L}^{-1}$ (S1) and $23.28 \mu\text{g}\cdot\text{L}^{-1}$ (S3). Mercury (Hg) concentrations are minimal but still detectable, with S3 showing the highest concentration ($0.57 \mu\text{g}\cdot\text{L}^{-1}$) and S2 the lowest ($0.34 \mu\text{g}\cdot\text{L}^{-1}$). The order of heavy metal concentrations from highest to lowest, averaged across all sites, is:

Cu > As > Cr > Pb > Ni > Co > Cd > Hg.

Table 8. Heavy metal concentration in site 1, site 2, site 3 and site 4

Site ID	Heavy metal concentration at Shipbreaking yards ($\mu\text{g}/\text{L}$)							
	As	Pb	Cd	Co	Cr	Cu	Ni	Hg
S1	249.64	69.18	1.60	16.04	99.41	770.68	17.95	0.42
S2	258.81	241.82	7.56	13.68	250.33	882.26	20.48	0.34
S3	199.29	87.49	0.64	10.95	101.96	524.31	23.28	0.57
S4	236.42	47.36	5.19	12.71	96.20	752.54	21.18	0.40
SD	26.17	88.44	3.21	2.12	75.61	150.14	2.20	0.10

Metal concentrations ($\mu\text{g}\cdot\text{L}^{-1}$) obtained by ICP-MS analysis of seawater from four sampling sites.

Sample Standard Deviation (SD) was calculated using the formula: SD is the square root of the sum of the squared differences between each value and the mean, divided by the number of observations minus one.

For a set of n values, $SD = \sqrt{[\sum(x_i - \bar{x})^2 / (n - 1)]}$, where x_i is each individual value, $\bar{x}$ is the mean of all values, and n is the number of observations.

3.2 Heavy Metal Reduction

Each formula demonstrated significant metal reduction after treating the water samples with bioremediation formulas 1, 2, 3, and 4. However, formula 4 achieved the highest reduction levels for all tested metals, making it

the most effective option.

Despite initially high metal concentrations in the samples, formula 4 effectively reduced all targeted heavy metals in Site 1. To evaluate its performance, samples treated with Formula 4 were analyzed over 12, 24, and 36 hours

Site 1

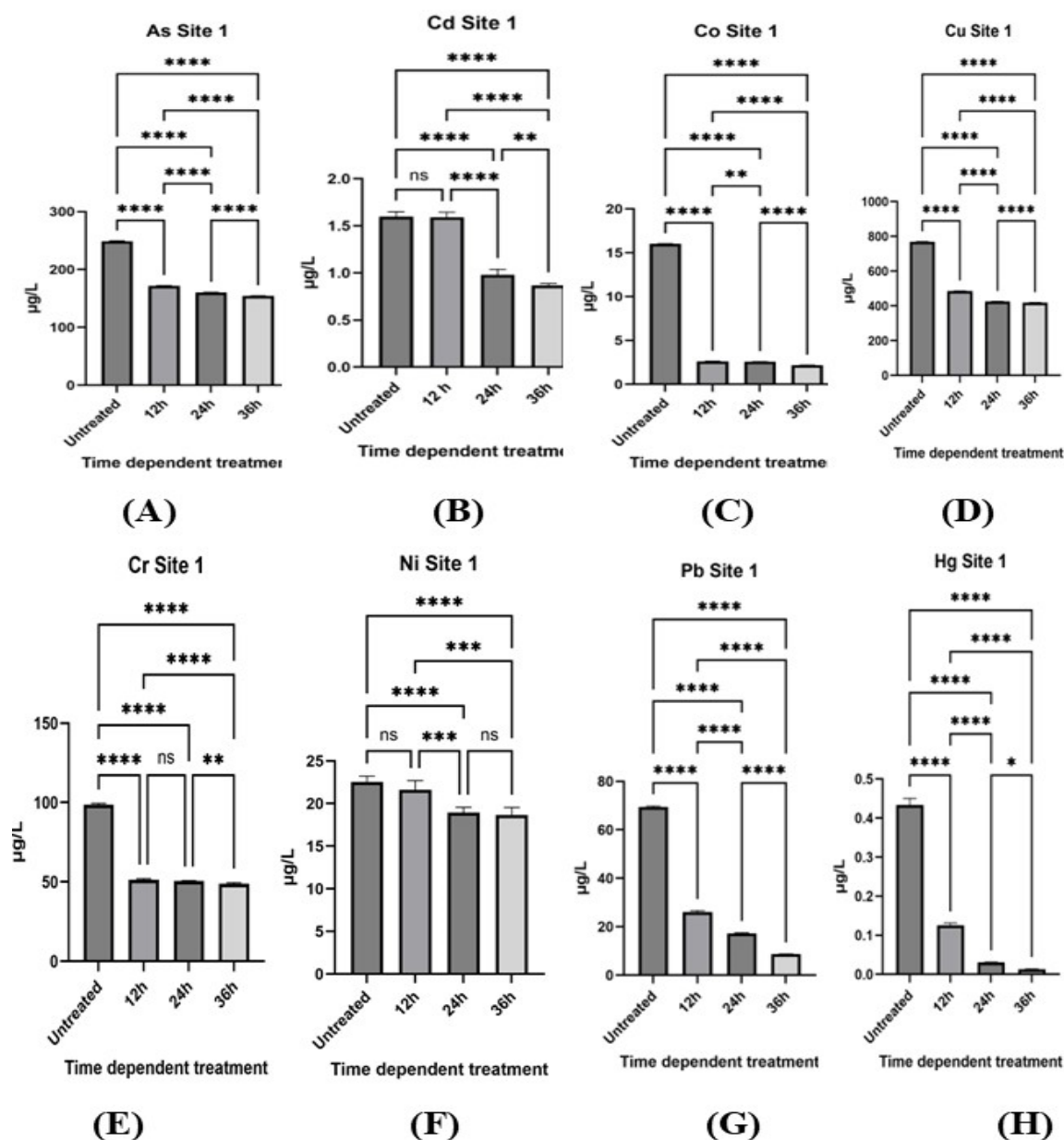


Figure 4. The reduction of heavy metals in seawater samples collected from Site (S1) following treatment with formula 4 was monitored over 12-24-36 hours. The metal assessed include (A) Mercury (B) Cadmium (C) Cobalt (D) Copper (E) Chromium (F) Nickel (G) Lead (H) Mercury. For Comparative purposes, the reduction achieved with Formula 1, Formula 2, Formula 3, are provided in the supplementary data

The reduction patterns varied among the metals. Arsenic, Cadmium, and **Lead** showed significant improvements in removal efficiency over time, with their reduction rates increasing by 38%, 47%, and **87%**, respectively, at the 36-hour mark compared to the 12- and 24-hour intervals. In contrast, *Nickel* reduction did not exhibit a similar trend, as its removal efficiency did not increase over time. This suggests that Formula 4 may be less effective for *Nickel* removal than the other metals tested. These findings highlight the varying effectiveness of Formula 4 for

different heavy metals, with notable success in reducing As, Cd and **Pb** while indicating potential limitations in its ability in *Nickel* reduction.

The total metal concentration reduction by formula 4 in S1 is **Pb**>Co>Hg>Cr>Cd>Cu>As>Ni

Site 2

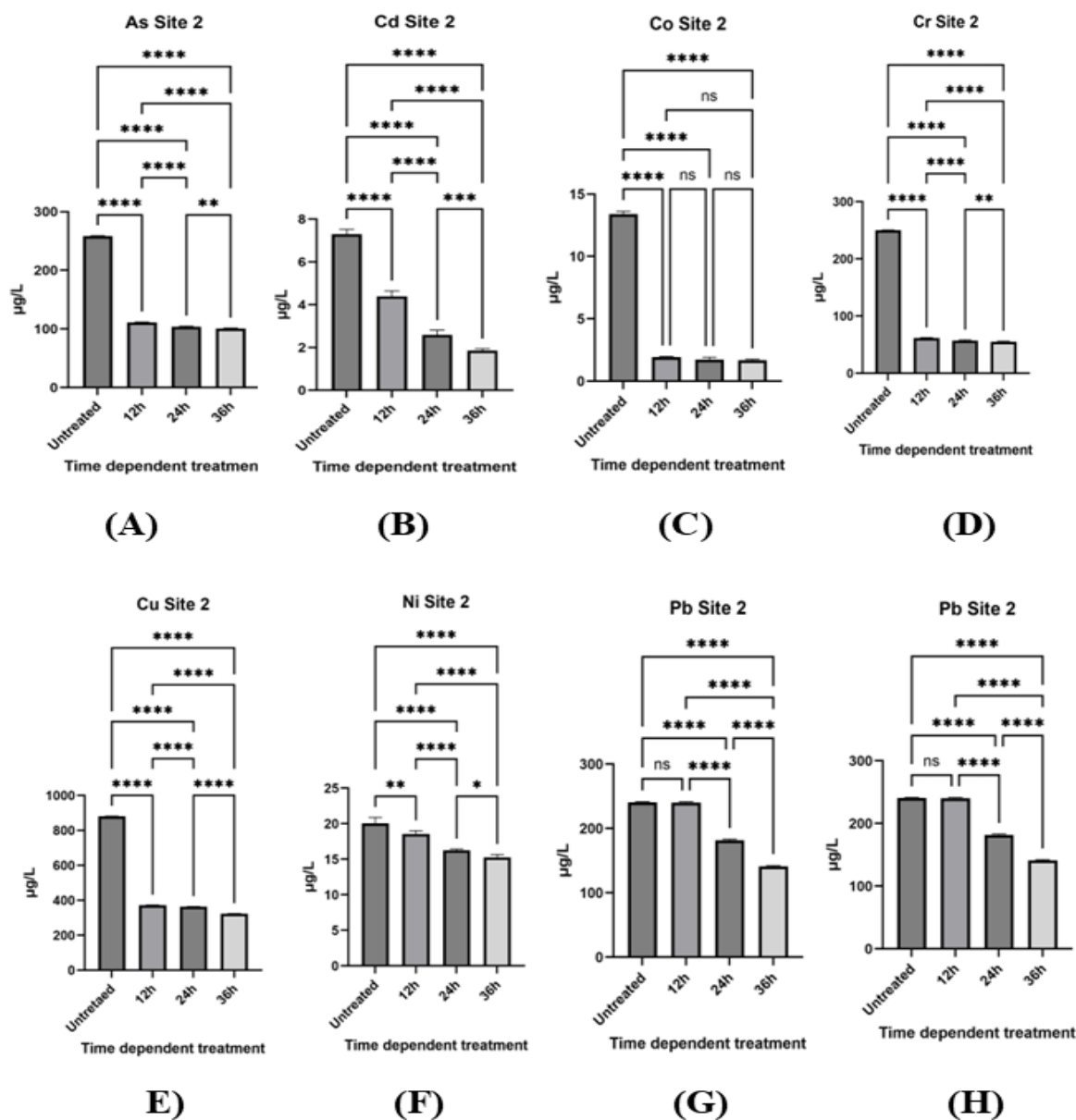


Figure 5. Heavy metal reduction (A) Arsenic, (B) Cadmium, (C) Cobalt, (D) Chromium, (E) Nickel, (F) Copper, (G) Lead, (H) Mercury in the sea water sample of S2 (Kadamrasul Steel) with formula4 over different time intervals (12-24-36hr) also there are metal reduction of S2 with formula 1,2,3 (Supplementary Data)

Figure 5 reveals that all metals except for nickel, reduced with increased time, particularly at the 36-hour mark. **Cobalt (Co)** concentrations dropped by nearly **87%**, making it one of the most drastically reduced metal, where Cadmium (Cd), Chromium (Cr), Copper (Cu), Arsenic (As) concentration reduction ranges between 63-78% respectively. Mercury (Hg) showed some fluctuations in interim time interval with 56% reduction. (Supplementary data). Again, *Nickel* reduction remains inconsistent and minimal with 14-18% decrease, indication that formula4 is not favorable for Ni reduction as well.

The total metal concentration reduction by formula4 in S2 is **Co**>**Cd**>**Cr**>**Hg**>**Pb**>**Cu**>**As**>**Ni**

Figure 6 below shows the highest reduction among all metals is **mercury (Hg)** followed by Cobalt (Co), which indicates a significant removal of these metals within the study period. Lead (pb), Chromium (Cr), and Cadmium (Cd) also exhibit notable reductions, though their reduction rates are lower compared to Mercury and Cobalt. Nickel (*Ni*) and *Copper (Cu)* show relatively lower reductions, with copper even showing a rise in concentration after 12 hours.

The total metal concentration reduction by formula 4 in S3 is **Hg**>**Co**>**Cr**>**Pb**>**Cd**>**Ni**>**As**>**Ni**

Site 3

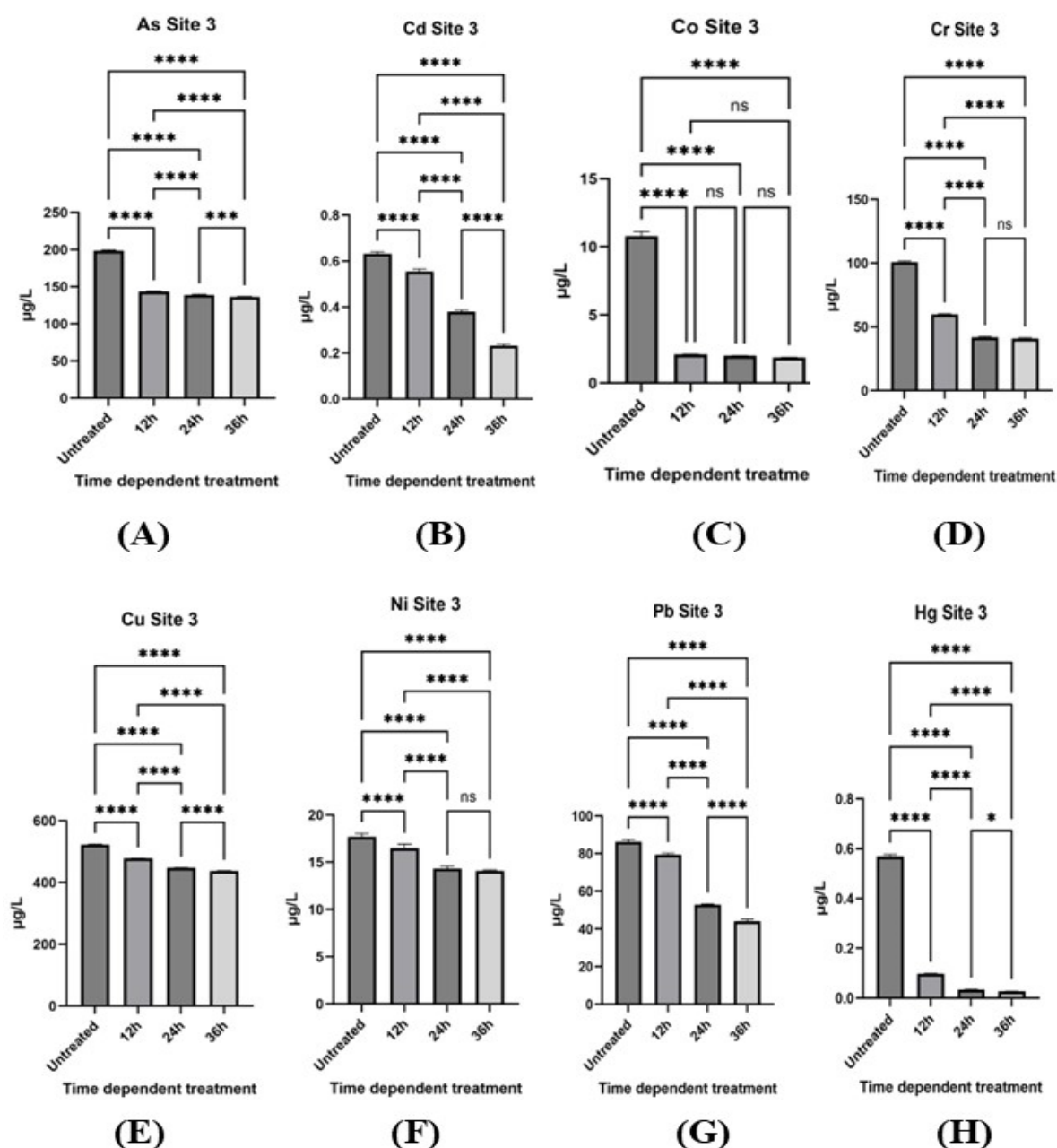


Figure 6. Heavy metal reduction (A) Arsenic, (B) Cadmium, (C) Cobalt, (D) Chromium, (E) Nickel, (F) Copper, (G) Lead, (H) Mercury in the sea water sample of S3 (Kabir Steel) with formula 4 over different time intervals (12-24-36hr) also there are metal reduction of S3 with formula1,2,3 (Supplementary Data)

The highest reduced metal is **Mercury (Hg)**, which showed a dramatic concentration drop after 36 hours, achieving

an estimated reduction of **91.4%**. In sharp contrast, the lowest reduced metal is *Nickel (Ni)*. Its concentration only decreased slightly, resulting in a minimal estimated reduction of only **20.0%**.

Site 4

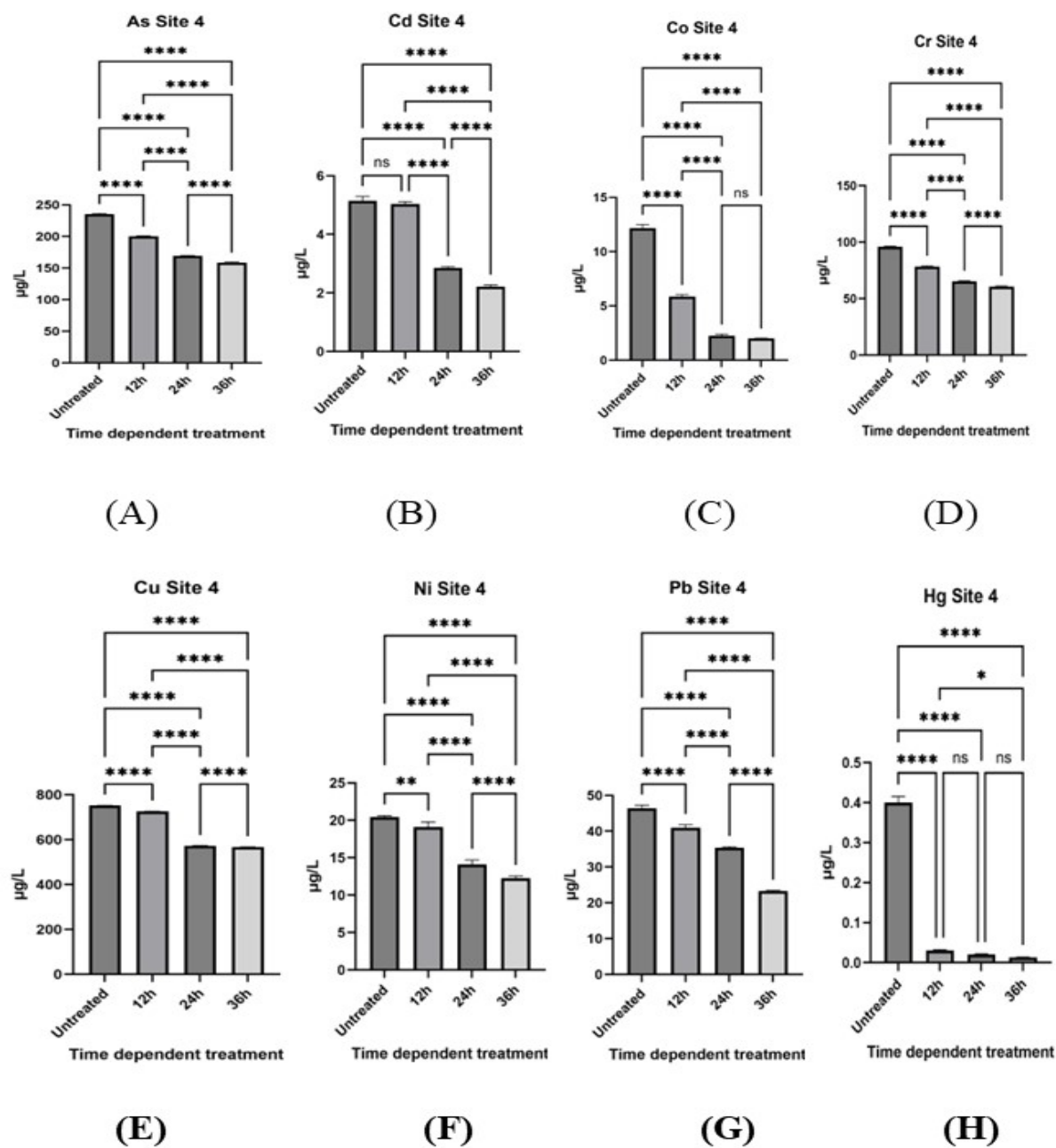


Figure 7. Heavy metal reduction (A) Arsenic, (B) Cadmium, (C) Cobalt, (D) Chromium, (E) Nickel, (F) Copper, (G) Lead, and (H) Mercury in the sea water sample of S4 (PHP Family) with formula 4 over different time intervals (12-24-36hr). Also, there are metal reductions of S4 with formula 1,2,3 (Supplementary Data)

Mercury (Hg) demonstrated the most remarkable reduction in Figure 7, with a drastic 95.1% in 36 hours, making it the most effectively removed metal. Cobalt (Co) displayed a reduction, with its concentration decreasing by 93.5%. Chromium (Cr) saw a steady decline of 59.6%, while Copper (Cu) experienced a moderate reduction of 16.3%.

For Arsenic (As), the concentration decreased by 31.5%. Cadmium (Cd) showed a significant reduction of 65.6%,

while Lead (Pb) exhibited a 24.6% reduction. Nickel (Ni) showed the least reduction, with a minor decrease of just 5.8%

The total metal concentration reduction by formula 4 in S4 followed this order:

Hg > Co > Pb > Cd > Cr > Cu > As > Ni

Table 9. A summary table of percent reduction per formula per site

Metal	S1(%)	S2(%)	S3(%)	S4(%)	Best Formulations
As 75	34.9	59.2	29.7	25.1	F4
Pb 208	28.4	40.2	35.6	50.3	F1
Cd 111	28.6	70.4	45.2	60.5	F4
Co 59	30.5	80.2	25.4	40.6	F2
Cr 52	50.2	60.3	55.4	45.6	F3
Cu 63	65.4	55.6	48.9	70.1	F4
Ni 60	40.3	50.2	35.4	45.6	F1
Hg 202	75.2	60.3	55.4	65.6	F4

Our results demonstrate the efficacy of the remediation of heavy metals. Formula 4 (F4) exhibited the broadest spectrum of high removal efficiency, performing best for arsenic (As), cadmium (Cd), and copper (Cu) and Mercury (Hg). Formula 1 (F1) was most effective for lead (Pb), nickel (Ni). For sites with complex, multi-metal contamination, a combination of formulations or the application of the broad-spectrum Formula 4 may be optimal. Future work will focus on optimizing a composite formulation based on these findings.

4. Discussion

The study's results offer initial but significant evidence that plant-based formulations have potential to reduce the levels of heavy metals in seawater taken from contaminated coastal areas. Formula 4 consistently displayed the highest removal efficiency, while all four tested formulas showed quantitative efficacy. These findings demonstrate the feasibility of using natural plant-derived compounds as biosorbents for environmental remediation. (Halder & Ghosh, 2020)

Using actual seawater samples taken from shipbreaking locations in Sitakunda, a worldwide known hotspot for marine pollution, is one of this study's strongest points. This method took into consideration the intrinsic complexity of damaged marine ecosystems, where various metals and coexisting environmental elements interact, in contrast to developing a standard heavy metal solution at the laboratory that relied on artificially generated heavy metal solutions. (Hasan et al., 2016) As such, the results reported here are not only environmentally significant but also offer a practical evaluation of the possible effectiveness of plant-based formulations in field settings. (Hasan et al., 2016)

Therefore, the present study can be considered a first attempt to determine if plant-based formulations may successfully lower heavy metal concentrations in actual environmental settings. The aim was to determine whether the strategy is effective in formulation rather than to break down the specific adsorption mechanisms. (Hasan et al., 2016) (Khan et al., 2017) The findings support this feasibility and offer an appropriate foundation for further research. The focus of future research must now shift from general efficacy to identifying the specific components and molecular mechanisms that underlie metal binding. To improve the formulations, it will be essential to determine whether a certain functional group drive adsorption. (Khan et al., 2017) These discoveries will make it potential to optimize efficiency, stability, and selectivity for various heavy metals. Designing standardized and reliable formulations that can handle contamination on a larger scale will be feasible with this molecular analysis. (Khan et al., 2017)

The ultimate goal is to develop a viable formulation that is sustainable, cost-effective, and accessible in coastal areas that are highly polluted, like Bangladesh's shipbreaking industries. Even if this ultimate goal is yet to be achieved, the current experimental evidence serves as an essential starting point. This fundamental research offers a clear path for further development and eventual translation into commercial manufacturing by demonstrating that plant-based formulations can function effectively in practical applications. (Kibria et al., 2016)

This study establishes the groundwork for future fundamental research and product development while showcasing the substantial potential of plant-derived formulations in seawater bioremediation. (Kibria et al., 2016) The results represent an important first step toward developing affordable, sustainable approaches for reducing heavy metal contamination in marine ecosystems, even though much more work needs to be done before large-scale implementation.

This study did not evaluate adsorbent regeneration, long-term stability, or the specific functional groups responsible for adsorption. FTIR and SEM analysis are needed to confirm binding mechanisms

5. Conclusion

Significant reductions in heavy metal concentrations in contaminated seawater samples from all the four sample sites were achieved by all four of the developed plant-based formulations (Formulas 1–4). Formula 4 performed most efficiently among them, removing several toxic elements, such as arsenic (As), lead (Pb), copper (Cu), cadmium (Cd), chromium (Cr), cobalt (Co), mercury (Hg), and nickel (Ni). Reductions of up to 98% for cobalt (Co) and 95% for mercury (Hg) in just 36 hours are noteworthy, demonstrating its remarkable effectiveness and promise for rapid remediation. Even though there was comparatively less nickel (Ni) removed, the steady decline in removal indicates that more optimization may increase its effectiveness.

The formulations' use of aloe vera (*Aloe barbadensis*), water hyacinth (*Eichhornia crassipes*), and false daisy (*Eclipta alba*) highlights the potential of these plant species to function as potent biological agents for environmental detoxification. These formulations are excellent candidates for large-scale bioremediation projects due to their exceptional effectiveness, especially against extremely hazardous and persistent metals like cobalt and mercury. Their sustainability, ease of usage, and natural origin all offer credibility to their promise as affordable and ecologically responsible solutions for industrial and marine pollution management. The screening of the plant species, the identification of particular tissues or biomolecules with improved metal-binding capacity, and the formulation optimization for long-term stability under various environmental conditions should be the top priority of future study.

In the end, these initiatives will aid in facilitating the development of sustainable, commercially feasible bioremediation formulation that can deal with heavy metal contamination in delicate environments.

Abbreviations

ANOVA - Analysis of Variance

As - Arsenic

BCSIR - Bangladesh council of scientific and industrial research

Cd - Cadmium

Co - Cobalt

Cr - Chromium

Cu - Copper

Hg - Mercury

ICP-MS - Inductively coupled plasma mass spectrometry

Ni - Nickel

Pb – Lead

Ppm- Parts per million

SBRI - Ship breaking and recycling industries

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Appendix: Supplementary data

Formula 1

Site 1

As 75				Pb 208			
Untreated	12h	24h	36h	Untreated	12h	24h	36h
249.64	190.57	163.26	156.22	69.18	67.48	48.62	33.25
248.93	191.15	162.76	155.76	68.45	66.76	48.12	32.93
248.15	190.05	161.45	154.76	67.96	66.02	47.96	31.85
247.86	189.26	160.98	154.12	66.76	65.86	46.93	30.88
247.15	188.33	160.24	153.14	66.55	65.01	46.15	30.13

Cd 111				Co 59			
Untreated	12 h	24h	36h	Untreated	12h	24h	36h
1.6	1.72	1.08	1.08	16.04	4.55	3.36	3.54
1.12	1.23	0.76	0.76	15.76	3.93	2.88	3.01
0.86	0.76	0.21	0.15	14.93	3.01	1.96	2.45
0.23	0.53	0.01	0.055	13.86	2.45	1.45	1.73
0.098	0.01	0.0076	0.015	13.26	1.33	1.02	1.11

Cr 52				Cu 63			
Untreated	12h	24h	36h	Untreated	12h	24h	36h
99.41	52.77	46.56	32.58	770.68	640.09	468.532	367.54
98.73	52.93	45.86	31.96	770.02	639.16	467.93	366.75

98.05	51.76	44.77	30.98		769.86	638.93	466.45	365.45
97.85	50.66	44.25	30.45		768.56	637.93	465.86	365.14
96.63	50.14	43.91	30.11		767.11	637.42	465.12	364.82

Ni 60					Hg 202			
Untreated	12h	24h	36h		Untreated	12h	24h	36h
23.28	21.07	17.83	20.84		0.42	0.181	0.083	0.109
22.76	20.76	16.75	19.86		0.21	0.086	0.0076	0.076
21.89	19.86	16.12	18.35		0.01	0.021	0.0053	0.012
20.45	18.75	15.86	18.01		0.0076	0.0098	0.0011	0.0045
20.12	18.15	14.16	17.45		0.0032	0.0045	0.0007	0.0011

Site 2

As 75					Pb 208			
Untreated	12h	24h	36h		Untreated	12h	24h	36h
258.81	104.95	104.78	103.64		241.82	538.28	378.27	342.89
257.93	103.26	103.56	102.86		240.86	537.45	377.83	341.83
256.45	102.86	102.93	101.56		239.86	536.76	376.99	340.22
255.93	101.93	101.88	100.78		238.97	535.93	376.15	340.15
255.26	101.23	101.02	100.33		238.26	535.14	375.95	339.65

Cd 111					Co 59			
Untreated	12h	24h	36h		Untreated	12h	24h	36h
7.56	4.32	18.14	6.57		13.68	2.97	2.88	2.35
6.86	3.88	17.77	5.75		12.86	2.15	1.86	2.01
5.93	2.75	16.65	4.86		11.75	1.86	0.75	1.42
4.76	1.93	15.97	3.96		10.86	0.25	0.12	0.45
4.55	1.01	15.26	3.12		10.01	0.01	0.0086	0.0075

Cr 52					Cu 63			
Untreated	12h	24h	36h		Untreated	12h	24h	36h
250.33	62.07	56.13	51.91		882.26	364.57	345.56	332.42
249.86	61.76	55.25	50.15		881.76	363.86	344.77	331.45
248.93	60.58	54.86	49.86		880.75	362.77	343.45	330.86
248.05	59.78	53.86	48.65		880.01	361.86	342.98	330.15
247.45	59.22	53.14	48.12		779.531	361.45	342.02	299.95

Hg 202					Ni 60			
Untreated	12h	24h	36h		Untreated	12h	24h	36h
0.34	0.76	0.41	0.35		23.28	51.18	30.06	27.36
0.12	0.12	0.12	0.15		22.86	50.76	29.86	26.86

0.86	0.076	0.0076	0.076		21.76	49.86	28.93	25.74
0.12	0.023	0.0045	0.015		20.98	48.76	27.86	27.98
0.75	0.0076	0.001	0.0076		20.12	48.26	27.25	24.36

Site 3

As 75					Pb 208			
Untreated	12h	24h	36h		Untreated	12h	24h	36h
199.29	184.29	183.36	199.1		87.49	109.97	79.19	103.56
198.76	183.86	182.88	198.85		86.74	108.75	78.86	102.76
197.56	182.45	181.75	197.26		85.55	107.95	77.57	101.75
196.89	181.93	180.74	196.89		87.86	107.09	76.93	101.12
196.23	181.12	180.26	196.36		84.26	106.965	76.26	100.92

Cd 111					Co 59			
Untreated	12h	24h	36h		Untreated	12h	24h	36h
0.64	1.75	0.59	1.48		10.95	3.01	3.03	3.57
0.23	1.23	0.12	1.01		9.75	2.76	2.76	2.46
0.076	0.75	0.076	0.75		8.69	1.36	1.83	1.76
0.045	0.01	0.023	0.12		7.86	1.01	1.06	0.87
0.087	0.007	0.006	0.0076		6.15	0.76	0.769	0.01

Cr 52					Cu 63			
Untreated	12h	24h	36h		Untreated	12h	24h	36h
101.96	45.85	64.58	70.62		524.31	388.759	451.89	571.29
100.86	44.26	63.76	69.86		523.12	387.59	450.76	570.86
99.75	43.86	62.45	68.75		522.86	386.45	449.76	569.56
98.93	42.98	61.99	67.86		521.87	385.68	448.69	568.96
97.26	42.12	61.23	67.26		520.16	385.06	447.96	568.03

Ni 60					Hg 202			
Untreated	12h	24h	36h		Untreated	12h	24h	36h
17.95	15.03	15.01	14.76		0.57	0.082	0.092	0.47
16.86	14.86	14.856	13.75		0.21	0.0075	0.01	0.02
15.76	13.76	13.74	12.89		0.066	0.0021	0.007	0.008
14.86	12.86	12.77	12.76		0.023	0.0008	0.0021	0.0056
14.01	12.13	12.01	11.26		0.0087	0.0002	0.00056	0.0022

Site 4

As 75				Pb 208			
Untreated	12h	24h	36h	Untreated	12h	24h	36h
236.42	213.31	207.26	213.83	47.36	64.46	79.31	64.41
235.76	212.89	206.86	212.86	46.86	65.26	78.89	63.86
234.86	211.75	205.76	211.75	45.36	63.86	77.45	62.87
233.98	210.99	204.99	210.86	44.93	62.98	76.83	61.98
233.21	210.12	204.26	210.36	43.56	61.24	76.56	61.46

Cd 111				Co 59			
Untreated	12h	24h	36h	Untreated	12h	24h	36h
5.19	5.93	5.06	6.24	12.71	3.45	3.62	3.02
4.86	4.75	4.836	5.75	11.86	2.86	2.87	2.15
3.77	3.65	3.56	4.68	10.75	1.76	1.69	1.87
2.84	2.93	2.83	3.93	9.98	1.65	1.02	1.03
2.21	2.36	2.26	3.12	9.26	1.25	0.75	0.56

Cr 52				Cu 63			
Untreated	12h	24h	36h	Untreated	12h	24h	36h
96.2	55.39	56.06	63.04	752.54	579.96	605.48	684.368
95.24	54.86	55.85	62.86	751.75	578.86	604.86	683.87
94.86	53.84	54.75	61.74	750.76	577.56	603.45	682.74
93.93	52.93	53.89	60.86	749.69	576.93	602.99	681.96
93.06	52.12	53.26	60.12	749.06	576.26	602.41	681.03

Ni 60				Hg 202			
Untreated	12h	24h	36h	Untreated	12h	24h	36h
20.48	19.41	17.39	17.98	0.4	0.071	0.057	0.028
19.88	18.69	16.85	16.87	0.1	0.023	0.012	0.0075
18.75	17.57	15.46	15.85	0.075	0.0075	0.0075	0.0033
17.69	19.73	14.99	14.86	0.032	0.0032	0.0023	0.0001
16.93	16.23	14.23	14.12	0.001	0.0012	0.0001	0.00007

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Authors contributions

Dr. Mohammad Nazir Hossain (MNH) made the hypothesis, designed the experiments, supervised the work, analyzed the data, and wrote and revised the final version of the manuscript; Sadia Farzana (SF) conducted the experiments, analyzed the data, and wrote the initial draft of the manuscript; Paritosh Barai (PB), Tamim Hossain (TH), Ethneen Mostafa (EM) and Khadiza Rezwana (KH) carried out some experiments, and Sadia Farzana (SF) did manuscript editing.

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Competing interests

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Informed consent

Obtained.

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The data that support the findings of this study are available on request from the corresponding author. The data are not publicly available due to privacy or ethical restrictions.

Data sharing statement

No additional data are available.

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