AN ASSESSMENT OF THE LEVEL OF SOIL CONTAMINATION WITH HEAVY METALS AROUND LETSENG AND KAO DIAMOND MINING IN LESOTHO

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DECLARATION

I declare the thesis hereby submitted by me for Master of Science Degree in Soil Science at the University of Lesotho as my own work and that to my best knowledge contains no work submitted previously as thesis for any degree at other university. I furthermore cede copyright of the thesis in favour of the National University of Lesotho.

Signature:…………………………… Date: ~~17-02~~ 17-01-2024

ABSTRACT

The study was aimed at determining the extent of soil contamination with heavy metals due to wastes from Kao and Letseng Diamond Mining and to determine the potential risks to the soil and crops in nearby farmlands. A number of physical and chemical soil parameters were tested namely; texture, pH, Organic matter, and CEC to find out their impact on bioavailability of heavy metals in the soils and crops around Kao, Letseng and Mokhotlong Agricultural Research. Furthermore, study was made to assess the effects of heavy metals on soil microbial activity, and to determine whether heavy metals in plants and soils are mild, sufficient or toxic. Heavy metals were tested whether available or not available (not detected) in three sites. The samples were collected from three sites, two of which are from contaminated (Kao and Lets'eng mining) sites and one from uncontaminated (Mokhotlong Agricultural Research Station) site in Lesotho. Basic cations and metals were extracted by DTPA method (Lindsay and Norvell, 1978) and later determined using an AAS 500 model.

The results indicated that there are higher concentrations of heavy metals around Letseng Diamond Mining in the order of Cr > Pb > Cd > Ni > Fe > Mn > Cu > Co > Zn. Around Kao Diamond site, three metals were in the toxic range in the order of Cd > Pb > Cr. Kao Diamond mining site is located within Kao Village, where the effluent from the mining drains into farmlands, which obviously poses health threat. Chlorosis and stunting of plants have been observed especially in the rangelands around Letseng Diamond Mining. This is to high concentrations of heavy metals, which could hinder plant growth and affect the food chain thus posing serious risk to health of animals, humans and microorganisms. Again the results showed the plants with high concentrations of heavy metals have stunt growth and soils with high concentrations of heavy metals showed lower microbial population which has negative impact on soil fertility.

Results of this study indicate that the heavy metal risk around the mining sites is alarming and hence appropriate measures need to be taken to rectify the situation by applying biochar. Whilst Mokhotlong Agricultural Research shows only essential heavy metals and their concentrations are not alarming with high population of microorganisms.

Keywords: heavy metals, potential risks, bioavailability, toxic, contaminated, health threat

DEDICATION

This thesis is dedicated to my parents:

The Chief Magistrate, Mathuso Tjakata

Who made it a point of duty to lay for me a sound educational foundation, encouraging myself to work hard until the completion of the thesis

CERTIFICATION

I the undersigned, certify that the work reported in the thesis was done by Motsamai Julius Tjakata under my supervision.

Supervisor: Prof Fisscha Itanna

Signature:......................................**Date:** 17.02.2024...................................

.... 8/10/24 ....

HEAD OF DEPARTMENT DATE

.... 10/10/2024 ....

DEAN OF FACULTY DATE

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ACRONYMS

MAR:	Mokhotlong Agricultural Research
LDM:	Letseng Diamond Mining
KDM:	Kao Diamond Mining
Cd:	Cadmium
Zn:	Zinc
Co:	Cobalt
Mn:	Manganese
Fe**:**	Iron
Ni:	Nickel
Pb:	Lead
Cr:	Chromium
CEC:	Cation Exchange Capacity
AAS:	Atomic Adsorption Spectrometer
WHC:	Water Holding Capacity
DTPA:	diethylenetriaminepentaacetic acid

Chapter 1

Introduction

Heavy metal contamination results from deposition of excess amounts of toxic metals in soil (Su et al. 2014). The occurrence of heavy metals in soils could be through natural or human activities. Some heavy metals are essential for organisms and plants in the environment because of their low concentrations. However, if they exceed normal limits, they could become toxic to soil organisms and plants as well (Monga et al. 2022). In nature, heavy metals accumulate in soils by natural processes such as volcanic eruption, weathering of rocks and pollution through the action of wind or water. Artificially, they could arise from human activities such as mining or discharging of agricultural wastes. These heavy metals are transported from their sources either by wind or water and later accumulate in the soil (Mirsal 2008).

The mining of diamonds was first introduced in Lesotho in the 1950s where Basotho were allowed to mine as artisanal miners until late 2005 when a new Act paved the way for small-scale miners (Lerotholi, 2021). Diamond mining forms only a small part of Lesotho GDP. The contribution to GDP of the mining sector is approximately 7% and in 2018 diamond mining generated almost 36% of export earnings. Deposits of coal, quartz, agate, galena and uranium have been found in Lesotho but are not yet commercially viable. The diamond mining is of biggest interest to international investors. The major players in Lesotho's mining industry are the foreign companies such as Gem Diamonds and Firestone Diamonds both having headquarters in London. The mining operators have put certain amount of money to fund projects aiming to develop living standards in local areas.

Opencast mining has serious impact on environment, such as the destruction of natural soils and the extraction of essential volumes of materials. The blasting of the earth causes the formation of new human induced soils, that accumulate wastes from the mines, known as anthropogenic soils. Mine soils are very young soils which have a thin A horizon that is normally developed on unstable materials and characterized by unstable and scarce cohesion which easily exposes the mine soils to water and air erosion (Legostaeva and Golobova 2021)

There are several mining sites in Lesotho mainly in connection to mining of diamonds. Lots of materials and chemicals such as hydrogen peroxide, alkaline reagent, mixture of concentrated sulphuric, hydrochloric and nitric acids, gasoline, iron silicates are used to extract these minerals of which some are very dangerous to humans as well as organisms and crops. Some metals after being used in the process of mining are transported either by wind or water and then stored in soil where they become dangerous to human life and organisms living near that region. The soil properties such as organic matter have significant effect on bioavailability of heavy metals (Olaniran et al. 2013). Clay content and pH also have an impact on metal availability in biological and biochemical processes/properties.

Heavy metals have an impact on soil enzymatic activities through association with the microbial community that creates enzymes. Uptake of heavy metals by plants and their accumulation in plants affects animals and plants which make part of the human food chain. Metal build-up in soils surely affects some microbial processes such as respiration rate (Singh & Kalamdhad 2011). Levels of heavy metals in soils need to be assessed so that the risk they are posing on plants, organisms and the environment should be taken into consideration.

The mining method used in Lesotho is open cast extraction which includes shafts, pipes and tunnelling. The diamondiferous pipes in Lesotho districts are as follows: Letseng-la-Terae and Mothae pipe in Mokhotlong; Kao, Liqhobong, and Lemphane pipes in Botha Bothe; Kolo and Sekameng in Mafeteng. The Letseng-la-Terae mine in Lesotho highlands produces the largest high-quality pink and black diamonds in the world. It is located in Maluti Mountains where the weather is very cold throughout the year and mostly snowing.

Kao mining is approximately 35 km far from Letseng-la-Terae by road (Thabane 2000). In the 1960s, individual diggers among Basotho and some South Africans came to mine in Kao Mining illegally. They called themselves Liphokojoe (which means jackals). They were mining or stealing the Company’s unprocessed ore and were also not welcomed by Kao Villagers. Kao mining site is within the village of Kao village. It has even taken some of the farm fields where farmers used to produce or cultivate; mainly, cereal crops such as wheat, sorghum, maize and beans.

Uptake of heavy metals by plants and their accumulation in plants affects animals and plants such as spinach, which make part of the human food chain. Levels of heavy metals in soils need to be assessed so that the risk they pose on organisms and the environment should be taken into consideration.

Heavy metals have a density greater than $5g/cm^3$, with atomic number more than 20. Some of the heavy metals include: Cadmium (Cd), Mercury (Hg), Arsenic (As), Copper (Cu), Manganese (Mn), Selenium (Se), Zinc (Zn), Iron (Fe) (Salomons et al. 2012: Bahiru and Yegrem 2021). Heavy metals such as Chromium (Cr) and Cadmium (Cd) are able to decrease oxidative damage and denaturation of microorganisms as well as weakening the bioremediation capacity of microbes. Moreover, heavy metals disturb metabolic functions in organisms (including humans) in two ways: they affect the function of main organs and glands such as brain, heart etc. The other effect is that they could displace main nutritional minerals from their original place hence interfering with the biological functions of plants (Singh et al. 2011). The presence of heavy metals such as Cu, may lead to the predominance of organo-metallic complexes instead of organic contaminant adsorption to the soil mineral (FAO & UNEP 2021).

There are other indirect impacts that are often rejected, leading to underestimation of the costs of soil pollution (Weissmannova & Pavlovsky 2017). Many ecosystem services are hindered by soil pollution, reducing productivity and resilience in the long term. Soil pollution causes loss of crop yields and food wastage due to high levels of contaminants, loss of biodiversity and increased incidence of pests, decreased water quality and eutrophication of the marine environment (Motuzova et al. 2014).

Heavy metals are special group of elements because of their toxic effect exerted on plants and soils upon their high concentrations. Some heavy metals such as Cu, Fe, Mn and Zn are required for growth and functioning of plants when they are in normal concentration (Itanna and Coulman 2003). Contamination of soil is mainly caused by toxic metals including Zn, Pb, Al, Cd, Ni, Fe, Mn, As which are major mining effluent tailings (Okereafor et al. 2020). There are nonessential heavy metals such as Hg, Cr, Cd, Pb and Al which means they are not needed by plants even in little amounts for any metabolic processes

There are plant species such as corn, barley etc that can tolerate excess heavy metals. Soil contaminants can get to plants through soil, dust, air, water or food. The degree of heavy metal sorption is influenced by environmental factors, soil components and properties as well as amount of heavy metals present in soil (Itanna et al. 2008). Higher degree of heavy metals in the soil hinders the microbial activity which plays significant role in the reclamation of contaminated soils.

Justification

Heavy metals are special group of elements because of their toxic effect exerted on plants and soils upon their high concentrations. Some of heavy metals such as Cu, Fe, Mn and Zn are required for growth and functioning of plants when they are in normal concentration (Itanna and Coulman 2003). There are plant species such as corn, barley etc that can tolerate excess heavy metals. Soil contaminants can get to plants through soil, dust, air, water or food. The degree of heavy metal sorption is influenced by environmental factors, soil components and properties as well as amount of heavy metals present in soil (Itanna et al. 2008). Higher degree of heavy metals in the soil hinders the microbial activity which plays significant role in the reclamation of contaminated soils.). Higher degree of heavy metals in the soil hinders the microbial activity which plays significant role in the reclamation of contaminated soils.

Problem statement

There are several mining sites in Lesotho mainly in connection to mining of diamonds. Lots of materials and chemicals such as gasoline, are used to extract these minerals of which some are very dangerous to humans as well as organisms. Some metals after being used in the process of mining are transported either by wind or water and then stored in soil where they become dangerous to human life and organisms living near that region. The soil properties such as organic matter have significant effect on bioavailability of heavy metals (Olaniran et al. 2013). Clay content and pH also have an impact on metal availability in biological and biochemical processes/properties.

Heavy metals have an impact on soil enzymatic activities through association with the microbial community that creates enzymes. Uptake of heavy metals by plants and their accumulation in plants affects animals and plants such as spinach, which make part of the human food chain. Metal build-up in soils surely affects some microbial processes such as respiration rate (Singh & Kalamdhad 2011). Levels of heavy metals in soils need to be assessed so that the risk they are posing on organisms and the environment should be taken into consideration.

Objective

To determine the extent of soil contamination with heavy metals in Kao, Letseng and Mokhotlong Agricultural Station and also the potential risks to the soil and crops in nearby farmlands as well as potential risks to microorganisms and environment itself.

Specific objective

1. To determine the impact on bioavailability of heavy metals in the soils and crops around Kao, Letseng and Mokhotlong Agricultural Research
2. To assess the effects of heavy metals on soil microbial activity
3. To determine whether heavy metals concentrations in plants and soils are insufficient, sufficient or toxic

Hypothesis

1. H_o: High bioavailability of heavy metals poses risks to soils, and crops around Kao, Letseng and Mokhotlong Agricultural Research.

   H_A: Low bioavailability of heavy metals in soils pose low risk of life to plants and soils around Kao, Letseng and Mokhotlong Agricultural Research
2. Ho: Heavy metals in soil negatively impact microbial activity by deactivating enzymes, decreasing microbial biomass, and altering the composition of microbial community

   H_A: Exposure to heavy metals in soil lead to decreased microbial population thus decreasing microbial activity and lower soil fertility
3. Ho: The heavy metal concentrations in soils and plants can either be insufficient, sufficient or toxic based on their concentrations, with higher concentrations corresponding with toxicity

   H_A: The heavy metals in soils and plants are toxic despite their concentrations

Chapter 2

Literature review

2.0. Interaction of soil contaminants with soil

Healthy soils are required for food safety and security. Therefore, there is always a need to cater for productivity and food safety. The interaction and movement of contaminants in the soil involve different physical, chemical and biological processes. There are three processes that take place (1) retention on and within the soil body (2) Infiltration, diffusion and transport by soil solutions (3) Alteration, transformation, and initiation of chemical changes within the soil (Mirsal 2008). In soil the transportation of heavy metals is through diffusion and dispersion in the form of active transport influenced by different factors such as rain intensity evaporation, e.t.c (Pisane 2004).

The interaction of toxic heavy metals with soil is quite strong and depends on element and its speciation (Saha et al. 2017) as well as the form they exist in the environment (Dube et al. 2000). Dube et al. (2000) concluded that the availability of heavy metals in soils indicate serious harmful environment that is very difficult to reclaim. They further indicate that chemical character of heavy metals makes them to biodegrade as well as soil matrix complexity.

2.1. Soil contamination due to heavy metals

Contaminants are chemicals that are available at higher levels than other sections in the environment. The toxicity of heavy metals on the soils relies mostly on the ability of soils to immobilize those pollutants on the specific environment. Heavy metals stay longer in the soil, the reason being that they are non-degradable either through microbial or chemical processes. Localization of high amounts of metals could cause an increase in toxicity of heavy metals. Some of the metals react with other metals to become toxic in the soil (Briffa et al. 2020). Forms and mobility of heavy metals mainly trace elements required by plants is affected by surface organic matter accumulation together with other properties such as redox potential and soil pH (Lepp 2012).

Li et al. (2014) found out that soils around mining sites are contaminated by heavy metals emitted from mining activities that pose risks to public as well as crops grown or vegetation around. Irrigating either by treated or untreated waste water from mines would still increase the heavy metal concentrations in the soil (Sharma et al. 2007). Krishna et al. (2013) investigated the level of soil contamination near Chromite mines in Chikkondanahalli of Nuggihalli Schist Belt, Karnataka India, where they found out that increased concentrations of Cr, Ni, Co in soils of the study site exceed Soil Quality Guideline limits.

Land use activities mainly diamond mining in Letseng around the catchment are sources of pollution particularly nutrients because heavy metals showed to appear naturally within the Klip River Catchment (KRC) and the geology of the area supported that. These Heavy Metals (HM) (Pb, Cu & Cr) and nutrients (NO_3-, NO_2- & NH_3) are threat to health implications for both human beings and biota (Li et al. 2019). Another study on determination of HMs in fish tissues such as liver and kidney could assist to assess possible bioaccumulation impacts of these HMs. Public health study on Patising catchment (near Letseng Diamond Mining village) community to assess residents' health status with respect to the pollution in the catchment could be essential. It could be essential to assess how the Patising stream could have affected livestock of the community residing in the area. Discussions with an area chief reveal that, the stream is no longer extracted for potable water instead the community utilize water from the upstream confluence of Khubelu River and Patising which carry contaminants from Letseng (Shakhane 2019).

Zn, Cd, As, and Pb are main anthropogenic accumulators of Potentially Toxic Elements (PTEs) in the diamond mining while Mn is mainly from natural sources (Gololobova et al. 2022). There are mobile forms of Ni, Mn, and Cd which are characterised by biogenic (produced by organisms) accumulation in the upper organic horizon and also the supra-permafrost horizon which accumulates the mobile forms of Cr, Ni, Co, Mn, and Cu in diamond mining soils.

2.2. Availability of heavy metals in soil and their forms

Organic matter has an ability to retain more metals in an exchange form by supplying organic chemicals to the soil solution that act as chelates that have potential to make more metal available to the plant (McCauley et al. 2009). Heavy metal adsorption decreases with decreasing organic matter, porosity and moisture content of soil. Furthermore, clay fractions, have high sorption capacity which enables them to bind heavy metals. The concentration of heavy metals is high in O horizon where there is high amount of organic matter and decreases with the profile, thus going down the profile, organic matter decreases and also heavy metal concentration decreases (Zaky and Abdel-Salam 2020).

The binding of heavy metals and speciation in soil solution is affected by the Soil Organic Matter. The organic amendments lowers redistribution of heavy metals into less available forms. Humic acids regarded as organic particles are responsible for the adsorbed heavy metals (Kwiatkowska-Malina 2018). Organic matter creates complexes with heavy metals thus has tendency of transition metal cations to create stable complexes with organic ligands. Soil organic matter recently has been studied because of heavy metal sorption, since the amount of organic matter has an effect on speciation of heavy metals in soil. For instance, concentrations of Cd and Ni in soil solution is decreased by high organic matter content (Skłodowski et al. 2006).

Changes in soil moisture content can alter metal availability. In acid soils, the availability of cadmium (Cd), zinc (Zn) and nickel (Ni) (but not copper) increased with drying from saturation to 50 % water holding capacity (WHC). Drying soil from 100 to 30 % WHC had weak or no significant effects on available metals. Availabilities of Cd, Ni and Zn correlated well with available Al for dry conditions. Available Cd and Zn were lower in dried soils from the cycle of saturation to 50 % WHC. Hence, soil water regimes could be manipulated to alleviate soil metal availability. Individual metals and different soil types, and soil moisture contents before drying should be considered when dried soils are used for evaluation of metal availability (Li et al. 2015)

The pH variations have an impact on the extent of the concentrations of Cu, Pb and Cd in the effluent. Moreover, complex formation restricts the sorption of the metals on the clay, with an increasing impact in the order: Mn $\leqslant$ Pb $\leqslant$ Cd $\leqslant$ Zn $<$ Ni $<$ Cu $<$ Cr (Abollino et al. 2003). The soil pH is usually the most important factor that governs uptake of heavy metals. For instance, low pH enables Cd accumulation. Soil pH should hence be taken into much consideration or deeper investigation since it is a main chemical factor affecting the availability of heavy metals (Kirkham 2006). Zn Cd and Pb increase exponentially in acidic regions. It is thus crucial to examine soil pH to observe the transportation of heavy metals within soil profile. Heavy metals appear in the soil in different oxidative states that determine mobility and capability of heavy metals (Ebenebe et al. 2015).

Xiao et al. (2019) used secondary phase fraction (SPF) to assess the potential availability of heavy metals at soil depth of 0-20 cm. They found out that total and SPF concentrations decrease as one sample is away from source of contamination. BCR sequential extraction results indicate that quantity of mobile fractions of heavy metals in acid solution, reducible and oxidizable Mn, Cd, and Pb were higher than that of the immobile fractions (Sungur et al. 2014).

Acidity is a major factor playing role in solubility of metals in soil while mobility of heavy metals in soil is determined mainly by clay content, amount of organic matter and soil pH. When clay content, organic matter and pH are high, the more firmly bound metals are the ones that stay longest in the soil (Stazi et al. 2015)

The uptake and accumulation of micronutrients is influenced by the soil pH; whereby, higher soil pH has low concentration of micronutrients. CEC plays a role in sorption of heavy metals. Cd has capability to replace Calcium in soil. Organic Carbon along with CEC has an influence on sorption of heavy metals and their bioavailability (Boss and Bhattacharyya 2008). Metal mobility, ion retention and leaching could be promoted by coarse texture, high CEC and high soil pH (Itanna et al. 2008). Cd concentration can be reduced by organic fertilizers such as faecal matter and cow manure (Woldetsadik et al. 2016). Heavy metal adsorption on clay minerals occur onto both planar and edge sites when at low ionic strength but an increase in the ionic strength donates more Na^+ ions, especially for the planar sites which are considered of lower affinity, but compete with the heavy metals for the adsorption (Malandrino et al 2006).

Cd availability is controlled by soil pH and organic matter, that is, as pH decreases, the amount of Cd in plants increases. Soil organic matter quality plays significant role by binding and accumulating Cd. Soil organic matter is the primary sorbent for heavy metals (Gray et al. 2004). Soil salinity has an influence on Cd mobility, thus does not mean it increases Cd availability. An addition of soil organic matter (SOM) does not have effect on total concentration, rather affects the availability. Thus, increasing SOM, decreases Cd soil availability and as the result lowers plant uptake (Filipovic et al. 2018).

The chromate ions become toxic to soil when present in hexavalent form. The detrimental effect on the plant is caused by higher concentrations of chromium which is toxic to the plants. The chromium immobilization is the result of its accumulation in root vacuoles and also its incapability to translocate from the root to the aerial shoot parts (Kumar and Maiti

2013). There are different forms of Cr, but toxicity of Cr is related to the exchangeable Cr and also controls the bioavailability of Cr (Garnier et al. 2006). Cr toxicity in plants causes impairment of photosynthesis, reduction of growth, decreased germination. Chromium can be absorbed through carriers such as sulfate by plants (Ertani et al. 2017). The plant uptake of heavy metal depends on mobility and availability of such metal in the soil.

Conceptual and quantitative model approaches have to be developed to predict fate and transport of heavy metals in soils. As the retention mechanism of metal ions at soil surfaces is the sorption which involves the loss of a metal ion from an aqueous to a contiguous solid phase and having three major processes: adsorption, surface precipitation, and fixation. Specific adsorption generate strong and irreversible binding of heavy metal ions with organic matter and variable charge minerals while nonspecific adsorption is an electrostatic phenomenon in which cations from the pore water are altered for cations near the surface. Cation exchange is a form of outer-sphere complexation with only weak covalent bonding between metals and charged soil surfaces (Liar et al. 2007)

The affinities of cadmium, lead, and nickel for organic matter are normally less than copper thus copper binds very well with organic matter. Zinc and nickel selectivity for organic matter is very less (Qiao et al. 2023). The dominance of organic bound fraction was determined for Cu, mainly in organic rich surface soil therefore speciation of adsorbed metals revealed less selectivity of Zn and Ni to soil organic matter. The surface soils normally having greater adsorption for heavy metals as well as provide greater stability to adsorbed metals, especially for Cu compared to underlying soil (Hossain et al. 2009). Different rates of Cu discharge from different size fractions is influenced mainly by different size fractions of soil particles indicating high heterogeneity in terms of morphology and chemical composition. The smallest clay size fraction (0.22–2 μm) carry highest concentrations of SOM and Fe/Al (hydr)oxides, being the major reactive fraction of soil particles driving Cu release (Shi et al, 2020).

Higher concentrations of Mn and Cu are typical in the soils of the Sredne-Markhinsky diamond-bearing region thus the accumulation of Cr, Ni, and Co illustrates the impact of kimberlite magmatism in general (Legostaeva et al. 2020). They represent natural geochemical anomalies associated with the mineral composition of rocks and groundwater, which carry a number of impurity elements with high toxic properties (Tl, Di, As, Cd, Hg), and increased concentrations of heavy metals (Cu, Zn, Pb, Ti, V and others). Geochemical risks for the Daldyn-Alakit diamond-bearing region seem as an accumulation of different trace elements, such as Cr, Co, Ni, Mn, Li, Be, and Sn on the day surface during mining operations. Biogenic accumulation of Ni, Mn, and Cd appear in the upper layer of the soil and Cr, Ni, Co, Mn, and Cu in the suprapermafrost soil horizon which are typical for the soils of the studied area.

Birim River is heavily polluted with heavy metals based on the result obtained from the experiment compared to the World Health Organization (WHO) guidelines for allowable amount of heavy metals in soil. Samples collected in different locations show high heavy metal concentrations in areas where small scale mining is dominant, showing that the major contamination source in the soil or environment is resulting from small scale mining activities than large scale minings. Afum et al. (2016) found out that heavy metal concentration measured as dissolved were less than WHO standards with the exception of Fe. There are increased accumulations of heavy metals in the suspended mineral fractions of the environment and diamond mining sediments were also greatly polluted with heavy metal sinks (Afum et al. 2016).

The major anthropogenic contribution in the heavy metals in the study area seem to be Zn, Cd, As, and Pb. The investigation indicates origin of Mn in the area is most likely to be a natural source. The content of Co, Cr, and Ni are driven by both lithogenic control and anthropogenic sources. Active accumulation of mobile forms of Mn, Zn, and Ni with anomalously high concentration coefficients can be searched in the soils in the affected zone of mining operations. Anthropogenic soil contamination has possibility of being distributed over an area of 260 km (Gololobova et al. 2022). In Lesotho, mining at Letseng Diamond minings have negatively affected the ecosystems of Mokhotlong District via soil erosion, site contamination, surface water pollution from mining waste, land degradation from non-biodegradable litter, and the accumulation of toxic waste landfills (Mukurunge & Bhila 2019).

However, the contaminated area can still be recovered. For instance, in pine bark amendment (could be used for adsorption of heavy metals like biochar) and ageing experiment for sorption of heavy metals had less impact on Cd, Ni and Zn fractionation, whereas essential alterations were detected for Cu and Pb in response to both pine bark amendment and ageing thus there were decrease in the soluble fractions, and increase in less mobile fractions. Desorption experiments indicated that both pine bark amendment and ageing declined heavy metal discharge from the mine soil. The results of this study indicate that pine bark amendment could be applied to raise heavy metal retention (especially in the case of Cu and Pb) in acid mine soils, thus decreasing the risks of metal transfer to uncontaminated environmental zones (Fernandez-Sajurjo et al. 2017).

2.3. The effects of heavy metals in the soil Microorganisms

Heavy metal increase in the soil affects enzyme activity which could affect the rate of heavy metal concentrations in soils by decreasing microbial population in the soil (Karaca et al. 2010: Chuo 2018). Ma et al. (2022) revealed that indigenous microorganisms such as Actinobacteria increase with the increase in heavy metal contamination. Chuo (2018) concluded that microbial community is a good indicator of the level of soil heavy metal contamination and impact of heavy metals on soil ecology. The availability of toxic metals in soils leads to serious problem since the productivity of the land will be decreased (e.g. lead poisoning makes other soil nutrients unavailable) (Ashraf and Ali 2007; Stazi et al. 2015).

Soil microbes such as bacteria, fungi and actinomycytes are reduced in contaminated site even becoming less sensitive. Increased heavy metal concentration in the soil has negative impact on microbial activity thus, lowering organic matter mineralization which will affect plants during growth (Oliveira & Pmpulha 2006). Lead is very harmful to the environment since it does not have biological role like other metals (Sobolev and Begonia 2008). The effect of high metal contamination in soil is that if it increases, total bioactivity, richness and microbe activity also decreases (Xie et al. 2016).

The resistance of bacteria to the heavy metal is usually encoded by genes located on plasmids. It is possible that growth of the bacterial community in polluted environment is due to gene transfer through plasmids that make the bacteria to be even tolerant to the environment. The increasing frequency of heavy metal tolerated species causes changes in the bacterial communities once exposed to the heavy metals on the environment. The time is very important in the effect of heavy metals on microbial activity with processes involved in soil. Thus too low toxicity level of metal affects the microorganisms but with time. The very same level will have an effect to the microorganisms in the soil. Pb forms more stable complexes with humic substances compared to Cd, thus major sensitive soil microbes are affected, such as N-transformations are affected (Baath 1989).

The pollution of soils with heavy metals has an influence on characteristics of soils, such as affecting enzyme activity that helps to decompose organic matter. Soil enzyme activity is regarded as very sensitive since its very essential in microbial processes including nutrient cycleing. Thus, soil enzyme activity responds rapidly to the changes in soil either naturally or through anthropogenic factors (Karaca at al. 2010). Soil microbes are very sensitive compared to macro-organisms in soil ecosystem once exposed to environmental stress. The soil microbes will show some alterations to soil environment as early as possible. Heavy metals are non-degradable affecting soil microbial population. The main heavy metals affecting microbial community are: Pd, Zn. Cd, As, Cr, (Chu 2018)

Heavy metal contamination could generate varying microbial pattern. There are many microorganisms in soil and the change of chemical or biological properties of soil due to metal contamination could affect microbial community (Perezdemora et al. 2006). Longer residence of heavy metals in the soil, will target those soil microbes adapted to the soil. This is accelerated by increased content of organic carbon in seriously polluted soils. Heavy metals destroy spatial structure of the active groups of the enzymes thus heavy metals contamination has direct effect on soil enzyme activity in the soil (Chu 2018). Metal toxicity to soil microorganisms causes decline in the soil microbial biomass due to the declined substrate utilization efficiency since microorganisms subject to metal stress require higher energy cost (Giller et al. 2009)

2.4. The effects of heavy metals in the plants/crops

Metal toxicity decreases the uptake and translocation of nutrients and water and allows oxidative damage thus restricting plant growth. Concentration of heavy metals can differ from plant and soil depending on the seasons. For instance, concentrations of Cd, Cu and Mn in soils are positively correlated with concentrations of Cd, Cu and Mn in plants only during summer (Sharma et al. 2007). Zinc and Nickel are essential metals (Itanna and Coulman 2003) but increased concentrations pose risk to plants and their roots whereby there will be stunted growth and oxidative damage in plants (Tahar and Keltoum 2011). An increased level of heavy metals in plants could cause the following symptoms in plants; chlorosis, necrosis, damage to plant organs and plant phenotype changes (Wang et al. 2019).

High concentrations of heavy metals have impact on plant growth, photosynthesis and phytoaccumulation efficiency. Root elongation for absorption of heavy metals can be inhibited by other heavy metals such as Mn due its meristematic cell division potential (Benzarti et al. 2008). The mobility of heavy metals is decreased when soil pH is above 5. Thus, plant growth is weakened and deficiencies of Fe and Mn will be there (do Nascimento et al. 2021). The level and amount of heavy metal accumulation in plants rely on soil type, pH, humidity and micronutrient content and as well as time of crop harvest. For example, in alkaline soils, leaching and bioavailability of heavy metals to plants are lower and presence of organic matter can also restrict metal uptake from soil solution (Kumari and Mishra 2021)

Soils, either in urban or agricultural areas resemble a significant sink for metals discharged into the environment from a wide variety of anthropogenic sources. Once in soil, some of metals are persistent and stay longer in soil because of their state of immobile nature. Other metals are very mobile therefore the potential of transport either through soil profile down to ground water aquifer or via plant-root uptake (bio available) is likely. One has to take into account the mobility and the bio-availability of heavy metals once it enters the food chain because plant uptake of metals parallels the bio available fractions of the metals in soil (Tserene 2010)

Soil management alters physical, chemical, and biological characteristics, and consequently, alternative responses by biological activities to heavy metal toxicity can be observed (Wani et al 2007). The effects of heavy metals on plant development differ with reference to soil characteristics, the type of plant and the metal. Finally, the effects of heavy metals on both plants and crops must take into account in order to act upon the risk of these contaminants being transported to the food chain (Guala et al. 2010)

The lower the leaf production rate and the plant mass the more metal contaminated soil substrate contained, and the more the phenological development would be delayed. The late and decreased reproduction may have large effects at population, community and ecosystem` level, even if the effect of low metal levels on plant growth may be small and attribute to increased evolution of metal tolerance. Flowering phenology is considered very active on response to metals. The latter being a result of advanced seed ripening due to heavy metals cause leaf life span to be decreased at the highest and the lowest degree of metals (Ryser and Sauder 2006)

Cd reduces hormetic effects on plants which is an adaptive response of cells and organisms to intermitted stress, and damage the potential mechanisms that enhance them to have a better performance under Cd (Carvalhon et al. 2020). Once there is Cd availability in the plant, it becomes toxic to the plant therefore the solution is to completely eliminate it from the environment. Plant and soil factors need to be assessed on how they affect Cd availability (Kirkham 2006). Incubation with Cd-tolerant plant growth-promoting rhizobacteria has the potential to increase the Cd toxicity in the plant and can also increase the availability of Cd to soil (Guo & Chi 2014). The availability of Cd disrupts the equilibrium state of nutrients in the plant and also in the soil. It does so, by affecting roots which absorb the nutrients and also by affecting how nutrients are distributed in the plant parts. Cd has several effects on the plant, one it competes with other nutrients for transport mainly absorption by roots, secondly the water uptake by plants is disturbed by Cd availability, thirdly in the transport process Cd affects key enzymes (Mourato et al. 2019).

High levels of Cd >3 mg/kg affect plant height, stem diameter, plant biomass, root volume, root surface area, and root biomass, as well as soil enzyme activities and microbial biomass (Pinto-Poblete et al. 2022). Cd availability exponentially reduces the shoot of the plant. Cd has several effects on plant photosynthesis (Krantev et al. 2008).

Chapter 3

METHODOLOGY

3.1. **Study site**

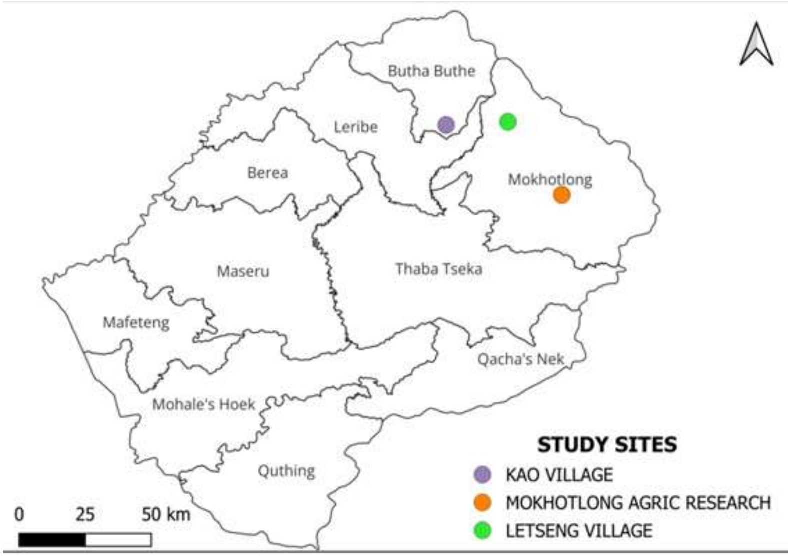

Fig 1: Map of Lesotho showing 3 study sites of the interest. Letseng Village and Kao Village are of interest and focus area since there are mining activities nearby

Soil samples were collected from three sites in Lesotho; namely, Letseng Diamond Mining outlet, Kao Village and the Agricultural Research Station in Mokhotlong. The two sites are along the wastewater drainage lines at the outlets of the mines. The site at Kao Village is near Storm Mountain Diamonds (Pty) Ltd ('SMD'), which operates at a Kimberlite Diamond mine at Kao, Botha Bothe, Lesotho. It is specifically located on agricultural fields directly affected by the wastewater from the mining plant, where more wastewater flow is observed during the rainy season. The Agricultural Station in Mokhotlong is taken as a control site, since it is not affected neither by the wastewater nor by airborne particles from the mining sites.

The dominant vegetation at Kao village is weeping willow, poplar and grasses (figure 1). The farmers there mainly grow vegetables including potatoes and cereals such as sorghum and maize. The soils in Kao are Mollic Heptosol and Vertic Cambisol (Schmitz and Rooyani 1987). Geographically, the Kao village sampling site is located between 29^{o}00’56’’ S and 28^{o}37’19’’ E coordinates, at an elevation of 2 378 m where the samples were collected for heavy metal analysis. The place is very cold and mostly snowing during winter. The soil series found in Kao is Popa Soil, which is a well-drained, shallow, medium texture soil. The epipedon is a very dark brown to black loam or sandy loam underlain by basalt similar to that of Letseng.

Letseng Mining is the biggest mining site in Lesotho at Mokhotlong District, which is located at 29^{o}00'45'' S and 28^{o}52'39'' E, at an elevation of 2885 m (figure 1). Soil samples for this study were collected alongside an outlet on the southern side of Letseng Diamond. The dominant vegetation in the region is mainly E*riocephalus punctulatus* and grass. The soils found in the region are Eutric Fluvisol and Humic Acrisol. The Popa soil series, because of its favourable characteristics for grassland production, is concentrated in that place. The place is very cold with highest temperature being 15 ^{o}C during the summer and regularly snowing during the winter.

The control site was in Mokhotlong Agricultural Research Station, which is located between 29^{o}17'17''S and 29^{o}04'47''E (Figure 1). The weather is unstable or unpredictable and more often there is snowfall with long winters or coldness. The geology of Mokhotlong Agricultural Research filled with igneous volcanic basalt with soils that has high fertility. The soils are slightly acidic. Unlike Letseng, the place is not cold as Letseng and highest temperatures reaching 25 ^{o}C. There is little of chemical weathering happening, only rapid physical weathering (action of frost and ice) takes place therefore soil is removed by wind and water due to high altitude. The soils found the region are Fluvisol and Humic Acrisol.

3.2**. Experimental unit**

Soil samples were collected at Letseng outside the Letseng mining diamond premises as you travel to Maloraneng Guest House (Figure 2). The soils from the contaminated sites were collected along wastewater drainage lines at the mines' outlets. The red pins on Figure 2 shows where the samples were taken and the sampling points are 50 meters apart.

Fig. 2: Soil sampling sites at Letseng (red dropped pins representing spots)

Soil samples were then collected at Kao at an interval of 20 cm (figure 3) alongside the slope from five different spots along the drainage route of contaminated wastewater that flows downward from the source during the rainy period. An independent seemingly a control sample was collected 50 m away from the drainage line. The red pins that are on the pictures below show where the samples were taken from.

Fig 3: Soil sampling sites at Kao, the mining is at position above the village and the agricultural land below the village (red dropped pins representing spots)

Mokhotlong Agricultural Research is located in north of Lesotho at Mokhotlong districts (fig. 4). The red dropped pins shows how the samples were taken and due to bedrock subsurface (20 – 40 cm) soil was not collected. The interval of 50 m was made like two other studies of interest (fig 3). The stones around sampling pins could easily be seen

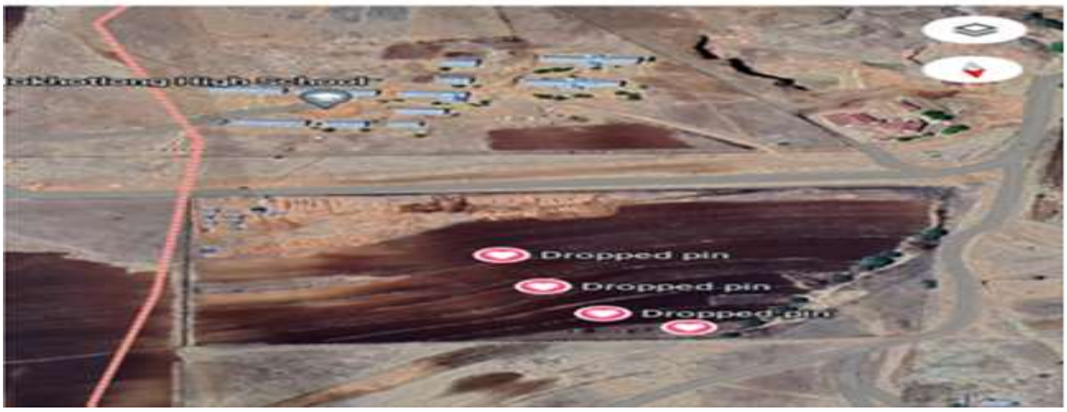

Fig 4: Soil sampling locations at Mokhotlong Agricultural Research (red dropped pins representing spots)

3.3. Pot experiment trial and layout

A pot experiment involving soils from mining sites at Kao, Letseng and a control from Mokhotlong town (Min of Agric) it was conducted at the National University of Lesotho. The greenhouse was used where growth conditions such as temperatures and moisture are controlled. The pots of 3 litres were filled with soil of 3 kg and there were three replications for each two cereals planted (fig. 5). Two cereal crops that were selected for plantation are sorghum (PAN 8625) and Pearl millet (Okashana 2). In each pot only three plants were allowed to grow, others were thinned and weeds thinned whenever available.

The pot experiment was conducted for period of three months then the samples were collected to be analyzed in the laboratory. There were no other agronomy measures that were taken except daily irrigation of the cereals. The plants in the greenhouse were monitored as daily by the researcher paying attention to any physiological changes.

						OK, L, R1, S1	OK, L, R2, S1	Ok, L, R3, S1	V8625, L, R1, S1	V8625, L, R2, S1	V8625, L, R3, S1
						OK, L, R1, S2	OK, L, R2, S2	Ok, L, R3, S2	V8625, L, R1, S2	V8625, L, R2, S2	V8625, L, R3, S2
						Ok, L, R1, S3	OK, L, R2, S3	Ok, L, R3, S3	V8625, L, R1, S3	V8625, L, R2, S3	V8625, L, R3, S3
						Ok, K, R1, S1	Ok, K, R2, S1	Ok, K, R3, S1	V8625, K, R1, S1	V8625, K, R2, S1	V8625, K, R3, S1
Ok, M, R1, S1	OK, M, R2, S1	Ok, M, R3, S1	V8625, M, R1, S1	V8625, M, R2, S1	V8625, M, R3, S1	Ok, K, R1, S2	Ok, K, R2, S2	Ok, K, R3, S2	V8625, K, R1, S2	V8625, K, R2, S2	V8625, K, R3, S2
Ok, M, R1, S2	OK, M, R2, S2	Ok, M, R3, S2	V8625, M, R1, S2	V8625, M, R2, S2	V8625, M, R3, S2	Ok, K, R1, S3	Ok, K, R2, S3	Ok, K, R3, S3	V8625, K, R1, S3	V8625, K, R2, S3	V8625, K, R3, S3
Ok, M, R1, S3	OK, M, R2, S3	Ok, M, R3, S3	V8625, M, R1, S3	V8625, M, R2, S3	V8625, M, R3, S3	Ok, K, R1, S4	Ok, K, R2, S4	Ok, K, R3, S4	V8625, K, R1, S4	V8625, K, R2, S4	V8625, K, R3, S4
Ok, M, R1, S4	OK, M, R2, S4	Ok, M, R3, S4	V8625, M, R1, S4	V8625, M, R2, S4	V8625, M, R3, S4	Ok, K, R1, S5	Ok, K, R2, S5	Ok, K, R3, S5	V8625, K, R1, S5	V8625, K, R2, S5	V8625, K, R3, S5

Fig. 5: Pot layout for the soils collected from Letseng, Kao and Mokhotlong Agricultural Research in greenhouse. OK represents Pearl millet Okashana 2, V8625 representing sorghum PAN 8625, M representing Mokhotlong, K representing Kao, L representing Letseng, S representing spot where the samples were taken

The soil samples were collected in two years which is 2022 and 2023 from all the sites for determination of heavy metals (Table 1). In 2022, soil samples were collected and analyzed for heavy metals, and then 2023 soil was collected for pot experiment mentioned in fig. 5. The soil samples were taken in same spots as the previous samples and GIS software was used to collect the coordinates that give the map in figure 1.

Table 1: Volumes of soil and soil sampling per site in 2022 & 2023

Sites	Study target	No. of Observation points	No. of samples
Kao Diamond Mining (2022)	Surface layer from depth of 0-20 cm & 20-40 cm	6	12
Letseng Diamond Mining (2022)	Surface layer from depth of 0-20 cm & 20-40 cm	3	6
Mokhotlong Agric Research (2022 & 2023)	Surface layer 0-20 cm for pot experimental in NUL	4	24
Letseng Diamond mining (2023)	Surface layer 0-20 cm for pot experimental in NUL	3	18
Kao Diamond Mining (2023)	Surface layer 0-20 cm for pot experimental in NUL	5	30
Kao Diamond mining	Maize and cabbage samples	5	5

3.4. Preparations and observations

Soil samples collected were air dried in the laboratory for analysis and were passed through a 2 mm mesh sieve. pH in H_2O and KCl was determined using calibrated pH meter. Organic matter was determined using Walkley-Black Titration method. Texture was determined using hydrometer method based on Stokes' law and CEC was determined using NH_4OAc extractable method.

For soil pH determination, 20g of soil samples were weighed in Erlenmeyer flask and two flasks were used as control. 50ml of distilled water was mixed with 20g of soil in the bottle then shaken for 15 minutes with a reciprocal shaker. Suspension of the soil was then let to settle. Thereafter, calibrated soil pH meter was used for each sample to measure pH in water of soil sample. After measuring soil pH in soil, 3.73kg of KCl was added in the soil mixed with water then shaken with a reciprocal shaker for 15 minutes. The soil pH was then taken for the soil pH in KCL and the KCl was added to soil solution to test for the presence of exchangeable aluminium.

For CEC determination, 5g of air-dried soil was weighed and transferred into the beaker. 20ml Ammonium acetate was added to the soil in the beaker and the mixture was allowed to stand for 10 minutes. 100ml Erlenmeyer flask was mounted with funnel and a filter paper and the mixture was washed with ammonium acetate to pass through the filter paper and 100ml of the filtrate was collected. Soil residue was washed with 100ml of sodium acetate to saturate sites with sodium and the filtrate was discarded. The excess sodium was washed with 100ml of pure alcohol and the filtrate was discarded. Lastly 100ml of ammonium acetate was added and filtrate 2 was collected.

For texture analyses, 50g of soil was weighed and 50ml of sodium hexametaphosphate (dispersing agent) and 50ml of distilled water were added to the soil to allow the mixture to be well mixed and shakable. The shaker was used to mix the samples well where they would be transferred to the 1000ml measuring cylinder. The water was filled to the mark of 1000ml measuring cylinder then plunging was made. The hydrometer and thermometer recordings were made after two 2hours. The second reading was made in the same manner, using hydrometer and thermometer.

For Organic Carbon determination, 0.5g of sieved soil was weighed and 10ml of Potassium dichromate and 20ml of concentrated H_2SO_4 were added respectively, to the soil in a 250ml Erlenmeyer flask. Each sample was determined in duplicates including the blank sample. After 30 minutes, 150ml of distilled water was added for dilution then six drops of ferroin indicator was added in solution. 0.5 Fe^{++} was then added for titration of solution until reddish brown colour appeared.

For heavy metal (Cr, Cd, Pb, Co, Ni, Mn, Fe and Cu) determination, DTPA extraction method was followed. AAS analyst 500 model was used to determine the content of heavy metals in the digested soil samples. 20.0g of soil was weighed then transferred into 100ml polythene bottle. 40.0 ml of DTPA extractant was added in each bottle for both the soil samples and the blanks. Samples were shaken for 2 hours using reciprocal shaker and filtered so as to determine heavy metal contents of the soils using AAS analyst 500 model.

Soil microbes were identified in the Crop Science Laboratory in the National University of Lesotho. 0.5 g of soil was transferred into each test tube. The serial dilution technique was used which is classical microbiological procedure. Nutrient Agar was used for isolating bacteria and potato dextrose agar was used for fungi isolation. 10^{-5} for bacteria and 10^{-3} for fungi soil dilution were utilized for plate count. Fungi were purified by two rounds so as to identify the genus.

Plant samples of Pearl millet and sorghum from the pot experiment were analyzed in the Soil Science Laboratory of NUL. For heavy metal determination in plants, plants were grind using leaf powder grinding machine. 0.5g of ground plant samples were weighed in Erlenmeyer flask for analysis. A combination of different reagents such H_2SO_4, HCl and H_2O_2 were added in glass tubes for solubilization of minerals. The samples were than evaporated on hot plate at 320^oC to reduce contamination of the sample with substance in air, vessel walls as well as local environment and solution was cooled at room temperature and lastly deionized water was added. AAS 500 model was used to read all heavy metals in plant tissues.

3.5. Analysis of data

Data was analysed using SPSS software.   Linear regression analysis was computed to assess the relationship between individual heavy metal concentration in the vegetable samples and in soil. Means, standard deviation and coefficient of variation (CV %) were determined for each treatment.

$$CV = \frac{standard\ deviation}{mean}$$

Significance differences between treatments from the mining sites and control were estimated using one-way ANOVA followed by Pearson's test, where p values less than 0.05 were considered as significant ($P<0.05$) allowing only a 5% chance error in the results.

Single contamination index of each metal pollutant was calculated using the following equation to assess the pollution degree of each metal in the area,

$$P_f = C_f / S_f$$

where P_f is the single contamination index of heavy metal pollutant, C_f is sample concentration, and S_f is regional value (Chukwu et al. 2008; Egaspin 2002: Adelegan 2002; Lacutusu 2000) (Table 2).

Table 2: Soil contamination index of heavy metal pollutant

Index	Description
<0	Not Contaminated
0-1	Uncontaminated to slightly
1-2	Moderately contaminated
2-3	Moderately to highly contaminated
3-4	Highly contaminated
4-5	Highly to very highly contaminated
>5	Very highly contaminated

Permissible level of heavy metals in soil was determined according to Ksheminska et al. 2003; WHO 1996; Sayadi and Sayyed 2011; Osmani et al. 2015; Bowen 1979; Singh and Steinnes 1994; WHO 2007 (Table 3). The values in table 3, have been taken from literature since they are the ones that help to identify whether there is contamination or not.

Table 3: Allowable heavy metals levels in soil and crops

Name of metal	Soil (mg/kg)	Crops (mg/kg)
Zn	50	0.60
Ni	35	10
Cu	36	10
Mn	1000	15-100
Cd	0.8	0.02
Pb	10-30	2
Cr	100	1.3
Fe	-	640-2486
Co	8	-

CHAPTER 4

RESULTS & DISCUSSION

4.1 SOIL TEXTURE

The Letseng Diamond Mining soils are sandy loam which implies that there is high amount of sand compared to clay and silt percentages (table 4). Sand has high drainage capacity because of large spaces or pores between them as the result the retention or adsorption of heavy metals would be less. Table 4 shows that the soil texture of the soil from Kao Diamond Mining site is loam soil. Loam soil is good at adsorption and retention of heavy metals and most of metals are likely to retain at the surface. Table 4 shows that the texture of the Mokhotlong Agricultural Research site is clay loam. The clay loam soil has a higher potential to retain high amounts of heavy metals on its surface.

Table 4: Particle size analysis of soils from the study sites

	Clay %	**Silt %**	**Sand %**
Letseng Diamond mining	10.7± 0.5	20.6± 7.3	68.7± 7.1
Kao diamond mining	26.5± 4.0	48.7 ± 5.4	24.8± 3.0
Mokhotlotong Agricultural research	34.7± 7.0	22.6± 4.1	42.7± 5.2

Clay has the greater adsorption capacity of heavy metals therefore there are larger amounts of heavy metals that are being adsorbed on the clay minerals. This means that the cations have capacity to replace other cations since the replacement depends on the strength of the ions held against the ions to be adsorbed (Xie et al. 2023). Clay minerals both natural and modified forms have very good feasibility in getting rid of different toxic aquatic metal pollutants. Clay is defined as fine-grained natural raw material, which is an effective adsorbent to trace heavy metal ions (Uddin 2017). Once the natural clay is activated, the adsorption capacity increases and this process also rapidly increases very fast. This also indicates that it is very important to know the adsorption or retention capacity of the clay for heavy metals (Djebbar et al. 2012). Copper and Zinc follow the second order reaction kinetic which they are well fitted when being adsorbed on the clay mineral and also mainly natural clay could be used as an effective removal of adsorbed metals (Veli and Alyüz 2007).

The heavy metal adsorption by sand is critical because it can only be described in particular with interaction between surface functional group of silicates (sand) and the metal ions. It has been found out that sand can be applied to reduced amount of heavy metals in the environment but notably lower concentrations of heavy metals. In the monolayer, the heavy metal adsorption follow this sequence Pb>Cr>Cu>Zn (Awan et al. 2003). Letseng Diamond Mining has 60% of sand and surface-modified quartz sand can be applied which has adsorption mechanism that can get rid of heavy metals notably from aqueous solutions through surface complexation, and electrostatic and gravitational forces. But in the soil, it is unclear how sand actually adsorbs heavy metals (Wang et al. 2023).

Silt is much contained in the Kao Diamond mining than any other places because the soil in Kao Village is frequently disturbed during planting deposits from the mining site. It should be understood that adsorption and pH work hand in hand, one factor influencing the other either alternatively or correspondingly.

4.2. SOIL PH

Soil pH of Letseng Diamond Mining (LDM) is between 4.4 and 6.0 which means the soil ranges from being acidic to slightly acidic (Table 5). This means that the first depth from 0-20 cm has the potential to retain more of heavy metals, due to soil acidity. The pH of the 0-20 cm depth is similar to the other spots sampled across the gradient. There is less variation on the surface layer in LDM dispute different elevations and distance apart from each spot. The soil pH of the 20-40 cm depth goes as low as 4.4 which implies that the soil is acidic with in the area.

The surface soil (0-20 cm) of the Kao Diamond Mining (KDM) site (Table 5) has a soil pH of 3.9 which indicates that this soil sampled in Spot one is very acidic compared to spot 2 to spot 5. Spot 2 to 5 have soil pH values that are acidic in which most nutrients may be available in the surface soil. The control that is 50 meters away from the stream has a soil pH of 5.5 which is acidic and which most crops prefer for growth. The soil pH of subsurface layer (20 – 40 cm) in Kao Diamond Mining (Table 5) from spot 2 to spot 5 including the control, is acidic except spot 1 which has lower pH implying that selective metals will not be available. The soil pH from surface soil (0-20 cm) to subsurface soil (20-40 cm) is similar, with the soil pH being acidic. The data variation between the spots sampled is likely similar from both the surface and subsurface layers with different spots at different positions. This tells that the soil in the area is acidic and more metals will be available in the area of the study.

The surface soil (0-20 cm) of Mokhotlong Agricultural Research (MAR) has soil pH of 5.3 to 5.9 from site 1 to site 5 implying that the soil is acidic at this surface layer (Table 5). The soil at the MAR site has pH that is favourable for many crops and more nutrients and metals could be available at greater amounts if they exist in the area of the study. The soil is acidic with low variability when the mean is centered in MAR thus the pH values across all the spots sampled in the surface layer are more similar.

Table 5: pH KCl of the KDM, LDM and MAR study site.

	KDM	LDM	MAR
Spot 1 (0-20cm)	5.2	6.0	5.4
Spot 2 (0-20cm)	5.4	5.5	5.1
Spot 3 (0-20cm)	5.5	5.6	5.9
Spot 4 (0-20cm)	5.6	-	5.3
Spot 5 (0-20cm)	5.6	-	-
Control (0-20cm)	5.5	5.7	-
Spot 1 (20-40cm)	5.2	5.6	-
Spot 2 (20-40cm)	5.5	5.5	-
Spot 3 (20-40cm)	5.4	4.4	-
Spot 4 (20-40cm)	5.5	-	-
Spot 5 (20-40cm)	5.7	-	-
Control (20-40cm)	5.6	5.5	-

Soils from LDM, KDM and MAR are all acidic and thus the decreasing of soil pH and increase of ionic strength of heavy metals decreases with the adsorption of metal ions (Malandrino et al. 2006). Soil pH is a key parameter that has an impact on the metal ion speciation. At high pH values, the precipitates of Zinc and lead start to dissolve as hydroxyl complexes, and the allowable pH is found to be within the range of 3–10 (Abollino et al. 2003).

The plants that can grow well on the soil (pH of 3.9 spot 1 first layer in Kao Diamond Mining) are acid loving plants. The mean of Mn is only significant to mean of soil pH ($p<0.05$). Studies have concluded that increased amount of adsorbed Zn in soil significantly increases with soil pH, soil organic carbon and CEC (Desta et al 2021).

Iron is essential for plant growth therefore it becomes more available as soil pH is declining from <7.5. Soil pH dramatically influences heavy metal desorption (Zeng et al 2011). Soil pH is directly related to solubility and availability of heavy metals in the soil (Berbecea et al. 2011). The improved adsorption capability and declining of the bioavailability of Cd depend on soil pH since pH increases the negative charge on edge of the clay minerals, hydrated oxides, and organic matter (Gu et al. 2022).

4.3: SOIL ORGANIC MATTER (SOM) OF THE STUDY SITES

Letseng Diamond Mining (LDM) in surface soil (0-20 cm) has lower Soil Organic Matter (SOM) % in spot 1 and spot 2 has 1.42% and 1.90% respectively which are lower than spot 3 and the control soil that have high SOM (Table 6). The subsurface soil (20 to 40 cm) in spot 1 (Table 6) shows that the SOM is less in surface layer than the subsurface layer and this is because soil is sandy but other spots have lower SOM in the subsurface layer. Generally, the SOM decreases with the increasing soil depths because there are fewer organic materials decomposing on 20 to 40 cm depth. High SOM indicates that soil has high potential to restore more nutrients in the soil and bind soil very well.

Kao Diamond Mining (KDM) has lower SOM in soil than LDM and MAR (Table 6) and this shows that the fertility status of the soil here is low and potential of soil to hold more nutrients and metals is low. The surface soil (0-20 cm) in all spots has low SOM but variability around means is high in spot 2 and in control which shows the presence of low to high SOM. The subsurface soil (20-40 cm) has lower SOM (Table 6) thus indicating that SOM decreases as with the depth and the potential of soil to hold heavy metals will decreases with the soil depth. Variability in many spots sampled is significantly different when the mean is centred. Soil in KDM has lower SOM thus amounts of heavy metals would be found in lower quantities.

Mokhotlong Agricultural Research (MAR) has lower SOM in spot 1 to spot 3 while spot 4 has high SOM. Spot 4 has high SOM because in the area sampled previous year, fodder was planted compared to other spots sampled. The variability in spot 1 to spot 3 is very high which indicates that the gap between minimum value obtained and maximum value are very significantly different in thus resulting in higher standard deviation. It appears that residues are trapped at MAR spot 4 due to topography more than other spots which have slopes that are slightly gently.

Table 6: SOM % of KDM, LDM and MAR site

	KDM	LDM	MAR
Spot 1 (0-20cm)	1.61	1.42	0.13
Spot 2 (0-20cm)	1.00	1.90	0.85
Spot 3 (0-20cm)	1.10	3.61	2.18
Spot 4 (0-20cm)	1.17	-	3.09
Spot 5 (0-20cm)	1.04	-	-
Control (0-20cm)	2.41	3.72	-
Spot 1 (20-40cm)	.23	2.25	-
Spot 2 (20-40cm)	.16	1.29	-
Spot 3 (20-40cm)	1.44	2.80	-
Spot 4 (20-40cm)	.73		-
Spot 5 (20-40cm)	.63		-
Control (20-40cm)	1.97	3.13	-

LDM has more Soil Organic Matter percentage than KDM and MAR this is because the environment is not an agricultural land has it has its vegetation mostly Sehala-hala throughout the year which make good soils underneath. Soil organic matter is that small portion of the soil that has been made from decomposition of plant and animal residues. Soil organic matter (which was found with lesser percentages mostly low in areas studied) is essential sorbent and is a binding agent of sand particles that regulate the activity of Cu^{2+}, Cd^{2+}, Zn^{2+}, and Ni^{2+} in these sandy soils. The contribution of clay silicates to metals binding become more essential factor mostly when metal loading is high in comparison with soil organic matter content and thus allow higher adsorption of heavy metals (Weng et al. 2001).

Where there is less amount of organic matter, the adsorption capacity is reduced and the Cr activity is increased (Yu et al. 2023). The soil organic matter (appendix 2.3) has moderate positive correlation with Zn, Co, Ni, and Fe except Pb, which has a weak correlation. Organic matter has an effect on the retention of heavy metals and also strong effect on the cation exchange capacity and as buffer capacity. This means there is little mobility and availability of heavy metals when there is combination with organic soils (Olaniran et al. 2013). There are two forms of metals found in the soil, that of cationic (problem causing) and that of anionic forms which react with soil first making them negatively charged. The metal adsorption in the clay minerals rise in the following order: $Cr^{3+} < Cu^{2+} < Ni^{2+} < Zn^{2+} \leq Cd^{2+} \leq Pb^{2+} \leq Mn^{2+}$ (Abollino et al. 2003: Olaniran et al. 2013). During the decomposition of soil organic matter, some of the nutrient elements are released to the environment and become toxic if they already existed in that particular area (Sophn and Berg 2023).

4.4: SIGNIFICANCE OF CALCIUM TO EXCHANGE WITH HEAVY METALS

Letseng Diamond Mining (LDM) calcium found in soil from surface soil (0-20 cm) in spot 1 to control ranges from 75.7 to 98.7 ppm in soil (Table 7). Soil calcium is low in the first layer since normally Ca levels in soils exceeds 400 ppm. The subsurface soil (20 – 40 cm) in LDM from spot 1 to control ranges from 92.4 to 137. 9 ppm. The soil has low calcium. The acidic soil normally has low content of calcium. The lower content of calcium in LDM mean that heavy metals could have replaced it in the soil.

Kao Diamond Mining (KDM) from (Table 7) has soil calcium ranging from 2233.2 ppm to 3126.2 ppm in the surface soil (0 – 20 cm). These imply that in the first layer there is adequate calcium since KDM has high amount of silt. The clay soil can reach as high as 2100 ppm of soil calcium. The subsurface soil (20 - 40 cm) of KDM site has calcium ranging from 2605.2 ppm to 3315.7 ppm indicating that calcium is also adequate. The variability of means in between the spots sampled is different. The higher amount of calcium would allow more or high amount of heavy metals to exchange with calcium with different species of heavy metals.

Mokhotlong Agricultural Research (MAR) have calcium ranging from 2783.0 ppm to 3422.2 ppm in table 7. The results obtained (Table 7) shows that calcium is sufficient and reaction with other metals would be very easy. MAR is the agricultural land its calcium in soil is greater than LDM and KDM and plants are have low chances of having calcium deficiency.

Table 7: Soil calcium levels of 3 experimental sites

ppm Ca^{2+}	KDM	LDM	MAR
Spot 1 (0-20cm)	2233.2	79.3	2783.0
Spot 2 (0-20cm)	3126.2	75.7	3422.2
Spot 3 (0-20cm)	2702.5	98.7	2926.0
Spot 4 (0-20cm)	2954.5	-	3029.7
Spot 5 (0-20cm)	2480.5	-	-
Control (0-20cm)	2715.2	129.0	-
Spot 1 (20-40cm)	3315.7	106.5	-
Spot 2 (20-40cm)	2981.5	108.1	-
Spot 3 (20-40cm)	3034.2	92.4	-
Spot 4 (20-40cm)	3061.2	-	-
Spot 5 (20-40cm)	2605.2	-	-
Control (20-40cm)	2980.7	137.	-

Heavy Metals ions, such as Pb^{2+}, $Zn^{2+,}$ Cd^{2+}, Cr^{3+}, and Cu^{2+}, could be well immobilized by Calcium while the pH of the soil could be increased. For instance, where Zn^{2+} precipitates as Zn(OH)2, ZnCO3, ZnO, Zn3(PO4)2·4H2O and CaZn2(PO2)2·2H2O (Cruz-Guzmán et al. 2006). Again, Zn^{2+} can replace Ca^{2+}, that means cations respect the rule of lyotropic series. It ranks the cations based on their strength with which cation is adsorbed on the exchange sites. The cation with less strength of adsorption would be replaced by cation with higher strength of adsorption on the exchange sites during the reaction (Leontidis 2016). Organic cations exchange with the metals for adsorption sites on the clay sites with respect to their affinity (Calace et al. 2002).

The large amount of calcium adsorbed on the edge of clay sites, prevents accumulation and toxicity of heavy metals to be absorbed. Usually Cd causes some interference with the biological functions of calcium ions in plant and as a result calcium detoxifies Cd when it is in oxide form (Xie et al. 2023). Heavy metals are positive cations that can easily exchange with calcium on the edge of the clay soils where it is retained or adsorbed in particular.

4.5: SIGNIFICANCE OF MAGNESIUM TO EXCHANGE WITH HEAVY METALS

Letseng Diamond Mining (LDM) has magnesium that ranges from 603.5 ppm to 639.5 ppm in surface soil (0-20 cm) that can be in table 8. Magnesium in table 8 is adequate and would allow heavy metals to exchange on the sites of soils. LDM subsurface soil (20-40 cm) has the magnesium ranging from 588.5 ppm to 442.5 ppm which indicated that it is adequate. The means of magnesium on the spots samples are likely the same from table 8, dispute different topography and positions.

Kao Diamond Mining (KDM) has magnesium ranging from 657.2 ppm to 671.0 ppm in the surface soil (0 -20 cm) from table 8 which shows magnesium is sufficient in the environment. Magnesium is cation and has a potential to exchange with certain heavy metals in soil. The subsurface soil (20-40 cm) has 651.0 ppm to 698.0 ppm of magnesium. The magnesium in this layer is sufficient and it will have the exchange with some of heavy metals.

Mokhotlong Agricultural Research (MAR) has sufficient magnesium which ranges from 728.7 ppm to 751.5 ppm in table 8. The MAR soil has clay content ranging from 30% to

40% and adsorption capacity is high and heavy metals present could easily exchange with magnesium in soil. MAR has high amount of the magnesium compared to KDM and LDM of the study area.

Table 8: Soil magnesium levels of 3 experimental sites

ppm Mg^{2+}	KDM	LDM	MAR
Spot 1 (0-20cm)	665.7	639.5	739.2
Spot 2 (0-20cm)	671.0	603.5	751.5
Spot 3 (0-20cm)	667.0	607.0	748.2
Spot 4 (0-20cm)	657.2	-	728.7
Spot 5 (0-20cm)	671.7	-	-
Control (0-20cm)	660.5	637.5	-
Spot 1 (20-40cm)	692.2	593.2	-
Spot 2 (20-40cm)	680.7	588.5	-
Spot 3 (20-40cm)	651.0	642.5	-
Spot 4 (20-40cm)	698.0	-	-
Spot 5 (20-40cm)	678.0	-	-
Control (20-40cm)	687.0	637.3	-

Magnesium has a tendency to alleviate the heavy metal toxicity in the soil mainly when it has the same radii with certain elements in soil (Rengel et al. 2015). Mg solubility decreases with increasing equilibrium pH and thus reducing the heavy metal contamination in the soil. It is further noticed that heavy metal toxicity can be reduced by the addition of magnesium in the soil (Zhang et al. 2020)

4.6: SIGNIFICANCE OF POTASSIUM WITH HEAVY METALS EXCHANGE

Letseng Diamond Mining (LDM) has the soil potassium (table 9) ranging from 1802.1 ppm to 5011.5 ppm in the surface soil (0 – 20 cm). The amount of potassium in the environment is adequate and there is high potential that there is high exchange of potassium with heavy metals in the environment. The subsurface soil has adequate potassium ranging from 2957.7 ppm to 4623.0 ppm (Table 9) and the control has higher potassium compared to other spots that have been sampled in both layers. LDM has high amount of sand so the trend of potassium down the depth could not be differentiated.

Kao Diamond Mining (KDM) has the potassium ranging from 1499.5 ppm to 5445.0 ppm (Table 9). The subsurface soil (0 – 20 cm) has the adequate potassium and the control has more of potassium in the soil. The subsurface soil (20-40 cm) has the potassium ranging from 1644.0 ppm to 3702.5 ppm which is adequate in soil and has higher potential to exchange with other cations such as heavy metals species.

Mokhotlong Agricultural Research (MAR) has very high amount of potassium ranging from 6109.0 ppm to 9163.7 ppm (table 9). The reason why K is higher in MAR is because there was fodder planted where the samples were collected. MAR has high amount of potassium compared to KDM and LDM of the study sites.

Table 9: Soil potassium levels of 3 experimental sites

ppm K^+	KDM	LDM	MAR
Spot 1 (0-20cm)	1499.5	4393.0	6109.0
Spot 2 (0-20cm)	2216.5	3417.7	7170.2
Spot 3 (0-20cm)	2256.7	1802.1	7698.2
Spot 4 (0-20cm)	3263.5	-	9163.7
Spot 5 (0-20cm)	3020.7	-	-
Control (0-20cm)	5445.0	5011.5	-
Spot 1 (20-40cm)	1644.0	3981.5	-
Spot 2 (20-40cm)	2672.0	4240.7	-
Spot 3 (20-40cm)	1870.7	2957.5	-
Spot 4 (20-40cm)	1830.5	-	-
Spot 5 (20-40cm)	2453.0	-	-
Control (20-40cm)	3702.5	4623.0	-

In some studies it is indicated that heavy metals in the following descending order, Ni > Cu > Zn > Cd could affect the uptake of primary minerals N,P,K in the soil. Ni is the most serious of all, and has the biggest impact of these on the uptake of N,P,K (Daghan 2013). K^+ homeostasis, transportation, and acquisition have important role in plant survival and Potassium in soil appears as a free or absorptive bound cation (Shamsi et al. 2008). Mainly under higher toxic heavy metal stress, K^+ loss from plant cells is very dramatic and can also be termed as K^+ efflux or electrolytic leakage which has a role in killing the cells that activate enzymatic activity (Schlenker 2014). K^+ efflux has essential role in stimulating catabolic processes that further assist in having “metabolic” energy for adaptation, repair, and restoration of plant’s natural health only when it is in moderate stress conditions from the heavy metal toxicity (Dhiman et al. 2022). The displacement of K^+ follows the sequence of Pb>Cu>Zn from the cation exchange sites and Potassium is reduced in soil cation exchange capacity because changes of K behaviour is let by heavy metals levels (Tu et al. 2000).

4.7: SIGNIFICANCE OF SODIUM TO EXCHANGE WITH HEAVY METALS

Letseng Diamond Mining (LDM) has the soil sodium ranging from 2350.2 ppm to 8261.9 ppm (Table 10) from the surface soil (0 – 20 cm). The amount of sodium is considered adequate in table 10 and that says it will allow high possible cation exchange of the heavy metals with sodium. In the subsurface soil (20 – 40 cm), LDM has the soil sodium ranging from 4949.9 ppm to 9345.4 ppm which is sufficient in the environment. Soil sodium in table 10 increased with soil depth but with the control decreased with soil depth.

Kao Diamond Mining (KDM) has sodium in the surface soil (0 - 20 cm) ranging from 1246.5 ppm to 2553.2 ppm (Table 10), which indicates that soil sodium is adequate in the environment. This allows available heavy metals to exchange. The subsurface soil (20 - 40cm) has soil sodium ranging from 1424.7 ppm to 2665.7 ppm (Table 10).

Mokhotlong Agricultural Research (MAR) has soil sodium ranging from 1404.0 ppm to 2577.5 ppm which indicates that the soil has adequate sodium in soil. Sodium content in soil is significantly similar to both mining sites (table 10), with MAR site showing sodium content fluctuations. LDM has high amount of sodium in soil compared to MAR and KDM.

Table 10: Soil sodium levels of 3 experimental sites

ppm Na^+	KDM	LDM	MAR
Spot 1 (0-20cm)	2232.2	8261.9	2046.5
Spot 2 (0-20cm)	2553.2	2350.2	2577.5
Spot 3 (0-20cm)	2238.2	8887.6	2031.7
Spot 4 (0-20cm)	1873.2	-	1404.0
Spot 5 (0-20cm)	1549.7	-	-
Control (0-20cm)	1246.5	5380.0	-
Spot 1 (20-40cm)	2369.2	8487.2	-
Spot 2 (20-40cm)	2665.7	8389.6	-
Spot 3 (20-40cm)	2129.2	9345.4	-
Spot 4 (20-40cm)	1672.0	-	-
Spot 5 (20-40cm)	1589.5	-	-
Control (20-40cm)	1424.7	4949.9	-

The Pb^{2+} can easily substitute the position of Na^+ on clay sites of soil because cation replacement is linked to the ionic radius. Sodium medium ionic radius has impact on immobilization of Pb^{2+} (Chen et al 2023). Competition between cations and protons for binding sites means the adsorption of metal ions like Cu^{2+}, Co^{2+}, Ni^{2+}, Cd^{2+}, and Zn^{2+} is usually decreased first by low pH values and secondly by cation and proton competition for binding sites. The protonation on the surface of adsorbents increases the metal uptake capacity at lower soil pH and this mostly happens when there are anionic metal species such as CrO_4^{2-} (Gadd 2009). The speciation of heavy metals among chloride and oxide species is affected by the changes in the moisture in soil and sodium content. A reduction in sodium content will result in high percentage of lead, thus as Pb toxicity increases alternatively the sodium content in the soil is reduced in the soil thus Pb becoming more toxic (Durla et al. 1997).

4.8: THE ROLE OF CEC IN THE ADSORPTION OF HEAVY METALS

Soil Cation Exchange Capacity (CEC) in Letseng Diamond Mining (LDM) table 11 ranges from 41.20 meq/100g of soil to 45.83 meq/100g of soil (Table 11) in the surface soil (0 – 20 cm). The subsurface soil (20 – 40 cm) has the CEC ranging from 40 meq/100g of soil to 47.24 meq/100g of soil thus soil has an adequate CEC in soil and allows heavy metals replacement.

Kao Diamond Mining (KDM) has CEC in the surface soil (0 -20 cm) ranging from 5.45 meq/100g of soil to 11.10 meq/100g of soil indicating that the CEC is low (Table 11). The exchange of heavy metals with soil cations will be very low. The subsurface soil (20 – 40 cm) in table 11 has the CEC ranging from 6.19 meq/100g of soil to 11.59 meq/100g of soil and CEC is low.

Mokhotlong Agricultural Research (MAR) has the sufficient CEC ranging from 35.4 meq/100g of to 36.4 meq/100g of soil (Table 11). The CEC is adequate at MAR since it can all available heavy metals to replace other cation. LDM has high amount of CEC followed by MAR and lastly KDM in soil.

Table 11: Soil CEC levels of 3 experimental sites

CEC (meq/100g of soil)	KDM	LDM	MAR
Spot 1 (0-20cm)	9.70	42.50	35.40
Spot 2 (0-20cm)	11.10	42.20	36.45
Spot 3 (0-20cm)	9.73	42,56	36.04
Spot 4 (0-20cm)	8.14	-	37.71
Spot 5 (0-20cm)	6.73	-	-
Control (0-20cm)	5.42	45.83	-
Spot 1 (20-40cm)	10.30	41.63	-
Spot 2 (20-40cm)	11.59	40.77	-
Spot 3 (20-40cm)	9.25	42.49	-
Spot 4 (20-40cm)	7.27	-	-
Spot 5 (20-40cm)	6.91	-	-
Control (20-40cm)	6.19	47.24	-

It should be noted that CEC greater than 25 meq/100g soil is found only in organic soils, muck soil or clays since CEC have retention of nutrients and also potential of available nutrient supply for the plants (Zgorelec et al. 2019). The expected amount of CEC differs from textural classes, for sandy soils 3-5 meq/100g soil, for loams 10-15 meq/100g soil, silt loams 15-25 meq/100g soil, clay and clay loams 20-50 meq/100g soil and lastly for organic soils 50-100 meq/100g soil (LaBarge et al. 2012). The sorption capacities for Cd^{2+} and Zn^{2+} are similar to their CECs, suggesting that the ion exchange being a major binding mechanism for these ions. Heavy metals leaching from the mining dumping site have continuous distribution downstream by water and wind because these materials have a low sorption capacity for metal ions due to their sandy texture, low organic matter content but the lower CEC could be implying that the adsorption of heavy metals being high.(Kyziol 2002; Jung 2008).

4.9: CONTRIBUTION OF ZINC TO THE CONTAMINATION OF SOIL

Letseng Diamond Mining (LDM) in both layers/depths has very low amount of Zn in the soil (Table 12) and thus shows there is no contamination of Zn in the environment. The pollution index of zinc in LDM is < 0 which imply that there is no toxicity of zinc on the environment or soil.

Kao Diamond Mining (KDM) has Zn (mg/kg) mean ranging from 3.85 mg/kg to 23.37 mg/kg from both depths or layers which is very low (Table 12). The pollution index is below zero which implies that at KDM zinc does not impose any problem to the environment.

Mokhotlong Agricultural Research (MAR) has lower zinc ranging from 10.28 mg/kg to 21.06 mg/kg in table 12. MAR has low zinc in soil but the zinc found in MAR is higher than the one found in LDM and KDM.

Table 12: Zinc content in the soil of KDM, LDM and MAR sites

Zn mg/kg	KDM	LDM	MAR
Spot 1 (0-20cm)	3.85	1.08	21.06
Spot 2 (0-20cm)	12.50	0.84	13.90
Spot 3 (0-20cm)	9.47	0.92	12.65
Spot 4 (0-20cm)	16.25	-	10.28
Spot 5 (0-20cm)	12.35	-	-
Control (0-20cm)	23.37	1.62	-
Spot 1 (20-40cm)	4.77	0.95	-
Spot 2 (20-40cm)	9.47	0.61	-
Spot 3 (20-40cm)	7.12	0.65	-
Spot 4 (20-40cm)	11.12	-	-
Spot 5 (20-40cm)	7.65	-	-
Control (20-40cm)	6.92	1.83	-

The allowable or maximum permissible level of Zn in soil is 50 mg/kg (Olobatoke and Mathuthu 2016; Yu et al 2022). The contamination index of Zn to the environment in Letseng diamond mining is less than zero which implies there is no pollution of Zn on the Mining site. Zn is likely to be found in the Earth's crust but also regarded as an essential for plant protein but mainly in the function of protein synthesis. Zn dispute being essential it goes parallel with its toxicity to the environment (Hussain et al. 2022).

The cold weather conditions and wet springs seasons could bring zinc deficiency as well as high availability of phosphorus in soil where the pH is high (>7.0). Organic matter found in all sites is small and that also can also bring small zinc to be deficient in soil. (Noulas et al. 2018). Soils with high amount of calcium most of the time have low content of zinc in the soil due to the exchange and zinc cannot place calcium.

Excessive levels of Zn from its sources such as weathering of rocks and mining activities can affect the bioavailability and absorption of other metals as well by altering soil and aquatic microbial diversity (Hussain et al. 2022). For plant growth, Zn content in soil recommended for both Kao and Letseng mining Diamonds and for Mokhotlong Agricultural Research, Zn should be added in the soil to meet the crops' need for an element (Ahmad et al. 2012).

4.10: CONTRIBUTION OF COPPER TO THE CONTAMINATION OF SOIL

Letseng Diamond Mining (LDM) has a very low amount of copper in the soil in both layers (Table 13). The pollution index is below zero which means that the area of the study is not contaminated by copper. There is variability on copper found in different elevation or position with different spots means. There is no contamination of copper at LDM environment.

Kao Diamond Mining (KDM) has very low amount of copper at both depths (Table 13). The content of copper in this study area is very low implying that the mining has no little contribution of contamination of copper. There is no contamination at KDM for copper and this is because the place has low organic matter.

Mokhotlong Agricultural Research (MAR) has Cu mg/kg ranging from 26.91 mg/kg to 32.89 kg/mg at different spots (Table 13). There is high amount of copper at MAR that shows no contamination rather the sufficient amount of copper at MAR. MAR has high amount of copper compared to KDM and LDM.

Table 13: amount of copper concentration in KDM, LDM and MAR sites

Cu (mg/kg)	KDM	LDM	MAR
Spot 1 (0-20cm)	3.20	6.48	26.91
Spot 2 (0-20cm)	3.40	6.45	32.89
Spot 3 (0-20cm)	3.33	6.67	29.48
Spot 4 (0-20cm)	2.90	-	25.53
Spot 5 (0-20cm)	2.35	-	-
Control (0-20cm)	1.89	7.64	-
Spot 1 (20-40cm)	1.677	3.12	-
Spot 2 (20-40cm)	3.37	5.59	-
Spot 3 (20-40cm)	2.86	6.15	-
Spot 4 (20-40cm)	3.26	-	-
Spot 5 (20-40cm)	2.42	-	-
Control (20-40cm)	2.93	5.26	-

The allowable or maximum permissible level of Cu in the soil is 36 mg/kg (Olobatoke and Mathuthu 2016; Yu et al 2022). The contamination index of Cu to the environment in MAR is 0.18 which implies there is very slightl or no contamination of Cu on the agricultural land while LDM and KDM show that their environments are not contaminated. Cu is also potentially toxic above critical concentration range level however Copper (Cu) is an essential metal for human, animals and plants. Cu being toxic on environment reduces oxidative stress within plants through enabled production of reactive oxygen species (Shabbir et al. 2020). The Cu content in the soil is very low and plants would likely have the deficiency of copper if grown in all study areas.

The critical concentration range of Cu in soil is 2 to 50 mg/kg, in this case MAR and Kao would show deficiencies of Cu. Usually (>90%) of the dissolved Cu in soil is complexed with dissolved organic matter except for acidic soils (Oorts 2013). The soil pH is low that means the Cu toxicity is increased thus an increase in pH will definitely have the toxicity of Cu^{2+} ions high in soil (Ginocchio et al. 2002). High amount of silt at Kao Diamond Mining (Table 4.1) imply that the ability of the desorption of Copper ions is decreased when there is reduction of H^+ (when the soil is becoming more acidic) could raise the adsorption activity of Copper. Under the condition of strong acidity and low concentration of Copper, thus the adsorption capacity of Copper is lower than the desorption capacity (Xie et al. 2018)

4.11: CONTRIBUTION OF MANGANESE TO THE CONTAMINATION OF SOIL

The surface soil (0-20 cm) at Letseng Diamond Mining (LDM), has manganese ranging from 43.26 mg/kg to 133.70 mg/kg in soil (Table 14). There is no contamination in LDM but manganese should be controlled before the levels could be increased higher than critical level. The subsurface soil (20 - 40 cm) in LDM has manganese ranging from 59.40 mg/kg to 138.71 mg/kg in soil (Table 14). There is no contamination in the area due to manganese toxicity in Letseng.

Kao Diamond Mining (KDM) from both depths and layer in table 14 has very low manganese. There is no contamination of manganese in this area of study since the values of manganese does not even approach the allowable amount of manganese in soil.

Mokhotlong Agricultural Research (MAR) at all spots has very low manganese ranging from 52.05 mg/kg to 63.81 mg/kg (Table 14). There is no contamination of manganese at MAR since this place should be noted that many experiments are conducted here; manganese toxicity will immediately be resolved

Table 14: Concentration of manganese in soils collected from KDM, LDM and MAR

Mn mg/kg	KDM	LDM	MAR
Spot 1 (0-20cm)	13.25	88.75	58.26
Spot 2 (0-20cm)	26.25	133.70	63.81
Spot 3 (0-20cm)	26.87	43.26	63.32
Spot 4 (0-20cm)	34.18	-	52.05
Spot 5 (0-20cm)	29.68	-	-
Control (0-20cm)	33.338	58.70	-
Spot 1 (20-40cm)	26.57	85.68	-
Spot 2 (20-40cm)	28.51	126.44	-
Spot 3 (20-40cm)	28.32	138.71	-
Spot 4 (20-40cm)	35.27	-	-
Spot 5 (20-40cm)	31.21	-	-
Control (20-40cm)	31.25	59.40	-

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Manganese oxides are capable of oxidizing some metal cations such as Pb, Co, Cu, and Ni on Mn mineral surface (Caporale et al. 2016). The manganese obtained in study areas has possibility that it come from different source with other heavy metals except Cd which could be from the same source of Mn. The soil pH of the three soils is above 5.0, which indicates that the availability of manganese to the plants was increased because if the pH would be reduced thus saying the Manganese availability will be increased or becoming more available to the plants which might become toxic to them if keep accumulating in the soil. Manganese is essential in Chloroplast formation and photosynthesis (Humphries et al. 2016).

In this context, Manganese does not pose risk in Kao Diamond Mining but can slightly pose the risk at the Letseng Diamond Mining therefore it should be taken care of since its low content is due to rapid mobilization. Manganese is likely to become available at soil pH 5 to 6, thus at very low pH (<5) there will be accumulation of Mn becoming toxic to plants. Mn could also be used to oxidise the Cr at certain pH, the pH increases as manganese oxide decreases Cr in the soil (Feng et al. 2006).

4.12: CONTRIBUTION OF CHROMIUM TO THE CONTAMINATION OF SOIL

Letseng Diamond Mining (LDM) has sufficient chromium in soil and can be considered as slightly contaminated by chromium in the environment (Table 15). The presence of chromium in soil can also be toxic to plants once available in the environment. The level of chromium in both depths is likely similar at LDM

Kao Diamond Mining (KDM) has chromium in the surface soil (0 – 20 cm) ranging from 34.85 mg/kg to 76.90 mg/kg (Table 15). The KDM is slightly contaminated with chromium however in few years the contaminated level in KDM would be moderate here, if control measures are not taken into consideration. The subsurface soil (20 – 40 cm) has chromium ranging from 24.16 mg/kg to 94.10 mg/kg indicating that KDM is slightly contaminated with chromium. Spot 3 in subsurface soil shows that chromium concentration is nearly going to exceed the allowable amount of chromium in soil.

Mokhotlong Agricultural Research (MAR) has very low amount of chromium therefore it is not contaminated by chromium (Table 15). KDM has more chromium in the soil followed by LDM and lastly MAR with low amount of chromium in soil.

Table 15: Concentration of chromium in soils collected from KDM, LDM and MAR

Cr mg/kg	KDM	LDM	MAR
Spot 1 (0-20cm)	34.85	32.30	2.17
Spot 2 (0-20cm)	48.73	37.06	2.70
Spot 3 (0-20cm)	66.61	37.82	2.32
Spot 4 (0-20cm)	64.06	-	2.71
Spot 5 (0-20cm)	74.81	-	-
Control (0-20cm)	76.90	38.36	-
Spot 1 (20-40cm)	84.36	35.87	-
Spot 2 (20-40cm)	88.78	35.91	-
Spot 3 (20-40cm)	94.10	34.49	-
Spot 4 (20-40cm)	19.87	37.17	-
Spot 5 (20-40cm)	24.16	-	-
Control (20-40cm)	31.31	-	-

KDM has a higher Chromium concentration than LDM and MAR, this means the contamination level of Chromium in the soil is 0.5184 which implies that the place is very slightly contaminated at KDM. Chromium is easily absorbed by plant roots since it may involve in action of insulin, the metabolism of proteins , starch and also to mention the carbohydrates (Kapoor et al. 2022).

The maximum allowable concentration of Chromium in soil is 100 (mg/kg) and both sites (LDM and KD) which are main study sites which have or within the range where Chromium does not pose that much risk to the plant. Due to its toxicity, chromium when its hexavalent it must be eliminated or controlled as early as possible since it causes oxidative stress due to production of free radicals because it appears in variety of different oxidation states (Zulfiqar et al. 2023). Since Chromium is very low in the sites studied, because Cr (VI) has tendency to be decreased to Cr (III) in soil when organic matter content is high (Ertani et al. 2017)

4.13: CONTRIBUTION OF IRON TO THE CONTAMINATION OF SOIL

Letseng Diamond Mining (LDM) both depths have very low concentration of iron content in soil from table 16. There is no contamination of iron in LDM and the plants in this region might also show some deficiency of iron if they were to be planted.

Kao Diamond Mining (KDM) in table 16 has very low amount of iron content in soil. There is no contamination of iron rather there is higher deficiency of iron on this environment.

Mokhotlong Agricultural Research (MAR) has very low amount of iron in table 16 and it must be taken into consideration. KDM have high amount of iron compared to MAR and LDM from the soil samples analysed in the laboratory.

Table 16: Concentration of iron in soils collected from KDM, LDM and MAR

Fe mg/kg	KDM	LDM	MAR
Spot 1 (0-20cm)	17.55	14.09	73.18
Spot 2 (0-20cm)	22.42	12.81	37.55
Spot 3 (0-20cm)	47.35	28.26	57.37
Spot 4 (0-20cm)	83.45	-	51.60
Spot 5 (0-20cm)	77.52	-	-
Control (0-20cm)	180.20	104.36	-
Spot 1 (20-40cm)	57.22	16.40	-
Spot 2 (20-40cm)	43.12	27.91	-
Spot 3 (20-40cm)	80.82	94.41	-
Spot 4 (20-40cm)	129.87	-	-
Spot 5 (20-40cm)	89.27	-	-
Control (20-40cm)	196.07	82.72	-

Iron content in the mining sites is very low but in LDM and KDM has higher iron compared to MAR (should be noted that MAR is used to perform experiment and every year its planted) because of the soil type which contribute to immobilization of heavy metals (Dwevedi et al. 2017) . At MAR the iron content is low, likely because high calcium may have contributed to low iron. Iron is essential in photosynthesis and chlorophyll synthesis in the plant but its availability to environment control the distribution of plant species, restricts yield and also has greater influence on nutrition and food quality (Schmidt et al. 2020).

There is no or slightly contamination of Iron in mining sites studied except MAR which is the control to mining sites which are the main targets. The maximum allowance percentage of iron in soil is very high amount and depends on the soil colours and regions as well.

4.14: CONTRIBUTION OF LEAD TO THE CONTAMINATION OF SOIL

Letseng Diamond Mining (LDM) has high amount of lead in soil and can be considered as highly contaminated by lead in the environment in table 17. The presence of lead in soil can also be toxic to plants once available in the environment. LDM in both layers in table 17 have values exceeding 30 mg/kg which is the allowable amount of lead in soil and the control (apart from stream by 50 m) as well exceeding limits that have to be there in the soil

Kao Diamond Mining (KDM) table 17 from spot 1-5 has lead ranging from 71 mg/kg to 88.10 which shows that the surface soil (0 – 20 cm) is very contaminated. The subsurface soil (20-40 cm) shows that the amount of lead from many spots are still within allowable limits except. The presence of lead in soil can also be toxic to plants once available in the environment. The surface soil has more lead than the surface soil in Kao Diamond Mining and the area is high amount of lead.

Mokhotlong Agricultural Research (MAR) has sufficient lead level in soil and can be considered as no or slightly contaminated by lead in the environment table 17. KDM has high amount of lead followed by LDM and lastly MAR.

Table 17: Concentration of lead in soils collected from KDM, LDM and MAR

Pb mg/kg	KDM	LDM	MAR
Spot 1 (0-20cm)	71.00	51.73	3.76
Spot 2 (0-20cm)	76.80	60.88	4.27
Spot 3 (0-20cm)	79.10	64.28	4.47
Spot 4 (0-20cm)	88.03	-	5.07
Spot 5 (0-20cm)	85.16	-	-
Control (0-20cm)	86.52	70.06	-
Spot 1 (20-40cm)	93.93	71.07	-
Spot 2 (20-40cm)	25.81	74.71	-
Spot 3 (20-40cm)	28.72	75.12	-
Spot 4 (20-40cm)	31.70	-	-
Spot 5 (20-40cm)	43.66	-	-
Control (20-40cm)	42.60	82.91	-

Lead is very high in the Letseng Diamond Mining and Kao Diamond Mining this is because the maximum allowable concentration of Lead naturally is 10 mg/kg to 30 mg/kg in soil (WHO 2007). The mining operating trucks can have linkages of fuel and add lead to soil, this is because Lead can be added to the petrol through organo-lead (Needleman and Gee 2013). The Letseng Mining Diamond is heavily contaminated with lead in the study area and this implies that animals and also shepards living near the mining site are likely to suffer from lead which reduce growth and reproduction in plants and animals, and neurological impact in vertebrates (Siddiqua et al. 2022).

Kao Diamond Mining is moderately to highly contaminated by Lead and it should be noted that it is not essential at all because if, found in the environment it is still toxic, but at higher amount can even kill (Bouida et al. 2022; Ren et al. 2022). Mokhotlong Agricultural Research shows no contamination of Lead since its way lower than the allowable limit. Lead is highly dangerous to living organisms mainly microorganisms and plants since it destroys biological function such as morphological, physiological, and biochemical dysfunctions in plants as well as deactivation or damaging enzymes of organisms (Fahr et al. 2013).

4.15: CONTRIBUTION OF COBALT TO THE CONTAMINATION OF SOIL

Letseng Diamond Mining (LDM) has low amounts of cobalt table 18 in both layers which shows slight or no contamination except control (0 – 20 cm) and spot 3 (20 – 40 cm) which exceeds limits of 8 mg/kg indicating moderate to high contamination. The presence of cobalt in soil can also be toxic to plants once available in the environment.

Kao Diamond Mining (KDM) in both layer/depths has low amounts of cobalt showing that that there is no contamination in the area table 18. The presence of cobalt in soil can also be toxic (when it exceeds allowable limits) or beneficial to plants once available in the environment. The level of cobalt in both depths is likely similar at LDM

Mokhotlong Agricultural Research (MAR) has insufficient amount of cobalt in table 18 showing that there is no contamination in the area. LDM has high amount cobalt in the soil and followed by MAR and KDM

Table 18: Concentration of cobalt in soils collected from KDM, LDM and MAR

Co mg/kg	KDM	LDM	MAR
Spot 1 (0-20cm)	2.46	3.28	2.82
Spot 2 (0-20cm)	2.275	6.52	1.17
Spot 3 (0-20cm)	3.00	3.35	3.38
Spot 4 (0-20cm)	1.75	-	2.70
Spot 5 (0-20cm)	2.02	-	-
Control (0-20cm)	2.67	10.6	-
Spot 1 (20-40cm)	1.50	4.06	-
Spot 2 (20-40cm)	1.65	7.65	-
Spot 3 (20-40cm)	6.62	52.05	-
Spot 4 (20-40cm)	1.98	-	-
Spot 5 (20-40cm)	1.67	-	-
Control (20-40cm)	2.27	6.38	-

According to Barałkiewicz, and Siepak (1999), most world soils have an average cobalt concentration in soil of 8 mg/kg in average and this implies that Letseng Diamond Mining is slightly less contaminated by cobalt with contamination factor of 0.721. However, in one or two of the spots where the samples were taken, the maximum concentration of cobalt shows that there is contamination of cobalt. Organic substances easily adsorb Cobalt since it is positively charged and forms organic chelates. Adsorption has a big influence on the mobility of Cobalt when its reactivity is increased under oxidative reaction since Cobalt exists in soil as free metal and found in forms of ores (Ponti et al. 2009). MAR and Kao diamond Mining have little contamination of Cobalt and it could also be insufficient to plants if grown in that particular environment.

4.16: CONTRIBUTION OF NICKEL TO THE CONTAMINATION OF SOIL

The amount of nickel in Letseng Diamond Mining (LDM) shows that there is no contamination at both depths (Table 19). The availability of nickel in soil can also be toxic to plants when it exceeds limited amounts. The level of nickel in both depths is similar at Letseng Diamond Mining (LDM).

Nickel in table 19 has low amounts indicating that at Kao Diamond Mining (KDM) some spots show there is no contamination. However, nickel in control both depths and spot 3 (20 – 40 cm) show there is high amount of nickel in KDM implying there is also contamination that need to be taken care of before it can make more harm.

Mokhotlong Agricultural Research (MAR) has very high amount of nickel according to Table 19, except spot 3. MAR is contaminated by nickel and plants that can be grown on the environment, should be ones that require more nickel. MAR has amount of nickel in soil followed by LDM and lastly KDM

Table 19: Concentration of nickel in soils collected from KDM, LDM and MAR

Ni mg/kg	KDM	LDM	MAR
Spot 1 (0-20cm)	3.45	17.36	55.08
Spot 2 (0-20cm)	5.47	28.09	77.23
Spot 3 (0-20cm)	14.52	18.17	70.90
Spot 4 (0-20cm)	20.43	-	43.08
Spot 5 (0-20cm)	25.61	-	-
Control (0-20cm)	35.27	34.41	-
Spot 1 (20-40cm)	3.21	6.65	-
Spot 2 (20-40cm)	6.27	20.27	-
Spot 3 (20-40cm)	65.46	26.64	-
Spot 4 (20-40cm)	13.78	-	-
Spot 5 (20-40cm)	20.97	-	-
Control (20-40cm)	41.95	28.81	-

The permissible concentration of Nickel in soil is 35 mg/kg according to WHO (2007). However Letseng Diamond Mining shows very high contamination index or factor which implies the contamination already exists allowable limits of nickel in the soil. It should be noted that in lighter soils, concentrations of Nickel should be lower and the permissible concentration is 15 mg/kg (Barałkiewicz, and Siepak 1999). Hence, Letseng soil that has high amount of sand has slight to moderate contamination of Nickel. However, taking the maximum value of Letseng Diamond Mining, Nickel concentration, the contamination factor becomes 2.29 implying that the environment is moderately contaminated. Nickel is essential in growth of higher plants. It should be noted that Nickel can be found in soil in different forms which are adsorbed or complex on organic cation surfaces or on inorganic cation exchange surfaces (Iyaka 2011)

4.17: CONTRIBUTION OF CADMIUM TO THE CONTAMINATION OF SOIL

Letseng Diamond Mining (LDM) (Table 20) has cadmium that is slightly contaminating the site except spot 1 from surface soil (0 – 20 cm) which shows there is greater contamination of cadmium on the environment. In recent years the environment will be contaminated with cadmium since other places in the very same environment are contaminated.

Kao Diamond Mining (KDM) is contaminated with cadmium (Table 20) except spot 1 in the first that shows no contamination. Cadmium exceeded 0.8 mg/kg which is allowable limit in the soil from both layers.

Mokhotlong Agricultural Research has no contamination of cadmium from both depths and shows there are no sources of cadmium at all in the environment. KDM has high amount of cadmium followed by LDM and then followed by MAR that has no amount cadmium at all.

Table 20: Concentration of cadmium in soils collected from KDM, LDM and MAR

Cd mg/kg	KDM	LDM	MAR
Spot 1 (0-20cm)	.00	1.15	0.00
Spot 2 (0-20cm)	1.15	0.71	0.00
Spot 3 (0-20cm)	.95	0.68	0.00
Spot 4 (0-20cm)	1.55	-	0.00
Spot 5 (0-20cm)	1.4	-	-
Control (0-20cm)	1.65	0.43	-
Spot 1 (20-40cm)	1.75	0.24	-
Spot 2 (20-40cm)	1.85	0.31	-
Spot 3 (20-40cm)	2.55	0.35	-
Spot 4 (20-40cm)	1.05	-	-
Spot 5 (20-40cm)	1.20	-	-
Control (20-40cm)	1.45	0.38	-

Kao Diamond Mining is facing extremely severe contamination of Cadmium, which needs to be taken care of as early as possible. Cd presence in the soil is non-essential and rather toxic to the environment in which it exists. The concentration of Cd in soil solution usually relies on the adsorption of Cd on the surface of soil particles and thus adsorption of Cd reduces the extent of mobility of the Cd in the soil (Abt & Robin 2020). The adsorption and desorption reactions of Cd regulate mainly the bioavailability and mobility of Cd on the surface of the soil colloids.

The factors that regulate the accumulation of Cd in soil and its availability are: organic material/organic matter, texture, Fe and manganese (Mn) oxide and hydroxides, Zn, Carbonates, chlorine, Cation Exchange Capacity and lastly the soil pH (Contaminants et al. 2019). Cadmium despite being very toxic is also carcinogenic and teratogenic which have effects of diseases such as Itai-itai which is short term disease but very acute. It also has effect on microorganisms such as bacteria where-by it will affect its diversity, community structure and its metabolic functioning (Duan et al. 2020)

4.18. SOIL MICROORGANISMS

4.18.1. SOIL BACTERIA FROM NUTRIENT AGAR

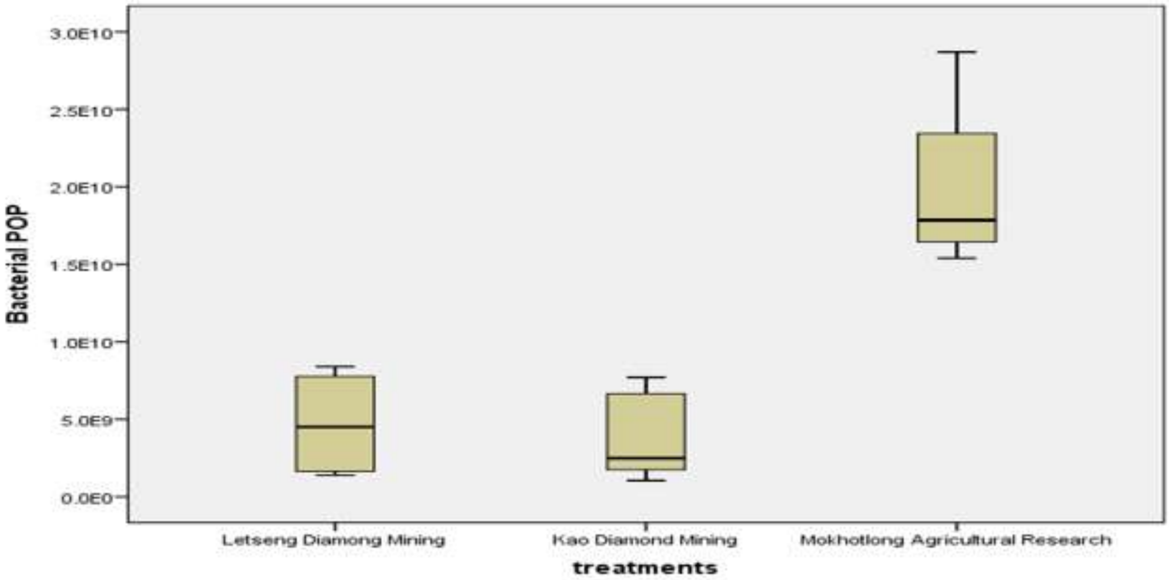

Fig. 6: Bacteria in soil community population detected from 3 different sites in nutrient agar's

Kao Diamond Mining is having the least bacteria population mainly because of the contamination level of some of heavy metals including the Cadmium in the area which might be the main cause of population decline. Letseng Diamond mining has less but not least bacteria population and the reason is because of Lead toxicity and Chromium toxicity on the environment. MAR has no heavy metal contamination therefore the bacterial population is very high and also the environmental conditions favour the microbes. Heavy metals mainly Cadmium have a very serious effect on genetic structure of bacteria and thus saying there are higher amount of genetic diversity in the environment that in group. Lead and Cadmium normally destroy cell membranes and totally damage the structure of DNA (Naz et al. 2022).

In Letseng Diamond Mining and Kao Diamond Mining the bacteria population is very low and main contributors of low bacteria population in the mining sites are heavy metals contamination therefore there might also be other factors such as organic matter that may be very low in the mining sites that is habitat of soil microorganisms. Few population of bacteria that are studied, could have developed the mechanisms to adopt in heavy metal conditions, becoming tolerant of Bacteria (Jarosławiecka and Piotrowska-Seget 2022).

The bacterial identification can be done in more advanced laboratories to further identify and select the bacteria's that are there in the plate agars. Therefore, only few populations were studied to observe the effect of heavy metals on the environment and to quantify the extent of contamination.

4.18.1. FUNGI DETECTION IN CONTAMINATED SOIL

Fungi were detected in all sites but the issue was the microscope used did not function so well. The fungi were detected in the potato nutrient agar making some moulds where some were darker in colour, some were green to grey colour. Cadmium, lead and some other heavy metals at higher concentrations actually restrict antioxidant enzymes to work properly. Toxic heavy metals could easily be phyto-stabilized by Mycorrhizal fungi in the environment (Bano and Ashfaq 2013). Fungi usually develop set of response mechanisms that will restrict the toxicity of the metal to their cells so that it will adapt to higher concentration of heavy metals.

Most fungi depend heavily on extracellular enzymes for nutrient acquisition, and some of fungi such as Saprotrophic fungi are highly sensitive to heavy metals and those enzymes usually are a target of heavy metal toxicity (Baldrian 2010). It was found that the fungus Trichoderma harzianum can easily survive where there is excessive Cd, Pb and Cu (Mohammadian et al. 2017).

4.19. PLANT ANALYSES FOR SORGHUM AND PEARL MILLET

Table 21: Heavy metal contents in sorghum and Pearl millet grown on soils from the three study sites under a pot experiment

		Cu mg/kg	Zn mg/kg	Fe mg/kg	Pb mg/kg	Ni mg/kg	Co mg/kg	Cr mg/kg	Mn mg/kg	Cd mg/kg
Sorghum for Letseng	Mean	8.43	26.23	105.33	7.00	6.43	14.33	13.63	7.73	2.86
	Std. Dev	5.42	1.85	56.87	2.85	1.23	.80	1.53	1.81	.90
Pearl millet for Letseng	Mean	2.03	23.20	85.83	8.53	6.43	13.20	10.06	4.53	2.10
	Std. Dev	.80	2.170	12.55	1.10	.66	.818	.92	1.10	.10
Sorghum for Kao	Mean	1.88	28.88	181.64	5.86	8.88	12.46	15.84	21.42	1.62
	Std. Dev	1.54	5.05	123.67	.58	1.60	.65	6.58	22.60	.25
Sorghum for Mokhotlong	Mean	2.17	26.67	217.40	8.05	7.22	12.80	2.80	8.90	.00
	Std. Dev	.59	1.57	76.39	2.88	1.46	.92	.66	3.43	.00
Pearl millet for Kao	Mean	.80	29.44	180.08	8.26	8.56	11.98	7.72	9.16	1.22
	Std. Dev	.43	2.04	88.03	.90	.740	.622	1.90	4.61	.19
Pearl millet for Mokhotlong	Mean	1.75	26.00	149.55	8.70	8.07	10.15	2.95	7.55	.00
	Std Dev	.79	2.42	47.66	.32	.79	3.68	1.40	1.48	.00

The permissible value of Cu in plants is 10 mg/kg according to (WHO 2007), therefore none of the crops tested in the pot experiment exceeded allowable limit of Cu in plants. However, Letseng Diamond Mining Sorghum, Kao Diamond Mining and MAR have similar amount of Cu level in plants approaching the permissible level, respectively. Copper assists to control plant diseases. The ingestion of Copper through food chain in plants can bring chronic exposure (Coelho et al. 2020; La Torre et al. 2018).

Zn permissible value is 0.6 mg/kg. All crops tested had Zn concentrations above the recommended concentration of Zn in plants. The plant molecular regulators are very sensitive in controlling adaptation of plants when Zn is in excess and also plants adapt to shortage of Zinc supply by enhancing the zinc uptake capacity (Assunçao et al. 2010). Zinc in soil is found to be very low but the plant is having higher amounts. This is because of the mechanisms the plants use to extract the elements from the soil and should be noted that crops tested are all heavy feeders. Zinc uptake is affected by numerous factors such as Sulphate and other soil factors such as soil pH. Zinc affinity becomes strong with soil pH. Thus the adsorption also increases with increasing Zn to become more available to plants (Recena et al. 2021). Zn is essential in physiology and biochemistry of plants however excessive amount could cause oxidative damage by allowing the levels of reactive radicals (Natasha et al. 2022). Iron is high in sorghum, because MAR, Kao Diamond Mining and Letseng Diamond Mining soils have 217, 180 and 105 mg/kg in the plant respectively. The values of concentrations are very low implying that the plants have absorbed less Iron from soil and they do not pose risk to plants and very low to accumulate in the food system. Zinc play many roles in the plant enzymes such as cytochromes mainly in transport chain through electrons, and it is needed in higher amount by plants and its deficiency will cause the plant to trample (Rout and Sahoo 2015).

It appears that the sorghum and Pearl millet experiments under a pot trial, they have both similar lead (>2 mg/kg) in the plants and it should be noted that the permissible concentration of Pb in the plant is 2 mg/kg according to WHO (2007). Letseng had soil contamination of mainly lead (table 17) that could have the significant contribution on affecting the crop directly since it exist allowable limit in soil. Lead presence to the environment is very toxic and should be taken care of because where it is available, it inhibits plant growth as it can be seen in Figure 8 where Letseng plants mainly sorghum and Pearl Millet did not grow well or had stunted growth and also including other heavy metals present in soil also contribute to poor growth of plants.

In Kao village, it can be clearly seen in Figure 8 where stunted growth of sorghum is happening; it seems that the plants are undergoing high stress (Sharma and Dubey 2005). It should be noted that lead, unlike other heavy metals, is easily absorbed by plants and this has a very big effect on plant growth. Plants to protect themselves should have strong cell wall to prevent accumulation of lead inside the cell (Nas and Ali 2018).

Nickel was found to be lower than maximum permissible level in the plants thus imposing no risk to any plants grown in the environments. Nickel strongly plays essential role in metabolic reaction in plants (Ahmad and Ashraf, 2011). In case where Nickel exceeds permissible values in plants, its effects are reduced or inhibition of growth, chlorosis induction, necrosis and wilting will happen (Bhalerao et al. 2015).

All sites and plants tested showed that cobalt is below the range expected in healthy plants and the permissible level of cobalt in the plant is 15-25 mg/kg in the plants (Hu et al. 2021). Cobalt like potassium helps in retardation of senescence of leaves whilst it makes seeds become tolerant to drought. It is mainly important in synthesis of B12 and inhibits nitrogen deficiency since it is needed by several enzymes that take part in nitrogen fixation (Roychoudhury and Chakraborty 2022). Co-induced Fe deficiency is the main effect of growth inhibition since Co effect on growth is very additive to that of Fe deficiency. Kao Diamond Mining and Letseng Diamond showed that the level of cobalt is low but when cobalt is high in soil, Co in the roots and leaves can disrupt a range of metabolic processes due to competitive interactions with other micronutrients cations (Liu et al. 2000).

Soils in all sites exceeded the allowable concentration and the permissible level of chromium in plants is 1.3 mg/kg according to (WHO, 2007). Chromium in the soils of the two mining sites was high and its concentration in the plants also shows that the plants are contaminated with Chromium. Chromium toxicity was observed in the plants because with chromium toxicity the major symptoms observed are decreased root growth, stems, and plants leaves but some other factors might cause deficiency also (Shadreck and Mugadza 2013). Chromium is a serious threat to the plants because at some certain stages it has totally killed the plants beside restricting the germination of seeds by triggering the protease activity (Saud et al. 2022). Chromium in the environment should be controlled totally.

It must be noted that the level of Manganese in plants is way too low in sites studied. The highest Mn content in the crops was 21.4 mg/kg for sorghum in the Kao village farm and small (4.1 mg/kg) in Cabbage sample that was taken from Kao Village. Manganese is very important in chloroplast formation, photosynthesis and nitrogen metabolism. Plants absorb heavy metals through plant root exudates as a soluble component or solubilized fractions. Therefore, the presence of heavy metals in soils imposes an impact on growing plants (Arif et al. 2016; Bahiru and Yegrem 2021)

The amount of cadmium found in the plants is as high as 20 mg/kg and the allowable amount of Cd in plants is 0.02 mg/kg according to (WHO, 2007). Pearl millet and sorghum had high amounts because they had more than 0.02 mg/kg of Cadmium in their tissues and thus Kao

Village has very high contamination of Cd. Unlike other heavy metals discussed, Cadmium toxicity in crop plants has a potential to decrease uptake and translocation of nutrients and water. There are also other effects that Cadmium toxicity could cause to crops such as; higher oxidative damage, plant metabolism interruption, and plant morphology and physiology restrictions (Haider et al. 2021). Cd has high potential to be absorbed by plant roots because it has similar behavior like Calcium. Cd accumulation in plants brings about severe phytotoxicity to plants, thus restricting the efficiency of phytoremediation. To correct Cd toxicity effect in plants, Ca could be used since both of them have similar chemical properties (Huang et al. 2017).

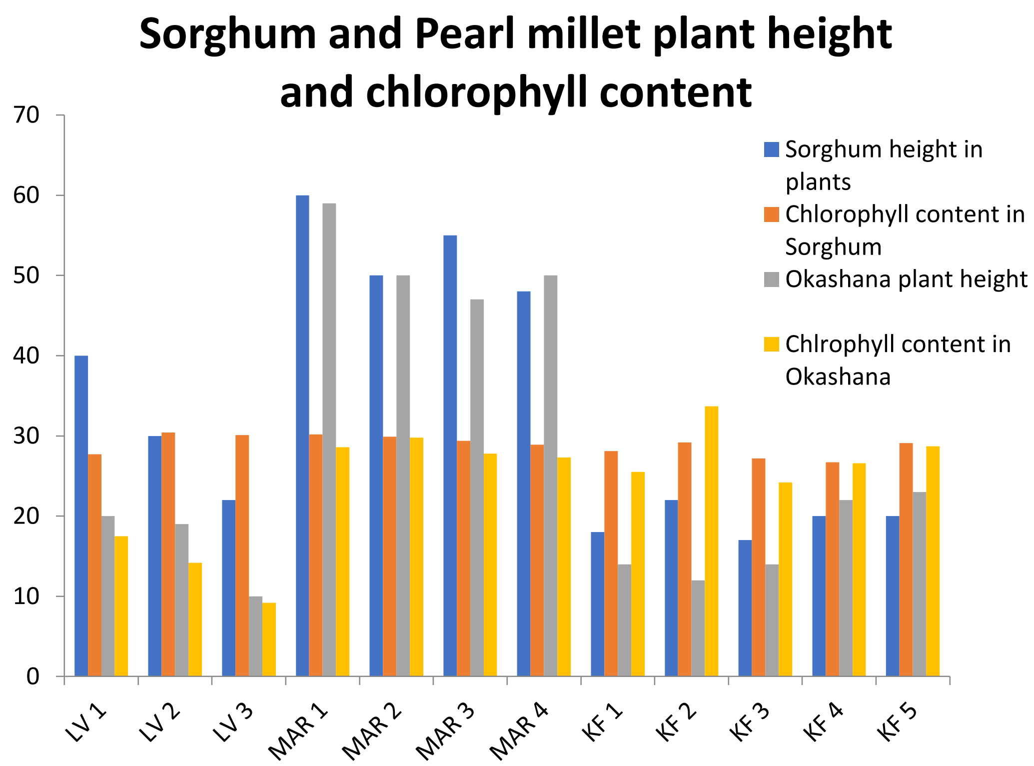

Fig 7: Plant height and Chlorophyll analyses in pot experiment for 3 sites LV, KF and MAR repesents Letseng Village, Kao Farmlands and Mokhotlong Agricultural Research repectively

The bar chart clearly gives a summary of how contaminants have affected the plants in the pot experiment that was conducted in the green house (Fig 7). MAR really performed well during the two to three months growth period. Both crops tested in the mining sites performed so badly because their plant height was increasing but very slowly because of the low chlorophyll measured since it was very low to allow the plant to grow to the optimum.

Heavy metals in the soil compete with other needed nutrients; therefore the growth of the plants is affected, as this could clearly be seen with the soils from Kao and Letseng Diamond Mining sites. At MAR, growth is high and rapid due to fact that there were no contaminants. Plant growth was not good in two mining sides but control which is Mokhotlong Agricultural Research shows that the growth was successful (Fig 8., Fig 9., and Fig 10).

Fig 8. Kao Diamond Mining plant height in pot experiment

Fig 9. Letseng Diamond Mining height in pot experiment

Fig 10. Mokhotlong Agricultural Research plant height in pot experiment for sorghum and Pearl millet

The heavy metals present in the plants in larger quantities are considered to be most toxic and could also affect human life by entering the food chain. The plant height in Kao Diamond Mining is very small, which implies that there is a severe contamination of Cadmium mostly and other heavy metals that affect plant growth and also affect the photosynthesis of plants. In the Letseng Diamond Mining (from pot experiment), what is mostly affecting the plant growth or height specifically is the Chromium which is non-essential as well as lead. It should be noted that at low concentration, non- essential heavy metals truly affect the plant growth, ecosystems, micro-organisms as well as human life (Bharti and Sharma 2022).

The behaviour of Cadmium in water is highly soluble and this makes it to be even more harmful to the plants even at lower concentrations. The symptoms that have been observed in Kao Village plant samples as well as with the pot experiment are stunting and chlorosis of plants, indicating the effect of Cd toxicity. The main reason of stunting in growth and chlorosis is due to Cd restricting the plant roots to absorb nutrients that that they require for growth (Dağhan 2013).

These none essential elements affect plant growth because of the inhibition of plants to absorb the nutrients in the soil and growth is highly affected at that moment. In both Letseng and Kao Diamond Mining, the chlorophyll content is very low implying that the synthesis of glucose and oxygen is very low, which results in the plants not growing to the needed optimum growth due to the effects of heavy metal concentrations in the soil (Zhang et al. 2020).

Chapter 5

CONCLUSION

Letseng Diamond Mining is mainly affected by high concentrations of Chromium in the soil as well as lead in the soil. These heavy metals are non-essential and their presence in the environment affected the plants in pot experiments since there was chlorosis and stunted growth compared to the experiment with soils from MAR which is the control site, that showed an excellent growth of Pearl millet and sorghum. The contamination level of heavy metals in Letseng Diamond Mining site is moderate with low microbial population and the soils from Letseng had higher amount of sand compared to Kao Diamond Mining and MAR. It was found that the soils are acidic which allow rapid movement or mobility of heavy metals and also had direct impact on heavy metal availability. Sorghum and Pearl millet have absorbed significant levels of heavy metals from the soils into their roots.

Kao Mining is highly contaminated with Cadmium and lead mostly and as a result, there is stunted growth and chlorosis observed in the plants. The microbial population in the soils from the mining sites are lower compared to the control soil from MAR, which is very low which indicates that the population is also reduced or affected. The sources of these heavy metals in the Kao Diamond Mining village are mainly from the tailings that reach the sampled points through the atmospheric dust. The study discovered that these heavy metals reach the sampled points through the wastewater from the industry, especially during the heavy rains which flow down the stream.

Recommendations

It is recommended that this study should be taken further because there is a need for identifying the heavy metals species since not all the species of heavy metals impose risk to the environment. The mining sites must have to research more on how to recover the contaminated sites.

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