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ARTICLE

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Journal of Environmental Chemistry and Ecotoxicology

Full Length Research Paper

Sulphur and heavy metals contents in soils and *Grewia* bicolor leaves around the Selibe Pikwe Cu-Ni mine (BCL), Botswana

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This study investigated environmental pollution due to mining activities at the BCL mine in Selibe-Phikwe town, Botswana and assessed the level of heavy metals and sulphur in soil and *Grewia bicolor* leaves adjacent to the mine. Soil and *G. bicolor* were contaminated with heavy metals on the downwind side, where also concentrations of sulphur in the soil and *G. bicolor* were found to be elevated when compared to the upwind side. The leaves samples, were significantly difference at P = 0.00 and soil were significantly different at P = 0.03 for both west and east side. A mean soil pH of 4.12 in the prevailing wind direction suggested that atmospheric deposition from the mine has caused acidification. With further distance from the mine concentration in soil and leaves appeared to reflect a decreasing deposition of heavy metals and sulphur. Elevated concentration of heavy metals in leaves represent a high risk of bioaccumulation and biomagnifications in the food chain because *G. bicolor* leaves are grazed on by farmed animals and are common ingredients of traditional medicines.

Key words: BCL, heavy metals, Grewia bicolor, sulphur, pH, environmental pollution

INTRODUCTION

Pollution due to mining activities is a great concern throughout the world since it has been recognized as a major contributor to environmental pollution over the past years (Zhou et al., 2007). Mining and smelting affect the air, terrestrial and aquatic ecosystems through discharge of solid and liquid waste, as well as particulate matter (Nagajyoti et al., 2010). Developing countries which are rich in minerals like Botswana are at a greater risk from mining pollution. Mining of minerals is associated with benefits such as economic development (Rashed, 2010), increased employment and foreign exchange reserves.

Notwithstanding the considerable economic benefits of mining, it should also be noted that mines have the potential to leave a long lasting legacy for the environment as evidenced by case studies in Sudbury, Canada and Kola Peninsula in Russia (Whitby et al., 1976; Poikolainen, 1997).

The presence of heavy metals in soils may pose risk and hazards to humans and the ecosystem through: direct ingestion or contact with contaminated soil, the food chain, drinking of contaminated water (Candeias et al., 2011), reduction in food quality via phytotoxicity,

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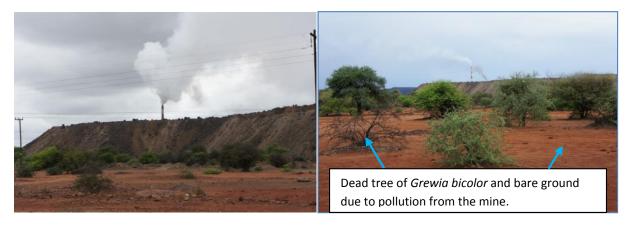


Figure 1. Mining pollution and its environmental impact from the BCL mine in Selibe-Phikwe (Western Side).

reduction in land usability for agricultural production causing food insecurity and land tenure problems (Wuana and Okieimen, 2011).

Although the extraction of metals such as copper-nickel is necessary for the economic or industrial development of societies (Rashed, 2010), their significant risk to the environment comes from the disposal of mining waste (Figure 1). If the mine waste is not properly disposed, it can contribute immensely to heavy metal pollution in the surrounding environment, which can result in the destruction of ecological landscape, ground water pollution and decrease in biological diversity (Sheoran et al., 2011; Rashed, 2010; Kiikkila, 2003).

Copper-Nickel mines releases not only potential toxic metals but also other particulate matter and gaseous materials such as dust, SO₂, CO, CO₂, N₂O, NO₂ and NO in the atmosphere (Nagajyoti et al., 2010). This can have grave consequences for the ecosystem as evidenced by the forests devastation and reduced diversity of vascular plants in Sudbury, Canada (Nkongolo et al., 2008).

The threat that heavy metals pose to humans and animal health is aggravated by their long term persistence in the environment (Yoon et al., 2006). Heavy metals such as Lead and Cadmium are non-biodegradable and have no known biological function. Copper, Zinc and Nickel are source of nutrients to plants at low concentrations but they become toxic at elevated concentrations.

Heavy metals tend to bioaccumulate and biomagnify in animal tissues. Since domestic animals such as cattle and goats do graze in the surrounding area of the BCL mine, they are likely to ingest soil and plants material contaminated with heavy metals which in turn can pose a risk to people consuming animal meat. Cadmium and Lead tend to accumulate in the liver and kidneys of animals (Kirkham, 2006; Wilkinson et al., 2003). These tissues are regarded as delicacies in Southern Africa (Ikenaka et al., 2010; Yabe et al., 2011) and their consumption may pose a risk in contaminated areas.

The Bamangwato Concessions Limited mine (BCL) at Selibe Phikwe is rich in copper and nickel deposits. The main concern with this particular mining and smelting operations is the release of sulphur dioxide emissions into the atmosphere which can damage the environment through acidification. Another concern is the significant volumes of wastes that are deposited on the soil which eventually affect plants and animals through exposure to elements contained in the residue (Figure 1).

Given the environmental impact caused by sulphur dioxide in the adjacent area of the BCL mine, the objective of the present study is to asses sulphur and heavy metals content in soil and *Grewia bicolor* leaves along a 4 km west and east transect in BCL mine. The soil pH of the area around the latter will also be investigated to further demonstrate the impact of mining on the environment.

MATERIALS AND METHODS

Study site

The study site is located in Selibe Phikwe which is about 400km north from the Capital city Gaborone in eastern Botswana (Figure 2). The area is known for mining and smelting of copper and nickel since 1960's but full operation started in 1973. The BCL mine is the most important anthropogenic polluter in the town and the country. A lot of sulphur dioxide is pumped directly into the atmosphere through a 153 m stack.

The soils are classified as Eutric Regosols and Haplic Luvisols (Dept of Surveys and Mapping, Botswana). The major vegetation is of hardveld dominated by *Colophospermum mopane*. The mean annual rainfall is 415 mm and maximum daily temperature is 31.6°C. The prevailing wind comes from south east and east of the town according to the 2000-2006 wind rose data, Dept of Meteorology (Figure 3).

Sampling

The soil and plants were sampled in mid February 2012. Samples were collected along a 4km transect running from the western

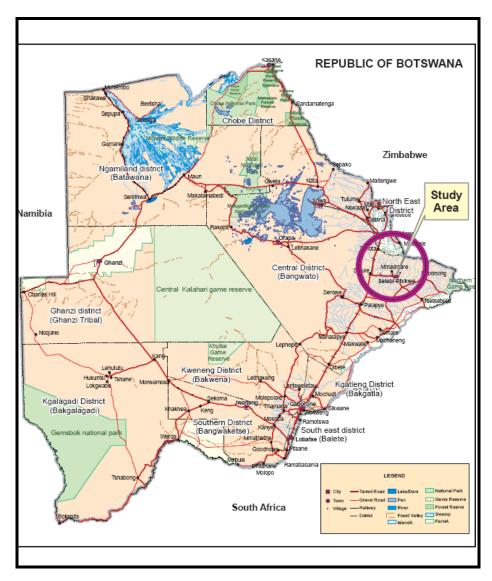


Figure 2. Map showing the study area in Selibe Phikwe (BCL mine), Botswana (Department of Surveys and Mapping).

(downwind of the mine) and eastern part of the BCL mine in Selibe Phikwe. The geographical coordinates for all sampled plots were taken by a Global Position System (Garmin GPS 76). The sampling plots (10 m x 10 m) were made every 0.5 km along each transect. Three soil samples were collected by a soil auger at a depth of 0-10 cm. They were then combined into one composite soil sample. Two composite soil samples were taken per plot and a total of thirty two soil samples were collected in the study area. Mature and healthy leaves of *G. bicolor* were randomly picked around each plant at breast height (1.4 m).

Sample preparation

Soil samples were air dried in the laboratory before chemical analysis was performed. After two weeks of air drying, the samples were sieved in a <2 mm stainless steel sieve. *G. bicolor* leaves (unwashed) were dried in an oven at a temperature of 70°C for 24 h. After the leaves were dried, they were ground to powder with

Fritsch pulverisette five machines. The pulverised leaves were stored in air tight plastic bottles until they were analysed.

Chemical analysis

For the determination of the total soil heavy metal concentration, the homogenized sample, about 0.500 g±0.05 g was digested in a closed microwave vessel system (Milestone Ethos EZ, equipped with Mutliprep-41 rotor) according to USEPA 3051 Method. Standard soil reference materials from Institute of Geological Sciences Japan (JSO-1) were used for the quality control and to verify the accuracy of the metal analysis.

To determine heavy metals in plants, 0.500 g±0.05 g plant materials was weighed into porcelain crucibles and incinerated in a muffle furnace (Thermolyne, Model F6000, USA) at a temperature of 550°C for 6 h until a white ash was obtained. Before loading with plant samples the furnace was conditioned for 2 h at a temperature of 800°C. After incineration, the ash was dissolved in 10 ml diluted

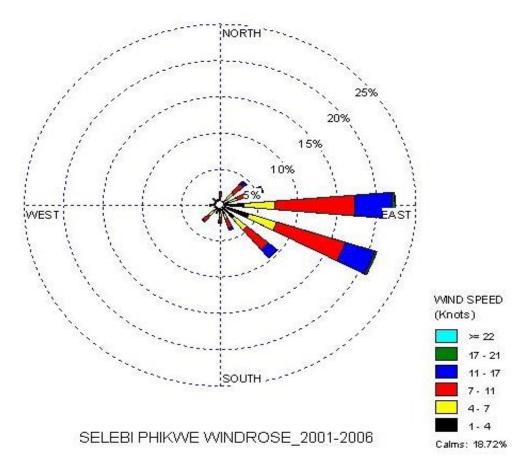


Figure 3. Prevailing wind direction in Selibe Phikwe from 2001-2006 (Department of Meteorology, Botswana).

hydrochloric acid (6N) and made up to a volume of 50 ml before being analysed for heavy metals with AAS. Reference material and blanks were included with the samples as a quality control measure. While analysing samples, a standard was analysed after every ten samples to monitor the instrument drift from the initial calibration. AAS analysis was performed in triplicates and reslope standard was performed to update the calibration during analysis.

An Atomic Absorption Spectrometer (Perkin Elmer 3110 with S10 auto sampler) was calibrated using quality standards from Merck Chemicals, German. Samples were performed under an air and acetylene flame gas mixture. Lead and Cadmium were determined by a Varian 220FS AAS.

Sulphur in soil and plant samples was analysed using a Leco Sulphur analyzer (S-144DR). A sample of 0.350 g was subjected to a temperature of 1350°C in an oxygen rich environment. The instrument was calibrated with appropriate Leco standards and samples were run in duplicate inclusive of reference material STD-2 and Tobacco leaves from Leco Corporation.

The soil pH was determined according to Soil Sampling and Methods of Analysis (Carter, 1993). The samples were run in duplicate.

Data analysis

The data was calculated using SPSS Statistical software version 19 and graphs were performed with Ms Excel 2010. Wilcoxon sign rank test was used to find out if there is any significant different

between samples from the west and east (Moreno-Jimenez et al., 2009). Spearman's rank correlation was used to determine the strength of correlation between different parameters in *G. bicolor* leaves and soil in the west and east transect respectively (Gauthier, 2001; Ogunkunle and Fatoba, 2014).

RESULTS AND DISCUSSION

Heavy metals and sulphur concentration in soil

Previous research has shown that soils around Copper-Nickel mines tend to have high concentrations of heavy metals (Nkongolo et al., 2008; Meadows and Watmough, 2012) especially Cu, Ni and Pb. In most cases, the downwind side is the most affected as depicted by the mean concentrations of heavy metals in Tables 3 and 5. Concentrations of heavy metals in soil and plants from the upwind and downwind sides were significantly different at P=0.00 and P=0.03, respectively.

Figures 4 and 5 further depicts an elevation of heavy metals concentration in the western side of the mine whereas it is opposite with the upwind side. This increase could be a result of dust particles emanating from mine waste, crushed stones and fumes from the smelter which

Table 1. Correlation coefficients (R^2) of each element in *G. bicolor* leaves.

	Zn	Cu	Ni	Cd	Pb	S
Zn	1					
Cu	-0.060	1				
Ni	-0.026	0.805**	1			
Cd	0.176	0.321	0.461**	1		
Pb	0.021	0.549**	0.631**	0.345	1	
S	-0.078	0.579**	0.569**	0.229	0.017	1

Correlation is significant at **0.01 and *0.05 level (2-tailored).

Table 2. Correlation coefficients (R²) of each element in soil.

	Zn	Cu	Ni	Cd	Pb	S
Zn	1					
Cu	-0.060	1				
Ni	-0.026	0.805**	1			
Cd	0.176	0.321	0.461**	1		
Pb	0.021	0.549**	0.631**	0.345	1	
S	-0.078	0.579**	0.569**	0.229	0.017	1

Correlation is significant at **0.01 and *0.05 level (2-tailored).

Table 3. Mean (±S.D) concentration of heavy metals & sulphur in G. bicolor leaves – West.

Distance (km)	Zn (µg/g)	Cu (µg/g)	Ni (μg/g)	Cd (µg/g)	Pb (μg/g)	S (%)
0.5	1.97±0.01	41.18±1.24	29.41±1.91	3.46±0.06	7.82±0.00	1.21±0.00
1.0	1.68±0.06	46.59±0.83	27.10±2.38	1.32±0.00	7.54±0.06	1.04±0.08
1.5	2.22±0.03	35.01±0.95	15.28±2.92	1.14±0.00	7.18±0.51	0.85±0.51
2.0	2.17±0.04	24.58±0.82	18.20±9.05	1.05±0.01	7.08±1.19	0.35±0.00
2.5	0.95±0.18	26.60±0.05	16.00±1.41	0.92±0.11	8.33±0.86	0.38±0.00
3.0	1.83±0.07	17.72±0.40	14.64±2.40	0.91±0.01	5.58±1.27	0.80±0.16
3.5	1.46±0.40	7.51±0.07	16.36±5.94	0.98±0.11	2.29±2.05	1.30±0.00
4.0	2.20±0.23	9.23±0.44	9.83±2.53	0.95±0.07	4.38±1.07	1.12±0.10

Table 4. Mean (±S.D) concentration of heavy metals & sulphur in *G. bicolor* leaves – East.

Distance (km)	Zn (μg/g)	Cu (µg/g)	Ni (μg/g)	Cd (µg/g)	Pb (μg/g)	S (%)
0.5	3.60±0.68	8.60±1.73	21.68±0.25	1.06±0.28	7.46±0.31	0.43±0.16
1.0	2.68±0.37	7.17±1.15	10.15±0.21	1.01±0.14	5.94±0.20	0.32±0.00
1.5	3.50±1.58	4.25±1.12	7.58±2.01	1.08±0.17	3.94±4.62	0.34±0.07
2.0	1.93±0.04	3.16±0.40	5.97±1.51	1.06±0.11	5.94±1.67	0.38±0.01
2.5	1.85±0.01	3.44±0.17	10.63±0.27	1.02±0.11	4.89±0.33	0.36 ± 0.05
3.0	1.95±0.21	3.77±0.64	6.50±0.42	1.00±0.08	6.35±1.29	0.29±0.003
3.5	1.44±0.08	3.32±0.42	4.27±0.47	0.97±0.042	5.13±0.96	0.24±0.03
4.0	2.21±0.69	3.60±0.11	2.90±0.59	0.91±0.07	3.44±0.71	0.35±0.06

has been deposited in the soil and vegetation adjacent to the mine by the wind. Figure 4 shows that Cu, Ni and Pb are elevated in the soils from the west, whereas Cd and Zn are elevated in soils from the eastern side. The elevation of Cd in soil from the east could be from the parent rock. Cu and Ni had 62.5 and 37.5% of samples

Distance (km)	Zn (µg/g)	Cu (µg/g)	Ni (μg/g)	Cd (µg/g)	Pb (μg/g)	S (%)	рН
0.5	3.10±0.11	46.67±0.12	15.05±0.16	0.51±0.13	9.91±1.40	0.05±0.01	3.95
1.0	1.95±0.01	46.19±2.28	7.30±1.16	0.73±0.04	7.11±1.26	0.05±0.01	4.12
1.5	4.46±0.03	42.43±3.01	10.31±0.01	0.78±0.03	7.90±0.28	0.03±0.001	4.22
2.0	3.17±1.48	42.33±0.69	7.12±0.54	0.63±0.07	11.75±1.46	0.04±0.00	3.85
2.5	3.33±0.01	20.24±0.57	19.33±2.42	0.59±0.07	7.35±0.21	0.02±0.00	4.30
3.0	2.03±0.10	17.36±0.42	15.97±7.62	0.74±0.03	6.71±0.86	0.01±0.002	4.10
3.5	1.64±0.00	10.46±0.59	17.64±0.28	0.53±0.01	5.93±0.04	0.01±0.001	4.34
4.0	1.90±0.08	20.39±0.78	9.22±0.11	0.39±0.13	9.70±0.62	0.02±0.002	4.06

Table 5. Mean (±S.D) value of heavy metals, Sulphur and pH in soil – West.



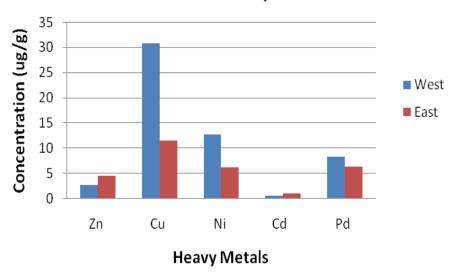


Figure 4. Comparison of heavy metals concentration in soil samples.

with concentration above three fold in the downwind side. Both heavy metals do not bio-magnify up the food chain; although at higher concentrations they can be detrimental to animals and people. On the other hand, 62.5% of samples had Pb content which exceeded one fold in the downwind side. Although Cd and Pb are within the world soil range, the concern is that they are highly toxic metals and can accumulate in body organs and further biomagnify through the food chain. In Figure 6A, most of the elements are elevated in the inner 0.5 km from the mine except Sulphur which peaked at 1.5-2.0 km. Cu and S peaked from 0.5-2 km, then flattened thereafter (Figure 6C). Overall, the western side has higher concentration of heavy metals in both soils and plants. The concentration of heavy metals further shows a decrease as you go away from the mine and this has been observed by other authors.

In Tables 5 and 6, the ranges for heavy metals and sulphur were Zn (1.64-9.14 μ g/g) Cu (4.55-46.67 μ g/g), Ni (2.63-19.33 μ g/g), Cd (0.39-1.06 μ g/g), Pb (5.40-9.91

μg/g) whereas Sulphur and pH ranged from 0.01-0.05(%) and 3.85-5.91 respectively. In this study, the soil heavy metals contents were within the world soil ranges (Rojo et al., 2004) except for Cd which is elevated on both sides. Cd could be emanating from the parent rock. The concentrations of Zn were very low, maximum of 14 ug/g, which could be the result of Sulphur as it is known to decreases soil pH and increases the solubility, availability and mobility of heavy metals, which can eventually leaches into streams (Skwierawska et al., 2012; Fijalkowski et al., 2012).

In Table 5, Sulphur content was higher in the first inner 2 km; depicting the impact of the mining pollution in the soil adjacent to it. The upwind side had lower mean concentration of 0.01% S while downwind side had 0.03% S. According to Linzon et al. (1979), average background Sulphur concentration for soil sampled in Southern Ontario, Canada ranged from 0.05-0.08% for rural and urban areas respectively. The Sulphur content in this study is below the background concentration

Mean values for heavy metals in G. bicolor leaves

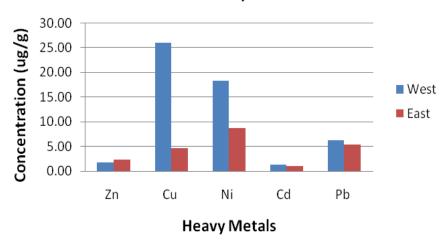


Figure 5. Comparison of heavy metal concentration in Grewia bicolor leaves.

compared to the latter region.

The mean soil pH in the downwind side is 4.12 whereas in the upwind side is 5.12. These acidic environment could be the consequences of sulphur dioxide emission from the smelter and as such it has negatively affected the vegetation therefore there is no grass and seedlings (Figure 1). The pH of 5.12 in the upwind side shows that the soil in the study area is acidic in nature; however 75% of samples from the same sampling site had pH values above 5 (Table 6). It should be noted that soil pH is considered as one of the most important factors that determine the concentration of metals in the soil solution, their mobility and availability to plants (Fijalkowski et al., 2012). In highly acid soils, the mobility of heavy metals is much higher than in soils with neutral and alkaline reactions or pH (Fijalkowski et al., 2012).

The correlation analysis was employed to determine the common source of pollution (Table 2). Positive correlations were found between Cu-Pb, Cu-Ni, Pb-S, Pb-Zn and Cd-pH. This positive correlation maybe showing that this elements emanate from the same source (Yoon et al., 2006), which is the mine. Since cadmium does not correlate well with other heavy metals, it may be coming from the geological parent material.

Heavy metals and sulphur in G. bicolor leaves

The results of *G. bicolor* leaves show that heavy metals and sulphur concentration followed a similar trend which occurred in soil samples from west transect. The samples from the western transect had elevated concentration of Cu, Ni, Cd and Pb compared to the eastern transect whereas Zn was the only one elevated in the eastern side Figure 5. Within the sampling sites, 87.5% of plant

samples in the downwind side have Cu and Ni concentrations above 2 fold compared to the upwind side. Cu and Ni are essential micronutrients for micro organisms, plants and animals; however at higher concentrations they can have toxic effects (Sheoran et al., 2011). Lead is usually concentrated in the roots of plants: however the observed elevated concentration in G. bicolor leaves could be due to aerial deposition. Generally, plants do not absorb or accumulate lead in their leaves except in the roots. The ranges of heavy metals and Sulphur in G. bicolor leaves on both transects were Zn (0.95-3.60 μg/g), Cu (3.16-46.59 μg/g), Ni (2.90-29.41 µg/g), Cd (0.91-3.46 µg/g), Pb (2.29-8.33 µg/g) and S (0.24-1.30%) (Tables 3 and 4). Since the plant samples were unwashed, the observed elevated concentrations could be due to aerial deposition. Looking at Figure 6B, the concentration of heavy metals is elevated at 0.5 km then it flattens. This is understandable because it is against the prevailing wind (Figure 3). In Figure 6D, Cu, Ni and S are elevated at 0.5-2 km and the rest of other elements does not shown any trend with regard to distance from the mine. The eastern side depicted a much lower heavy metal concentration in soils and plant samples.

However looking at the heavy metals concentrations, there is a potential risk of toxicity to animals grazing in the vicinity of the mining area. Yabe et al. (2011) has conducted a study in animals grazing in a Pb-Zn mining area (Kwabe, Zambia) and he found that the animals accumulated higher concentrations of Pb and Zinc.

Sulphur is an essential element in plants because it serves as a macronutrient; however it becomes detrimental above threshold values. Long term exposure to elevated concentration of Sulphur in plant foliage could cause chronic injuries. A study carried by Linzon et al. (1979) showed that average background Sulphur

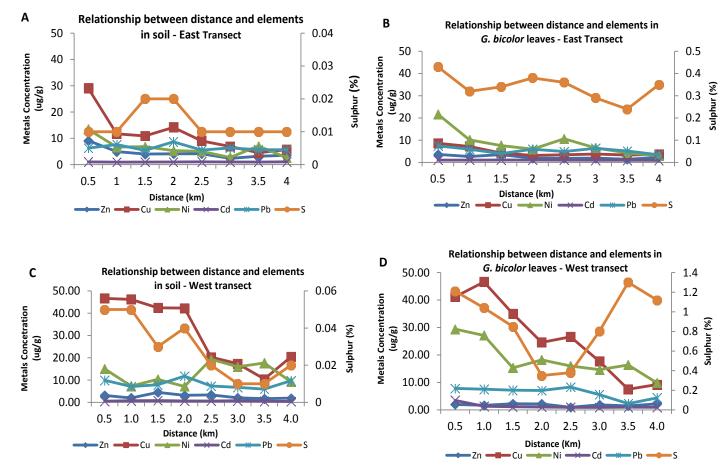


Figure 6. Changes in heavy metals and sulphur concentrations in relation to distance from the BCL mine.

Table 6. Mean (±S.D) value of heavy metals, Sulphur and pH in soil – East.

Distance (km)	Zn (μg/g)	Cu (µg/g)	Ni (μg/g)	Cd (µg/g)	Pb (μg/g)	S (%)	рН
0.5	9.14±1.22	29.01±5.16	13.38±1.87	1.03±0.04	6.29±1.06	0.09±0.001	4.28
1.0	4.95±0.07	11.72±3.42	6.65±7.99	0.94±0.11	7.61±1.23	0.01±0.003	4.53
1.5	4.03±0.35	10.89±3.42	6.83±0.61	0.96±0.00	5.40±1.24	0.01±0.008	5.24
2.0	4.11±1.34	14.15±4.88	5.32±1.24	1.01±0.01	8.65±0.24	0.02±0.01	5.91
2.5	4.13±1.23	9.00±2.18	5.02±0.06	1.05±0.07	5.52±1.10	0.01±0.00	5.60
3.0	2.37±0.98	6.88±1.66	2.72±0.03	0.99±0.10	6.35±0.15	0.01±0.004	5.46
3.5	3.16±1.53	4.55±1.29	7.06±0.57	0.97±0.16	5.68±0.37	0.01±0.03	5.68
4.0	3.55±0.07	5.67±0.64	2.63±0.10	1.06±0.03	5.64±0.42	0.01±0.001	5.14

concentrations for 33 different plant species in Southern Ontario-Canada were within the range of 0.13-1.72% thus higher than the concentrations in my study area. Elevated Sulphur concentration of above 0.80% was found in the downwind side whereas 0.43% (max) was detected in upwind side (Tables 3 and 4). The concentration of Sulphur in the downwind side was more than 1.5-3 fold (min and max respectively) in the

upwind wide (Tables 3 and 4). This shows that the western side is more polluted with sulphur as compared to the eastern side. In plants samples, positive correlations were found between Cu-Ni, Cu-Pb, Cu-S, Ni-Cd, Ni-Pb and Ni-S (Table 1). Cu, Ni and S correlated well with most of the elements and this trend is also showing in soil samples. Cu, Ni, S clearly demonstrates that they originate from the same source, however Zn did

not correlate with this three elements and it also depicts that it doesn't originate from the mining pollution (Tables 1 and 2).

Conclusion

This research study shows that significant amount of heavy metal and sulphur has been deposited in the western side of the mine (downwind) on both soil and *G. bicolor* leaves. The soil and plants samples from the western side of the mine had high concentration of Cu, Ni and Pb. The statistical test further show that the western side is significantly more polluted than the eastern side, at P=0.00 and P=0.03 (2-tailed test) on both soil and *G. bicolor* respectively. The study also revealed that the soil in the western side of the mine is acidic, with mean pH of 4.12. Furthermore, for the first time, the mean background pH has been found to be 5.12 which demonstrate that the soils in the study area are acidic in nature. Concentration of soil parameters in the downwind side were in the following order Cu>Ni>Pb>Cd>S.

Assessment of the results of the study area against the world range depicts that heavy metals were below the maximum allowable limits. However persistence of the latter in *Grewia bicolor* and soil continues to be a treat to animals and people of Selibe-Phikwe and the surrounding areas due to potential biomagnifications and bioaccumulation, especially with regard to Pb and Cd.

The recommendation of this study therefore is that more research needs to be done on heavy metals in edible and medicinal plants and also in animals grazing in the mining area.

Conflict of Interests

The authors have not declared any conflict of interest.

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