CSIR COLLEGE OF SCIENCE AND TECHNOLOGY

IMPACT OF AUTOMOBILE WASTE ON HEAVY METAL

ACCUMULATION AND MICROBIAL ACTIVITY AT TWO SELECTED

AUTO-MECHANIC WORKSHOPS IN KUMASI, (ASHANTI REGION) -

GHANA

SAMUEL KWASI BENEFO
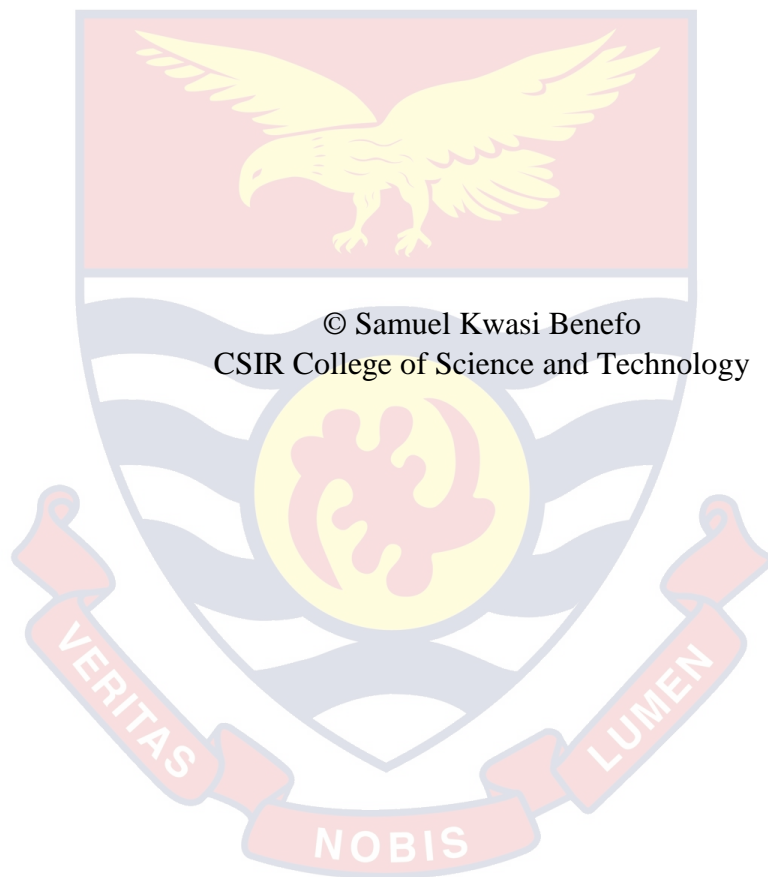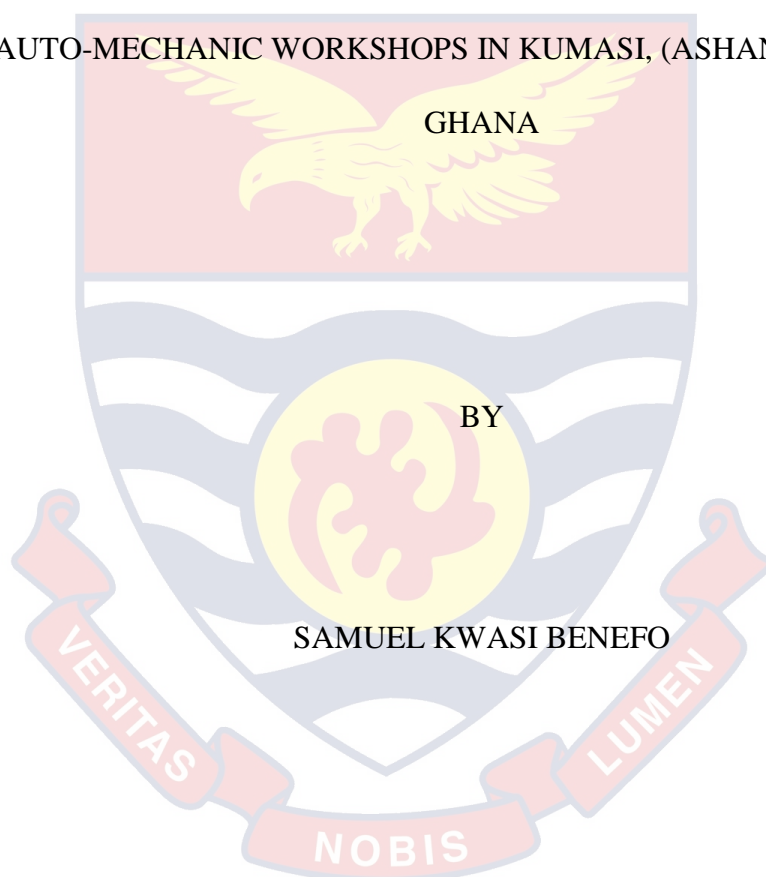
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BY

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Thesis submitted to the Department of Soil Resources Management of the
CSIR College of Science and Technology, in partial fulfilment of the
requirements for the award of Master of Philosophy degree in Soil Health and
Environmental Resources Management.

SEPTEMBER 2020

DECLARATION

Candidate's Declaration

I hereby declare that this thesis is the result of my own original research and that no part of it has been presented for another degree in this College or elsewhere.

Candidate's Signature…………….………….………Date …………...…………

Samuel Kwasi Benefo

Supervisors' Declaration

I hereby declare that the preparation and presentation of the dissertation were supervised in accordance with the guidelines on supervision of thesis laid down by the CSIR College of Science and Technology.

Principal Supervisor's Signature……………….….Date ……………………

Name: Dr. Edward Yeboah

Co- Supervisor's Signature………………………… Date …………………….

Name: Dr. Emmanuel Dugan

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ABSTRACT

Auto mechanics have over the years played a major role in the transport industry in Ghana by providing services to many road users including; heavy duty cars, tankers, salon cars, commercial vehicle among others. In the process of discharging their duties, they intentionally and unintentionally deposit various waste from vehicles on the soil within the workshops and the adjoining fields. A survey was conducted to assess the impact of automobile waste on soil heavy metal levels and soil microbial activity. Two auto mechanic workshops at Edwenase and South Suntreso in Kumasi Metropolis (Ashanti Region of Ghana) were considered. The experimental design used was Randomized Complete Block Design. Composite soil samples were taken at depths of 0-10 cm and 10-30 cm from the upper part through to the middle part and to the valley bottom and analyse for heavy metals including; Pb, As, Zn and Cd. Selected parts of maize and plantain at the adjoining fields were also randomly sampled for heavy metal accumulation analysis. Soil samples were subjected to microbial activity analysis using the basal respiration method. The levels of Zinc recorded in the soil sample were highly above the FAO acceptable limit of $3.5 – 6$ mg/kg. The levels of arsenic at South Suntreso were within the FAO acceptable limit of 20 mg kg-1. Pb of plant samples had 0.03 mg/kg level. Zinc levels emerged the highest at 15.47 mg/kg in plant biomass. At South Suntreso, the valley bottom recorded the highest microbial activity of 65.24 $mgCO_2$/kg soil/day. After the study, it was established that there were some traces of heavy metals in soil and plant samples at the two study sites; and the presence of the heavy metals had an effect on soil microbial activities.

KEY WORDS

Auto mechanic workshop

Heavy metal uptake

Microbial activity

Significant difference

Soil pollution

Soil samples

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DEDICATION

To my late Father, Mr. Yaw Benefo, for his love, care, security and above all
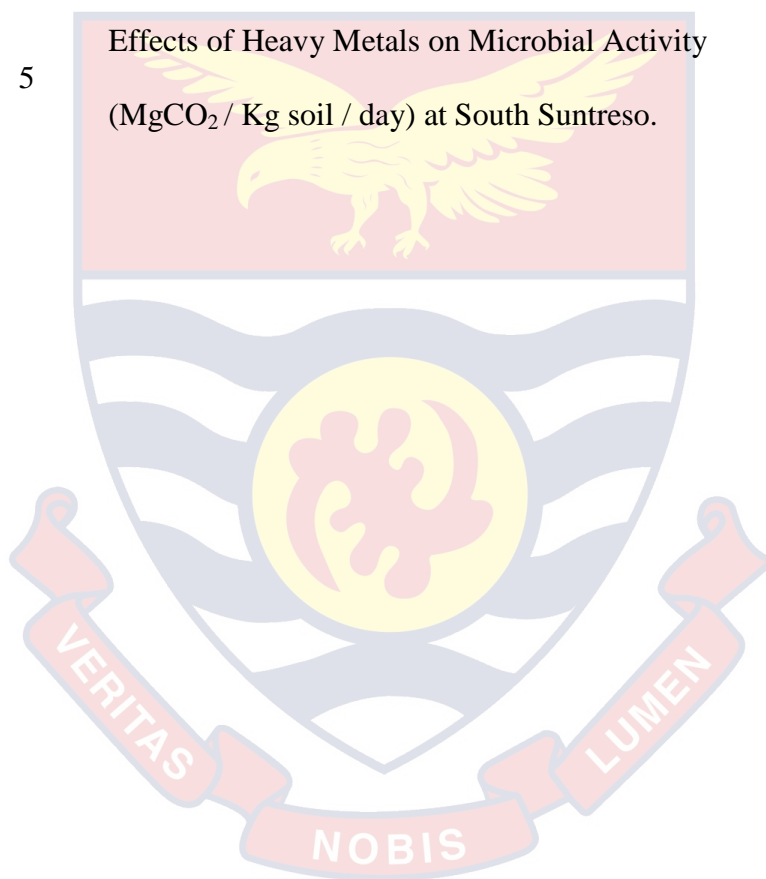
his dream to push me up the education. Rest well, Daddy.

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LIST OF ACRONYMS

FAO	-	Food and Agriculture organization
CSIR	-	Council for Scientific and Industrial Research
SRI	-	Soil Research Institute
EDTA	-	Ethylenediaminetetraacetate acid
SPSS	-	Statistical Package for the Social Science
CEC	-	Cation Exchange Capacity
WHO	-	World Health Organization
AAS	-	Atomic Absorption Spectroscopy
Cd	-	Cadmium
Pb	-	Lead
Zn	-	Zinc
As	-	Arsenic
Ppm	-	part per million
ANOVA	-	Analysis of variance

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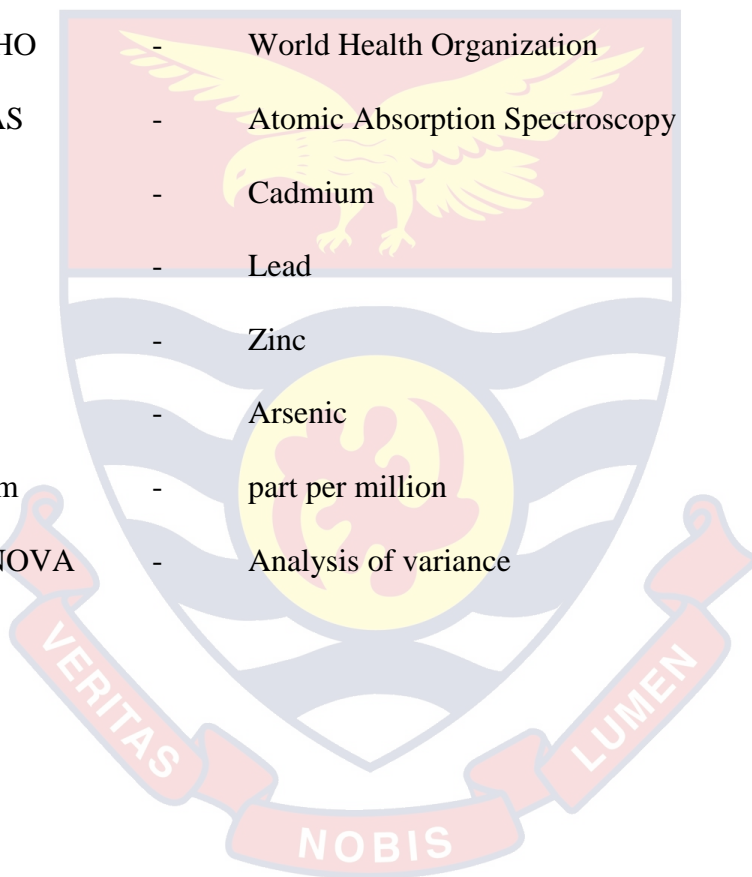
CHAPTER ONE

INTRODUCTION

Heavy metals, such as Fe, Mn, Cd, Hg, and Co, are introduced into soil on a large scale through human activities, such as municipal solid waste, mining activities, use of fertilizers and pesticides in agriculture, and industrial emissions. Soil pollution arises when the amount of these heavy metals becomes marginally higher than that of the background which in-turn cases deterioration the ecology and environment (Masindi & Muedi, 2018).

In Ghana, auto mechanics /artisans in the automobile industry play a major role in the maintenance of vehicles. Research has revealed that the activities of artisans in the automobile industry generate varieties of waste materials. They include; disposal of dirty engine oil, vehicle battery acid water, carbide from welding, metal scraps. (Marahatta, Gautam, Paudel & Yadav, 2018). The automobile wastes are made up of a mixture of chemicals including petroleum hydrocarbons, chlorinated biphenyls, heavy metals etc. Heavy metals are released into the soil through the disposal of engine oil, engine and gear box overhaul, battery charging, welding and soldering, automobile body work. Other sources include spraying, painting, and combustion processes (Pam, Sha'Ato, & Offem, 2013). The heavy metals that are found in the above-mentioned waste include; copper (Cu), lead (Pb), cadmium (Cd), zinc (Zn), manganese (Mn) and nickel (Ni), which may lead to risks in human health and the environment especially when they enter the food chain (Adelekan & Alawode, 2011).

When heavy metals are released into air, water and soil, it results in huge environmental problems. The presence of heavy metals has been

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considered as useful indicators for assessing contamination in surface soil, sediment and dusty environment. Where such activities are not properly monitored and regulated, they may give rise to higher levels of metals and hydrocarbons in the environment. Soil being a universal receptor; contains large amounts of heavy metals and hydrocarbons with different concentration ranges depending on sources, both natural and anthropogenic (Adelekan & Alawode, 2011).

Background to the Study

Studies have shown that used engine oil is less viscous than unused oil; and when disposed off into the soil, it adsorbs to the soil particles, reducing porosity and therefore reduces aeration of soil (Shukry, Al-Hawas, Al-Moaikal, & El-Bendary, 2013). Pollution of the soil with used engine oil leads to a build-up of metals in the soil and the eventual translocation into plant tissues such as leaves (Vwioko, Anoliefo, & Fashemi, 2006). Sejkorová, Hurtová, Glos & Pokorný, 2017) found that used engine oil easily migrates into the environment and eventually seeps into water bodies. Waste oil has severe adverse environmental and health impacts and is more toxic than virgin oil due to the presence of degraded additives and other contaminants (Meybeck & Redfern, 2013).

Cultivation of crops on abandoned fields around most auto-mechanic workshops is common in Kumasi. The presence of these heavy metals negatively affects soil's physicochemical properties and crops grown around the workshops. (Adelekan, 2011).

Soils surfaces found within auto-mechanic workshops are normally compact which favour erosion due to reduced activities of soil organism, soil aeration and water infiltration. Metals on the surface of such soil may be carried by runoff water or leached into the groundwater. Higher levels of metals and hydrocarbons in the environment can give rise to; water pollution, soil contamination, reduction in the activities of living organisms. The study was focussed at Edwinase and South Suntreso cluster of auto mechanic workshops and its surroundings. The auto-mechanics at the study site consist of; Welders, Auto electricians, Vehicle sprayers, Straighters and Vulganizers. These wastes may also affect the soil physico-chemical properties which may also have an effect on crops grown around the workshops.

Among the heavy metals, Cd pollution is the most serious situation. It retards the biological activity of soil micro-organisms, which affects crop yield and quality of agricultural produce. Again, Cd is difficult to be removed from the soil. It remains in the soil for a long period, which then accumulates in soil or plant edible parts. It is also a source of poison to animals and humans through the food chain (Nriagu, Wong, Lawson, & Daniel, 1998). Too much intake of Cd may result in prostate cancer, kidney cancer, and other diseases (Fan, 1995).

Microorganisms are essential component of soil; their activities reflect the intensity and trend of various biochemical reactions in soil. The changes in microbial community settings is a biological indicator for evaluating the quality and status of soil (Emily, Mailund, Hein, Schauser, & Schierup, 2009). Soil respiration facilitates exchange process between soil and CO_2 in the atmosphere. Its intensity is an important indicator for measuring total

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microbial activity (Smirnova et al., 2014). According to Neilson (2003), Soil enzyme serves as a biological chart of soil quality and as an indicator for evaluating soil fertility. It is normally used to observe the pollution and fertility of soil, and to check the quality of soil and the environment (Ramesh, Nibi, Kurup, Mohan, Aiswarya, Arsha, & Sarang, 2017). The reaction of catalase and urease in soil is susceptible to heavy metals, and they can in-turn reflect the harmful effects of heavy metals on soil microorganisms.

Many reports indicate that heavy metals interfere with the biochemistry of diverse group of microorganisms isolated from their natural environments (Igiri et al., 2018). However, information relating to the sensitivity of whole soil bacterial communities to heavy metals is not common. Microorganisms do not live in isolation but in complex biological communities within which exist complex interactions arising from biotic and abotic influences (Petersen and Klug, 1994; Chefetz, Hatcher, Hadar, & Chen, 1996). It is difficult to relate single species behaviours to the overall ecosystem. Since traditional methods of microbial ecology require that organisms from an environment be cultured in the laboratory before they can be identified and studied, monitoring the effect of pollutants on the activities of the overall communities poses a great challenge, because less than 10% of microorganisms from any environment is culturable (Bååth, Frostegård, & Fritze, 1992; Díaz-Raviña, Bååth, & Frostegård., 1994; Frostegard & Baath 1996).

Soil microbiology plays a vital role in nutrient cycling, organic matter decomposition and soil remediation (Johns, 2017; Kelly & Tate, 1998) and contributes heavily to the resiliency of a disturbed soil system. In a disturbed

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soil, microorganisms regulate restoration processes which make it possible for floral and faunal life to recolonize the soil.

Release of hydrocarbons into the environment whether accidentally or due to human activities is a main cause of water and soil pollution (Adeniji, Okoh & Okoh 2017). Soil contamination with hydrocarbons causes extensive damage of local system since accumulation of pollutants in animals and plant tissue may cause death or mutations in human. Considering the contribution of microorganisms to the soil, the potential effects of crude oil contamination on soil microbiology are important ecologically.

Moreover, petroleum hydrocarbons are absorbed by plant roots and accumulated. From there, they could potentially get into human's body through the food chain and pose a threat to human health. Metals accumulate in ecological food chain through uptake at primary producer level and then through consumption at consumer levels and plants roots are the primary contact site for heavy metal ions. Whereas, in aquatic systems, plant body exposed to these ions and heavy metals are absorbed directly to the leaves due to particles deposited on the foliar surfaces (Wuana & Okieimen, 2011).

It is well known that a lot of soil bacteria and fungi can utilize petroleum hydrocarbons as a carbon source. At the same time, some aboriginal microbes have gradually adapted to the long-term oil contaminated soil and developed a superior community which can make use of oil contaminants through special substrate enrichment. Therefore, bioremediation of oil contaminated soil has broad prospects because of its low cost, no secondary pollution, processing in situ and so on (Yan, Wang, Qu, & Li, 2013). Soil enzymes fulfil a critical role in many biochemical processes and may serve as

process level indicators of soil quality. Dehydrogenase is a widely studied oxidoreductase. Based on evidence including correlation with oxygen consumption, an assay of dehydrogenase activity may serve as a measure of general metabolic activity of the soil microbial community (Wolińska & Małachowska-Jutsz, & Matyja, 2012). Dehydrogenase activity has been suggested as a potential assessment tool for contaminated soils (Małachowska). Its close association with living microorganisms in soil may make dehydrogenase a sensitive responder to disturbance of, or amendments to, the soil system, and therefore a potential indicator of soil biological quality (Wolińska). Elevated dehydrogenase activity in a crude-oil-contaminated soil may indicate the stimulation of the soil microbial community in its biological role in the breakdown of contaminants.

Biodegradation by natural populations of microorganisms represents one of the primary mechanisms by which petroleum and other hydrocarbon pollutants can be removed from the environment and is cheaper than other remediation technologies (Adeniji, 2017). The success of oil spill bioremediation depends on one's ability to establish and maintain conditions that favor enhanced oil biodegradation rates in the contaminated environment. Numerous scientific review articles have covered various factors that influence the rate of oil biodegradation: Biodegradation rates of petroleum hydrocarbons and the development of oil degraders depend on the oil composition and environmental conditions (Dubinsky, Conrad, Chakraborty, Bill, Borglin, Hollibaugh, & Andersen 2013). The basic information such as residual oil concentration, population density of hydrocarbon degrading microorganisms and the biodegradation potential, environmental factors such as pH,

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temperature etc. are some of the key factors to be considered for bioremediation. One important requirement is the presence of microorganisms with the appropriate metabolic capabilities. If these microorganisms are present, then optimal rates of growth and hydrocarbon biodegradation can be sustained by ensuring that adequate concentrations of nutrients and oxygen are present and that the pH is between 6 and 7 (Adeniji).

Microorganisms have the following advantage on the degradation of heavy oil pollution: There are widely distributed numerous microorganisms with a strong reproductive capacity, adaptability and faster metabolic rate. The studies on degrading heavy oil pollution by microorganisms have shown that the degradation efficiency of heavy oil pollution has been able to reach about 70% under certain conditions (Tahseen et al., 2016).

There are many kinds of microorganisms that degrade oil pollution, and they are mainly screened from different oil-contaminated environment. Generally, dozen or several kinds of different bacterial species can be obtained in the same environment (Adeniji, 2017). Some microorganisms that are found to degrade heavy oil pollution include: Pseudomonas, Bacillus, Ochrobactrum, Acinetobacter, Proteobacteria, Firmicutes, Microbacterium etc. (Liu, Wu, Lin, et al., 2019). Bacteria have been found to be the microorganisms actively involved in the degradation of organic pollutants and many bacteria mentioned above are capable of effectively reducing the polycyclic aromatic hydrocarbons (Hamad, Moubasher, Moustafa & Mohamed, 2021).

Statement of the Problem

Artisans in the automobile industries generate a lot of waste at the workshop. The used lubricants mentioned contaminate the soil and its environs (underground water and crops). These lubricants contain heavy metals. The metallic elements are considered systemic toxicants that are known to induce multiple organ damage, even at lower levels of exposure (Ali, 2013).

Some of the used oils drain directly into the soil, others move through the drainage lines into streams. The levels of heavy metal accumulation in the soil at the selected sites are unknown although they have been in operation for a long time. At Edwinase, mechanic activities on the field started 10years ago while South Suntreso study area started about 19 years ago. It is suspected that crops grown downslope of the workshop may absorb some of the toxic elements (heavy metals) from the waste. It may also affect the soil's physico-chemical properties. Work has been done by Sadik et al. (2015) at Edwenase on the heavy metal accumulation in only the soil inside the workshops and its effects on the soil's physico-chemical properties. It didn't probe further to the effects on the plant biomass and soil microbial activity. There is limited information on the above subjects to help safeguard our soil and also consume healthy crops grown from such areas. Information that will be gathered will also contribute to knowledge especially to soil and crop researchers.

Also, the levels of microbial activities in the polluted soils at the selected study sites are not known. These need to be identified as far as bioremediation is concerned. The experimental fields have a number of clustered auto mechanic workshops. It is therefore possible that these shops

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will contribute heavy metal contaminants to the experimental site thereby polluting the soil as well as uptake by existing plants. It became imperative to monitor the levels of these heavy metals and nutrient status of soil and to access the microbial activities leading to the degradation of the oils and other contaminants. Crops from such fields if contaminated can cause a lot of health related issues to consumers of those crops. These metallic elements are known to be systemic toxicants that cause multiple organ damage, even at lower levels of exposure (Ali, Khan, & Sajad, 2013).

Research Objectives

The aim of this study is to determine the impact of automobile waste in the soil environment. Specifically, the study sought to:

1. Examine the variation of heavy metals from the workshops downslope.

2. Examine the levels of heavy metals in crops at the adjoining fields to the workshop

3. Measure the levels of microbial activities in heavy metal polluted soils.

Research Questions

1. Are there high levels of heavy metals downslope from the workshop?

2. Are there high levels of heavy metals in plants at the adjoining fields to the workshop?

3. What is the effect of heavy metals on soil microbial activity?

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Hypothesis

1. There are higher levels of heavy metals in soils around auto-mechanic workshops than soils at the control plots.

2. Plants at the adjoining fields to auto-mechanic workshops have higher levels of heavy metals than plants at the control plots.

3. Soils around auto-mechanic workshops have lower soil microbial activity than soils at the control plots.

Significance of the Study

This research will provide timely alert and environmental policy direction activities of auto-mechanics in Ghana; It is important for the policy makers to impose some form of restrictions on the activities of auto-mechanics. This research will provide the levels of heavy metals accumulation at the study areas which will determine the kind of policies to be implemented at all auto-mechanic workshops. It is also important to safeguard the lives of people who will be consuming crops from such fields. If the levels of heavy metals in the soil and plant biomass is known, consumers will be informed whether to patronize crops from such fields or not.

Also, food security is key in crop production. The fact that farmers are expected to produce crops to feed the nation does not necessarily mean food security should be overlooked. This research, through its recommendations, will provide ways that will maximise food security at the auto-mechanic workshops. Again, the study will inform policy makers on the needed preparations to be made before allowing artisans to site their various workshops. Although a number of people are aware of the activities auto-

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mechanics/artisans at the two fields of study, the level of contamination in the soil as well as the existing plants are not known. It became necessary for this information to be generated to aid policy makers to come out with the best remedies to safeguard the soils found around auto mechanic workshops.

Delimitation

This research focused on the heavy metal levels in soils, plant tissues and soil microbial activity around selected auto-mechanic workshops in Kumasi. The research was limited to Edwinase and South Suntreso cluster of auto mechanic workshops all located in Kumasi. The study would have been expanded to some other clustered of auto mechanic workshops, but due to time, financial and other constraints.

Limitations

Due to the emergence of covid-19 pandemic, most of the farmers and the artisans at both study sites were not ready to help in terms of giving vital information due to the spread of virus and the social distance protocols instituted by the Government of Ghana. During the collection of soil samples, most of the artisans tried to resist the researcher to pick the soils samples due to superstitious beliefs. The researched was also constrained financially which limited the work at only two auto-mechanic cluster of workshops instead of at least three in Kumasi and its surrounding areas.

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Organisation of the Study

This study is organized into five main chapters. Chapter One presents the introduction to the study, outlining the problem statement, the justification, the objectives of the study in its present form, research questions and their corresponding hypotheses. Chapter Two presents a review of relevant theoretical and empirical literature. It discusses the concepts outlined in the study citing relevant literature sources. Chapter Three presents the methodological framework of the study outlining data sources, tools for data collection and data analysis. It also gives a brief background to the study area. Chapter four presents and discusses results of the study organized along its specific objectives. Chapter five, which is the final chapter presents a summary of the study findings, conclusion and recommendations.
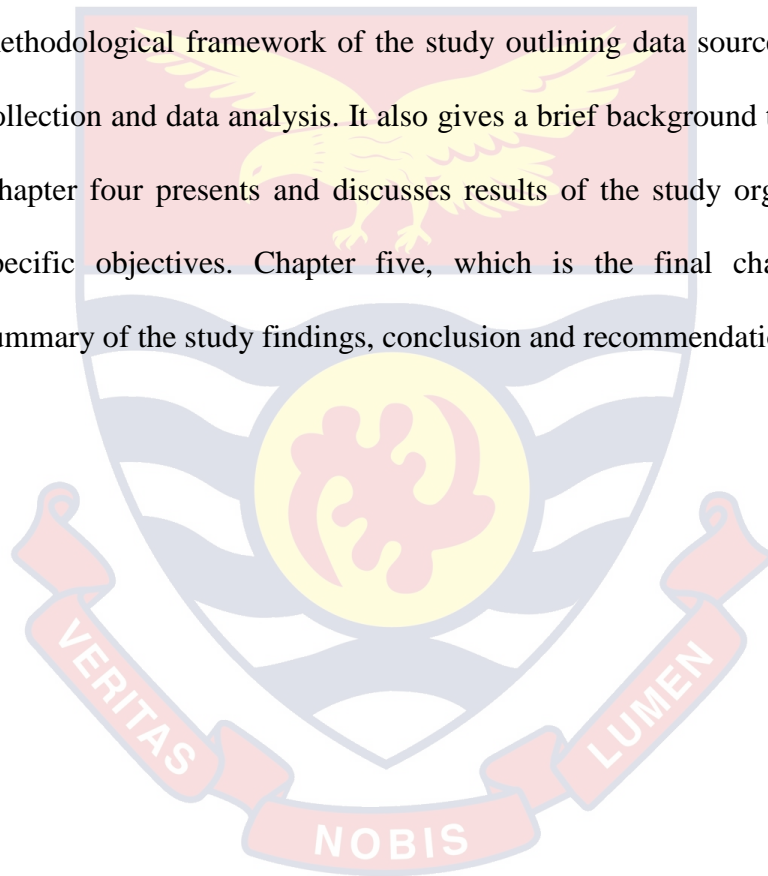
CHAPTER TWO

LITERATURE REVIEW

This chapter highlights on the existing literature linked to the study. It includes; the general idea of automobile waste in Ghana and other parts of the world. It also touches on heavy metals associated with auto mobile waste and its effects on the soil and existing plants found around the workshops. The chapter also highlights on micro-organisms in the soil and their importance to the ecosystem. It again highlights the effects of heavy metals on the activities micro-organisms in the soil.

Automobile Waste

Automobile waste such as used-lubricating oil, used grease, contaminated petrol and diesel, acid and calcium carbide are generated from auto-mechanic workshops that repair and service auto-mobiles. These wastes are not properly disposed of in many developing countries, since there is hardly any stipulated law regulating their location, activities and operations. (Ogundapo & Tobinsin, 2018) These workshops create and discharge automobile wastes into the environment (i.e. Soil and water bodies and sometimes air so that it is no longer pleasant or safe to use. These wastes are possible and available sources of environmental pollution if not properly disposed off and managed. The common method used by operators of automobile workshops to get rid of these wastes is by pouring them on the ground which either penetrate into the soil or washed by rainstorm into drains. When used-lubricating oil is poured down drains or onto the ground, the oil

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persists on the surface. Oil that is not properly disposed off accumulate the soil (Ogundapo & Tobinsin).

Disposal of automobile waste

Haphazard disposal of engine oil into gutters, drains, open vacant plots and farms is a common practice in Nigeria especially by motor mechanics. Heavy metals such as Vanadium, Lead, Aluminium, Nickel and Iron usually below detectable limits in unused lubricating oil have been reported to give high values (mg/kg) in used oil (Vwioko et al., 2006). These metals may be retained in soils in the form of oxides, hydroxides, carbonates, exchangeable cations, and/or bound to organic matter in soil. There is a growing global concern because of the multiple health risks to animals and humans following exposure (Zurayk, Sukkariyah, & Baalbaki, 2001).

Oil-contaminated soils are of environmental concern because they are unsuitable for agricultural and recreational uses and are potential sources for surface and ground water contamination. Oil polluted soil could also become unsuitable due to a reduction in the level of available plant nutrients or a rise to a toxic level of elements such as manganese. The heavy metal content of oil-contaminated soil imposes metabolic disorders and growth inhibition on most plant species (Ikhajiagbe, Ogedegbe & Anoliefo, 2013).

Heavy Metals

Heavy metals are chemical elements mostly with density greater than 4 g/cm3 found in all kinds of soils, rocks and water in terrestrial and freshwater ecosystems. The very low general level of their content in soils and plants as

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well as the definite biological roles of some of them makes them microelements (Lacatusu, 2000). They occur in typical background concentrations in these ecosystems. However anthropogenic releases can result in higher concentrations of these metals relative to their normal background values. When these occur, heavy metals are considered serious pollutants because of toxicity, persistence and non-degradable conditions in the environment, thereby constituting threat to human beings and other forms of biological life (Chinedu & Chukwuemeka, 2018)

Essential heavy metals

Some of the heavy metals (eg. Fe, Cu and Zn) are essential for plants and animals, their availability in medium varies, and metals such as Cu, Zn, Fe, Mn, Mo, Ni and Co are essential micronutrients, whose uptake in excess to the plant requirements result in toxic effects. The various ranges of few important heavy metals in plants include: As 0.02-7; Cd 0.1-2.4; Hg 0.005-0.02; Pb 1-13; Sb 0.02-0.06; Co 0,05-0.5; Cr 0.2-1; Cu 4.15; Fe 140; Mn 15-100; Mo 1-10; Ni 1; Sr 0.30 and Zn 8-100 in mg/kg weight on plants (Alloway, 2013).

Background on heavy metals

Heavy metal pollution refers to cases where the quantities of these elements in soils are higher than the maximum allowable concentrations, and this is potentially harmful to biological life at such locations. As noted by Gazso (2001), heavy metals come from a variety of sources but human economic activities such as coal and metal ore mining, chemical

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manufacturing, petroleum mining and refining, electric power generation, melting and metal refining, metal plating and to some extent domestic sewage are principally responsible. Some of the heavy metals such as Cu, Ni and Zn are essential to plants and animals in very low concentrations by serving as components of enzymes, structural proteins, pigments and also helping to maintain the ionic balance of cells (Kosolapov et al., 2004). These and other trace elements are important for proper functioning of biological systems and their deficiency or excess could lead to a number of disorders. Food chain contamination by heavy metals has become a burning issue in recent years because of their potential accumulation in biosystems through contaminated water, soil and air.

Assessment of Pollutant Concentrations

As observed by Begum et al. (2009), large quantities of pollutants have continuously been introduced into ecosystems as a consequence of urbanization and industrial processes. Metals are persistent pollutants that can be biomagnified in the food chains, becoming increasingly dangerous to human beings and wildlife. Therefore, assessing the concentrations of pollutants in different components of the ecosystem has become an important task in preventing risk to natural life and public health.

How Heavy Metals Enter the Environment

Heavy metals enter into the environment mainly via three routes namely: deposition of atmospheric particulate, disposal of metal enriched sewage sludges and sewage effluents and by-products from metal mining

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process. Soil is one of the repositories for anthropogenic wastes. Biochemical processes can mobilize them to pollute water supplies and impact food chains. Heavy metals such as Cu, Cr, Cd, Ni, and Pb are potential soil and water pollutants. Globally, the problem of environmental pollution due to heavy metals has begun to cause concern in largest cities since this may lead to geoaccumulation, bioaccumulation and biomagnifications in ecosystems. Heavy metal contaminants in the environment are eventually deposited in soils in some form of a low solubility compound, such as pyrite (Huerta-Diaz & Morse, 1992) or sorbed on surface-reactive phases, such as Fe and Mn oxides (Cooper, Neal, Kukkadapu et al., 2005; Hamilton, Smith, Davison & Sugiyama, 2005).

Anthropogenic Sources of Heavy Metals

Heavy metals are released into environment by various anthropogenic activities. The introduction of heavy metals due to continuous input of pesticides and fertilizer for food production is transported to surface water by erosion (Masindi & Muedi, 2018). Zn and Cd are commonly present in phosphate fertilizers and the input of these fertilizers is directly proportional to the concentration of heavy metals. In addition to Zn and Cd, pesticides used in agriculture have elements such as Hg, As and Pb. Though the metal based pesticides are no longer in use, the earlier unregulated pesticide application has led to increased accumulation of heavy metals in various environmental matrices. Added to these, various industrial activities such as mining, coal combustion, effluent streams, and waste disposal has increased the heavy

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metal contamination in the environment (Alengebawy, Abdelkhalek, Qureshi, & Wang, 2021).

Effects of Heavy Metals

Heavy metal contaminants in the environment are eventually deposited in soils in some form of a low soluble compound, such as pyrite (Huerta-Diaz & Morse, 1992) or sorbed on surface-reactive phases, such as Fe and Mn oxides (Cooper et al., 2005; Hamilton-Taylor et al., 2005). Lead (Pb) is the most common environmental contaminant found in soils. Unlike other metals, Pb has no biological role, and is potentially toxic to microorganisms (Sobolev & Begonia, 2008). Its excessive accumulation in living organisms is always detrimental. Furthermore, Pb exposure can cause seizures, mental retardation, and behavioural disorders in human beings. Heavy metal exposure to human beings occurs through three primary routes namely inhalation, ingestion and skin absorption. All these occur in myriads of places including auto-mechanic workshops.

Generally, toxic metals cause enzyme inactivation, damage cells by acting as antimetabolites or form precipitates or chelates with essential metabolites. According to USDA (2000), acute (immediate) poisoning from heavy metals is rare through ingestion or dermal contact, but it is possible. Chronic problems associated with long-term heavy metal exposures are mental lapse (lead); toxicological effects on kidney, liver and gastrointestinal tract (cadmium); skin poisoning and harmful effects on kidneys and the central nervous system (arsenic). There is a link between long term exposure to

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copper and decline of intelligence in young adolescents (Jaishankar et al., 2014).

Chronic cadmium exposures result in kidney damage, bone deformities, and cardiovascular problems (Goyer & Clarkson, 2001). Human diseases have resulted from consumption of cadmium contaminated foods (Satarug et al, 2010). The threat that heavy metals pose to human and animal health is aggravated by their low environmental mobility, even under high precipitations, and their long term persistence in the environment (Mench, Didier, Löffler, Gomez, & Masson, 1994). For instance, Pb, one of the more persistent metals, was estimated to have a soil retention time of 150 to 5000 years (Sobolev & Begonia, 2008). Also, the average biological half-life of Cd, another accumulation poison similar to lead, has been estimated to be about 18 years (Forstner, 1995).

Effects of heavy metals on soil organisms

As a result of low environmental mobility of those metals, a single contamination episode could set a stage for a long-term exposure of human, microbial, fauna, flora and other edaphic communities to heavy metal, necessitating long-term monitoring effort to assess effects of the metals. Studies have shown that long-term heavy metal contamination of soils has harmful effects on soil microbial activity, especially microbial respiration and enzyme activity (Begonia et al., 2004). Toxic effects of heavy metals on microorganism's manifest in numerous ways such as decrease in litter decomposition and nitrogen fixation, less efficient nutrient cycling and impaired enzyme synthesis (Baath, 1989). Aside from long-term metal-

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mediated changes in soil enzyme activities, many reports have shown large reductions in microbial activity due to short-term exposure to toxic metals (Doelman & Haanstra, 1979; Hemida et al., 1997).

Bacterial activity, measured by thymidine incorporation technique, has been shown to be very sensitive to metal pollution both under laboratory (Diaz-Ravina & Baath, 1996) and field conditions. Moreover, habitats that have high levels of metal contamination show lower numbers of microbes than uncontaminated habitats (Kandeler et al., 2000). All these have deleterious effects on agriculture, and ultimately human beings. Short-term and long-term effects of pollution differ depending on metal and soil characteristics (Kádár, 1995; Németh & Kádár, 2005). In the after-effect of heavy metal pollutions, the role of pollutant binding or leaching increases, which determine their bioavailability and toxicity.

Health effects of heavy metals on human

Heavy metals are non-degradable and their accumulations not only contaminate the surface environment but also contribute to air pollution, as they may become airborne or eroded and eventually enter the drainage system to affect aquatic ecosystems (Comfort et al, 2013). In general, the presence of heavy metals in high concentrations in the environment results in health hazards such as adverse effects of the nervous system, blood forming, renal and reproductive systems. Others include reduced intelligence, attention deficit and behavioural abnormality, as well as its contribution to cardiovascular disease in adults (Christoforidis & Stamatis, 2010). Heavy

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metal can cause serious health effects with varied symptoms depending on the nature and quantity of the metal ingested (Momodu & Ayankora, 2010)

Effects of heavy metals on plants

Heavy metal pollution of the soil also has negative side effects on plants. Anoliefo et al. (2001) showed a phytotoxic effect of soil collected from abandoned mechanic village and reported that the soil depressed and inhibited plant growth.

Effect of zinc on plants

The phytotoxicity of Zn and Cd is indicated by decrease in growth and development, metabolism and an induction of oxidative damage in various plant species such as The repeated paragraph has been removed Cd and Zn have reported to cause alternation in catalytic efficiency of enzymes in *Phaseolus vulgaris* and pea plants. Concentrations of Zn found in contaminated soils frequently exceed to those required as micro-nutrients and may cause phytotoxicity. Zn concentrations in the range of 150–300 mg/kg have been measured in polluted soils. High levels of Zn in soil inhibit many plant metabolic functions; result in retarded growth and cause senescence. Zinc toxicity in plants limited the growth of both root and shoot. Zinc toxicity also causes chlorosis in the younger leaves, which can extend to older leaves after prolonged exposure to high soil Zn levels. The chlorosis may arise partly from an induced iron (Fe) deficiency as hydrated Zn^{+2} and Fe^{+2} ions have similar radii. Excess Zn can also give rise to manganese (Mn) and copper (Cu) deficiencies in plant shoots.

Such deficiencies have been ascribed to a hindered transfer of these micronutrients from root to shoot. This hindrance is based on the fact that the Fe and Mn concentrations in plants grown in Zn rich media are greater in the root than in the shoot. Another typical effect of Zn toxicity is the appearance of a purplish red color in leaves, which is ascribed to phosphorus (P) deficiency. Zinc in excess reduces the germination, chlorophyll, carotenoid, sugar, amino acid and growth of cluster beans (*Cyamopsis tetragonoloba*). Whereas, in pea (*Pisum sativum*) reduces chlorophyll, photosynthesis and plant growth. In rye grass (*Lolium perenne*) it reduces the growth, nutrient content and photosynthetic energy conversion.

Effects of cadmium on plants

The permissible limit of cadmium (Cd) in agricultural soil is 100 mg/kg soil (FAO, 2001). Plants grown in soils containing high levels of Cd show visible symptoms of injury reflected in terms of chlorosis, growth inhibition, browning of root tips and finally death. The inhibition of root Fe (III) reductase induced by Cd led to Fe (II) deficiency, and it seriously affected photosynthesis. In general, Cd has been shown to interfere with the uptake, transport and use of several elements (Ca, Mg, P and K) and water by plants. Cd also reduced the absorption of nitrate and its transport from roots to shoots, by inhibiting the nitrate reductase activity in the shoots. Appreciable inhibition of the nitrate reductase activity was also found in plants of *Silene cucubalus*. Nitrogen fixation and primary ammonia assimilation decreased in nodules of soybean plants during Cd treatments (Ohyama, Sato, T., Yamamoto, Arai & Satsuma, 2013).

Metal toxicity can affect the plasma membrane permeability, causing a reduction in water content; in particular, Cd has been reported to interact with the water balance. Cadmium treatments have been shown to reduce ATPase activity of the plasma membrane fraction of wheat and sunflower roots (Haider, Liqun, Coulteret al., 2021**)**. Cadmium produces alterations in the functionality of membranes by inducing lipid peroxidation and disturbances in chloroplast metabolism by inhibiting chlorophyll biosynthesis and reducing the activity of enzymes involved in CO_2 fixation. In wheat (*Triticum spp*.) excessive of cadmium reduces the seed germination; decrease in plant nutrient content; reduced shoot and root length. Whereas in garlic (*Allium sativum*) Cd accumulation reduced shoot growth. Lastly in Maize (*Zea mays*) it reduces shoot growth and inhibition of root growth (Wang, Zou, Duan, Jiang, & Liu, 2007).

Effects of lead on plants

Plants on land tend to absorb lead from the soil and retain most of this in their roots. There is some evidence that plant foliage may also take up lead (and it is possible that this lead is moved to other parts of the plant). The uptake of lead by the roots of the plant may be reduced with the application of calcium and phosphorus to the soil. Lead (Pb) is one of the ubiquitously distributed most abundant toxic elements in the soil. It exerts adverse effect on morphology, growth and photosynthetic processes of plants. Lead is known to inhibit seed germination of *Spartiana alterniflora*, *Pinus helipensis*. Early seedling growth was also inhibited by lead in soyabean, rice, maize, barley,

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tomato and certain legumes. Lead also inhibited root and stem elongation and leaf expansion in *Allium species* barley and *Raphanus sativas.*

The degree to which root elongation is inhibited depends upon the concentration of lead and ionic composition and pH of the medium. Concentration dependent inhibition of root growth has been observed in *Sesamum indicum*. A high lead level in soil induces abnormal morphology in many plant species. For example, lead causes irregular radial thickening in pea roots, cell walls of the endodermis and lignification of cortical parenchyma (Wang et al, 2007). Lead also induces proliferation effects on the repair process of vascular plants. Lead administrated to potted sugar beet plants at rates of 100–200 ppm caused chlorosis and growth reduction Anoliefo et al. (2001). High Pb concentration also induces oxidative stress by increasing the production of ROS in plants. In maize (Zea mays) reduction in germination percentage; suppressed growth; reduced plant biomass; decrease in plant protein content has been noticed. Whereas in portia tree (*Thespesia populnea*), reduction in the number of leaves and leaf area; reduced plant height; decrease in plant biomass and in Oat (Avena sativa), there inhibition of enzyme activity which affected CO_2 fixation.

Effects of Soil Microbes on Soil Ecosystems

Microbes, as the most active part of the soil, have an important role in the transformation and storage of various nutrients. At the same time, they play a very significant role on the decomposition of organic matter, mineral decomposition and the release of nutrient. In the agroecosystem, soil microbes mainly have two effects. First, the microorganisms themselves contain a

certain amount of elements such as C, P and N, which can be regarded as an effective C, P, N source repository, thus it can adjust soil nutrients and store soil nutrients. Secondly, micro-organisms through the transformation and promotion of the system's metabolic process can promote inorganic element to flow (Swift & Anderson, 1994). Carbon and other nutrients mineral decomposition and cycling in soil ecosystems are dominated by activities of microbes.

Effects of soil microorganism on crop growth

Microbes in the activities of life can transform inert nitrogen from the air into the ionic nitrogen that can be absorbed by the plants directly, it can ensure that the plant nitrogen nutrition. Microbes can decompose insoluble minerals in the soil during their life activities and convert them into soluble mineral compounds to help plants absorb various mineral elements. Some fungi hyphae and the root system of higher plants can form a union (Khan, 2005). The union is conducive to improving the resistance of plants in the adverse environment, and promotes plant growth. Soil microbes can degrade inorganic and organic pollutants, reduce the toxicity of pollutants to plants, and provide a good ecological environment for plant growth. Soil microbes can promote the organics to form humic acids around the roots, and promote plant growth and development.

Soil microbes can produce some secondary metabolites that have some stimulating effects on the growth and development of plants. Rhizosphere soil microbes form a physical barrier around the roots of plants, protecting plant roots in this micro-ecological environment, reducing the invasion of pathogens

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and pests (Wu & Lin, 2003). Plant rhizosphere-promoting bacteria can control crop susceptibility by fixing atmospheric nitrogen, producing plant hormones, promoting specific enzyme activities, and by producing antibiotics and some other substances that inhibit pathogens such as iron carriers and chelating media. Some microbial and microbial fertilizers, including dissolved phosphorus, can promote plant growth by increasing the fixation efficiency of biological nitrogen and the availability of substances that promote plant growth to increase the availability of trace elements such as iron and zinc.

Effects of soil microbes on soil structure

The soil environment is a very complex system, including a variety of small environments with different physical and chemical gradients and discontinuous environmental conditions. Microbes adapt to the micro-environment and interact with other parts of the soil or sensitive boundaries to produce various interactions. Soil microbes play an important role in the formation of soil structure. Actinomycetes produce mycelia that allow soil particles to bind. Therefore, the content of actinomycetes in fertile soil is higher than that in barren soils (Waldrop et al., 2000). The leading role of microbes is also different in different soils (Johns, 2017). When microbial extracellular polysaccharides, microbes and soil are separated, it makes the combination of soil particles, which contributes to the formation of soil structures. The activity of organic matter during the polymerization process is conducive to the formation and production of soil humus. This effect can reduce the invasion of soil water, so that the soil can maintain a good penetration and contain enough air.

Effects of Heavy Metals on Soil Microbial Ecological Function

Heavy metals are common and important refractory pollutants. The toxicity of heavy metals is primarily concerned with the bioavailability of metals, that is, the amount of organisms that are eventually absorbed into the body by absorption, migration and transformation (Leyval et al., 1997). High concentrations of heavy metals on the toxicity of microorganisms may have two reasons: heavy metals and microorganisms have a strong affinity, and it is easy with some biological macromolecules such as enzyme activity center, and electron-donating groups such as nucleic acid base and phosphate combination, resulting in the inactivation of these biological macromolecules, more than the ability of organisms to bear, resulting in biological disease and death (Igiri et al., 2018).

From a short-term perspective, heavy metal pollution will lead to the loss of microbiological diversity as those which are less adapted will decrease in population and those which can adapt will increase. A large number of metallothionein and small molecules such as glycine and taurine are easy to accumulate with the food chain of enrichment and transmission, and will endanger all biological, especially human health and life safety.

Effects of heavy metals on soil microbial activity

Some heavy metals in the soil also have an effect on the growth of soil microbes (Chu, 2018). Almost all soil biochemical reactions involve soil microbes, which play an important role in maintaining soil quality through the formation of soil organic matter and its decomposition of harmful substances, biochemical cycles and the formation of soil structure. Heavy metal

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contaminated soils have a negative effect on soil microbial properties, such as the underlying soil respiration rate and enzyme activity that depends on soil pH, organic matter and other chemical properties. Studies have shown that, in most cases, low concentrations of heavy metal contaminated soil are conducive to the release of CO_2, high concentrations of heavy metal pollution conditions, significant inhibition of soil respiration, severe heavy metal pollution can inhibit soil microbial activity, seriously threatening the soil ecosystem function.

Effects of heavy metals on soil enzyme activities

As the concentration of heavy metals increases, the activity of most enzymes is significantly reduced and the decrease in their activity may be caused directly by the interaction between the enzyme and the heavy metals, which is not associated with a reduction in microbes (Chu, 2018). Heavy metals have a significant effect on soil enzyme activity. On the one hand, heavy metals have a direct effect on soil enzyme activity, so that the spatial structure of the active groups of the enzyme is destroyed. On the other hand, the growth and reproduction of microorganisms are inhibited, thus reducing the synthesis and metabolism of the microbial enzyme. There is a very close relationship between soil enzymes and soil microbes, and some microorganisms and enzymes secreted by microorganisms participate in the circulation of soil ecosystems and energy together.

Effects of heavy metals on soil microbial community composition

After the heavy metal gets into the soil, the primary impact is the amount of soil bacteria, fungi, actinomycetes and other microbial population. Heavy metals affect the microbial quality and quantity in the soil. It also affects decomposition rate by soil microbes. For example, Hg in the environment, there are a variety of valence (elemental mercury, inorganic Hg2+, organic mercury compounds), organic synthesis of mercury in addition to artificial synthesis, some bacteria also have the ability to synthesize organic mercury. Heavy metal contamination can produce different microbial community patterns. Even if many of the chemical and biological properties of the soil have changed greatly, there are many original microorganisms in the soil that are present in the microbial community (Perezdemora & Burgos, 2006). Long-term heavy metal contaminated soil will choose those who can specifically adapt to polluted soil microbial population. The higher the content of organic carbon in severely polluted soils, the lower the efficiency of microbial populations in organic mineralization. This can be a simple indication of the impact of heavy metal.

Activities of Auto Mechanics

Auto repair shops offer miscellaneous repair services ranging from simple and fast oil change to complex engine rebuilding. They provide auto-body repair, electrical, welding and spraying services as needed. The operational processes at auto workshops often involve the use of toxic and hazardous materials such as solvents, paints, primers, etc. The car refinishing process results in the improved look of vehicles but generates hazardous

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wastes that require appropriate disposal. Petrol, diesel, solvents, grease, and lubricants can either be accidentally or deliberately released into the environment. Many of these petroleum products are organic chemicals that can be highly toxic and hazardous to soil, fauna and man. The use of automobiles and their repair has generally led to heavy metal-contamination in soil, which has grave consequences on soil dwelling organisms (Adewoyin et al., 2013). As a result of frictional wear, the hydraulic fluid in an automobile collects heavy metal debris such as Pb, Cd, Zn, Fe, Cu, etc. Usually, automobile waste will consist of auto body scraps (mainly of Al), pieces of mild steel, electrical components and wires (mostly Cu) (Nwachukwu, Feng & Alinnor, 2010).

Most auto repair shops in Ghana are located by the road side in the open and operated on bare soil. They normally have electrical, mechanical, welding, and spraying sections. Because they are operated on bare soil, the heavy metals from the vehicle fuel, lubricants, batteries, etc. are easily released into the environment and carried to distance locations by rain water. They can also be carried into the ambient air by resuspended dust particulates. This can have adverse effect on soil, water and air quality in the general environment even distant from the vicinity of the auto repair shops.

Agriculture

Less than 10 percent (8.5%) of households in the Metropolis are engage in agriculture. Of those in Agriculture, 91.6 percent are into crop farming and 10.7 percent are into livestock rearing. Poultry (chicken), goat, sheep and pigs are the animal mostly reared in the Metropolis. Urban

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agriculture is main agriculture practice in the Kumasi. It focuses on the cultivation of vegetables such as carrot, cabbage, lettuce and green onions (MOFA, 2016)

CHAPTER THREE

MATERIALS AND METHODS

This chapter describes the study sites including location maps, georeferenced of the soil and plant sample points, climate, vegetation, agriculture, relief and drainage among others. It also describes the soil and plant sampling techniques employed in the study. This chapter shows how the microbial activity levels were determined. In addition, it describes all the laboratory processes employed during routine analysis of soil samples, heavy metals analysis in soils and plants as well as microbial activity processes. It unveils the software that was used to analyse the data.

Study Area

The first study area is located in Edwenase-Kwadaso, a suburb of Kumasi in the Ashanti region of Ghana (Figure1). This used to be trial grounds for agricultural research activities. The geographical location of Edwenase lies in Latitude 6.6784° N and Longitude 1.6530°W with an altitude of 225m above mean sea level. The topography of the area is partly flat plane and undulating surface, and lies within tropical rain forest. The soil type is made up of Cutanic Lixisol (World reference base of soil resources WRB, IUSS working group WRB, 2006). The soil at Edwenase falls within Kumasi – Asuansi / Nta – Offin Association.

The presence of the Agricultural College and two research institutes in the town attract a large population of people and vehicles, which in turn gives a boost to automobile workshop activities. The coordinates for the study area is Latitude 06° 40' 15'' N and Longitude -01° 38' 55''W with altitude 256m

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and The GPS coordinate for the control experimental field is Latitude 06° 40'

37'' N to 06° 40' and Longitude 01° 39' 23'' W with altitude 275m
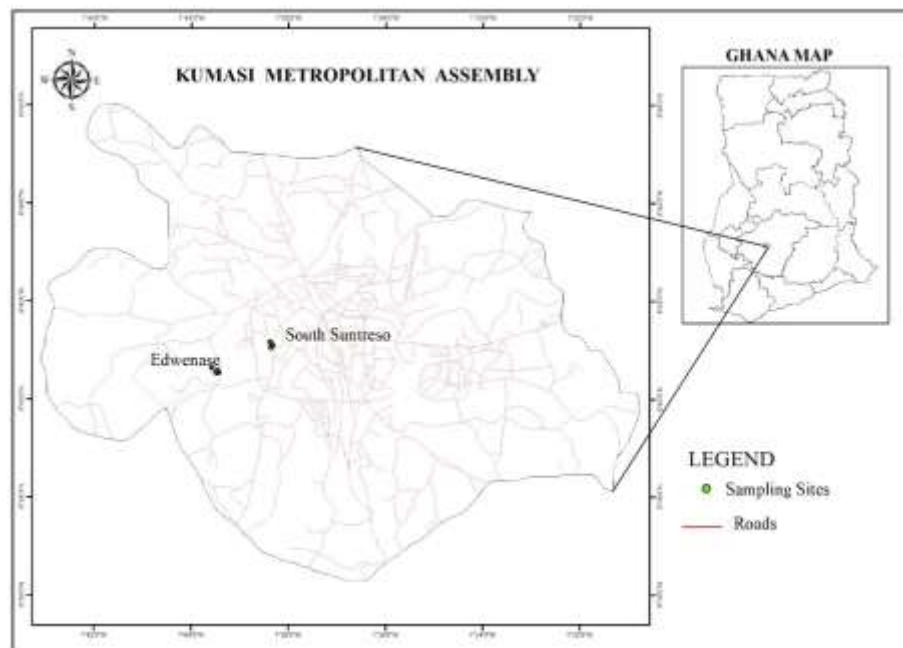
Figure 1: Shows GPS Locations where Soils Were Sampled Using the GPS
Magellan Explorist 210.

The second study area was at South Suntreso under Kumasi

Metropolitan Assembly (KMA). The soil at South Suntreso falls within

Akumadan-Bekwai/Oda Association. The sample area lies within latitude

6.6945^o N and longitude $1.6409\ ^o$ W with altitude 229m above sea level. The

Metropolis shares boundaries with Kwabre East Municipal and Afigya

Kwabre District to the north, Atwima Kwanwoma District and Atwima

Nwabiagya North District to west, Asokore Mampong Municipal and Ejisu

Municipal to the east and Bosomtwe District to the south. The population of

the Metropolis according to the 2010 Population and Housing Census stands at

730,249 with 826,479 males. 903,770 females.

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Climate

The study area falls within the tropical monsoon and is characterised by average temperatures ranging from 21.5°C to 30.7°C. Average annual rainfall is 1,500mm with peaks of 1,663mm 1,921mm and in 2013 and 2017 respectively. The rainfall pattern is bimodal. The average humidity is about 84.16% at 09.00 GMT and 60% at 15.00 GMT. The highest maximum temperature on record for Kumasi is 37.8°C (100°F), while the record for the lowest minimum temperature is 10.6°C (51.1°F) in 2016 (MOFA, 2016).

Vegetation

The natural vegetation of the study area was semi-deciduous forest but currently used for the production of vegetables – cabbage, carrots, spring onions, green pepper, lettuce, cucumber, french beans; Root and tubers – plantain, cassava, yam; Cereals – rice, maize. Non-traditional agriculture including – mushroom, grass cutter and snails are also produced (MOFA, 2016).

Sampling location

Samples were collected from two clusters of mechanic workshops, one at Edwenase and the other at South Suntreso in Kumasi. The study area for the control experiment is located 100meters away from the main study site at Edwenase which is not influenced by any auto mechanic activities.

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Initial Soil Characterization

Samples of soil from different locations at the control experimental field were collected. These fields are 100 m away from the main study area which were not affected by auto mechanic activities. The soil samples were collected into polythene bags, sent to the laboratory, dried and sieved through 2mm mesh to obtain the fine earth for routine analysis This served as control to the main experimental fields where soil pollution was suspected to be higher.

Soil Sampling Design

The experimental design used for the study was Randomized Complete Block Design. The study focussed on the adjoining fields at each location. Each of the two fields was divided into three portion including the upper part, middle part and along the valley bottom. Within the workshops under study where the mechanics actually perform their mechanic activities, special soil samples were also taken. This was done because there was an assumption of abnormal accumulation of all forms of auto-mobile waste. Mechanics sometimes deliberately pour off oils and used car barriers on the soil which later leach into the soil. Soil samples were taken at two depths (0-10cm and 10-30cm). A total of nine (9) sampling points were randomly sampled in each of the study sites including the control plots at each site totalling 72 composite samples.

Soil sampling started from upper part (workshop area) through the middle part and to the soils along the valley or valley bottom using Gouge Auger, at the depths of 0-10 cm and 10-30cm representing the top soil and the

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sub-soil respectively. Triangulation soil sampling method was used across the study areas. Three (3) triangular points were marked (Figure 2) and soil samples were taken at each vertex at the two depths explained above. The discrete soil samples collected at each vertex of the triangle were mixed up according to depth class to form composite samples 1, 2 and 3. This means that at each part, six (6) composite samples were collected. The geo-reference and elevation of each selected site was recorded with a global positioning system receiver (GPS, Magellan Explorist 210). After taking the soil samples, the soil was placed in polythene bags, labelled and transported to the laboratory.
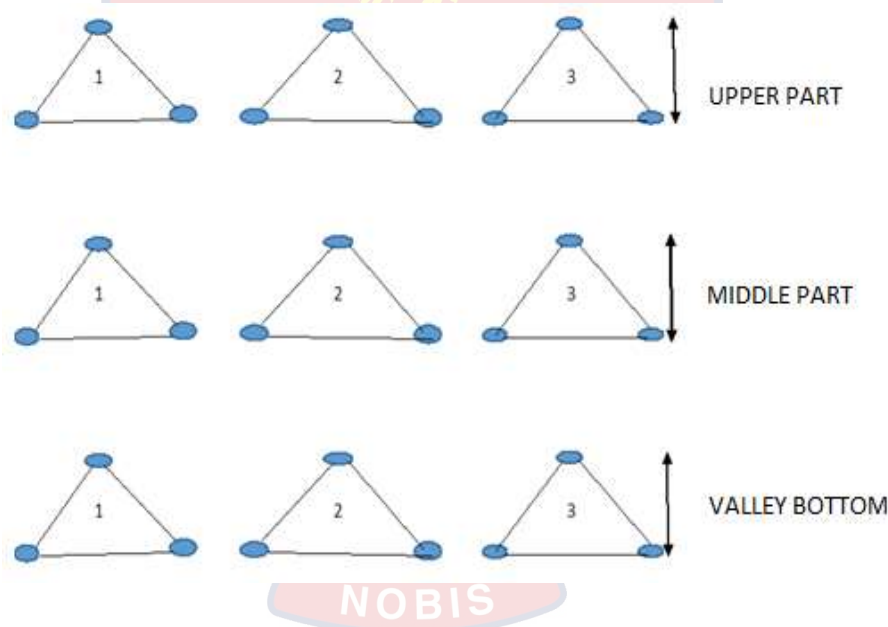

Figure 2 : The experimental design

Crop Sampling

Some of the existing plant biomass or crops at the adjoining fields were sampled at 10 m interval for heavy metal accumulation analysis. The crops were maize plants, plantain and plantain leaves. These crops were

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chosen because they were the most dominant crops at the study sites. At Edwinase, eighteen (18) plants maize were sampled from upper slope through to middle and to valley bottom including control plot. At each slope three plants were sampled. At South Suntreso, eighteen (18) plantain fruits and leaves were sampled from upper slope through to middle slope to valley bottom including the valley bottom.

Crops samples were taken at regular distance of 10m from each other. The GPS location at each sample collected was recorded. The plants selected were placed in transparent polyethylene bags and labelled according to the location it was sampled. They were sent to Soil Research Institute Laboratory at Kwadaso, Ghana.

Laboratory Procedures

Sample preparation and analysis

Samples that were sent to the laboratory were moist. This was due to the previous rains which made the soil very moist. Soil samples were labelled and later given lab numbers for easy identification. The moist soils were air-dried in enamelled trays. Care was taken to maintain the identity of each sample at all stages of preparation. During drying, the trays were numbered. Alternatively, the trays were placed in racks in a hot-air cabinet, whose temperature did not exceed 35 °C and relative humidity was between 30–60 percent.

Post-drying care

Air-dried samples were gently grounded with a wooden pestle and mortar so that the soil aggregate is crushed but the soil particles do not break down. Samples of heavy clay soils were also ground with an end-runner grinding mill fitted with a pestle of hard wood and rubber lining to the mortar. Pebbles, concretions and stones were not broken during grinding. After grinding, the soil was screened through a 2-mm sieve.

Since soil was to be analysed for trace elements, containers made of copper, zinc and brass were avoided during grinding and handling. Soil samples were mixed thoroughly after it was passed through the sieve. The soil samples were stored in cardboard boxes in wooden drawers. These boxes were numbered and arranged in rows in the wooden drawers, which were in turn fitted in a cabinet in the soil sample room prior to analyses.

Plant samples

Plant and fruit samples were chopped into smaller pieces and placed in brown envelopes. Each envelope was labelled accordingly for easy identification during the drying process. The labelled samples were collected and placed in an oven at a temperature of 70°C. It was allowed to dry for the period of 24 hours where the oven temperature was reduced to 45°c for 12 hours. Dried leaves and crops were removed from the oven and allowed to cool. The samples were later grinded using a miller into smaller particles. The samples were then placed back into their various labelled envelopes for analysis.

Soil chemical analysis

The materials used for Bulk Density Were: Weighing scale Weighing cans with soil samples, Core sampler with diameter 5cm and height of 5cm and Oven.

Procedure

Each of the study field was divided into three portions; the upper part, middle part and valley bottom. In each portion, three soil samples were taken at three different points at the same gradient using steel core of diameter 5 cm. 2 cm of surface soil from the spot where samples were taken were removed and the samples were then levelled. The steel core with a known volume was driven into the soil. The soil was excavated from around the cylinder and removed beneath the bottom. Excess soil was trimmed from the ends. Same was done at the other portions. In all, nine soil samples were taken from each fields totalling 18 core soil samples for the study. The soils were placed in sampling bags and labelled.

At the laboratory, all the soil samples were re-labelled for easy identification. Eighteen weighing cans of known weight were arranged and labelled corresponding to the labels of the soil samples. Bulk soil samples were placed into a weighing can of known weight (W1) and weight with its content of moist soil (W2) and recorded. The oven was set at a temperature of 105°C and dried for 24 hours. After oven-drying, the samples together with the cans were allowed to cool in a desiccator to prevent absorption of water. The samples were weighed (W3) and recorded using the weighing scale using the weighing scale. Using Microsoft excel, the data was recorded. Results were

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generated which include; bulk density, % moisture content and porosity and % porosity. The volume of the cylinder used in taking samples was calculated as V(W3-W2) / V.

Calculations

Actual weight of Sample W = W2-W1

Volume of Cylinder V= $\prod r^2 h$

Bulk Density = $\dfrac{W}{V}$ (g/cm^3)

Total porosity (%) = 1- $\left(\dfrac{\text{bulk density}}{\text{particle density}}\right)$ X 100 = Soil porosity (%) = 1-

$\left(\dfrac{\text{soil bulk density}}{2.65}\right)$ x100

Soil Moisture content (g/g) = $\dfrac{\text{(weight of moist soil – weight of oven dry soil)}}{\text{Weight of oven dry soil}}$

Particle size distribution

   Procedure: The determination of particle size distribution was carried out with a densimeter, using Calgon as a dispersing agent. The particle size distribution classes refer to I.S.S.S standards: sand 2 – 0.02 mm; silt 0.02 – 0.002 mm; clay < 0.002 mm.

Materials and Reagents:

DENSIMETER: Model ASTM 152 h.

500 ml glass cylinders

10% CALGON solution: Dissolve 15.9 gr of anhydrous Na_2CO_3 in a 500 ml beaker and add 71.4 gr of sodium hexametaphosphate; make volume up to 1L.

Determination

25 g of soil was placed in a 250 ml plastic bottle and 10 ml of Calgon and 100 ml of water added. The suspension was agitated overnight using mechanical shaker at 120 r.p.m. The following morning, the samples in the bottles were transferred into 500ml cylinders, then the volume was topped up to 500 ml. Each cylinder was hand-agitated for 1 minute; densimeter readings started started 5 minutes later. Each reading was increased by 0.5 to account for meniscus. A second reading was taken 16 hours later.

A blank reading was taken as well; it was also prepared as (10 ml of calgon in a volume made up to 500 ml). The temperature of the samples was also measured. The first reading provides the sum of clay and silt (C + S).

Calculation

First reading (C + S)   42.5   B1 (blank) 1.5

Second reading (C)   23.0   B2 (blank) 1.0

Clay % = (2nd read. − B) X2

= (23 − 1) x2

= 44%

Silt = [(1st read. − B1) − (2nd read. − B2)] x 2

= [(42.5 − 1.5) − (23 -1)] x 2

= (41 -22) x2

= 38%

Sand % = 100 – C% - S%

= 100 – 44 – 38

= 18%

N.B. Results are expressed by integers. If temperature is above 20 x C the densimeter may give negative values; in these cases the reading of the blank should be added not subtracted.

TEXTURE: a texture class is assigned to the soil by using the given triangle
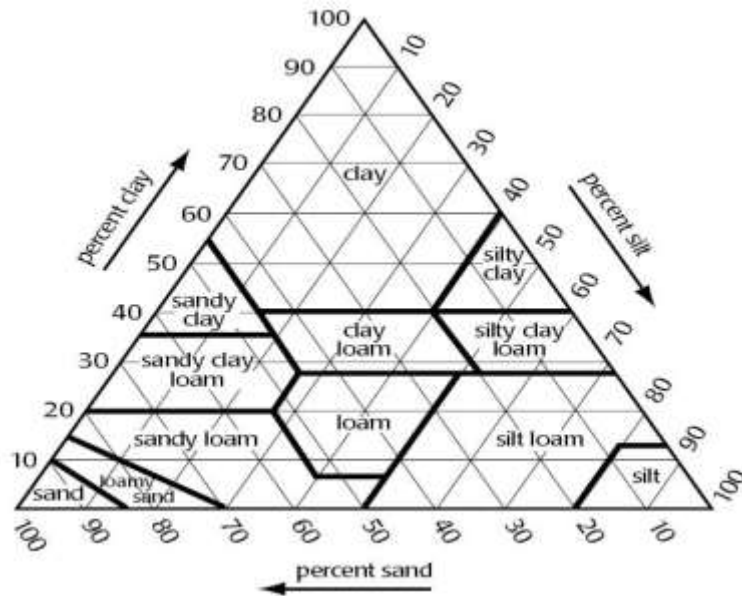
Figure 3: Textural triangle

Chemical analysis

Soil pH

Principle: Soil pH is measured with a pH-meter on a 1:2.5 soil/water suspension. It can also be measured using 1:1, 1:2, 1:5 based on the type of test to be carried.

Reagents: Distilled water

Buffer solutions: pH 4.0, 7.0, 10.0

Calibration of the pH Meter and Checking the Sensitivity of the Electrode

The electrode of the pH meter was placed into a pH solution of buffer 4, calibrate (cal) was entered on the pH meter. Same was done for pH buffer 7 and pH 10.

After calibration, their electron volts were measured to check the sensitivity of the pH meter. If the sensitivity is less than 70% it means the electrode needs to be change.

Determination of pH (water pH)

10 gr of soil was placed in a 50ml plastic beaker together with 25 ml of distilled water and mixed with a glass rod. The 72 composite soil samples were stirred for 30 minutes intermittently. The soil pH reaction was measured on the suspension after calibrating the instrument with the buffered solutions of pH 4.00 and 7.00. The pH was read by immersing the electrode into the supernatant and the pH value recorded.

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Organic carbon

Walkey and black method

Reagents:

1 N $K_2Cr_2O_7$: Dissolve 98.07g of potassium dichromate (oven-dried for 2 hours at 120 C) in a 2 litre volumetric flask containing 1500 ml of water. When the dissolution is complete make up the volume. Transfer this solution into a dark glass bottle Concentrated H_2SO_4 (98%)

0.025 N ferroin: weigh 1.485g of monohydrate orthophenantroline ($C_{12}H_8N_2$. x H_2O) and 0.695g of ferrous sulphate ($FeSO_4$ .7H_2O) and dissolve with deionized water in a 100ml volumetric flask. The glass containing the solution was stored in a dark environment. 0.5 N MOHR'S SALT: dissolve 196.1g of ferrous ammonium sulphate [$FeSO_4(NH_4)_2(SO4)_2.6H_2O$)] in a 1000 ml volumetric flask containing 600 ml of water; add 20 ml of concentrated sulphuric acid and when dissolution is completed make up the volume.

Determination

One gram (1g) of soil was weighed and placed it in a 250 ml. Erlenmeyer flask. Then, under the hood, 5 ml of potassium dichromate and 10 ml of concentrated sulphuric acid were added. The solution was allowed to rest for 3 hours. Then 75-100 ml of deionized water, 2-3 drops of ferroin and titrate with Mohr's salt were also added. At the same time a blank with 5 ml of dichromate and 10 ml of sulphuric acid were prepared.

Calculation: the result can be expressed as organic carbon or as organic matter.

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O.C% $= \frac{(b-a) \times N \times f \times 0.39}{W}$

Where: b = ml of Mohr's salt used for the blank

a = ml of Mohr's salt used for the sample

N = normality of Mohr's salt

F = normality correction factor

W = weight of the sample

O.M. % = O.C. x 1.724

In other specific case, operating with 5 ml of dichromate and 10 ml of concentrated sulphuric acid, the calculation is the following:

O.M. % = (b − a) x $\frac{6.7}{b}$

Total nitrogen

Reagents:

Concentrated H_2SO_4 (d 1.84)

40% NaOH in water

0.1N H_2SO_4

0.1N NaOH

Mixed indicator: methyl red – blue methylene. 0.1 gr of methyl red and 0.05 gr of blue methylene were dissolved in 100 ml of alcohol (store in a dark bottle)

Determination (mineralization)

Five grams of soil which was sieved at 2 mm was weighed and placed in a digester tube. A tea spoon of copper sulphate pentahydrate and 20 ml of concentrated H_2SO_4 were added. The solution was placed in the digester using

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the following mineralization procedure: started at room temperature, temperature ramp up to 200°C; the temperature was constantly kept for 30 minutes.

During the distillation process, 25 ml of 0.1N H_2SO_4 was added in a 500 ml Erlenmeyer flask; about 200ml of distilled water was also added; two / three drops of mixed indicator was added as well. The Erlenmeyer flask was placed on the distiller and checked whether the end of the condensation pipe was covered by the acid.

Distilling using the following program was used during the distillation process:

- 5ml water

- 60 ml of 40% NaOH

- Pause 0 minutes

- Steam flow rate 100%

- Distillation time, 5 minutes

- Suction residues NO

After the distillation, the Erlenmeyer flask with 0.1 NaOH was titrated until the final color change from red to green

Calculation

$$N\% = \frac{((25 - a) \times 14)}{W(gr)} \times 100$$

Where:

25 = ml of 0.1 N H_2SO_4 used in the beaker

A = ml of 0.1 NaOH used in the titration

W = weight of the soil in grams

14 = molecular weight of nitrogen

Available phosphorus (Bray and Kurtz's method No. 1)

PRINCIPLE: Available phosphorus is extracted with a solution of HCl and NH_4F; the colorimetric determinations are carried out with ammonium molybdate ammonium and ascorbic acid as a reducing agent.

Reagents

2 M HCl solution: 167 ml of concentrated HCl (d=1.18) was diluted in 1 L of distilled water. The morality was verified with 1 N NaOH.

Extracting solution

(0.03 M NH_4F + 0.025 M HCl). 2.22 gr of NH_4F was dissolved in a 2 litre volumetric flask containing 1 litre of distilled water; 25 ml of the 2 M HCl solution was added (with a graduated pipette), agitated to make up the volume. The pH of the solution usually varies between 2.6 ± 0.05. 10 liters of solution was prepared.

Ammonium molybdate solution

70 ml of concentrated sulphuric acid was added to a 1 litre volumetric flask containing 500 ml of water (d=1.84); it was left to cool at room temperature. 6g of ammonium molybdate was dissolved in 100-150 ml of distilled water; 0.3g of antimony and potassium tartrate was also dissolved in 100ml of distilled water. The solutions were transferred to the volumetric flask

containing the sulphuric acid while topping was done to make up the volume. All the solutions were at room temperature before mixing. It was stored in a dark bottle.

Working solution: 0.3g of ascorbic acid was dissolved in a 200 ml volumetric flask containing 100 ml of distilled water; 50 ml of the ammonium molybdate solution was added and topped to make up the volume. (This solution should be prepared on the spot).

Std phosphorus solution (P):

0.4394 gr of KH_2PO_4 (previously oven-dried at $105^{o}C$ for 2 hours) was added to a 1 L volumetric flask containing 500 ml of distilled water; 5 ml of concentrated H_2SO_4 and topped to make the volume up. It was stored in a plastic bottle. The solution contains 0.1 mg/ml of phosphorus as P.

Distilled water

Determination

5g of soil was weighed and place in a 100 ml plastic bottle; 35 ml of extracting solution was added; it was then agitated for 5 minutes in the horizontal shaker; an adequate amount of extract was filtered. 1 ml of extract was collected with a pipette and placed it in a test tube, 9 ml of working solution was added; it was agitated and waited for 1 hour. The absorbance was measured on the spectrophotometer at 882 nm or 720 nm 660 nm (lower sensitivity) using the blank as reference.

Calculation

The concentration of phosphorus in the extract is determined using the calibration curve and is then related to the phosphorus in the soil using the following formula:

P ppm/soil = P mg/1 x $(\frac{35}{W(gr)})$ x f

Where: W (gr) = weight of the sample in grams

f = dilution factor

If phosphorus is expressed as P_2O_5 the calculation is as follows:

P ppm/soil = P mg/1 x $\frac{40}{W(gr)}$ x f x 2.2914

Calibration curve: The calibration curve is obtained by applying the described procedure to Std solutions of phosphorus prepared by diluting the 100 ppm Std solution. Std solutions with 1, 2 ,3, 4 5 and 6 ml of 100 ppm Std solution in 100 ml volumetric flasks and making the volume up with extracting solution (0.03 M NH_4F + 0.025 M HCl). Quality of reagents permitting, the instrument can be zeroed with blank or distilled water.

Exchangeable cations

Determination of exchangeable cation (Ca, Mg, K, Na) with atomic absorption

Apparatus

   i.   Rack

  ii.   Leaching tubes /falcon tubes

 iii.   Conical flasks

    iv.   Burette

    v.   Centrifuge

Reagents

    i.   Ammonium acetate [$NH_4CH_3CO_2$]

    ii.   Potassium hydroxide [KOH]

    iii.   Ammonium buffer [NH_3/NH_4Cl]

    iv.   Potassium cyanide [KCN]

    v.   EDTA

    vi.   Eriochrome Black T

    vii.   Murexide

Procedure

In this process, 2.5g of each soil sample was weighed into a rubber container with a lid. 40ml of ammonium acetate at pH of 7 was added to the sample. The mixture was shaken for about 5 minutes at 4000 rpm. The extract was centrifuged for the determination of Ca and Mg.

For Ca and Mg

15ml of the leachate was pipetted into a conical flask. 5ml of ammonium buffer and 1ml of potassium cyanide was added. The mixture was titrated against 0.02N EDTA using Eriochrome Black T as an indicator.

Determination of Ca only

    i.   An aliquot of 15ml from the extract was placed in a conical flask.

ii.   1ml of potassium cyanide (2%) was added.

iii.   5ml of potassium hydroxide (8N) was added.

iv.   A pinch of indicator (potassium sulphate + murexide) was added.

v.   Titration was done with EDTA until a purple colour was attained as an end point.

Determination of Na and K by AAS

Method

Flame Photometry

Reagents

Neutral Ammonium acetate solution (1N)

Dilute 570ml of glacial acetic acid (99.5%) with deionised water to a volume of approximately 5 litres. Then add 690ml of concentrated ammonium hydroxide (NH_4OH) and add deionised water to obtain a volume of about 9.9 litres. Check the pH of the resulting solution; add more NH_4OH if necessary, to obtain a pH of 7 and dilute the solution to a volume of 10 litres with deionised water.

Standard Potassium Solution: 2000 ppm

Accurately weigh 1.907g of potassium chloride previously dried for 2 hours at 105°C. Dissolve in about 50ml of the ammonium acetate/acetic acid solution. Transfer to a 500ml volumetric flask and dilute to 500ml with the ammonium acetate/acetic acid solution. 1000 ppm sodium standard solution

Prepare a 5ppm sodium standard as follows: add 5ml of the 1000 ppm standard to a 1 litre volumetric flask. Add 1N ammonium acetate to the mark.

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Prepare 4, 3, 2 and 1ppm sodium standards in a similar way adding 4, 3, 2 or 1ml of the 1000 ppm standard to a 1 litre volumetric flask together with 1, 2, 3 or 4ml deionised water (to make a volume of 5ml). Add 1N ammonium acetate to the mark. Prepare a blank solution by adding 5ml deionised water to a 1 litre volumetric flask and make up the volume to the mark with 1N neutral ammonium acetate.

Method

2.5g of soil sample was weighed into a rubber container with a lid. 10ml of ammonium acetate at pH of 7 was added to the sample and the mixture shaken for 5 minutes. The mixture was centrifuged and the supernatant decanted. Potassium standard solutions were prepared to cover the range 0 to 100 ppm potassium. The flame photometer was set up at 100 using the 100 ppm potassium solution. A calibration graph was prepared by successively aspirating 20, 40, 60 and 80 ppm potassium standard solutions. The potassium and sodium content of the soil extract was determined by aspirating the solution, diluting as necessary. The concentration was calculated by reference to the calibration graph, taking into account any dilution factor (1:100).

Calculation

i. Considering a 1:20 extraction ratio (2.5g and 50 ml) and a 1:25 dilution we have the following coefficients:

ii. Potassium meq/100 g = reading x 1.28

iii. Potassium K_{20} ppm = reading x 620

iv.   Potassium K_{20} ppm = K meq x 470

v.   Magnesium meq/100 g = reading x 4.11

vi.   Magnesium MgO ppm = reading x 829

vii.   Magnesium MgO ppm = reading x 829

viii. Magnesium MgO ppm = Mg meq x 202

ix.   Sodium meq/100 g = reading x 2.17

x.   Calcium meq/100 g = reading x 2.50

Calculation

CEC = Concentration meq/100g (Ca + Mg + K + Na)

Where CEC = Cation Exchange Capacity

$$CEC = 4.6\frac{meq}{100g} + 2\frac{meq}{100g} + \frac{62ppm}{390} + \frac{36ppm}{230}$$

Where $390 = \frac{39}{1} \times 10$, $\frac{Atomic\ weight\ of\ K}{Valence} \times 10$ (Conversion to meq/100g)

$230 = \frac{39}{1} \times 10$, $\frac{Atomic\ weight\ of\ Na}{Valence} \times 10$ (Conversion to meq/100g)

CEC = 7.19 meq/100g

Determination of metals in soil samples (Zn, Cd, Pb, As)

Ethylenediaminetetraacetic acid (EDTA) with ammonium acetate was used for the extraction of the element. The estimation of the elements in extract was done with the help of an AAS.

Apparatus:

Zinc, Lead, Cadmium and Arsenic Ethylenediaminetetraacetic acid (EDTA) with ammonium acetate is commonly used for the extraction of many elements. Diethylenetriaminepentaacetic acid (DTPA) is another common

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(universal) extractant and it is widely used for the simultaneous extraction of elements such as Zn, Cu, Fe and Mn (Lindsay & Norvell, 1978).

Procedure

0.5M EDTA was prepared by weighing 18.64g of the EDTA into a 2L flask. 1.0 N Ammonia Acetate Solution was added to the solution. The solution was dissolved and topped to the mark with DI.

The main procedure started by weighing 5g of the soil sample and addition of 25mls of EDTA solution. The mixture was shaken for two (2) hours with a mechanical shaker at 180 rpm. The solution was filtered using Whatman filter. The filtrate was sent to the AAS for reading of As, Cd, Zn and Pb. The concentrations on AAS for Metals like (As, Zn, Pb and Cd) were determined.

A standard calibration curves was prepared for each metal. The AAS and the Graphical Furnace were well set and all aligned. All readings were in ppm. The readings were then multiplied by the dilution factor.

Determination of heavy metals in plant samples

Apparatus

Oven

Crucibles

Metal hot plate

100ml volumetric flask

Furnace

AAS

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Reagents

1: 2 nitric acid

Distilled water

Procedure

Samples were placed in the oven and dried for 24 hours at 105°. Dried plant samples were removed from the oven and milled using the electrical milling machine. 1g of the milled sample was weighed and placed in crucibles. They samples were then ashed using the furnace. 1: 2 Nitric acid was added to the ashed for digestion to take place. The solution was heated on a hot plate for about four (4) minutes until digestion is over. The digested samples were filtered into a 100ml volumetric flask. An aliquot of the filtrate was sent to the Atomic Absorption Spectrophotometer (AAS) for the various readings (As, Pb, Cd and Zn)

Soil microbial activity (basal respiration method)

Reagents

1 M HCl, 1 M $BaCl_2$, 1 M NaOH, phenolphthalein indicator, Incubator, plastic bottles and bottles made with glass

Procedure

As a measure of antecedent biological activity was measured using static incubation of field-moist soil adjusted to 60% water holding capacity in a temperature-controlled incubator at $25 \pm 1°C$ for 21 d (Islam and Weil 2000).

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Briefly, 20-g field moist soil samples were incubated in Mason jars sealed with plastic lids. A vial containing 20 mL of 1 M NaOH was placed in each of the Mason jars to trap the evolved CO_2 from the soil. After 21 days of incubation, 2.5 mL of 1 M $BaCl_2$ and 3 drops of phenolphthalein indicator at 1 % were added to the NaOH containing vials that had trapped the released CO_2. The amount of CO_2 evolved from the soil was determined by titration of excess NaOH with 1 M HCl solution.

Soil BR (mg CO_2 kg^{-1} d^{-1}) was calculated as follows:

$$BR = \left(CO_{2Soil} - CO_{2Air} \right) 21 d^{-1}$$

Data Analysis

The data collected was statistically analysed to done to separate significant means using a two-way Analysis of Variance (ANOVA) and Tukey test. parameters include; Nitrogen, pH, electrical conductivity, particle size distribution (PSD), ECEC, Organic carbon, microbial boimass carbon (MBC) and that of the heavy metals including Cd, Zn, Pb and As. The significant means of the parameters of crop samples were also be analysed statistically. Whereas the data attained from microbial activity was also analysed. SPSS software version 23 was used to analyse all the parameters on the soil properties, that of the heavy metals and microbial activity. While Microsoft excel was used to analyse the data from microbial activity and the graphical presentations.

CHAPTER FOUR

RESULTS AND DISCUSSION

This chapter highlights the various results obtained after laboratory analysis of the soil samples and the plant samples. It describes the trend in heavy metal accumulation at both fields, the accumulation levels are being compared to FAO/WHO acceptable limits in soils. The chapter also unveils the levels of heavy metal accumulation in the existing plants at both study fields including that of the control plots at South Suntreso and Edwenase. Results for microbial activity are highlighted in this chapter. In all, the various results have been discussed thoroughly in this chapter; providing some form of reasons for some observations in the data provided.

Nutrient Content in the Soil

Soil pH

Considering the effect on site which was in evidence Table 1, the exact point of significant effect has been unveiled at Figure 1 and 2. Irrespective of the depth, pH at South Suntreso auto mechanic shop was higher than that of Edwenase shops. Paying much attention to the above graphs, the average pH levels at south Suntreso valley bottom was higher than that of south Suntreso. The level of pH at South Suntreso and Edwenase stood at 7.57 and 6.87 respectively. At the same time the average pH for the control at south was higher (less acidic) to that of Edwenase which showed acidic properties. At each site, the average pH for control was much lower than the study fields at both sites (figure 4 and 5).

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The pH levels of valley bottom at South Suntreso were higher than that of the upper slope of the same site. The difference was significant from each mean. The reverse occurred at Edwenase where the pH was higher at the upper slope followed by the middle slope and the least recorded at the valley bottom. In general, while the pH levels kept rising from upper slope to through to the middle slope down the valley bottom at South Suntreso, the reverse was recorded at Edwenase were showed a decreasing trend from upper slope through to the valley bottom. Again, all the controls were lower than its corresponding study fields irrespective of the depth. All in all, the level of pH at the two fields was good enough for crop growth (Figure 4 & 5).

It was predicted that there will be higher levels of pH at both study sites since all of them had a sloppy adjoining fields which prevented automobile waste, especially, the metals in it from making the soil acidic. Again, all the two fields of study had enough organic materials to keep the pH at the right level. The artisans dump most of their refuse example polythene bags, left-over food, human faeces among others, so the prediction was right since the soil was in good conditions even with the presence of automobile waste on the fields.

Figure 4 : pH content of soils sampled from South Suntreso and Edwenase study sites depth (0-10cm). Column area means and error bars are standard errors of the means at $p < 0.05$.

■ 10-30cm Upper slope ■ 10-30cm Middle slope ■ 10-30cm Valley bottom

Figure 5: pH content of soils sampled from South Suntreso and Edwenase study sites depth (10-30cm). Column area means and error bars are standard errors of the means at $p < 0.05$.

% Organic carbon

Organic carbon content was not significantly different in terms of the two sites in general with mean of 2.12. Considering the various depths, % 0rganic matter was significantly different from upper slope through to middle slope and valley bottom. The levels of organic carbon with respect to site and depth interaction were also significantly different (Figure 6 & 7).

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Delving into the above information at site specific, organic carbon content in the soil at South Suntreso was much lower on average than that of Edwenase in average irrespective of the depth. The same situation was realized with the controls where South Suntreso rather dominated with higher organic carbon content as against Edwenase. There were significant differences in % OC levels at depth of 0-10 cm (top soil) and 10-30 cm (sub-soil), % OC was much lower at 0-10cm depth to that of 10-30 cm depth at South Suntreso considering all the slopes in the study. Control at South Suntreso had the overall higher % OC from both fields. There was significant difference between the two depths of upper at South Suntreso. At 0-10 cm % OC was 0.99 but depth 10-30 cm recorded 1.25 which showed significant difference. At Edwenase the significance was realized at the middle slope; at depth 0-10 cm the % OC was 1.49 while at depth 10-30 cm the significance was realized as low as 0.55 % OC. Control at south showed massive significance level of % OC; the difference affected all the slopes where depth 0-10cm took the highest mean as against depth 10-30 cm (Figure 7).

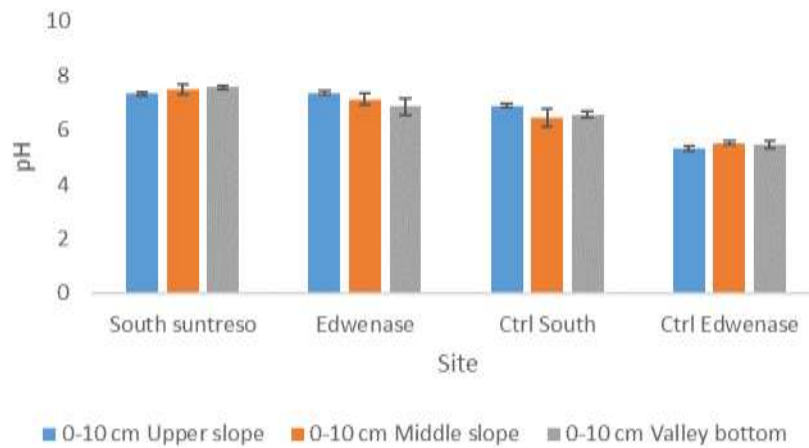*Figure 6* : % Organic carbon content of soils sampled from South Suntreso and Edwenase study sites depth (0-10cm). Column area means and error bars are standard errors of the means at $p < 0.05$.
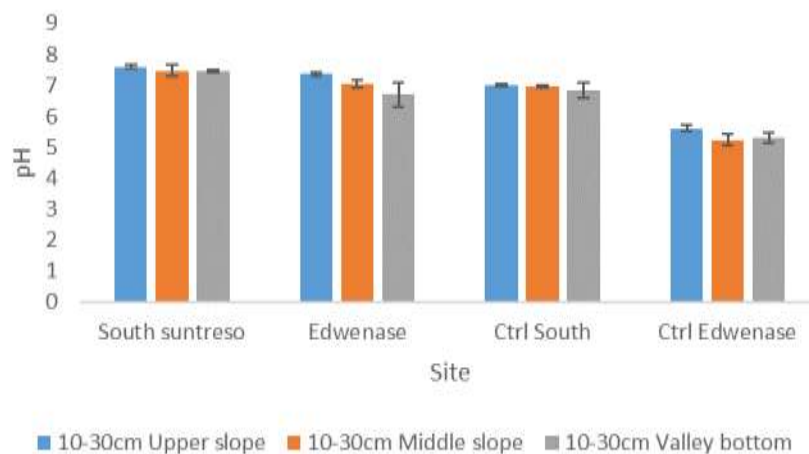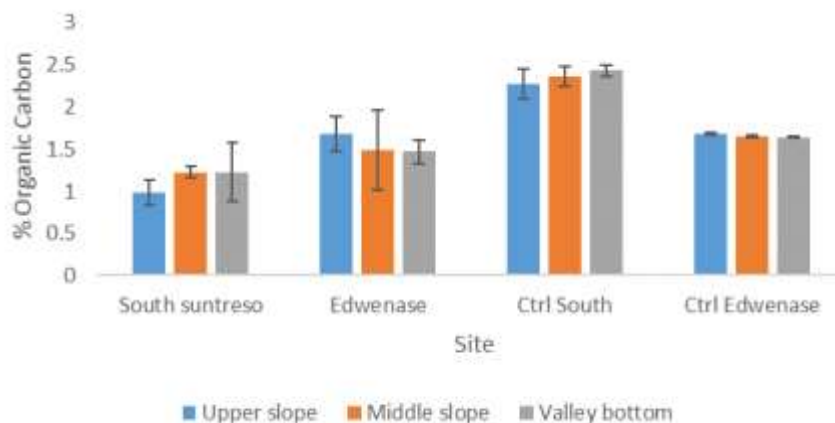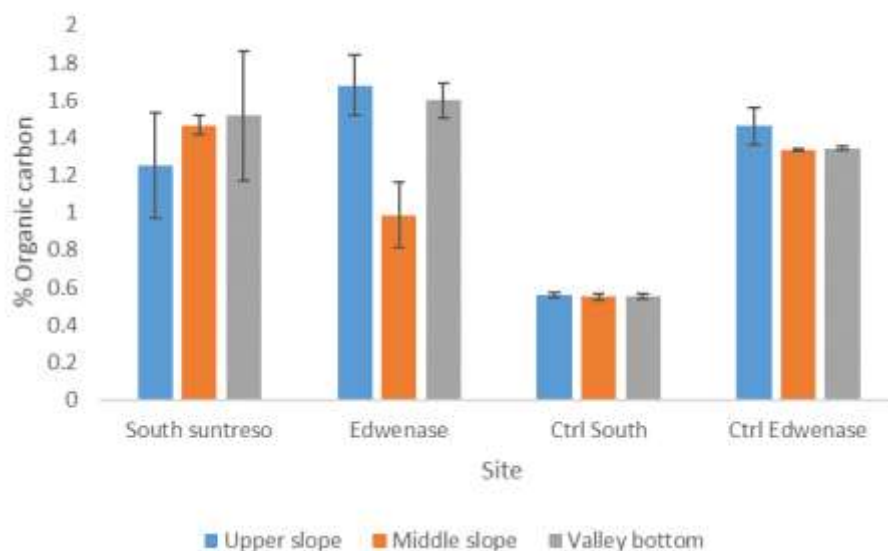

Figure 7: % Organic carbon pH content of soils Sampled from South Suntreso and Edwenase study sites depth 10-30cm. Column area means and error bars are standard errors of the means at $p < 0.05$.

Total nitrogen

Apart from looking at the site in general, none of the other treatments or its interactions showed any significant difference between their average means. Control soils at South Suntreso irrespective of the depth had the highest nitrogen. The rest had fair distribution of nitrogen. South Suntreso upper slope with depth 0-10 cm and Edwenase middle slope depth 10-30 cm recorded the same means (0.09) which was the least nitrogen discovered. The highest mean was found at South Suntreso control middle slope at depth 0-10 cm. there was significant difference between South Suntreso valley bottom of depth 0-10 cm and 10-30 cm at control (Figure 8 & 9).

Figure 8: % Nitrogen content of soils sampled from South Suntreso and Edwenase study sites depth 0-10cm. Column area means and error bars are standard errors of the means at $p < 0.05$.

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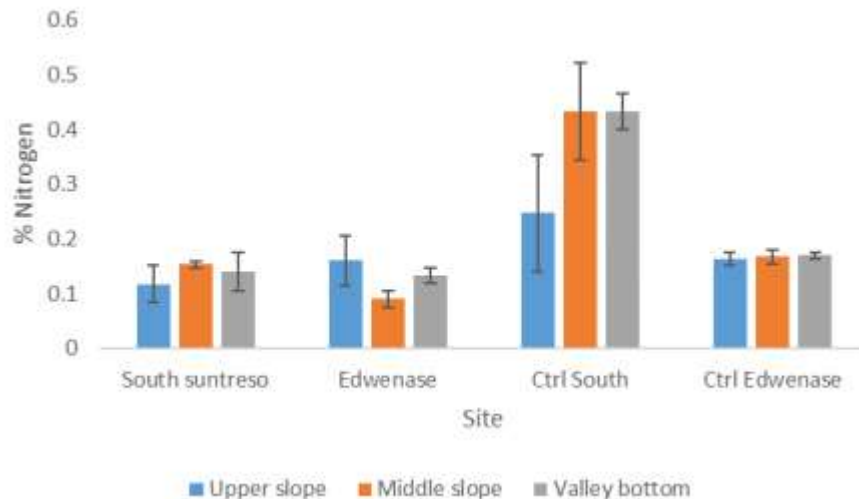
Figure 9: % Nitrogen content of soils sampled from South Suntreso and Edwenase study sites depth 10-30cm. Column area means and error bars are standard errors of the means at $p < 0.05$.

**Phosphorus**

Site and depth had significant effect on phosphorus levels in the soils sampled at the two sites. The valley bottom of both South Suntreso and Edwenase at depth (0-10 cm) was significant to each other (figured 11). The same situation was found at 10-30 cm depth of South Suntreso and Edwenase. Control for south Suntreso was significantly different from the main study field at South Suntreso all at 0-10 cm depth. Similar situation was found at depth 10-30 cm. considering the two control sites alone, Edwenase had higher phosphorus content than South Suntreso; there were significant differences between their means (Figure 10 & 11). The highest phosphorus was found at Edwenase upper slope depth 0-10cm with an average mean of 48.65 mg/kg. The least phosphorus content was recorded at South Suntreso control middle slope at depth 10-30 cm with an average mean of 1.73 mg/kg. South Suntreso and Edwenase upper slope depth 0-10 cm also had significant means between each other.

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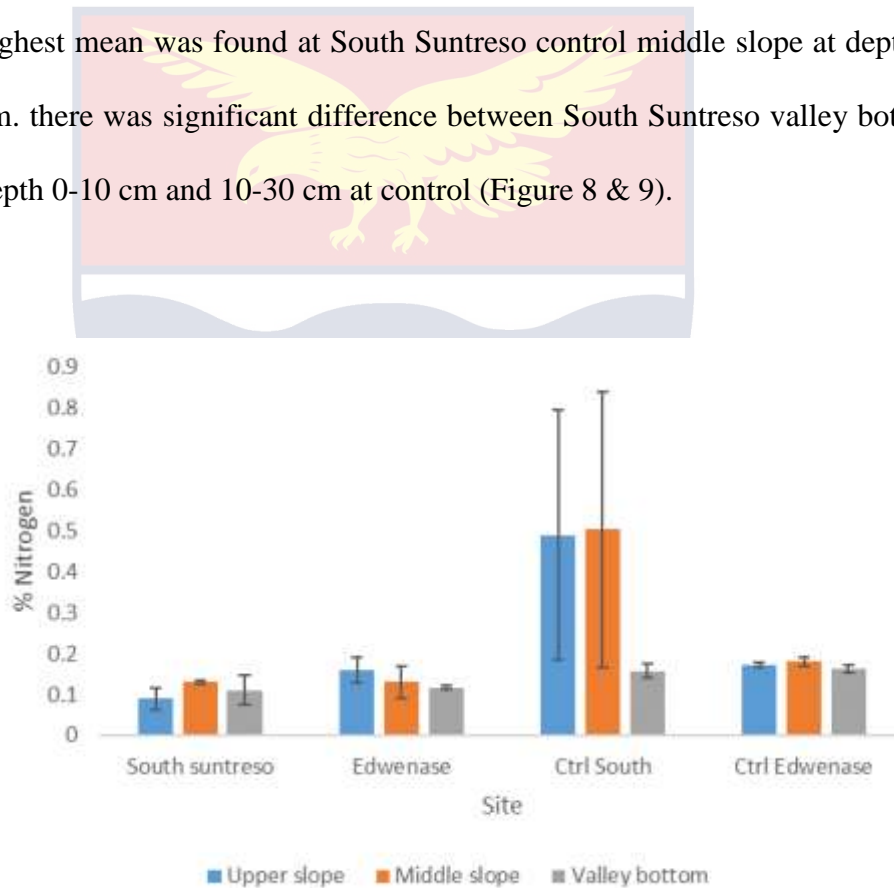*Figure 10*: Phosphorus content of soils sampled from South Suntreso and Edwenase study site depth 0-10cm. Column area means and error bars are standard errors of the means at $p < 0.05$.
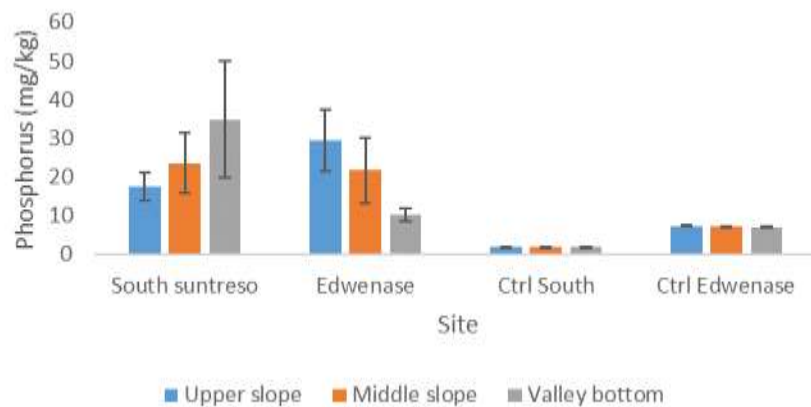
Figure 11: Phosphorus content of soils sampled from South Suntreso and Edwenase study sites depth 10-30cm. Column area means and error bars are standard errors of the means at $p < 0.05$.

Total exchangeable base (T.E.B)

Considering the site, above graph (fig. 12 and 13) show there was significant effect on T.E.B. This was evident at the two study fields; the means for South Suntreso middle slope was much higher and significant to that of Edwenase middle slope all at depth 0-10 cm. with respect to the control, South Suntreso recorded higher levels of T.E.B irrespective of the depth.

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At Edwenase depth 0-10 cm, T.E.B levels decreased from upper slope through the middle slope and to the valley bottom. South Suntreso recorded the highest level of T.E.B from both depths. South Suntreso and Edwenase had significant differences between them at both depths. The control for south from the three slopes was lower than the corresponding means at the main field which revealed much significant differences. The control for Edwenase also recorded much lower levels as compared to the corresponding study field which revealed some form of significance between the means. On the average, South Suntreso recorded higher T.E.B levels than Edwenase.
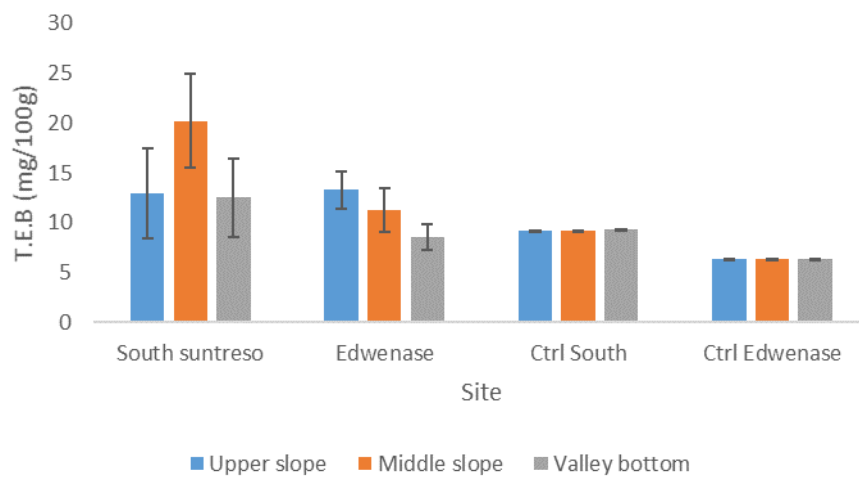
Figure 12 : T.E.B Content of soils sampled from South Suntreso and Edwenase study sites depth 0-10cm. Column area means and error bars are standard errors of the means at $p < 0.05$.
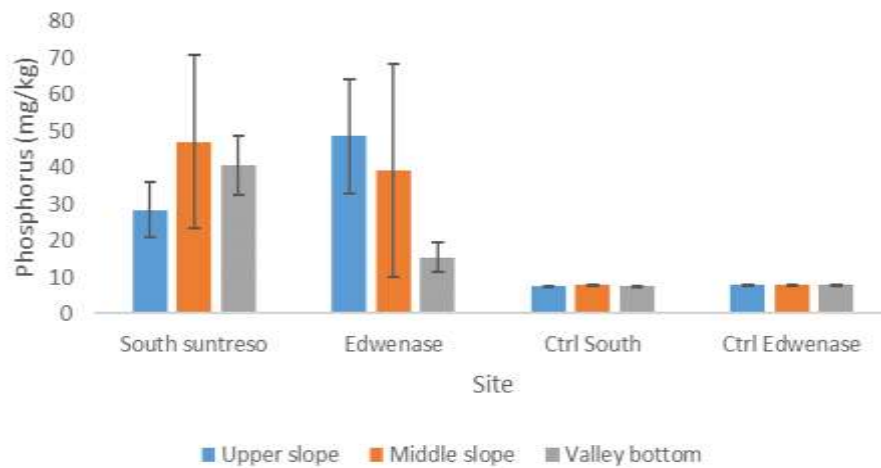
65
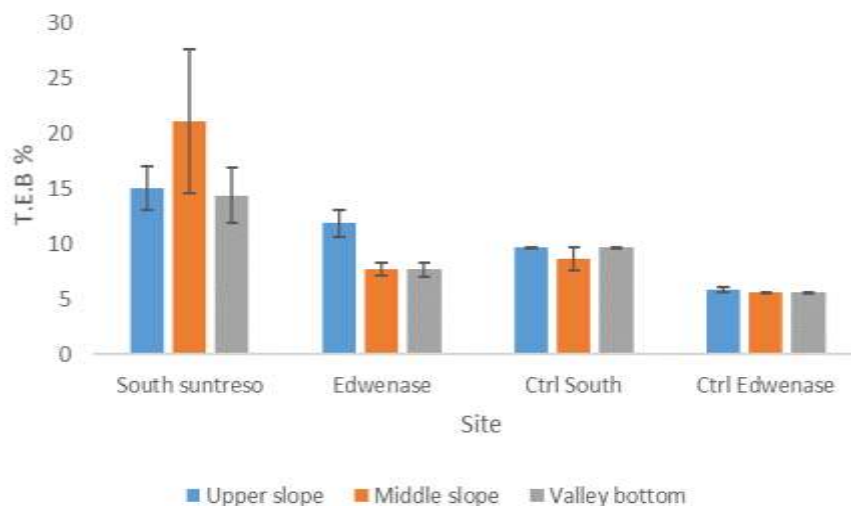
Figure 13: T.E.B Content of Soils Sampled from South Suntreso and Edwenase Study Sites Depth 10-30cm. Column Area Means and Error Bars are Standard Errors of the Means at P < 0.05.

Heavy Metal Levels in the Soil

Cadmium (Cd)

From Figure 14, the highest mean recorded at South Suntreso was 0.125mg kg-1 at valley bottom (10-30cm). The lowest mean 0.00 mg kg-1 was also recorded at the control plot upper part. The overall level of Cadmium (Cd) in the soil at the workshops showed significant difference to the control plot. Cadmium levels at the affected field increased from upper part down the slope. Plots at South Suntreso showed some significant difference in cadmium at both depths of the upper part and valley bottom. However, there was no significant difference in means at the middle part of the slope. Considering the control plot, both the slope and depth did not show any significant difference at P< 0.05 (Figure 14). Cadmium level was high at the valley bottom at both depths. At the control plot, cadmium was evenly distributed between the slopes and both depths. All the levels recorded at the workshops and control plots were below FAO / WHO (2001) permissible limit.

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Cadmium was found to be higher at both depths of the valley bottom. This was attributed to run-off because of the sloppy nature of the field. Sediments were being deposited at the valley bottom resulting in its highest accumulation. These results are similar to the findings of Adelekan and Alanwode (2011); where Cd levels were below the allowable limit. Cadmium movement is as a result of its mobility. It is also soluble in water. It may have also contributed to its highest level found at the valley bottom of the workshop. Cadmium tends easily move through soil systems than many other heavy metals (Alloway, 1995). Kuo, Heilman, & Baker (1983) made similar conclusions while stressing on higher vertical mobility of Cd as compared to Cu and Zn in soils in a study conducted in Montana, USA.

The overall mean cadmium content at South Suntreso was significantly higher than Edwenase. The control was also significantly different at both sites; the mean control at south was higher than that of Edwenase. The highest mean was recorded at South Suntreso valley bottom depth 10-30cm which was 0.13 mg/kg, whereas the least cadmium level was recorded at Edwenase middle slope at both depths which also recorded 0.001 mg/kg. From the graph, it shows Edwenase at both depths including control had little or no cadmium content. However, South Suntreso upper slope at both levels had very low cadmium levels recording 0.002 which was also similar to that of Edwenase. Considering South Suntreso main field and control which had an appreciable cadmium levels, there were some significant levels showing between them at both depths; at depth 10-30 cm the means from upper slope through middle slope down the valley bottom showed significant differences from each other

where valley bottom was higher followed by middle slope and upper slope recording the least (Fig. 14).

Similar situation was realized at South Suntreso depth 1-10cm in terms of the slopes, but significant level was only recorded between upper slope and valley bottom. The control at south had evenly distribution of cadmium from the three slopes. Considering the two depths under study, South Suntreso dominated. The cadmium levels recorded at both sites were all within the FAO limit which is 3mg/kg. These results have been confirmed by Adelekan (2011) where levels of Cd were below the allowable limit. This finding is in agreement with the work done by Mensah, Vlek & McCarthy (2017), where some cadmium values recorded at a clustered automechanic shop were below the permissible limit by WHO/FAO (2001). Cadmium is very much linked with non-residual fractions of heavy metals.

Cadmium tends easily move through soil systems than many other heavy metals (Alloway, 1995). Kuo et al. (1983) made similar conclusions while stressing on higher vertical mobility of Cd as compared to Cu and Zn in soils in a study conducted in Montana, USA. This makes them moveable and potentially bio-available for uptake by plants (Zhang et al., 2009). It is also in agreement with work done by Pam et al. (2013) who investigated heavy metals in soils of auto-mechanic shops and refuse dump sites in other parts of Makurdi, Central Nigeria, as well and reported a range of 0.6 - 3.5 mg/kg of cadmium content. Cadmium was predicted to have come from lubricating oils which were poured on the field directly while servicing vehicles. Another prediction was on the vehicle wheels which are sometimes burnt at the workshops to ease space. Masindi (2018) asserted that the ferrous steel

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industry is the main source of environmental Cd pollution. Naskret (2004) also found out that the accumulation of Cd in the study locations is predicted to come from lubricating oils, vehicle wheels and metal alloys used to harden engine parts.

Figure 14 : Heavy Metal (Cd) Levels in the Soil at South Suntreso
   *Bars with the same letters are not significantly different at p < 0.05 LSD

Lead (Pb)

From Figure 15, it indicates that at least all the soils sampled at the auto-mechanic workshop at South Suntreso had some levels of lead in them but at different concentrations. Considering the main field of study and the control plots, there were significant differences in the means at p < 0.05. Between the two sites, South Suntreso recorded the highest lead (Pb) content at both depths and slopes. The highest lead level of Pb found at South Suntreso was upper part depth 0-10cm cm with a mean of 32.43mg kg-1 followed by the middle part at depth of 0-10 cm with mean of 32.09mg kg-1.

The lowest level of lead at the main study field was found at the valley bottom depth 0-10 cm with a mean of 10.9mg kg-1. For the control, the upper, middle and the valley bottom recorded minute levels of Pb showing no significant differences among each other. Even at depth 10-30cm, there was no record of Pb in the soil.

This result is similar to work done by Fosu-Mensah et al. (2017) where the ANOVA showed significant differences ($p < 0.05$) in the mean concentration of Pb between the main study field at auto mechanic workshops and the control plots. His finding was attributed to low mobility of Pb in the soil. Most of the concentrations were limited to upper and middle part at depth 0-10cm. Critical study of the levels of lead at the two depths indicates higher levels were found at depth 0-10cm except for the valley bottom. This means lead was much accumulated at the top soil (Fig. 15).

This finding is in agreement with work done by Adelekan (2011), where the highest concentrations of Pb were recorded in the 0 to 15 cm layer while the least were recorded in the 45 to 60 cm of the soil in most cases and this shows a linear correlation of reduction with depth through the soil layers. This observation of higher reservation of Pb in the top layers of the soil affirms the finding of Davies (1995) which stated that lead is especially liable to accumulation in surface horizons of soil because of its low water solubility resulting in very low mobility. Sam (2015) also recorded higher levels of Pb in soils sampled from oil polluted soils. With reference to FAO (2001), Pb levels at upper and middle depth 0-10 were all above the permissible limit of 15 mg kg-1. The rest were within the acceptable limits.

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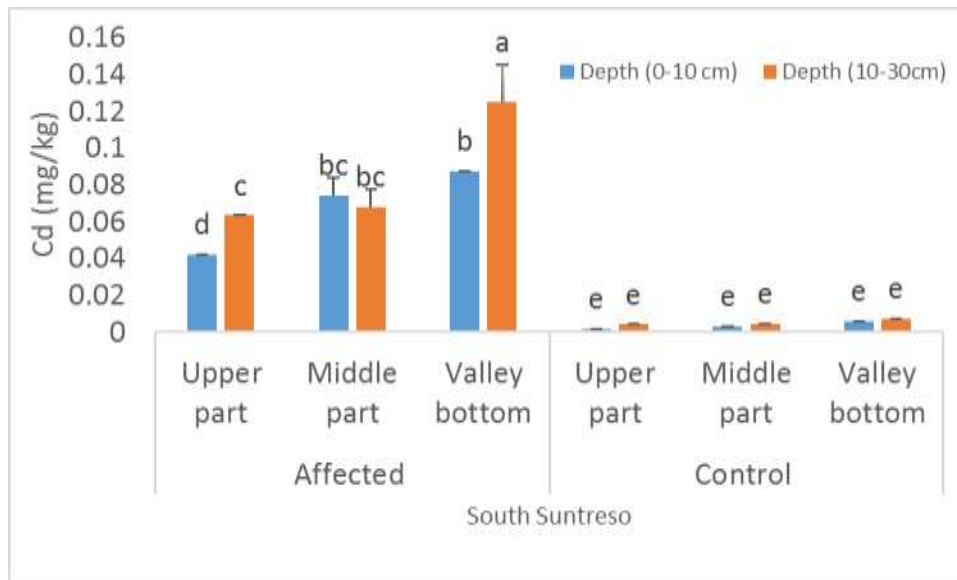*Figure 15* : Heavy Metal (Pb) Levels in the Soil at South Suntreso
        *Bars with the same letters are not significantly different at p <
        0.05 LSD.

Zinc (Zn)

Zinc was found in all the soil samples collected for the study. There were significant differences between the main auto-mechanic fields and the control plot. The control plots recorded the least of the means while the main field had an accumulation of higher zinc levels, this was attributed to the anthropogenic activities of the artisans at the clustered auto-mechanic workshops. There were no significant differences in depth at both the main field and the control plots. The highest mean was recorded at valley bottom depth 10-30cm with a record of 32.5mg kg-1 accumulation level followed by middle part depth 0-10cm at a mean of 32.2mg kg-1. Heavy metals are mostly of anthropogenic origin (Comfort, Ofosu, Bamford, Wordson, Atiemo, Aboh, & Adeti, 2013).

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The fact that there were little or low accumulation of zinc the control affirms the above assertion that heavy metal presence was not from a natural deposit but from the activities of artisans at the auto-mechanic workshops which had a repining effect on the soils. Figure 16 shows an increasing trend in the accumulation of zinc from upper slope along the catena to the valley bottom especially at depth 0-10cm. Results from the affected fields ie. Auto mechanic cluster were highly above FAO / WHO (2001) permissible limit of 3.5-6 gm kg-1. This was confirmed by Abenchi et al. (2010); the increasing level of Zn emanated from the auto mechanic clusters, since this element is found as part of many additives to lubricating oils, metal scraps, burning of tyres, welding and soldering.

The reverse happened at Edwenase where zinc levels showed a decreasing trend from upper slope through the middle down the valley bottom. Zinc was fairly distributed at various depths of the two study sites. All the levels of zinc recorded in the soil samples were highly above the FAO permissible limit of 3.5-6 mg/kg. The higher level of zinc found in the soil was attributed to the anthropogenic activities by auto mechanics. This was confirmed by Abenchi et al., (2010); the increasing level of Zn emanated from the auto mechanic clusters, since this element is found as part of many additives to lubricating oils.
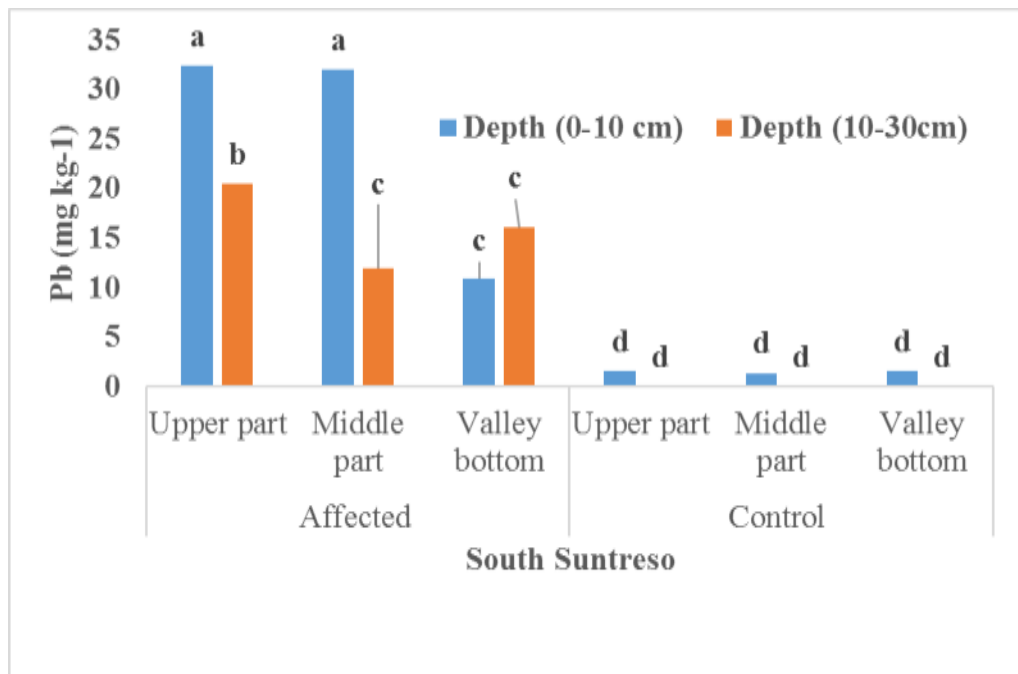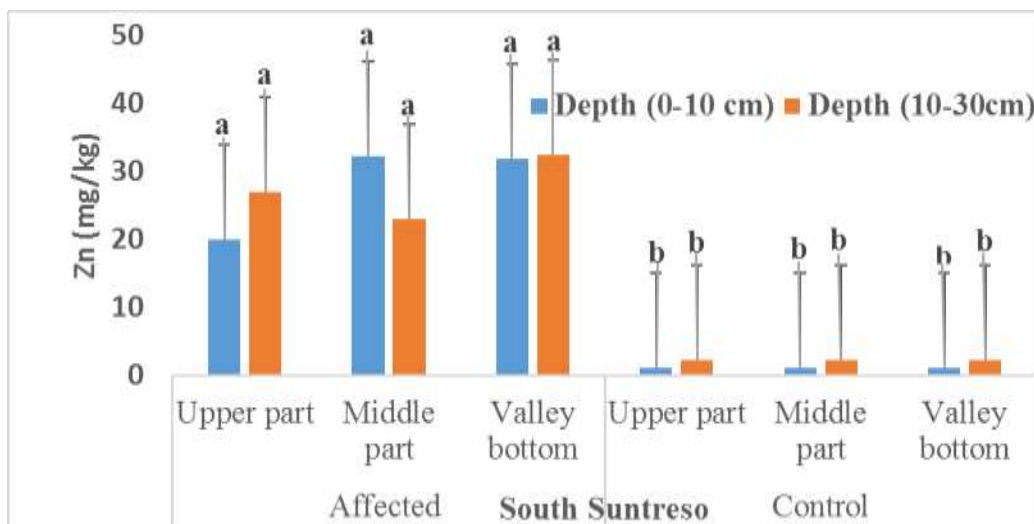
Figure 16 : Heavy Metal (Zn) Levels in the Soil at South Suntreso
        *Bars with the same letters are not significantly different at p <
        0.05 LSD

Arsenic (As)

At South Suntreso, arsenic (As) was also found in all the soils sampled for the study. The highest level arsenic was recorded at the valley bottom depth 10-30cm with mean of 11.72mg kg-1; while the lowest level being 0.82mg kg-1 at control plot at the upper part depth 0-10cm. Comparing the control plot to the main field, these was significant differences between the means. However, considering only the workshops, there was no significant difference between the means at both depths but there was significant difference between the valley bottom to the upper part and middle part of the field. Also, there was no significant difference in depths and slope at the control plots. The valley bottom at the workshop recorded the highest accumulation of arsenic. This was attributed to run off activities from the upper part of the slope down to the valley bottom. The run-off is able to deposit sediments at the valley bottom.

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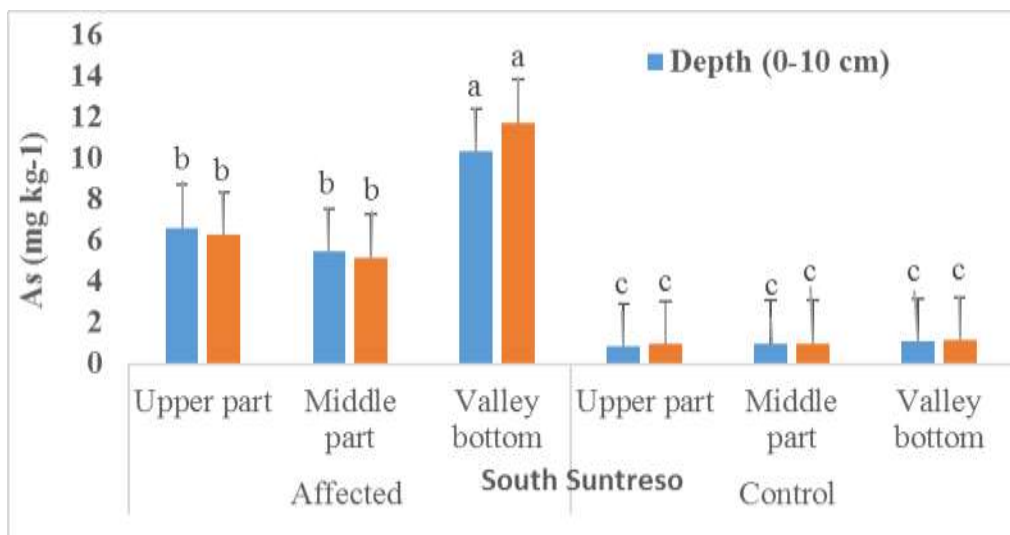
Figure 17: Heavy Metal (As) Levels in the Soil at South Suntreso
　　　　　*Bars with the same letters are not significantly different at p <
　　　　　0.05 LSD

Heavy metal levels in soils at Edwenase

Arsenic (As)

From Table 1, arsenic content recorded at Edwenase showed significant difference between all the soils sampled at the workshop and the control plot. Considering the slope, soils at the workshop recorded some significant differences between the means; the valley bottom of the upper part depth 0-10cm was significantly different from the upper and middle parts of the slope. Also, at depth 10-30, each of the slope was significantly different from each other i.e. Upper, middle and valley bottom. There was no significant difference between the means in terms of slope (upper, middle and valley bottom) at the control plot. The highest level of arsenic was recorded at the valley bottom of the workshop depth 0-10cm with mean concentration of 49.19 ± 1.01, followed by the same valley bottom at depth 10-30cm at the workshop with mean of 46.82 ± 4.55. There was no record of arsenic level at the control field. There was an increasing trend of arsenic levels from the

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upper part through the middle and to the valley bottom of the slope at both depths.

The workshop area experienced this gradual increase in the arsenic levels (Table 1). It was attributed to run-off and subsequent deposition of sediments downslope. Arsenic seems to have accumulated more at the sub soil at depth 10-30cm. This was due to seepage as a result of the nature of the soil which was sandy loam. In general, arsenic levels at Edwenase were above the FAO permissible limit which is 20 mg/kg. These results affirm the work done by Fosu Mensah *et al*. (2017) in a related work done in soils closer to auto-mechanic clustered workshops. Arsenic binds strongly with soil which is absorbed easily in the soil; its mobility in the soil is high. The most dominant site for arsenic was Ewdenase, including the various slopes and depths (Table 1).

Lead (Pb)

From Table 1 there was accumulation of lead (Pb) in the soils sampled at Edwenase auto mechanic workshop and the control plots at different levels. Considering the workshops and the control plot, there was significant difference between them at $P < 0.05$. The highest level of Pb was recorded at the upper part depth 0-10cm of the main workshop study area and the least Pb level showed at upper part depth 0-10cm at the control plot (Table 1). Considering the slope, there were significant differences between the valley bottom, middle slope and the upper part of the study area all at depth 0-10cm. same significant difference was realized at depth 10-30cm. Results from the

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control plot at Edwenase did not show any significant difference the slope and depth.

With respect the depth as a factor, the workshop recorded significant difference in the means at along the slope. Only the valley bottom of the workshop did not experience any significant difference in depth. The upper and middle part of the slope recorded significant differences in depth. Critical study of the trend in Pb indicates a different trend unlike arsenic and zinc where their levels increased as it moved down from upper, middle to the valley bottom. In effect the valley bottom of As and Zn had higher levels of accumulation. Pb rather showed a decreasing trend from upper, middle through to the valley bottom. The upper slope recorded the highest levels, this was as a result of poor absorption of lead in solution. Again, lead mobility is very slow thereby accumulating more at the upper part of the slope.

The levels of Pb recorded at Edwenase were below FAO/WHO (2001). This report is consistent with Nwachukwu et al. (2011) who also reported that the values of Pb obtained in his study were lower than the 1162 mg/kg for auto mechanic workshop area in Owerri, South-East Nigeria. Allowable limits of Pb concentrations differ widely with countries (Lacatusu, 2000). The concentration of lead (Pb) was predicted to have emanated from the waste vehicle batteries and other used oils that are poured on the soil by the mechanics at the study site. It was reported that Pb has the highest composition of heavy metals in waste oils (Wuana, 2011). It is possible that these levels of Pb is increased by the amount of waste oil, presence of automobile emissions, and expired motor batteries indiscriminately dumped by battery chargers and auto mechanics in the surrounding areas. Pb at auto-

electrical section are mainly from the expired acid Pb batteries. Both sites have auto-electricians who sometimes deliberately pour the acid in car batteries while servicing cars. This is the more reason why Pb was present at Edwenase.

Zinc (Zn)

From Table 1, Zinc (Zn) content recorded at Edwenase recorded significant difference between all the soils sampled at the workshop and the control plot. From the slope, soils at the workshop recorded some significant differences between the means; the valley bottom of the upper part depth 0-10cm was significantly different from the upper and middle parts of the slope. Also, at depth 10-30, each of the slope was significantly different from each other i.e. Upper, middle and valley bottom. On the contrary, there was no significant difference between the means in terms of slope (upper, middle and valley bottom) at the control plot. The highest level of Zinc was recorded at the valley bottom of the workshop depth 0-10cm with mean concentration of 58.5 ± 4.60, followed by the valley bottom at depth 10-30cm at the workshop with mean of 48.9 ± 9.40. There was an increasing trend of Zn levels from the upper part through the middle and to the valley bottom of the slope at both depths.

The workshop area experienced this gradual increase in the Zn levels but the control plots did not record this increasing pattern along the slope. (Table 1). It was attributed to run-off and subsequent deposition of sediments downslope. Higher levels of Zn were found at the sub soil at depth 10-30cm. This was due to seepage as a result of the nature of the soil which was sandy

loam. In general, Zn levels at Edwenase were highly above the FAO permissible limit which is 3.5mg kg-1.

The progressive pollution of the sites is mainly due to the unprofessional approach to auto repair works and the inappropriate disposal of the wastes. Zinc is an essential microelement which plays a very vital role in enzyme reactions but its content differs with the type of soil (Abidemi, 2011). High concentration of Zn can, however, pose health threats to human. Heavy metals are mostly of anthropogenic origin (Comfort et al., 2013). Results from Adelekan (2011) are in agreement with the presence of Zn at the workshop at different levels in this study. He also attributed the presence of Zn to the activities of artisans at the workshops affecting the soils around the cluster.
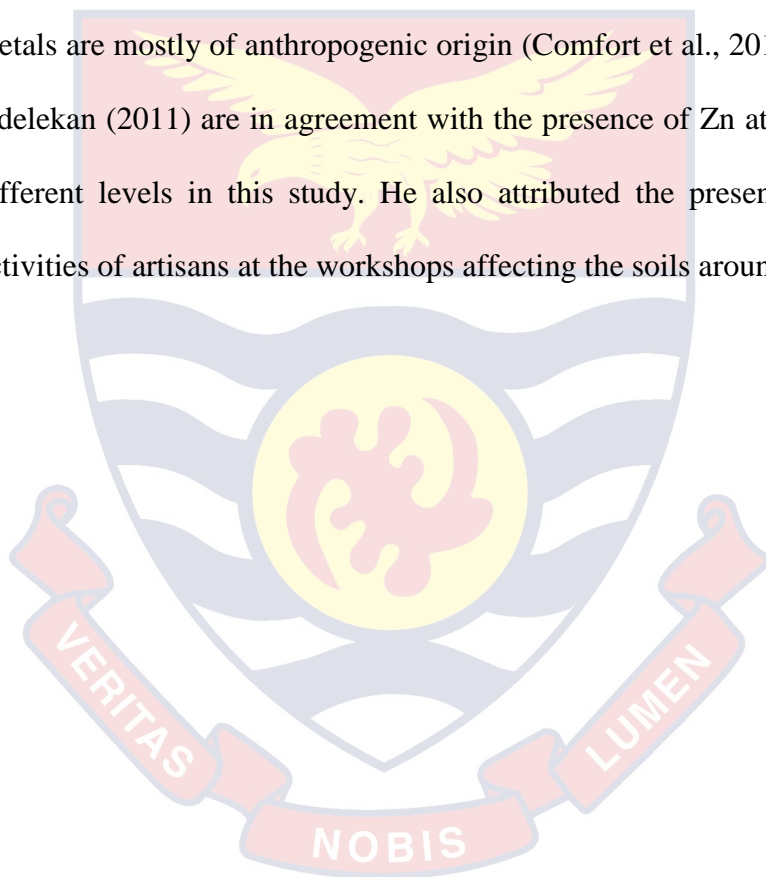
Table 1 - *Heavy Metal Levels in Soil Samples at Edwenase*

State of field / Depth	Part of slope	As (mg kg-1)	Pb (mg kg-1)	Zn (mg kg-1)
Affected (0-10cm)	Upper part	20.31 ± 1.87^c	17.33 ± 1.01^a	12.6 ± 3.27^{cd}
	Middle part	22.8 ± 2.65^c	15.37 ± 0.51^a	14.6 ± 5.26^c
	Valley bottom	49.19 ± 1.01^a	6.26 ± 1.24^c	58.5 ± 4.60^a
Affected (10-30cm)	Upper part	12.86 ± 2.2^d	11.41 ± 1.70^b	11.2 ± 1.58^{cd}
	Middle part	35.08 ± 4.53^b	7.47 ± 2.84^c	36.4 ± 6.70^b
	Valley bottom	46.82 ± 4.55^a	7.42 ± 0.30^c	48.9 ± 9.40^a
Unaffected (0-10cm)	Upper part	0.00 ± 0.00^e	1.83 ± 0.01^d	2.2 ± 0.00^d
	Middle part	0.00 ± 0.00^e	1.86 ± 0.01^d	2.2 ± 0.00^d
	Valley bottom	0.00 ± 0.00^e	2.02 ± 0.01^d	2.2 ± 0.00^d
Unaffected (10-30cm)	Upper part	0.00 ± 0.00^e	1.94 ± 0.01^d	3.7 ± 0.00^{cd}
	Middle part	0.00 ± 0.00^e	1.98 ± 0.01^d	3.7 ± 0.00^{cd}
	Valley bottom	0.00 ± 0.00^e	2.05 ± 0.01^d	3.7 ± 0.00^{cd}
FAO /WHO P L (2001 (mg kg-1)		20	15	3.5
LSD		6.68	3.10	10.60

*Mean with the same letters are not significantly different at $p < 0.05$ LSD

Heavy Metals in Plants (Maize) Samples at Edwenase

Arsenic

From Table 2, Arsenic (As) content recorded at Edwenase showed significant difference between all the plants sampled at the workshop and those at the control plot. From the slope, plants at the workshop recorded some significant differences between the means; Arsenic level at the valley bottom of the upper part was significantly different from the upper and middle parts of the slope at the workshop. However, there was no significant difference between the means in terms of slope (upper, middle and valley bottom) at the control plot. The highest level of arsenic was recorded at the valley bottom of the workshop with mean concentration of 23.81 ± 0.50, followed by the middle part at 22.06 ± 1.36 and the last accumulation level showing at the workshop with mean of 18.07 ± 0.36. There was an increasing trend of As levels in maize samples from the upper part through the middle and to the valley bottom of the slope.

The presence of arsenic in the maize plants at Edwenase comes as a result of the presence of high arsenic levels in the soils located at the workshop. Mobility of arsenic in maize plant is faster through the roots of the plant. This observation is consisted with work done by Fosu-Mensah et al., (2018). The mean concentration of arsenic in his work was above the FAO/WHO (2007) permissible limit of arsenic in plant samples. High concentration of arsenic in plants can cause nausea, vomiting, diarrhea, cough, headache and cardiovascular disease to animals especially livestock (Tchounwou, Yedjou, Patlolla & Sutton, 2012). The workshop area

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experienced this gradual increase in the as levels but the control plots did not record this increasing pattern along the slope.

Lead (Pb)

Lead (Pb) content in maize samples recorded at Edwenase showed significant difference between all the plants sampled at the workshop and those at the control plot. From the slope, plants at the workshop recorded some significant differences between the means; Pb levels at the valley bottom of the upper part were significantly different from the upper and middle parts of the slope at the workshop. On the contrary, there was no significant difference between the means in terms of slope (upper, middle and valley bottom) at the control plot (Table 2). The highest level of Pb was recorded at the upper part of the workshop with mean concentration of 0.423 ± 0.01, followed by the middle part at 0.421 ± 0.00 and the last accumulation level showing at the valley bottom of the workshop with mean of 0.416 ± 0.00.

There was a decreasing trend of Pb levels in maize samples from the upper part through the middle and to the valley bottom of the slope. This was a reflection of the trend of Pb levels in the soil. Lead mobility of Pb caused the levels recorded at the workshop. Pb has little plant motion (Batista et al., 2017). Plant roots can also uptake small amounts of Pb (Celik et al., 2005). This finding is also in agreement with work done by Amoakwah et al. (2020) in a related work. There were high levels of Pb in plants as a result of plant uptake. All the levels of Pb found in maize samples were a little above FAO/WHO (2010) permissible level of 0.3mg kg-1. In a related work done by Fosu-mensah et al. (2018), the concentration of lead in plant samples exceeded

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the WHO/FAO (2010) permissible limit of 0.3mg kg-1. The consumption of high amount of lead can cause anaemia, headache, brain damage, and nervous system disorder to humans and animals (Shi, Chen, Bi, et al., 2011).

Zinc (Zn)

In this study, there were accumulations of zinc at various levels in maize sampled at the clustered workshop. Zinc content in maize samples at Edwenase also recorded significant difference between all the maize plants sampled at the workshop and those at the control plot (Table 2). From the slope, plants at the workshop recorded some significant differences between the means; Zn levels at the valley bottom were significantly different from the upper and middle parts of the slope at the workshop. In addition, there was no significant difference between the means in terms of slope (upper, middle and valley bottom) at the control plot where maize samples were also taken and analysed for heavy metals.

The highest level of Zn was recorded at the valley bottom of the workshop with mean concentration of 47.0 ± 0.00, followed by the middle part at 45.5 ± 0.01 and the last accumulation level showing at the upper slope of the workshop with mean of 28.2 ± 0.01. There was an increasing trend of Zn in maize samples from the upper part through the middle and to the valley bottom of the slope. Zn results from (Table 2) recorded some levels of Zn highly above the FAO/WHO (2010) permissible limit of 27.4mg kg-1.

High level of zinc in the plants is in agreement with a previous study showing that lettuce and maize accumulated zinc to a greater extent (Sauerbeck, 1991) in a related work where maize plants recorded high levels

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of zinc above the FAO/WHO permissible limit. Although Zn is one of the essential nutrient elements that play a vital role in physiological and metabolic processes of many living organisms, it can result in adverse effects when its concentration reaches above permissible limits (Fosu-Mensah et al. 2018). It was observed from the report that each metal differed considerably in uptake from each other in the Table 2.

This observation is consistent with Intawongse (2006), who had similar results of differed metal uptake in plants from polluted soils. Soil pH is considered one of the most relevant factors that influence the metal transfer from soil to plants, and higher pH was reported to decrease the bioavailability and toxicity of metals such as Cd and Pb (Wang et al., 2006). The pH of the studied soil was 7.4, which could not have contributed much to the metal absorption mechanisms of plant.

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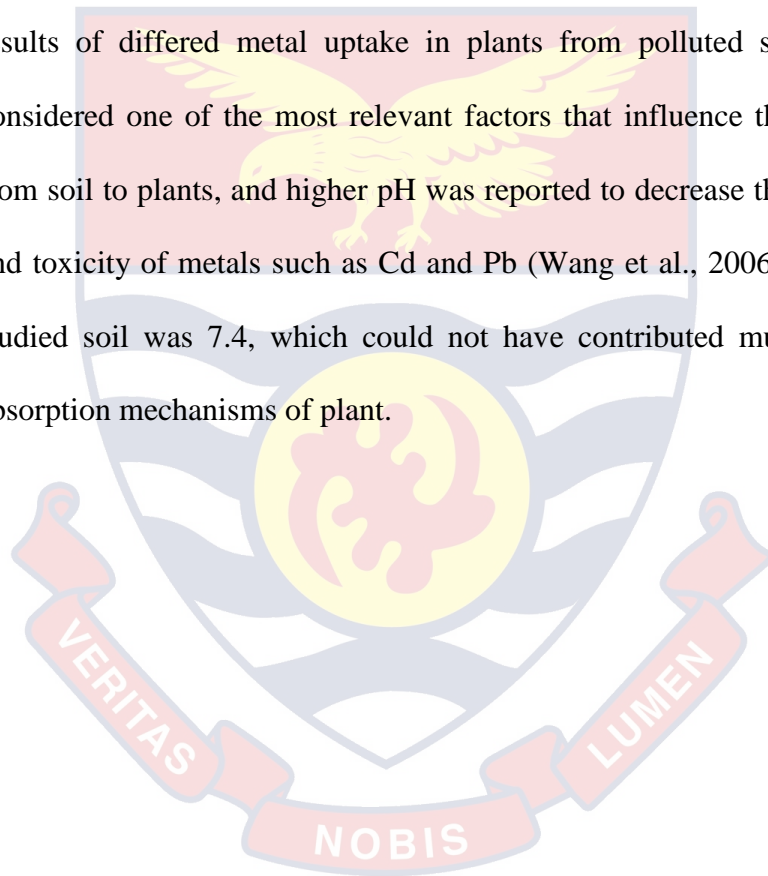
Table 2 - *Heavy Metal Levels in Maize Plants at Edwenase*

State of Field	Part of Slope	As (mg kg-1)	Pb (mg kg-1)	Zn (mg kg-1)
Affected / Edwenase	Upper part	18.07 ± 0.36^c	0.423 ± 0.01^a	28.2 ± 0.01^b
	Middle part	22.06 ± 1.36^b	0.421 ± 0.00^b	45.5 ± 0.01^a
	Valley bottom	23.81 ± 0.5^a	0.416 ± 0.00^c	47.0 ± 0.01^a
Unaffected / Edwenase	Upper part	1.42 ± 0.01^d	0.002 ± 0.00^d	19.0 ± 1.42^b
	Middle part	1.42 ± 0.02^d	0.003 ± 0.00^d	19.0 ± 9.70^b
	Valley bottom	1.43 ± 0.00^d	0.003 ± 0.00^d	19.0 ± 6.74^b
FAO / WHO (2001) permissible limit (mg kg-1)		0.15	0.3	27.4
LSD		1.71	0.002	1.62
% CV		5.8	4.3	10.3

*Means with the same letters are not significantly different at $P < 0.05$ LSD

Heavy Metals in Plants (Plantain) Samples at South Suntreso

Arsenic

Table 3, Arsenic (As) content recorded at South Suntreso showed significant difference between all the plants sampled at the workshop and those at the control plot. From the slope, plants at the workshop recorded some significant differences between the means; Arsenic levels at the valley bottom of the upper part were significantly different from the upper and middle parts of the slope at the workshop. However, there was no significant difference between the means in terms of slope (upper, middle and valley bottom) at the control plot. The highest level of arsenic was recorded at the valley bottom of the workshop with mean concentration of 24.04 ± 0.01, followed by the middle part at 23.23 ± 0.16 and the last accumulation level showing at the upper slope of the workshop with mean of 21.32 ± 0.08. There was an increasing trend of as levels in maize samples from the upper part through the middle and to the valley bottom of the slope. From Table 3, all the plantain samples had different accumulation levels of as above the FAO/WHO (2010) permissible limit of 0.15mg kg-1. The soils had high levels of arsenic at the workshop, this resulted in plant uptake at the same field.

The presence of arsenic in the maize plants at South Suntreso comes as a result of the presence of high arsenic levels in the soils located at the workshop. Mobility of arsenic in maize plant is faster through the roots of the plant. This observation is consisted with work done by Fosu-Mensah et al., (2018). The mean concentration of arsenic in his work was above the FAO/WHO (2007) permissible limit of arsenic in plant samples. High concentration of arsenic in plants can cause nausea, vomiting, diarrhea, cough,

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headache and cardiovascular disease to animals especially livestock (ATSDR 2007; Tchounwou et al., 2012).

Lead (Pb)

Lead (Pb) levels in plantain samples at South Suntreso showed significant difference between all the plants sampled at the workshop and those at the control plot. From the slope, plants at the workshop did not record any significant differences between their means from the upper part, through to the middle and to the valley bottom (Table 3). At the control plot, there were also no significant differences in Pb between the means of plantain samples in terms of slope (upper, middle and valley bottom). The highest level of Pb was recorded at the workshop with a general mean of 0.425 ± 0.00. All the the parts of the slopes recorded the same level of Pb as mentioned above which constitutes the highest as compared to the control plot which also recorded 0.014 ± 0.00 as the least Pb level. Pb has little plant motion (Guntherdt, 2007). Plant roots can also uptake small amounts of Pb (Celik, Kartal, Akdogan and Kaska, 2005). All the levels of Pb found in Plantain samples were a little above FAO/WHO (2010) permissible level of 0.3mg kg-1. The consumption of high amount of lead can cause anaemia, headache, brain damage, and nervous system disorder to humans and animals (Shi et al., 2011). In a related work done by Fosu-Mensah et al., (2018), the concentration of lead in plant samples exceeded the WHO/FAO (2010) permissible limit of 0.3mg kg-1.

Zinc (Zn)

Zinc was recorded some levels in plantain sampled at the clustered workshop. Plantain samples at South Suntreso for heavy metal analysis also recorded significant difference between all the samples at the workshop and those at the control plot (Table 3). From the slope, plants at the workshop recorded some significant differences between the means; Zn levels at the valley bottom were significantly different from the upper and middle parts of the slope at the workshop. In addition, there was no significant difference between the means in terms of slope (upper, middle and valley bottom) at the control plot where plantain samples were also taken and analysed for heavy metals.

The highest level of Zn was recorded at the valley bottom of the workshop with mean concentration of 15.45 ± 0.02, followed by the middle part at 10.83 ± 2.50 and the least accumulation level showing at the upper slope of the control plot with mean of 1.03 ± 0.01. There was an increasing trend of Zn in plantain samples from the upper part through the middle and to the valley bottom of the slope. Zn results from Table 3 recorded some levels of Zn below the FAO/WHO (2010) permissible limit of 27.4mg kg-1.

Although Zn is one of the essential nutrient elements that play a vital role in physiological and metabolic processes of many living organisms, it can result in adverse effects when its concentration reaches above permissible limits (Fosu-Mensah et al. 2018). Soil pH is considered one of the most relevant factors that influence the metal transfer from soil to plants, and higher pH was reported to decrease the bioavailability and toxicity of metals such as Cd and Pb (Wang, 2007). The pH of the studied soil was 7.4, which could not

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have contributed much to the metal absorption mechanisms of plants. It was observed from the report that each metal differed considerably in uptake from each other in the table above. This observation is consistent with Intawongse (2006), who had similar results of differed metal uptake in plants from polluted soils.

Table 3 **-** *Heavy Metal Levels in Plantain at South Suntreso*

State of Field	Part of Slope	As (mg kg-1)	Pb (mg kg-1)	Zn (mg kg-1)
Affected / South Suntreso	Upper part	21.32 ± 0.08^c	0.425 ± 0.00^a	8.03 ± 1.35^b
	Middle part	23.23 ± 0.16^b	0.425 ± 0.00^a	10.83 ± 2.50^b
	Valley bottom	24.04 ± 0.01^a	0.425 ± 2.45^a	15.45 ± 0.02^a
Control / South Suntreso	Upper part	1.42 ± 1.01^d	0.016 ± 0.00^b	1.03 ± 0.01^c
	Middle part	1.42 ± 0.01^d	0.016 ± 0.00^b	1.05 ± 0.01^c
	Valley bottom	1.43 ± 0.01^d	0.014 ± 0.00^b	1.05 ± 0.01^c
FAO/WHO (2010) permissible limit (mg kg -1)		0.15	0.3	27.4
LSD		0.212	0.002	3.351
% CV		0.6	1.7	17.6

*Means with the same letters are not significantly different at p< 0.05 LSD

Effects of Heavy Metals on Microbial Activity (mgCO$_2$/kgSoil/day)

Table 7 represents the data obtained at Edwenase for microbial activity of position and depths of (0-10 cm and 10-30cm). There were significant differences in means between microbial activity in soils at the workshop and the control plot at $P < 0.05$. Considering Table 4, the control plot had the highest microbial activity with respect to the samples from the workshop with mean of 30.2 ± 0.17. This was attributed to the fact that there was low presence of heavy metals at the control plots from the data provided. Taking into consideration data from the workshop, the upper slope had the highest microbial activity of 21.43 ± 8.57(mg CO$_2$/kg soil/ day), middle slope also recorded 10.00 ± 3.30(mg CO$_2$/kg soil/ day) while the valley bottom produced the least with average mean of 5.24 ± 2.52(mg CO$_2$/kg soil/ day). There was a decreasing trend of microbial activities from the upper slope through the middle slope and to the valley bottom at depth (0-10cm).

The sub-soil (10-30cm) data are as follows: upper slope was 12.86 ± 7.56 being the highest, middle slope 5.24 ± 1.90 and valley bottom recorded 5.24 ± 0.95. There was a decreasing trend of microbial activities at depth of 10-30cm from the upper slope along the soil catena, though the middle slope and the valley bottom recorded the same values.

The results from both depths indicate there was some form of microbial activities in the soil at different levels due to heavy metal pollution in the soil samples. Soil microorganisms require a conducive soil environment to be able to decompose organic material in the soil. It was predicted that the existence of auto mechanic waste in the soil would cause soil pollution which will in turn reduce the activities of soil microorganisms. From the figures in

the Table 4, there was a decreasing trend of microbial activities from the upper slope through the middle slope and to the valley bottom at both depths (0-10 cm and 10-30 cm). This was due to the nature of the adjoining study fields.

The steepness of the slope at Edwenase quickly deposits all these wastes (lubricants) at the valley bottom, leaving some few pollutants at the upper slope and middle part. Not much of the heavy metals remain at the upper slope especially when it rains because of the steep nature of the fields. This is the reason why there was higher microbial activities at the upper slope since microorganisms are not disturbed much by the pollutants.

The above data was confirmed by soil health guide for educators (2016). At the valley bottom, all the pollutants sink into the soil making it heavily polluted with heavy metals from the waste deposited. This is the reason why microbial activities at the valley bottom recorded the lowest considering the upper and middle slope. Low concentration of heavy metals helps in the release of CO_2 high concentration inhibits soil respiration and disturbs the ecosystem. Respiratory rate reduces in higher polluted soils due to the interactions involving metals in a combined ionic state (Nwuche, 2008).

Table 4 - *Effects of Heavy Metals on Microbial Activity (MgCO$_2$/ Kg soil / day) at Edwenase*

State of field	Part of slope	Depth (0-10cm) / (mg CO$_2$/kg soil/ day)	Depth (10-30cm) / (mg CO$_2$/kg soil/ day)
Affected / Edwenase	Upper part	21.43 ± 4.95[ab]	12.86 ± 7.56[bc]
	Middle part	10 ± 3.30[c]	5.24 ± 1.90[c]
	Valley bottom	5.24 ± 2.52[c]	5.24 ± 0.95[c]
Control / Edwenase	Upper part	29.17 ± 0.26[a]	29.53 ± 0.20[a]
	Middle part	28.4 ± 0.29[a]	29.17 ± 0.23[a]
	Valley bottom	30.2 ± 0.17[a]	29.97 ± 0.33[a]
LSD		6.17	6.17
% CV		6.4	6.4

*Means with the same letters are not significantly different at $p < 0.05$ LSD

Microbial Activity at South Suntreso

Microbial activities at South Suntreso are presented Table 5 above at a depth of (0-10 cm) and (10-30cm). The overall highest microbial activity was recorded at upper part depth 0-10cm at the control plot. There was significant difference between the activity levels in soils sampled at the workshop and the control plot. At the workshop, the slope recorded no significant differences between the upper part, middle and the valley bottom. There was same situation at the control plot. At the workshop, the upper part recorded the highest microbial activity at depth 0-10cm with mean of 65.2 ± 8.12, the middle slope is also represented by 33.8 ± 5.60 as its microbial activity, and the lowest activity 21.4 ± 1.72 was recorded at the valley bottom all from the top soil (Table 5). At South Suntreso top soil (0-10 cm), microbial activity rather showed a decreasing trend of activity as that of Edwenase.

Microbial activity from the sub-soil at South Suntreso was also represented by 33.8 ± 1.10 recorded from the upper slope, the middle slope shows 2.4 ± 0.95 and the valley bottom being 4.3 ± 0.00. Soil samples were taken from the same location at table 8. At depth of 10-30 cm, microbial activity increased marginally at the upper part of the slope. This was attributed to the higher level of heavy metals present at the upper part of the slope. Abdu (2016) asserts that higher metal concentration reduces the production of CO_2 which leads to reduction in microbial activity. His findings were similar to the results of this study where microbial activity was higher at low heavy metal concentrated part of the slope. The low levels of microbial activity at the workshop as compared to the control is attributed to anthropogenic activities

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by the artisans at both study areas; especially disposing off poisonous waste full of heavy metals.

These findings are consistent with work done by Amoakwah, Ahsan, Rahman et al. (2020) where similar observation was unveiled. Higher levels of heavy metals in the living tissue of micro-organisms causes severe organ impairment, neurological disorder which results in death of micro-organism in the soil (Murrieta et al., 2006). Whenever there is any discrepancy at such high metal levels it is often due to a community shift, in which case the tolerance of the dominant microbial group will determine the respiration (Giller et al., 1998).

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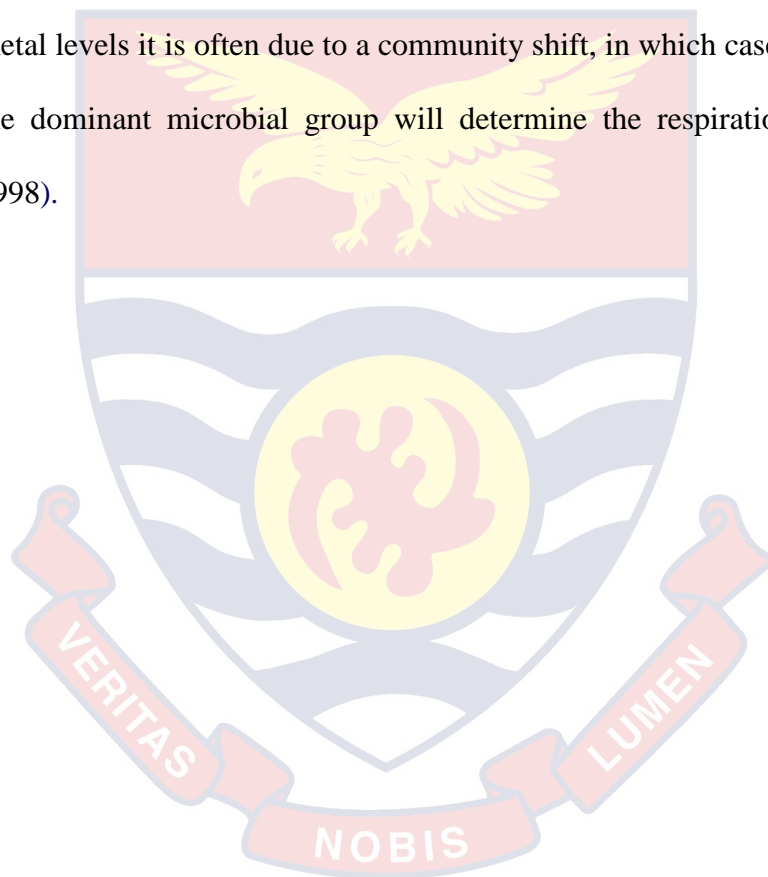
Table 5 - *Effects of Heavy Metals on Microbial Activity (MgCO₂ / Kg soil / day) at South Suntreso*

State of Field	Part of Slope	Depth (0-10cm) / (mg CO_2/kg soil/ day)	Depth (10-30cm) / (mg CO_2/kg soil/ day)
Affected / South Suntreso	Upper part	65.2 ± 58.12^{ab}	33.8 ± 28.10^{b}
	Middle part	33.8 ± 29.60^{b}	2.4 ± 0.95^{b}
	Valley bottom	21.4 ± 8.72^{b}	4.3 ± 0.00^{b}
Control / South Suntreso	Upper part	125 ± 2.27^{a}	119.4 ± 0.24^{a}
	Middle part	111.6 ± 3.60^{a}	107.9 ± 0.32^{a}
	Valley bottom	109.1 ± 0.32^{a}	107.7 ± 0.52^{a}
LSD		42.41	42.41
% CV		16.5	16.5

*Means with the same letters are not significantly different at $p < 0.05$ LSD

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CHAPTER FIVE

SUMMARY, CONCLUSIONS AND RECOMMENDATIONS

Summary

Auto mechanics have over the years played a major role in the transport industry in Ghana by providing services to many road users including; heavy duty cars, tankers, salon cars, commercial vehicle among others. In the process of discharging their duties, they intentionally and unintentionally deposit various waste from vehicles on the soil within the workshops and the adjoining fields. Their activities have been monitored for a long time and they seem not to consider the effects of the waste they dispose on soil properties, existing plants and soil microbial activities. An experiment was conducted to assess the impact of automobile waste on soil properties and micro-organisms. Two cluster of auto mechanic workshops at Edwenase and South Suntreso in Kumasi Metropolis (Ashanti Region of Ghana) were considered for the study.

The study sought to examine the variation of heavy metals from the workshops downslope, also to examine the levels of heavy metals uptake in plants at the adjoining fields to the workshop and to measure the levels of microbial activities in heavy metal polluted soils. The study dwelled on some hypothesis including; There are high levels of heavy metals in soils around auto-mechanic workshops than control soils. Plants at the adjoining fields to auto-mechanic workshops have high levels of heavy metals than unaffected soils. Soils around auto-mechanic workshops have lower soil microbial activity than control soils.

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In the soil analysis for heavy metals, all the soil samples showed some levels of heavy metal accumulation (Cd, Pb, Zn and As). The highest mean of Cadmium recorded at South Suntreso was valley bottom depth 10-30cm 0.125mg kg-1, whereas the least was 0.00mg kg-1 at control upper part (0-10cm). The levels were all within FAO/WHO (2001) acceptable limit of 3mg kg-1. Lead (Pb) levels at South Suntreso also recorded its highest at upper part (0-10cm) of the workshop with mean of 32.43mg kg-1 and the lowest accumulation was 0.00mg kg-1 at control upper, middle and valley bottom depth 10-30cm. The levels of accumulation at upper and middle part depth 0-10cm were all above FAO/WHO (2001) acceptable limit of 15mg kg-1; the rest were below the limit.

Zinc at South Suntreso had the highest level at the workshop valley bottom depth 10-30mg kg-1 and the least was 1.11mg kg-1 at control upper, middle and valley bottom depth 0-10cm. The levels at South Suntreso workshop were all above FAO/WHO (2001) acceptable limit of 3.5mg kg-1. Arsenic was also present at various levels in the soil at South Suntreso. The highest level was 11.72mg kg-1 at valley bottom 10-30cm, the lowest level was 0.82mg kg-1 at control upper part 0-10cm. The levels of Arsenic were all below FAO/WHO (2001) acceptable limit of 20mg kg-1. The levels of metals under study were all below FAO/WHO (2001) acceptable limit at the control plot.

At Edwenase, Cadmium recorded its highest level of 0.002mg kg-1 at the workshop at valley bottom depth 0-10cm the least had 0.00 at the upper, middle and valley bottom 'of the control plot at both depths. Lead (Pb) had 17.33mg kg-1 as the highest level at Edwenase upper part depth 0-10cm; its

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corresponding lowest level recorded was 1.83mg kg-1 at control plot upper part depth 0-10cm. The levels of zinc at the workshop were all above FAO/WHO (2001) acceptable limit with the highest being 58.5mg kg-1 at upper part 0-10cm at the workshop. The lowest Zn level was 2.2mg kg-1 which was recorded at the upper, middle and valley bottom of the control plot. Arsenic was also found in the soil samples at Edwenase. The highest level of 49.19mg kg-1 was recorded at valley bottom depth 0-10cm at the workshop with its corresponding least level of 0.00mg kg-1 which was found at the upper, middle and valley bottom of the control plot. The levels of arsenic at the workshop were all above FAO/WHO (2001) acceptable limit of 20mg kg-1.

Additionally, plant samples which were analysed for heavy metals also revealed some levels of heavy metals at both study fields. Arsenic level at South Suntreso had its highest level at the valley bottom with mean 24.04mg kg-1. The levels were highly above FAO/WHO (2010) acceptable limit of 0.15mg kg-1 in plants. Lead also had its levels a little above FAO/WHO (2010) acceptable limit of 0.3mg kg-1. Highest Pb level was 0.425mg kg-1 recorded at upper, middle and valley bottom of the workshop. Zinc in effect had all the levels within FAO/WHO (2010) acceptable limit of 27.4mg kg-1 with the highest level being valley bottom 15.45mg kg-1. At Edwenase. Arsenic, lead and Zinc all had it highest levels at the valley bottom of the slope with mean 23.81mg kg-1, 0.423mg kg-1 and 47.0mg kg-1 respectively.

Soil samples recorded at both fields had some levels of microbial activity. At South Suntreso, the control plot had the highest microbial activity of 125 ± 2.27mg kg-1. And the least microbial activity was 21.4 ± 8.72 mg

kg-1. Edwenase also recorded its highest activity   30.2 ± 0.17mg kg-1 at the control plot at the upper part depth 0-10cm was recorded at valley bottom of the workshop. The least microbial activity 5.24 ± 2.52mg kg-1 was recorded at the valley bottom at both depth of the workshop.

Conclusions

Results from the analysis showed Arsenic (Ar), Lead (Pb), Cadmium (Cd) and Zinc (Zn) were present in the soil samples at various levels. There were variations in the level of these metals in the soil. Most of them, as indicated in the results, were above FAO/WHO (2001) acceptable limit in soils. This affirms the hypothesis "There are higher levels of heavy metals in soils around auto-mechanic workshops than the control plot". Also there was plant uptake as a result of the presence of heavy metals in the soil. The levels were very high especially at the workshop due to the activities of the artisans. However, the heavy metal uptake at the control plot was very low. The heavy metal levels in maize and plantain were above FAO/WHO (2001) acceptable limit in plants.

This confirms the hypothesis of the study "Plants at the adjoining fields to auto-mechanic workshops have higher levels of heavy metals than plants at control plots.

The study was also extended to the effect of heavy metals on microbial activity. Soil samples at the workshop recorded the least of microbial activity as compared to its corresponding control plots at South Suntreso and Edwenase. There was high microbial activity at the control plots because low levels of heavy metals did not have a ripple effect on microbial activities in the

soil. This indeed affirms the hypothesis of the study "Soils around auto-mechanic workshops have lower soil microbial activity than soils at the control plot.

Recommendations

The artisans at auto mechanic workshops need to be educated in the ripple effects of their activities on the soil and the environment when they improperly dispose fuels, engine oils, acid in car batteries etc on the soils when servicing vehicles. Additionally, special chambers should be created within the workshops where all the waste generated will be deposited to reduce the spread of heave metals on the field especially the adjoining fields where cultivation is mostly done.

Farmers should be advised to avoid cultivating crops close to auto mechanic shops. Cropping at the affected fields will increase the health risk of consumers including the artisans as heavy metal levels exceeded the tolerable rate peculiar by FAO/WHO. The authorities of the Municipality should monitor the activities of the artisans at their various auto mechanic shops and sanction those who defile the orders. Again, soils at these mechanic shops should occasionally be sampled for laboratory analysis to know the heavy metal status of the soil and also to monitor the microbial environment for healthy soils.

Policy makers should ban commercial farmers from cultivating crops around existing auto-mechanic workshops to maintain food safety. Again, Government should set up Automobile villages where auto-repairs are kept at safe distanced from human inhabitation for good health.

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REFERENCES

Abe, F., Akimoto, H., Akopian, A., Albrow, M. G., Amendolia, S. R., Amidei, D., & Handler, R. (1995). Observation of top quark production in collisions with the Collider Detector at Fermilab. *Physical Review Letters*, *74*(14), 2626

Abenchi, E. S, Okunola, O. J, Zubairu, S. M. J, Usman, A. A, & Apene, E. (2010). Evaluation of Heavy metals in roadside soils of major streets in Jos Metropolis*, Nig. J. Environ. Chem. Ecotoxicol, 2*(6), 98 - 102.

Abidemi, O. O. (2011). Levels of Pb, Fe, Cd and Co in soils of automobile workshop in Osun State, Nigeria. *Journal of applied sciences and environmental management*, *15*(2).

Adelekan, B. A, & Alawode, A. O. (2011). Concentrations of Municipal refuse dumps to heavy metals concentrations in soil profile and ground water Ibadan, Nigeria. *J. Appl. Biosci., 40,* 2727-2737.

Adeniji, A. O., Okoh, O. O., & Okoh, A. I. (2017). Petroleum hydrocarbon fingerprints of water and sediment samples of Buffalo River Estuary in the Eastern Cape Province, South Africa. *Journal of Analytical Methods in Chemistry*, *2017*. https://doi.org/10.1155/2017/2629365

Adewoyin, O. A., Hassan A. T., & Aladesida A. A., (2013). The impact of auto-mechanic workshops on soil and groundwater in Ibadan metropolis. *African Journal of Environment Science and Technology, 7*(9), 891-898.

Alengebawy, A., Abdelkhalek, S. T., Qureshi, S. R., & Wang, M. Q. (2021). Heavy metals and pesticides toxicity in agricultural soil and plants: Ecological risks and human health implications. *Toxics*, *9*(3), 42.

101

Ali, H., Khan, E., & Sajad, M. A. (2013). Phytoremediation of heavy metals concepts and applications. *Chemosphere*, *91*(7), 869-881.

Alloway, B. J. (1995). Cadmium. In B. J. Alloway (ed.), *Heavy metals in soils* (2nd Ed.)*. Blackie, New York, pp. 122-151.

Alloway, B. J. (2013). Heavy metals and metalloids as micronutrients for plants and animals. (pp. 195-209). Springer, Dordrecht.

Alloway, B. J. (Ed.). (2012). Heavy metals in soils: trace metals and metalloids in soils and their bioavailability *(Vol. 22). Springer Science & Business Media.*

Amoakwah, E., Ahsan, S., Rahman, M. A., Asamoah, E., Essumang, D. K., Ali, M., & Islam, K. R. (2020). Assessment of heavy metal pollution of soil-water-vegetative ecosystems associated with artisanal gold mining. *Soil and Sediment Contamination: An International Journal*, *29*(7), 788-803.

Asamoah, D. K., Mursheda, A. & Khandakar, R. I. (2020). Assessment of Heavy Metal Pollution of Soil-water-vegetative Ecosystems Associated with Artisanal Gold Mining, Soil and Sediment Contamination: *An International Journal,* DOI: 10.1080/15320383.2020.1777936

Avila P. F., Ferreira, S. E., & Candelas, C. (2017). Health risk assessment through consumption of vegetables rich in heavy metals: the case study of the surrounding villages from Panasqueira mine, Central Portugal. *Environmental Geochemistry and Health*, *39,* 565–589.

Baath, E. (1989). Effects of heavy metals in soil on microbial processes and populations. *Water Air Soil Pollut., 47*(3-4): 335–379.

Bååth, E., Frostegård, Å., & Fritze, H. (1992). Soil bacterial biomass, activity, phospholipid fatty acid pattern, and pH tolerance in an area polluted with alkaline dust deposition. *Applied and Environmental Microbiology*, *58*(12), 4026.

Batista, A. A., Santos, J. A., Bomfim, M. R., Moreira, F. M., Leal, E. F., & Conceição, J. N. D. (2017). Induced changes in the growth of four plant species due to lead toxicity. *Revista Brasileira de Engenharia Agrícola e Ambiental*, *21*(5), 327-332.

Begonia, M. T., Begonia, G. B., Miller, G., Gilliard, D., & Young, C. (2004). Phosphatase Activity and Populations of Microorganisms from Cadmium and Lead Contaminated soils. *Bull. Environ. Contam. Toxicol., 73*, 1025-1032.

Brookes, P. C. (1995). The Use of Microbial Parameters in Monitoring Soil Pollution by Heavy Metals. *Biol. Fertile Soils*, *19*(4), 269-279.

Celik, A., Kartal, A. A., Akdogan, A. and Kaska, Y. (2005). "Determining the heavy metal pollution in Denizli (Turkey) by using Robinio pseudo-acacia L. *Environmental International, 31, 105-112*.

Chander, K., & Brookes, P. C. (1993). Residual effects of zinc, copper and nickel in sewage sludge on microbial biomass in a sandy loam. *Soil Biology and Biochemistry*, *25*(9), 1231-1239.

Chefetz, B., Hatcher, P. G., Hadar, Y., & Chen, Y. (1996). Chemical and biological characterization of organic matter during composting of municipal solid waste. *American Society of Agronomy, Crop Science Society of America, and Soil Science Society of America. 25*(4), 776-785.

Chinedu, E., & Chukwuemeka, C. K. (2018). Oil spillage and heavy metals toxicity risk in the Niger Delta, Nigeria. *Journal of Health and Pollution*, *8*(19).

Chu, D. (2018, February). Effects of heavy metals on soil microbial community. In *IOP Conference Series: Earth and environmental science* (Vol. 113, No. 1, p. 012009). IOP Publishing.

Comfort, G. A., Ofosu, F. G., Bamford, S. A., Wordson, D. A., Atiemo, M. S., Aboh, J. K. I. & Adeti, P. J. (2013). Heavy metal contamination in surface soil dust at selected fuel filling stations in Accra, *Ghana American Journal of Scientific and Industrial Research*, ISSN: 2153-649X.

Cooper, D. C., Neal, A. L., Kukkadapu, R. K., Brewe, D., Coby, A., & Picardal F. W. (2005). Effects of Sediment Iron Mineral Composition on Microbially Mediated Changes in Divalent Metal Speciation: Importance of Ferrihydrite. *Geochim. Cosmochim. Acta., 69*, 1739-1754.

Davies, B. E. (1995). *Heavy metals in soils* (2nd Ed.). Department of Botany/Microbiology, pp. 206-223. University of Lagos, Akoka, Yaba, Nigeria,

Davies, B. E. (1995). Lead. Heavy metals in soils (2nd ed). London: Blackie Academic & Professional, pp. 206–223.

Alloway, B. J. (1995) Heavy Metals in Soils. Blackie Academic and Professional, Chapman and Hall, London, 368 p. https://doi.org/10.1007/978-94-011-1344-1

104

Diaz-Ravina, M., & Baath, E. (1996). Development of metal tolerance in soil bacterial communities exposed to experimentally increased metal levels. *Appl. Environ. Microbiol., 62,* 2970-2977.

Diaz-Ravina, M., & Baath, E. (1996). Thymidine and leucine incorporation into bacteria experimentally contaminated with heavy metals. *Appl. Soil. Ecol., 3,* 225-234.

Díaz-Raviña, M., Bååth, E., & Frostegård, Å. (1994). Multiple heavy metal tolerance of soil bacterial communities and its measurement by a thymidine incorporation technique. *Applied and Environmental Microbiology*, *60*(7), 2238.

Doelman, P., & Haanstra, L. (1984). Short-term and long-term effects of Cd, Cr, Cu, Ni, Pb, and Zn on microbial respiration in relation to abiotic soil factors. *Plant Soil, 79*, 317-321.

Doncheva, S., Stoynova, Z., & Velikova, V. (2001). Influence of succi-nate on zinc toxicity of pea plants. *Journal of Plant Nutrition, 24(*6), 789–804.

Dubinsky, E. A., Conrad, M. E., Chakraborty, R., Bill, M., Borglin, S. E., Hollibaugh, J. T., & Andersen, G. L. (2013). Succession of hydrocarbon-degrading bacteria in the aftermath of the deepwater horizon oil spill in the Gulf of Mexico. *Environmental Science and Technology*, *47*(19), 10860-10867.

Ebbs, S. D., Kochian, L. V. (1997). Toxicity of zinc and copper to Brassica species: implications for phytoremediation. *J Environ Qual, 26,* 776–781. https://www.elsevier.com

Emily, M., Mailund, T., Hein, J., Schauser, L., & Schierup, M. H. (2009). Using biological networks to search for interacting loci in genome-

wide association studies. *European Journal of Human Genetics*, *17*(10), 1231-1240.

Amoakwah, E., Ahsan, S., Rahman, M. A., Asamoah, E., Ali, M., & Islam, K. R. (2020). Assessment of heavy metal pollution of soil-water-vegetative ecosystems associated with Artisanal Gold Mining, Soil and Sediment Contamination. *An International Journal, 12*(8), 1134-1141.

Fagbote, E. O., & Olanipekun, E. O. (2010). Evaluation of the status of heavy metal pollution of soil and plant (*Chromolaena odorata*) of Agbabu bitumen deposit area, Nigeria. *American-Eurasian Journal of Scientific Research, 5*(4), 241–248.

Fernandes, J. C., & Henriques, F. S. (1991). Biochemical, physiological and structural effects of excess copper in plants. *Bot Rev.*, 57, 247–273.

Forstner, U. (1995). Land contamination by metals: Global scope and magnitude of problem. *Metal Speciation and Contamination of Soil*, 1-33.

Fosu-Mensah, B. Y., Ofori, A., Ofosuhene, M., Ofori-Attah, E., Nunoo, F. E., Darko, G., & Appiah-Opong, R. (2018). Assessment of heavy metal contamination and distribution in surface soils and plants along the west coast of Ghana. *West African Journal of Applied Ecology*, 26, 167-178.

Frostegård, A., & Bååth, E. (1996). The use of phospholipid fatty acid analysis to estimate bacterial and fungal biomass in soil. *Biology and Fertility of Soils*, *22*(1), 59-65.

106

Giller, K. E., Witter, E., & Mcgrath, S. P. (1998). Toxicity of heavy metals to microorganisms and microbial processes in agricultural soils: a review. *Soil Biology & Biochemistry, 30*(10–11), 1389-1414.

Goyer, R. A., & Clarkson, T. W. (2001). Toxic effects of metals. In, *Casarett and Doullis Toxicology*: *The Basic Science of Poisons*, (*CD Klaassen, ed*.) (pp 81) Mc-Graw Hill. New York.

Gülser, F. & Erdoğan, E. (2008). The effects of heavy metal pollution on enzyme activities and basal soil respiration of roadside soils. *Environmental Monitoring and Assessment, 145*(1-3):127- 133.

Gülser, F., & Erdoğan, E. (2008). The effects of heavy metal pollution on enzyme activities and basal soil respiration of roadside soils. *Environmental Monitoring and Assessment 145*(1-3), 127-133.

Gupta, S., & Jena, V. (2013). Assessment of heavy metal contents of green leafy vegetables, *Croatian Journal of Food Science and T0echnology, 5(2*), 53-60.

Haider, F. U., Liqun, C., Coulter, J. A., Cheema, S. A., Wu, J., Zhang, R., & Farooq, M. (2021). Cadmium toxicity in plants: Impacts and remediation strategies. *Ecotoxicology and Environmental Safety, 4(*2), 33-40.

Hamad, A. A., Moubasher, H. A., Moustafa, Y. M., & Mohamed, N. H. (2021). Petroleum hydrocarbon bioremediation using native fungal isolates and consortia. *The Scientific World Journal*, *2021*.

Hamilton-Taylor, J., Smith, E. J., Davison, W., & Sugiyama, M. (2005). Resolving and modeling the effects of Fe and Mn redox cycling on

107

trace metal behavior in a seasonally anoxic lake. *Geochimica et Cosmochimica Acta*, *69*(8), 1947-1960.

Hemida, S. K., Omar, S. A., & Abdel-Mallek, A. Y. (1997). Microbial populations and enzyme activity in soil treated with heavy metals. *Water Air Soil Pollut., 95,* 13-22.

Holtan-Hartwig, L., Bechmann, M., Høyås, T. R., Linjordet, R., & Bakken, L. R. (2002). Heavy metals tolerance of soil denitrifying communities: N2O dynamics. *Soil Biol. Biochem., 34,* 1181-1190.

Huerta-Diaz, M. D., & Morse, J. W. (1992). Pyritization of trace metals in anoxic marine sediments. *Geochim. Cosmochim. Acta., 56,* 2681-2702.

Hunt N & Gilkes R. (1992). Farm monitoring handbook. The University of Western Australia, WA.

Igiri, B. E., Okoduwa, S. I., Idoko, G. O., Akabuogu, E. P., Adeyi, A. O., & Ejiogu, I. K. (2018). Toxicity and bioremediation of heavy metals contaminated ecosystem from tannery wastewater: a review. *Journal of Toxicology. 6(3*), 49-54.

Ikhajiagbe, B., Ogedegbe, A. U., & Anoliefo, G. O. (2013). Growth response of *Alternanthera brasiliana* (L.) Kuntze in a waste engine oil-polluted soil. *Journal of Emerging Trends in Engineering and Applied Sciences*, *4*(2), 322-327.

Intawongse, M., & Dean, J. R. (2006). Uptake of heavy metals by vegetable plants grown on contaminated soil and their bioavailability in the human gastrointestinal tract. *Food Additives and Contaminants*, *23*(1), 36-48.

Jaishankar, M., Tseten, T., Anbalagan, N., Mathew, B. B., & Beeregowda, K. N. (2014). Toxicity, mechanism and health effects of some heavy metals. *Interdisciplinary Toxicology*, *7*(2), 60.

Jiang, W., Liu, D., & Hou, W. (2001). Hyperaccumulation of cadmium by roots, bulbs and shoots of garlic. *Bioresource Technology, 76(1)*, 9–13.

Johns, C. (2017). Living soils: the role of microorganisms in soil health. *Fut Direct Intl*, 1-7.

Joint FAO/WHO Expert Committee on Food Additives. Meeting. (2001). Safety evaluation of certain mycotoxins in food (No. 74). Food and Agriculture Organization.

Kádár, I. (1995). Effect of heavy metal load on soil and crop. *Acta Agron. Hung., 43,* 3-9.

Kalagbor, I. A., Dighi, N. K., & James, R. (2015*).* Levels of some heavy metals in cassava and plantain from farmlands in Kaani and Kpean in Khana Local Government Area of Rivers State.   Department of Science Laboratory Technology, Rivers State Polytechnic. Bori. *J. Appl. Sci. Environ. Manage., 19* (2), 219 - 222

Kandeler, E., Tscherko, D., Bruce, K. D., Stemmer, M., Hobbs, P. J., Bardgett, R. D., & Amelung, W. (2000). Structure and Function of the Soil Microbial Community in Microhabitats of a Heavy Metal Polluted. Soil. *Biol. Fertil. Soils, 32,* 390-400.

Kelly, J. J., & Tate III, R. L. (1998). Effects of heavy metal contamination and remediation on soil microbial communities in the vicinity of a zinc smelter. *American Society of Agronomy, 27*, 609-617.

Khan, A. G. (2005). Role of soil microbes in the rhizospheres of plants growing on trace metal contaminated soils in phytoremediation. *Journal of Trace Elements in Medicine and Biology, 18*(4), 355-364.

Knezevic, M., Stankovic, D., Krstic, B., Nikolic, M. S., & Vilotic, D. (2009). Concentrations of heavy metals in soil and leaves of plant species. Paulownia elongata and Paulownia fortunei. *African Journal of Biotechnology, 8*(20), 5422–5429.

Knezevic, M., Stankovic, D., Krstic, B., Nikolic, M. S., & Vilotic, D. (2009). Concentrations of heavy metals in soil and leaves of plant species Paulownia elongata and Paulownia fortunei. *African Journal of Biotechnology, 8*(20), 5422–5429.

Kosolapov, D. B., Kuschk, P., Vainshtein, M. B., Vatsourina, A. V., Wiessner, A., Kästner, M., & Müller, R. A. (2004). Microbial processes of heavy metal removal from carbon- deficient effluents in constructed wetlands. *Engineering in Life Sciences*, *4*(5), 403-411.

Kostyantyn, U. (1997). Should auto body repair and paint shops in the State of Rhode Island be regulated under industrial pre-treatment program. Unpublished master's thesis submitted to Centre for Environmental Studies at Brown University, University of Kyiv.

Krstić, B., Stanković, D., Igić, R., & Nikolic, N. (2007). The potential of different plant species for nickel accumulation. *Biotechnology & Biotechnological Equipment, 21*(4), 431– 436. doi:10.1080/13102818.2007.10817489

Kuo, S. A., Heilman, P. E., & Baker, A. S. (1983). Distribution and forms of copper, zinc, cadmium, iron, and manganese in soils near a copper smelter. *Soil Science*, *135*(2), 101-109.

Kuperman, R. G., & Carreiro, M. M. (1997). Soil heavy metal concentrations, microbial biomass and enzyme activities in a contaminated grassland ecosystem. *Soil Biology and Biochemistry, 29*(2), 179-190.

Lacatusu, R. (2000). Appraising levels of soil contamination and pollution with heavy metals. *European Soil Bureau Research Report*, *4*, 393-402.

Leyval, C., Turnau, K., & Haselwandter, K. (1997). Effect of heavy metal pollution on mycorrhizal colonization and function: physiological, ecological and applied aspects. *Mycorrhiza, 7*(3), 139-153.

Liu, J., Wu, J., Lin, J., Zhao, J., Xu, T., Yang, Q. & Song, X. (2019). Changes in the microbial community diversity of oil exploitation. *Genes*, *10*(8), 556.

Luo, C., Liu, C., Wang, Y., Liu, X., Li, F., Zhang, G., & Li, X. (2011). Heavy metal contamination in soils and vegetables near an e-waste processing site, south China. *Journal of Hazardous Materials*, *186*(1), 481-490.

Małachowska-Jutsz, A., & Matyja, K. (2019). Discussion on methods of soil dehydrogenase determination. *International Journal of Environmental Science and Technology*, *16*(12), 7777-7790.

Manivasagaperumal, S., Balamurugan, S., Thiyagarajan, G., & Sekar, J. (2011). "Effect of zinc on germination, seedling growth and biochemical content of cluster bean (Cyamopsis tetragonoloba(L.) Taub*).Current Botany, 2*(5), 11–15.

Marahatta, S. B., Gautam, S., Paudel, G., & Yadav, U. N. (2018). Awareness of occupational hazards and associated factors among automobile repair artisans in Kathmandu Metropolitan City, Nepal. *Indian Journal of Occupational and Environmental Medicine*, *22*(1), 49.

Masindi, V., & Muedi, K. L. (2018). Environmental contamination by heavy metals. *Heavy Metals*, *10*, 115-132.

Masquez-Murrieta, I. Miguueles-Garduno, O. Franco-Hernandez, B. Govaerts, L. Dendooven (2006). C and N mineralization and microbial biomass in heavy-metal contaminated soil, *Eur. J. Soil Biol., 42, 89–98.*

Mench, M., Didier, V., Löffler, M., Gomez, A., & Masson, P. (1994). A mimicked in-situ remediation study of metal-contaminated soils with emphasis on Cadmium and Lead. *J. Environ. Qual., 23,* 58-63.

Fosu-Mensah, B. Y., Vlek, P. L., & MacCarthy, D. S. (2012). Farmers' perception and adaptation to climate change: a case study of Sekyedumase District in Ghana. *Environment, Development and Sustainability*, *14*(4), 495-505.

Meybeck, A., & Redfern, S. (2013). Voluntary standards for sustainable food systems: challenges and opportunities. A Workshop of the FAO/UNEP Programme on Sustainable Food Systems, Rome, Italy, 11-12 June 2013. In *Voluntary standards for sustainable food systems: challenges and opportunities.* Food and Agriculture Organization of the United Nations (FAO).

Mohanpuria, P., Rana, N. K. & Yadav, S. K. (2007). Cadmium induced oxidative stress influence on glutathione metabolic genes of Camella sinensis (L.). O Kuntze. *Environ Toxicol., 22,* 368–374.

Momodu, M. A. & Anyakora, C. A., (2010). Heavy metal contamination of ground water: The Surulere case study. *Research Journal Environmental and Earth Sciences 2*(1), 39-43

Moral, R., Pedreno, J., Gomez, I., & Mataix J. (1995). Effects of chromium on the nutrient element content and morphology of tomato. *Journal of Plant Nutrition, 18*(4), 815–822.

Németh, T., & Kádár, I. (2005). Leaching of microelement contaminants: A longterm field study. *Z Naturforschung, 60*, 260-264.

Nogawa, K., Honda, R., Kido, T., Tsuritani, I., & Yamanda, Y. (1987). Limits to protect people eating Cadmium in rice, based on epidemiological studies. *Trace Subst. Environ. Health, 21*, 431-439.

Nriagu, J. O., Wong, H. K., Lawson, G., & Daniel, P. (1998). Saturation of ecosystems with toxic metals in Sudbury basin, Ontario, Canada. *Science of the Total Environment*, *223*(2-3), 99-117.

Nwachukwu, M. A, Feng, H., & Alinnor, J. (2011). Trace metal Deposition in soil from Auto-mechanic village to urban residential areas in Owerri, Nigeria. *Proc. Environ. Sci. 4,* 310-322.

Nwachukwu, M. A., Feng, H. & Alinnor, J., (2010). Assessment of heavy metal pollution in the soil and their implications within and around mechanic villages. *Int. J. Environ. Sci. Tech, 7* (2), 347-358.

Nwachukwu, M. A., Feng, H., & Alinnor, J. (2010). Assessment of heavy metalpollution in the soil and their implications within and around mechanic villages. *Int. J. Environ. Sci. Tech, 7(2), 347 – 358.*

113

Nwuche, C. O., & Ugoji, E. O. (2008). Effects of heavy metal pollution on the soil microbial activity. *International Journal of Environmental Science & Technology*, *5*(3), 409-414.

Ogundapo Tayo O. & Tobinson A. (2018). Assessment of the impact of automobile waste on the environment Department of Environmental Engineering, University of Port Harcourt, Rivers State, Department of Mechatronic Engineering, University of Port Harcourt, Nigeria. *International Journal of Engineering Inventions*, *7*(7), 01-15

Oguntimehin, I., & Ipinmoroti, K. O. (2008). Profile of heavy metals from automobile workshops in Akure, Nigeria. *J. Eviron. Sci. Technol., 1*(7),19 - 26.

Ohyama, J., Sato, T., Yamamoto, Y., Arai, S., & Satsuma, A. (2013). Size specifically high activity of Ru nanoparticles for hydrogen oxidation reaction in alkaline electrolyte. *Journal of the American Chemical Society*, *135*(21), 8016-8021.

Ohyama, T., Minagawa, R., Ishikawa, S., Yamamoto, M., Hung, N. V. P., Ohtake, N., & Takahashi, Y. (2013). *Soybean seed production and nitrogen nutrition.* A comprehensive survey of international soybean research-Genetics, physiology, agronomy and nitrogen relationships, 115-157.

Okonokhua, B. O., Ikhajiagbe, B., Anoliefo, G. O., & Emede, T. O. (2007). The effects of spent engine oil on soil properties and growth of maize (*Zea mays* L.). *Journal of Applied Sciences and Environmental Management*, *11*(3), 102-110.

Onder, S., Dursun, S., & Demirbas, A. (2007). Determination of heavy metal pollution in grass and soil of city Centre Green Areas (Konya, Turkey Polish). *J. of Environ. 16*(1),145 - 154.

Pam, A. A., Sha'Ato, R., Offem, J. O. (2013). Contributions of automobile mechanic sites to heavy metals in soil: A case study of North Bank Mechanic Village Makurdi, Benue State, Central Nigeria. *Journal Chemistry Biol. Physical Sci., 3*(3), 2337-2347.

Perelomov L, & Kandeler E (2006). Effect of soil microorganisms on the sorption of zinc and lead compounds by goethite, *Journal Plant Nutrition. Soil Science, 169, 95–100.*

Perezdemora, A., & Burgos, M. (2006). Microbial community structure and function in a soil contaminated by heavy metals: effects of plant growth and different amendments. *Soil Biology and Biochemistry, 38*(2), 327-341.

Pérez-de-Mora, A., Burgos, P., Madejón, E., Cabrera, F., Jaeckel, P., & Schloter, M. (2006). Microbial community structure and function in a soil contaminated by heavy metals: effects of plant growth and different amendments. *Soil biology and biochemistry*, *38*(2), 327-341.

Petersen, S. O., & Klug, M. J. (1994). Effects of sieving, storage, and incubation temperature on the phospholipid fatty acid profile of a soil microbial community. *Applied and environmental microbiology*, *60*(7), 2421.

Pradhan, J. K., & Kumar, S. (2014). Informal e-waste recycling: Environmental risk assessment of heavy metal contamination in Mandoli industrial area, Delhi, India. *Environmental Science and*

Pollution Research, 21(13), 7913–7928. doi:10.1007/s11356-014-2713-2

Pradhan, J. K., & Kumar, S. (2014). Informal e-waste recycling: Environmental risk assessment of heavy metal contamination in Mandoli industrial area, Delhi, India. *Environmental Science and Pollution Research*, *21*(13), 7913-7928.

Prasad, K. V. S. K., Saradhi, P. P., & Sharmila, P. (1999). Concerted action of antioxidant enzymes and curtailed growth under zinc toxicity in Brassica juncea. *Environmental and Experimental Botany*, *42*(1), 1-10.

Ramesh, M. V., Nibi, K. V., Kurup, A., Mohan, R., Aiswarya, A., Arsha, A., & Sarang, P. R. (2017). *Water quality monitoring and waste management.* In *2017 IEEE Global Humanitarian Technology Conference (GHTC)* (pp. 1-7). IEEE.

Romero-Puertas, M. C., Serrano, M., Corpas, F. J., Gomez, M., Del Rio, L. A., & Sandalio, L. M. (2004). Cadmium- induced subcellular accumulation of O2and H2O2in pea leaves. *Plant Cell Env 27*, 1122–1134.

Rossiter, D. G. (2006). Classification of urban and industrial soils in the world reference base for soil resources (5 pp). *Journal of Soils and Sediments*, *7*(2), 96-100.

Salt, D. E, Blaylock, M., Kumar, N. P. B. A., Dushenkov, V., Ensley, D., Chet, I., & Raskin, I. (1995). Phytoremediation: A novel strategy for the removal of toxic metals from the environment using plants. *Biotechn 13,*468–474.

116

Sam, R. A., Ofosu, F. G, Atiemo, S. M., Aboh, I. J. K., Gyampo, O., Ahiamadjie, H., Adeti, J. P., & Arthur, J. K. (2015). Heavy metal contamination levels in topsoil at selected auto workshops in Accra. *International Journal of Science and Technology*, *4*(5), 222-229.

Satarug, S., Garrett, S. H., Sens, M. A., & Sens, D. A. (2010). Cadmium, environmental exposure, and health outcomes. *Environmental Health Perspectives*, *118*(2), 182-190.

Sauerbeck, D. R. (1991). Plant, element and soil properties governing uptake and availability of heavy metals derived from sewage sludge. *Water, Air and Soil Pollution 57/58*, 227–237.

Sejkorová, M., Hurtová, I., Glos, J., & Pokorný, J. (2017). Definition of a motor oil change interval for high-volume diesel engines based on its current characteristics assessment. *Acta Universitatis Agriculturae et Silviculturae Mendelianae Brunensis, 65*(2).

Sharma, P., & Dubey, R. S. (2005). Lead toxicity in plants. *Brazilian Journal of Plant Physiology*, *17*(1), 35-52.

Shi,G.T., Chen, Z.L., Bi, C.J., Wang, L., Teng, J.Y., Li, Y.S., Xu, S.Y. (2011). A comparative study of health risk of potentially toxic metals in urban and suburban road dust in the most populated city of China. *Atmospheric Environment*, *45*, 764-771.

Shukry, W. M., Al-Hawas, G. H. S., Al-Moaikal, R. M. S., & El-Bendary, M. A. (2013). Effect of petroleum crude oil on mineral nutrient elements, soil properties and bacterial biomass of the rhizosphere of jojoba. *British Journal of Environment and Climate Change*, *3*(1), 103-118.

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Smirnova, I. E., Tregubova, P. N., Koptsik, G. N., Stepanov, A. A., Kadulin, M. S., Turbaevskaya, V. V., & Zakharenko, A. I. (2017). Application of humic substances in the remediation of heavy-metal-polluted soils of the subarctic zone of the kola peninsula. *Moscow University Soil Science Bulletin*, *72*(5), 207-214.

Sobolev, D., & Begonia, M. F. T. (2008). Effects of Heavy Metal Contamination upon Soil Microbes: Lead-induced Changes in General and Denitrifying Microbial Communities as Evidenced by Molecular Markers. *International Journal Environ. Res. Public Health, 5*(5), 451.

Soil Health Guide for Educators (2019). *Soil bulk density/ moisture / aeration*. USDA Natural resources conservation service *stability*. Air Force Aero Propulsion Laboratory. United States Bureau of Mines. United States of America.

Suciu, I., Cosma, C., Todică, M., Bolboacă, S. D., & Jantschi, L. (2008). Analysis of soil heavy metal pollution and pattern in central Transylvania. *International Journal of Molecular Sciences*, *9*(4), 434–453. doi:10.3390/ijms9040434.

Swift, M. J, & Anderson, J. M. (1994). Biodiversity and ecosystem function in agricultural systems. *Springer*, 15-41.

Szili-Kovács, T., Anton, A., & Gulyás, F. (1999). Effect of Cd, Ni and Cu on some microbial properties of a calcareous chernozem soil. In J. Kubát (ed.), P*roc. 2nd symposium on the pathways and consequences of the dissemination of pollutants in the biosphere* (pp. 88-102). Prague.

Tahseen, R., Afzal, M., Iqbal, S., Shabir, G., Khan, Q. M., Khalid, Z. M., & Banat, I. M. (2016). Rhamnolipids and nutrients boost remediation of

crude oil-contaminated soil by enhancing bacterial colonization and metabolic activities. *International Biodeterioration and Biodegradation*, *115*, 192-198.

Tayo, O., Tobinson. A., & Briggs, A., (2018). Assessment of the impact of automobile waste on the environment. *International Journal of Engineering Inventions*, *17*(7), 1-15.

Tchounwou, P. B., Patlolla, A. K., & Centeno, J. A. (2003). Carcinogenic and systemic health effects associated with arsenic exposure-a critical review. *Toxicologic Pathology*, 31(6), 575–588.

Tchounwou, P. B., Yedjou, C. G., Patlolla, A. K., & Sutton, D. J. (2012). Heavy metal toxicity and the environment. *Molecular, Clinical and Environmental Toxicology*, 133-164. dOI: 10.1007/978-3-7643-8340-4_6.

United States Department of Agriculture (USDA) (2000). Heavy metals Contamination. Soil Quality Urban Technical Note No. 3. Natural Resources Conservation Service, USDA. https://dl.sciencesocieties.org/jeq.

Vwioko, D. E, Anoliefo, G. O., & Fashemi, S. D. (2006). Metal concentration in plant tissues of Ricinus Communis L. (Castor Oil) grown in Soil contaminated with Spent lubricating oil. *Journal of Applied Sciences and Environmental Management, 10*(3), 127-134.

Waldrop, M. P., Balser, T. C., & Firestone, M. K. (2000). Linking microbial community composition to function in a tropical soil. *Soil Biology and Biochemistry, 32*(13), 1837–1846.

Wang, G., Su, M. Y., Chen, Y. H., Lin, F. F., Luo D., & Gao, S. F. (2006). Transfer characteristics of cadmium and lead from soil to the edible parts of six vegetable species in south eastern China. *Environmental Pollution, 144,*127-35. doi:10.1016/j. envpol.2005.12.023.

Wang, M., Zou, J., Duan, X., Jiang, W., & Liu, D. (2007). Cadmium accumulation and its effects on metal uptake in maize (Zea mays L.). *Bioresource Technology, 98*(1), 82–88.

Wang, Y. B. (2007). Effect of probiotics on growth performance and digestive enzyme activity of the shrimp Penaeus vannamei. *Aquaculture*, *269*(1-4), 259-264.

Whisman, M. L, Goetzinger, J. W., & Cotton, F. O. (1971). *Radiotracer study of turbine aircraft fuel.* https://www.cdc.gov/niosh/nioshtic-2/10008729.html.

WHO/FAO (2007). Joint FAO/WHO food standards programme Codex Committee on contaminants in foods 5th Session: Working document for information and use in discussions related to contaminants and toxins in the GSCTFF. Rotterdam, Japan and Netherlands. http://www.fao.org/tempref/codex/Meetings/CCCF/CCCF5/cf05_INF. pdf.

Wolińska, A., & Stępniewska, Z. (2012). Dehydrogenase activity in the soil environment. *Dehydrogenases*, *10*, 183-210.

Wu, J. F., & Lin, X. G. (2003). Effects of soil microbes on plant growth. *Soils, 35*(1), 18-21.

Wuana, R. A., & Okieimen, F. E. (2011). Heavy metals in contaminated soils: A review of sources, chemistry, risks and best available strategies for

120

remediation. *International Scholarly Research Network*, *2011*. https://doi.org/10.5402/2011/402647.

Yan, S., Wang, Q., Qu, L., & Li, C. (2013). Characterization of oil-degrading bacteria from oil-contaminated soil and activity of their enzymes. *Biotechnology and Biotechnological Equipment*, *27*(4), 3932-3938.

Yourtchi M. S., & Bayat H. R. (2013). Effect of cadmium toxicity on growth, cadmium accumulation and macronutrient content of durum wheat (Dena CV.). *International Journal of Agriculture and Crop Sciences, 6*(15) 1099–1103.

Zurayk, R., Sukkariyah, B., & Baalbaki, R. (2001). Common hydrophytes as bioindicators of nickel, chromium and cadmium pollution. *Water, Air and Soil Pollution, 127,* 273-288.