POTENTIAL ENVIRONMENTAL AND HUMAN HEALTH RISKS CAUSED BY HEAVY METALS AND PATHOGENS FROM ILLEGAL LANDFILL SITES IN BOSNIA AND HERZEGOVINA

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ABSTRACT

In Bosnia and Herzegovina, several illegal waste dumps pose a significant threat to soil and water contamination. The aim of this study was to determine the levels of heavy metal contamination of soils and microbiological contamination of water near investigated landfills. The goal was to identify the harmful effects of illegal landfills on the environment and public health, as well as assessing the potential of contaminants to pollute soil and water resources. Using an Atomic Absorption Spectrometer, the amounts and distribution of four heavy metals (Cd, Co, Pb, and Cr) in soil at four illegal landfills in Central Bosnia were studied. Three water samples collected near the dumpsites studied were tested for microbes. Bacteriological analyses of water included determination of total mesophilic aerobic bacteria, aerobic heterotrophic bacteria, total coliform and fecal coliform bacteria and fecal enterococci. Quantitative results were analyzed using an analysis of variance and Tukey HSD post hoc test. Concentrations of Cd (4.96 mg/kg) and Pb (206.97 mg/kg) recorded in soil at particular sites were above the limits of maximum allowable concentration. Cr and Pb values in soil samples were relatively higher on average than that of Cd and Co. Enrichment factor and pollution load index indicated high concentration of heavy metals in soil, especially Cd. Based on these results, waste from illegal landfills may release considerable amounts of harmful metals and microbes into the environment.

Keywords: heavy metals; illegal landfills; pathogens; pollution

Introduction

Rapid population growth, intensive industrialization and urbanization have adversely affected the environment, primarily in terms of pollution of soil and water. The production of solid waste is increasing rapidly and is an important environmental problem worldwide (Williams and Hakam 2016). This has resulted in a dramatic increase in waste and numerous, often illegal and uncontrolled landfills (Biotto et al. 2009). Soil contamination caused by human activity is damaging the environment and ecosystems around the world (Bai et al. 2013; Adamcová et al. 2016; Chen et al. 2016). Studies indicate that exposure to toxic chemicals, such as heavy metals in waste or contaminated water is likely to adversely affect human health (Agusa et al. 2003; Nguyen et al. 2003).

The recent contamination of soil with heavy metals is of major global concern (Dahija et al. 2019). Lead, chromium, arsenic, zinc, cadmium, copper, mercury and nickel are the most common heavy metals that contaminate soil worldwide. Heavy metals are the dominant causes of environmental pollution due to the method of production, toxicity, possibility of uptake by plants and inclusion in the human food chain (Dumitrel et al. 2013). Accumulations of heavy metals are highly toxic for biological organisms, such as humans, animals, microorganisms and plants (Njagi et al. 2016).

In addition to these chemical contaminants, landfills are of major concern because they might contain viable pathogenic microorganisms or their toxins that are known to cause disease in animals or humans (Anand et al. 2021). The decomposition of food waste can also result in the production of organic leachate that can significantly change the composition of bacteria and initiate the growth of pathogenic bacteria such as *Salmonella*, *Pseudomonas*, *Enterobacteria* and *Clostridium perfringens* (Wu et al. 2018), which could be dispersed in the environment via water and wind.

Unfortunately, there is less and less time to develop an integrated waste management system in Bosnia and Herzegovina (B&H), as large amounts of waste are generated every day and landfills are the primary way of disposing of waste. Moreover, this is occurring in unregulated landfills, which is likely to have a long-lasting effect on human health and the environment. Thus, the objective of this study was to determine the concentrations of lead (Pb), chromium (Cr), cadmium (Cd), copper (Cu) in soil from four illegal landfills in Central Bosnia and determine the degree of microbiological contamination of water close to these sites.

Materials and Methods

Description of the study area

The area studied included four illegal landfills sites, namely: "Cipalo" (S1), "Dolovi" (S2), "Malkin Most" (S3) and "Paljike-Hum" (S4) and is located in Central Bosnia, Bosnia and Herzegovina (Fig. 1). The most common content of these waste dumps is domestic and household waste and construction material and they are

Bešta-Gajević, R., Gajević, M., Pilić, S., Adžaip, Z., Đug, S., Dahija, S.: Potential environmental and human health risks caused by heavy metals and pathogens from illegal landfill sites in Bosnia and Herzegovina
European Journal of Environmental Sciences, Vol. 12, No. 2, pp. 74–79
https://doi.org/10.14712/23361964.2022.8

close to streams. These landfills have an area of 4,355 m^2. A map of the area studied is shown in Fig. 1. Water samples were collected from “Cipalo”, “Dolovi” and “Malkin most”, but not “Paljike-Hum”, as there are no nearby streams there.

Collection of soil samples

At each of the sites, three quadrats were marked out. In each quadrat, four core soil samples were collected randomly at depths of from 0–20 cm using a soil auger and mixed together to form a composite sample. Three soil samples were collected at each site, making a total of 12 soil samples from the area studied. The control samples (S5) were collected 700 m away from each dumpsite.

At each site, the surface debris was removed and a sample of soil collected using a clean shovel. The soil samples were transferred to clean zip-locked plastic bags, labelled carefully and brought to the laboratory for further treatment and analysis. The samples were mixed, gently homogenized and sieved through a 2-mm-mesh sieve to remove any coarse components. The samples were first air-dried at room temperature, then placed in an electric oven at a temperature of 40 °C for approximately for 30 minutes. The resulting fine powder was stored at room temperature.

Heavy metals analysis

Approximately, 0.25 g of the dry soil was placed in a Teflon vessel (100 ml) to which 6 ml HCL (Merk 30%), 2 ml of HNO_3 (65% Merck) and 2 ml of HF (Merk 40%) were added and then mineralized using a Milestone, Start D microwave digestion system. After 26 minutes of digestion, samples were cooled for 30 minutes and diluted to 25 ml with distilled deionized water. A blank digest was carried out in the same way. The final solutions were subsequently analyzed for Cd, Co, Pb and Cr by flame atomic absorption spectroscopy (FAAS) using Atomic Absorption Spectrometer Agilent AA 220. The metal concentration is reported as mg/kg dry weight.

Methods of assessment of contamination of soils

We used the geoaccumulation index (I_{geo}), enrichment factor (EF) and pollution load (PL) to assess the degree of heavy metal contamination at each site.

The index of geoaccumulation (I_{geo}) is widely used for assessing contamination by comparing the levels

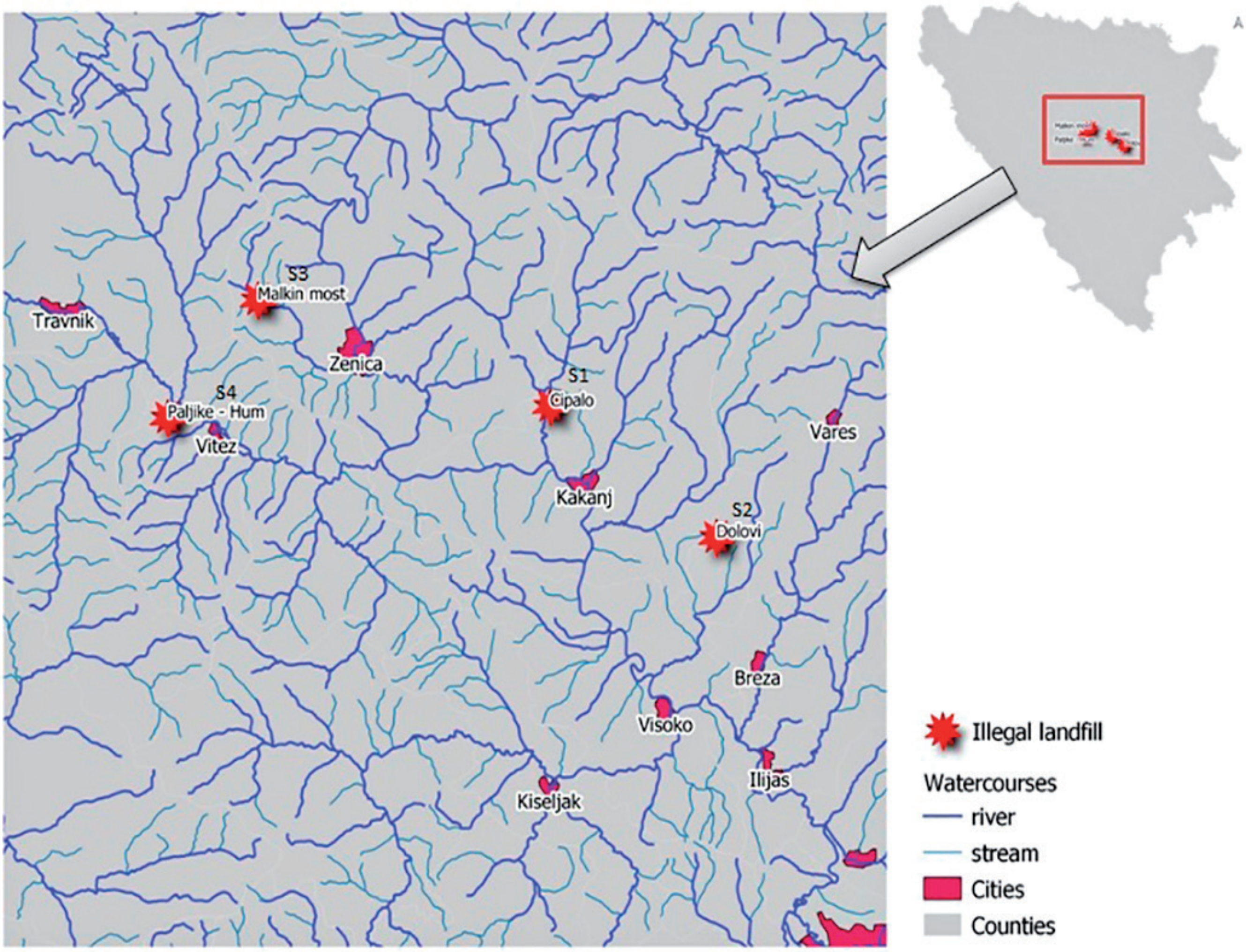

Fig. 1 A map of the area studied: “Cipalo” (S1), “Dolovi” (S2), “Malkin Most” (S3) and “Paljike-Hum” (S4).

of heavy metals with background levels (Muller 1969; Atiemo et al. 2011). Geoaccumulation index was calculated using the equation:

$$I_{geo} = log_2\left(\frac{C_n}{1.5B_n}\right) \quad (1)$$

where C_n is the measured concentration of the each element studied, and B_n is the geochemical background value of the element in fossil clay sediment (shale). If the value of I_{geo} is: < 0 = the soil is practically unpolluted, 0–1 = unpolluted to moderately polluted, 1–2 = moderately polluted, 2–3 = moderately to strongly polluted, 3–4 = strongly polluted, 4–5 = strongly to extremely polluted and > 5 = extremely polluted (Lu et al. 2009).

The enrichment factor (EF) of an element in soil samples are based on standardization of the measured element relative to the reference element. Mangan was used as a reference element in this study. The enrichment factor was calculated using the equation:

$$EF_x = \frac{(C_x/C_{ref})\, sample}{(B_x/B_{ref})\, Background} \quad (2)$$

where C_x is the concentration of the element of interest and C_{ref} represents the concentration of the reference element for normalization, B_x is the concentration of the element in the crust and B_{ref} is the concentration of the reference element used for normalization in the cortex (Ato et al. 2010). Based on enrichment, we distinguish five categories of contamination: $EF < 2$ = deficiency to minimal enrichment; EF = 2–5 = moderate enrichment; EF = 5–20 = significant enrichment; EF = 20–40 = very high enrichment; and $EF > 40$ = extremely high enrichment (Yongming et al. 2006).

The pollution index (PI) was defined as the ratio of the concentration of elements and the background content of the abundance of chemical elements in the continental crust. This index is often used to assess environmental pollution (dos Anjos et al. 2000). PLI is calculated as a geometric average of PI based on the following formula:

$$PLI = \sqrt[n]{PI_1 \times PI_2 \times PI_3 \times \ldots PI_n} \quad (3)$$

The pollution index for each heavy metal in this paper was rated as low ($PI \leq 1$), medium ($1 < PI \leq 3$) or high ($PI > 3$) according to Chen et al. (2005).

In this paper, for the assessment of soil pollution by Cd, Co, Pb and Cr we used a table in which are listed the maximum allowable concentrations of heavy metals in soil according to standard regulatory bodies such as World Health Organization, Food and Agricultural Organization and Standard guidelines in Europe.

Microbiological analysis

Water sampling was carried out according to standard methods for the examination of water and wastewater (APHA 2005). Surface water samples were collected in pre-sterilized polypropylene bottles, each of 1 l capacity. After collection, samples were transported at 4 °C prior to analysis. The water was collected from three sites in the immediate surroundings below the waste dumps. The total numbers of aerobic mesophilic bacteria and heterotrophic bacteria were determined using the serial dilution method. The results were expressed in terms of colony-forming units, (cfu)/ml. Furthermore, water samples were analysed for total coliforms and fecal coliforms and intestinal enterococci using the membrane filtration (MF) technique and 0.45 mm pore-size membrane filters (Millipore Corp., Berdford, MA). The results were expressed in colony-forming units (cfu) per 100 ml.

Statistical analysis

Experimental data were presented as mean values ± standard deviation (S.D.). To verify the statistical significance of the difference between various treatments, the data were analysed using a one-way analysis of variance and Tukey HSD post hoc test ($p < 0.05$). All statistical analyses were carried out using Statistica 10 software package (StatSoft. Inc).

Results and Discussion

Heavy metal concentration of soils at the different sites in the area studied

The mean concentrations of heavy metals (Pb, Cu, Cd and Co) recorded at landfill sites in Bosnia and Herzegovina are given in Table 1. Analysis of variance (ANOVA) revealed a significant ($p < 0.05$) variation in the concentrations of the four elements at the different sites (Table 1) and indicate the extent of metal pollution of the soils. The Tukey test revealed that the concentrations of lead were significantly ($p < 0.01$) higher at waste site (S4) than the other sites (Table 1).

The concentration of cadmium (Cd) ranged from 2.7 mg/kg at S2 to 4.96 mg/kg at S4, with a mean value of 4.04 mg/kg. The mean concentrations of Cd recorded in soil from S1, S3 and S4 sites were above the limit set by WHO/FAO (2001) of 3 mg/kg. However, the value for the soil from site S2 was 2.70 mg/kg, which is below the recommended limit. Concentrations of cadmium in the environment are constantly increasing due to continuous anthropogenic mobilization around the world, which is of great concern. According to Zheng et al. (2021) a large amount of Cd is discharged into the environment, greatly endangering the stability of the ecological environment and human health. Cd is an extremely toxic element that exhibits various toxic effects, including nephrotoxicity, carcinogenicity, teratogenicity and endocrine and reproductive toxicity (Wuana and Okieimen 2011; Genchi et al. 2020).

According to our results, high concentrations of lead (Pb) were recorded at all the sites. The concentrations

Table 1 Summary of heavy metal mean concentration in soils at different sites in Bosnia and Herzegovina.

Heavy metal (mg/kg)	Sample sites				
	S1	S2	S3	S4	S5
Cadmium (Cd)	4.80 ± 0.30a	2.70 ± 0.10b	3.74 ± 0.05c	4.96 ± 0.55a	2.45 ± 0.07b
Lead (Pb)	31.18 ± 0.68d	42.48 ± 0.61c	51.68 ± 0.58b	206.97 ± 0.47a	12.36 ± 0.55e
Chromium (Cr)	31.97 ± 0.38c	37.82 ± 0.73b	52.62 ± 0.94a	51.59 ± 1.42a	12.99 ± 0.58d
Cobalt (Co)	20.26 ± 1.21a	27.19 ± 0.70b	19.25 ± 0.43a	15.46 ± 0.57c	7.04 ± 0.22d

Notes: Values are means (± SD) of three replicates, S1 = Cipolo, S2 = Dolovi, S3 = Malkin most, S4 = Paljike-Hum, S5 = control. Means followed by a different letter(s) in the same row differ significantly (P = 0.05) according to the Tukey's multiple range test.

ranged from 31.18 to 206.97 mg/kg, with a mean value of 83.07 mg/kg. The highest mean value was recorded at site (S4) and the lowest at S1. ANOVA revealed significant differences ($p < 0.05$) in the mean concentration of Pb among the sites. The mean concentration of Pb at S4 was above the WHO/FAO (2001) permissible limit for soils of 100.00 mg/kg. As the worldwide Pb concentration for surface soil averages 32 mg/kg and ranges from 10 to 67 mg/kg (Pendias and Pendias 2001; Kinuthia 2020) the levels recorded at the sites studied are above this limit.

Lead and cadmium are generally considered the most toxic to humans and animals. In addition to exposure to lead through the food chain, ingestion of soil and dust can also be an important source (Kinuthia 2020). The highest concentration of Pb recorded at site (S4) can be attributed to the burning of waste such as refrigerators, cables, car tyres, batteries, air-conditioners etc. This study revealed that the concentration of chromium (Cr) in the soil ranged from 31.97 mg/kg at S1 to 51.59 mg/kg at S4, with a mean value of 41.48 mg/kg. The high concentration of Cr recorded might also be a result of the waste including refrigerators, computers, cables, printers, photocopying machines, car tyres and batteries. In landfills in the USA, Cr is the third-largest pollutant and the second largest among the inorganic pollutants, after Pb (Vodyanitskii 2016). According to Kinuthia et al. (2020) hexavalent Chromium compounds, including chromates of Ca, Zn, Sr and Pb, are highly soluble in water, toxic and carcinogenic. Similarly, the concentration of cobalt ranged from 15.46 at S4, to 27.19 at S1, with a mean value of 20.54 mg/kg. However, waste site (S2) had the highest and S4 the lowest mean concentration of Co. Factors that affect cobalt speciation in soil and sediments include their nature, the concentration of chelating and complexing agents, pH and the redox potential.

High levels of these metals in the soil at the landfills studied may be associated with anthropogenic impact. The different types of waste disposed of in these landfills over the years are most likely the main sources of high concentrations of these metals. All of the concentrations of heavy metals except cadmium recorded at the sites studied differed significantly from those recorded in the control. This is in line with the results of similar studies (Ogundiran and Osibanjo 2008; Cortez and Ching 2014; Murtić et al. 2021).

The results of the I_{geo} analysis indicate that S1, S2 and S3 were practically unpolluted with Pb, Cr and Co (Table 2). This may be attributed to the horizontality of the spread of metallic pollutants in these areas and the absence of e-waste. Moreover, these sites were strongly to extremely heavily contaminated with Cd. These findings corroborate the report of Pradhan and Kumar (2014) and Fossu-Mensah et al. (2017) that high concentrations of cadmium and other heavy metals may be due to e-waste. S4 was practically uncontaminated with Cr and Co, but extremely heavily contaminated with Cd and moderately to strongly contamination with Pb.

Table 2 Recorded values of Geoaccumation index I_{geo}.

Heavy metal	S1	S2	S3	S4
Cd	5.029146	4.199071	4.670307	5.076452
Pb	0.055797	0.501821	0.784644	2.786387
Cr	−0.7156	−0.47317	0.003458	−0.05347
Co	0.434099	0.858432	0.359896	0.043578

The enrichment Factor (EF) and pollution load index for all the heavy metals recorded in the soil at the different sites are presented in Table 3. Soil from S1 and S4 was extremely highly enriched with Cd, whereas S1 was minimally enriched with Pb, Cr and Co, and L4 significantly enriched with Pb and minimally so with Cr and Co.

Similarly, S2 and S3 were highly enriched with Cd, while S2 was moderately enriched with Co and minimally deficient in Pb and Cr. A comparison of the results for the different sites in terms of the EFs revealed that S1 and S4 were the most contaminated. However, the study also revealed that the concentration of cadmium (Cd) present in soil at all the sites was extremely high.

The PLI values recorded for the sites were in the high category, with PLI > 3. According to Chen et al. (2005) this means that the concentration of these heavy metals is high. The highest value of PLI was recorded for site 4, which was 5.84. These high values of PLI indicate the soils at the sites studied are heavily contaminated with heavy metals (Varol 2011).

Table 3 Results recorded for the Enrichment factor and PLI.

Heavy metal	S1	S2	S3	S4
Cd	41.5	23.48	32.67	42.89
Pb	1.32	1.8	2.19	8.76
Cr	0.77	0.91	1.27	1.22
Co	1.71	2.3	1.63	1.31
PLI	3.44	3.61	4.11	5.84

Microbiological analysis

The results of microbiological examination of water samples from S1, S2 and S4 are in Table 4. The total number of aerobic mesophilic bacteria is expressed in terms of cfu/ml and this group includes opportunistic pathogens. Based on the maximum values, S3 stands out with 1168 cfu. Concentrations of heterotrophic bacteria, which usually correspond to organic matter contamination (Kamjunke et al. 2020) ranged from 34 cfu/ml at S2 to 4458 at S3.

Furthermore, results in Table 4 indicate that total coliforms varied between 28 cfu/100 ml at S2 to 3566 at S4. Total coliforms indicate the general sanitary condition of water, since this group includes bacteria of fecal origin resulting from the disposal of animal waste, sewage, land and urban waste and domestic wastewater. Mean values of fecal coliforms ranged from 6 cfu/100 ml at S1 to 2028 at S3 as indicated in Table 4. The numerous fecal coliforms recorded at S3 may be due to landfill runoff and solid waste containing fecal material of domestic animals and humans. Similar observations are recorded by Nartey et al. (2012) who determined the effect of solid waste dumpsites on surface water systems. In our study, intestinal enterococci in water varied from 3 cfu/100 ml at S1 to 266 at S3 and the greatest fecal pollution was recorded in the watercourse located near S3. However, the water sample for S1 was less contaminated. Numerous epidemiological studies in different aquatic environments have shown a relationship between coliform counts and incidence of infectious diseases in humans (Lugo et al. 2021). The results of our microbiological analyses are in line with the findings of Pepper et al. (2006) that municipal solid waste may contain a variety of pathogens.

Conclusions

In conclusion, we found that soil samples from illegal landfills in Bosnia and Herzegovina contained heavy metals. Thus, there is an urgent need for banning illegal landfills in B&H in order to minimize environmental pollution as the results indicate that this waste contains significant levels of toxic metals and microorganisms that could pollute the environment. Future research should also concentrate on improving the efficiency of removing these pollutants by using environmentally efficient and cost-effective technologies such as bioremediation in order to reduce the potential adverse effect of this waste on the environment.

Acknowledgements

The authors are thankful to the Environmental Fund of the Federation of Bosnia and Herzegovina and in part the Federal Ministry of Education and Science and Institute for Biomedical Diagnostic and Research “Genom”, Travnik.

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Table 4 The results of the microbiological analysis of water samples.

Microorganisms	Sample sites		
	S1	S2	S3
Aerobic mesophilic bacteria	18 ± 1.732b	7.3 ± 1.1547c	1168.33 ± 7.6376a
Aerobic heterotrophic bacteria	169.333 ± 2.082b	34.666 ± 2.309c	4458 ± 7.000a
Total coliform bacteria	349.333 ± 8.021b	27.666 ± 1.528c	3566.333 ± 5.508a
Fecal coliform bacteria	6.333 ± 1.155b	7.333 ± 1.155b	2028.666 ± 3.215a
Intestinal enterococci	2.333 ± 1.1547c	24.000 ± 2.645b	266.666 ± 2.516a

Notes: Values are means (± SD) of three replicates, S1 = Cipolo, S2 = Dolovi, S3 = Malkin most. Means followed by different letter(s) in the same row differ significantly (p = 0.05) according to the Tukey’s multiple range test.

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