EFFICIENCY OF *PHRAGMITES KARKA* ON THE PHYTOREMEDIATION OF POULTRY LITTER CONTAMINATED SOIL

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CERTIFICATION

The undersigned certify that he has read and hereby recommend for acceptance by the Open University of Tanzania a thesis titled **"*Efficiency of Phragmites karka on the Phytoremediation of Poultry Litter Contaminated Soil*"** in fulfillments of the requirements for the degree of Master Science (Chemistry) by Thesis of the Open University of Tanzania.

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DECLARATION

I, **Seif Abdallah Nyati**, declare that this thesis is my own original work and that it has not been presented and will not be presented to any other University for a similar or any other degree award.

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Seif Abdallah Nyati

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Date

DEDICATION

My dedication goes to my family members: my wife Swaum and My Children Abdallah, Sozy and Samia for their patience during the whole period of Thesis work.

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I would like to thank all the many people who assisted me with information. I will not be able to mention them by names, but I will mention the farmers who grow crops along the shores of Lake Victoria and as well shopkeepers who sell a variety of goods for broiler chickens especially antimicrobials and growth boosters and many other goods.

Special thanks go to my family wonderful wife Swaumu and my children Abdallah, Samia and Sozy for their continued patience during the whole period of my project.

ABSTRACT

This study aimed to investigate the efficiency of *P. karka* as phytoremediator in removal of Nitrogen, Phosphorus, and Arsenic in poultry litter contaminated soil. The plant species was grown in the laboratory settings in two beds, bed_1 and bed_2 and 500g of poultry manures were added in each bed. Analysis of nitrogen was done using Kjedahl distillation method whereas phosphorus was determined by using spectrophotometric methods as described in the standards methods. Arsenic was determined by using atomic absorption spectrophotometer with vapour generation accessory. Results showed a decrease in P (2.5% to 1.3%), N (3.7% to 2.4%), As (0.32% to 0.001%), for just after two weeks from contamination. From these results, *P. karka* are efficient macrophytes in phytoremediation. It is recommended for use in phytoremediation of contaminated areas like the lake shores of Lake Victoria.

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LIST OF ABBREVIATIONS AND SYNONYMS

N	Nitrogen
P	Phosphorus
As	Arsenic
Pb	Lead
Cd	Cadmium
Zn	Zinc
Cr	Chromium
Cu	Copper
K	Potassium
NO_3^-	Nitrate ion
NH_4^+	Ammonium ion
NO_2^-	Nitrite ion
pH	measure of acidity or alkalinity of a medium.
IARC	International Agency of Research on Cancer
BCC	Basal cell carcinoma.
SqCC	Squamous cell carcinoma
BFD	Black foot Disease.
NH_3	Ammonia gas.
CO_2	Carbon dioxide.
PM	Particulate matter
EDCs	Endocrine Disrupting compounds.

WHO World Health Organization

USEPA - United States Environmental Protection Agency.

ERS - Estrogen Receptors.

PCBs - Polychlorinated biphenyls.

OCPs - Organochlorine Pesticides.

OSC - Osteoclasts

OSB - Osteoblasts

AAS-VGA – Atomic Absorption Spectrophotometer- Vapour Generation Accessory

NO_2 – N - Nitrite Nitrogen.

CHAPTER ONE

GENERAL INTRODUCTION

1.1 INTRODUCTION

Poultry industry is a fast growing industry due to an increase demand for meat and as a result this sector has resulted into an environmental burden due to an increased accumulation of wastes produced by the boiler chickens called Poultry litter (Bolan et al., 1992). The main contaminants present in poultry litter include Nitrogen (N), Phosphorus (P), Nickel (Ni), Antimicrobials, Excreted Estrogenic Compounds (Estriol, Estradiol and Estrone) and as well pathogens called *Salmonella* spp.Mishandling of poultry litter pose threats to the environment forinstance if someone swallow drinking water rich with high concentrations of Nitrates will acquire a blood disorder in infants called methemoglobinemia (Avery, 1999; manassaram, 2010).

Nitrates and Phosphorus may also lead to overgrowth of algae forms sucha as blue – green algae also called cyanobacteria which threates aquatic life for fishes and other aquatic organisms by releasing a variety of toxins such as hepatoxins, cytotoxins, neurotoxins and respiratory toxins (Babica, Blaha and Marsalek, 2009). While contamination by Arsenic present in poultry litter causes skin cancer for human being. Other threats posed by poultry litter include spread of various diseases such as dysentery, diarrhea, Abdominal pain, typhoid fever brought by a pathogen present in poultry litter called salmonella

spps which is spread during mishandled meat preparations (Corry et al., 2002; Boonsanong et al., 2002; sams, 2000). Other contaminants present in poultry litter are antimicrobials which are given to Broiler chicken for various purposes forinstance, spectinomycin for treating chronic respiratory diseases, gentamycin for prevention of early mortality, penicillin for promoting feed efficiency and when these antibiotics are given in a feed where by most of these are excreted in the feaces and spread in the environment hence cause health problems to aquatic creatures (EPA, 2013).

Due to many threats posed by poultry litter in the environment, this research article has come up with the concept called phytoremediation in order to remediate contaminated soils used for agricultural activities along the shores of lake Victoria by using wet land plants called *Phragmites karka* and these kind of plants have been chosen since are hollow and it is expected that a wide range of contaminants especially N, P and Ni will be eliminated from the contaminated areas.

1.2 .RESEARCH PROBLEM

The main concern of this thesis is to address a problem which has existed for several years for farmers who practice agricultural activities on the shores of lake Victoria for growing various crops such as spinach, maize, cabbage and many others and unfortunately these farmers have been using poultry manure for the improvement of their yield without understanding the risks posed by this kind of manure.

Based on literature review there are several hazards found in poultry manure that need to be addressed by relevant authorities to these local farmers so that they could not proceed with this habit of growing crops by using poultry manure on these lake shores. And unfortunately most farmers do accumulate hills of manure in their farming areas as a reserve stock which in some occasions some of the manures are carried downstream by runoff into the lake which results into an increase overgrowth of water blooms. Among threats posed by poultry litter include spread of cancers through swallowing of food rich with Arsenic (Bolan et al., 1992). Either swallowing nitrates through drinking water causes methemoglobinemia (Avery, 1999; Manassaram, 2010). High levels of N and P causes overgrowth of algae, and if blue green algae grows (cyanobacteria) then toxins will be released in water such as neurotoxins, endotoxins, respiratory toxins, cytotoxims (Babica, Blaha and Marsalek, 2009).

While pathogens called salmonella spps present in poultry litter causes several diseases such as dysentery, diarrhea, vomiting, typhoid fever and Abnominal pain (Corry et al., 2002; Boonsanong et al., 2002; Bisgaard et al., 2003. &Sams, 2000). Either the excretory products called Estrogenic Endocrine Disruptors which are the secreted hormones in the bodies of Broiler chickens also pose environmental impacts once they enter into water bodies by causing body deformations, abnormal colour, missing of certain features such as fins, tails, operculum (Mbuthia et al., 2014).

1.3. RESEARCH OBJECTIVES

1.3.1 .GENERAL OBJECTIVE

The main objective of this thesis is to examine the effects of *Phragmites karka* on the phytoremediation of poultry litter contaminated soil.

1.3.2. SPECIFIC OBJECTIVES

The specific objectives of this study are as follows:-

(i) To determine the levels of N, P and As in uncontaminated and contaminated soil farm with poultry litter.

(ii) To determine the levels of N, P and As in soil contaminated with poultry litter in a laboratory setting.

(iii) To determine the efficiency of *Phragmites karka species* in removing *N* and P and As .

1.4. HYPOTHESES

This Thesis used the following hypotheses:-

(i) The levels of N, P and As in soil contaminated with poultry litter is lower after phytoremediation in the laboratory setting.

(ii) The levels of N, P and As in a contaminated soil farm with poultry litter is higher than in the uncontaminated soil farm.

(iii) Phragmites karka species can significantly remove N, P and As in poultry litter contaminated soil

1.5. SIGNIFICANCE OF STUDY

This study will enhance policy makers and government officials as well understand hazardous effects of using poultry manures along the shores of water bodies for instance on lake shores as these poultry manures favours growth and excessive accumulations of water blooms as these manures carry a lot of plant nutrients such as nitrogen (N) and Phosphorus (P).

Other threats posed by using these poultry manures to be addressed to policy makers include:-

- Cause neurological effects to aquatic organism such as fishes.
- Cause body deformations effects to aquatic creatures for instance causing tail deformation, deformed head, deformed heart, abnormality in colour.

Based on these findings will enhance policy makers to come up with the enacted laws on how to protect the lake shores such as Lake Victoria

- Prohibiting the growing of crops by farmers along the shores of the lake. Hence the study will make the government benefit because the environmental resources such as water will be conserved.

- Secondly, the government could adopt the use of bioremediation technique in remediating the contaminated areas of the lake shores

Due to civilization and urbanization in urban areas a large quantity of wastes is generated which is dumped in the environment annually. Solid waste management is a major challenge in urban areas throughout the world and without an effective and efficient solid – waste management program, the waste generated from various human activities can result in health hazards and have a negative impact on the environment.

In this paper, it has been seen that excessive use of poultry manures as a source of plant nutrients pose an environmental threat and hence solid – waste management is needed in order to protect the environment against environmental contamination brought by poultry manures which add excessive Nitrogen and Phosphorus elements in the soil.

Thus, Bioremediation is an effective process which is not only a process of removing the pollutants from the environment but also it is cheap and environmental friendly as it does not pose environmental hazards.

There are several kinds of bioremediation techniques including;- bioventing, biosparging, bioagumentation, biopiling and Phytoremediation.

Bioventing is a technique to degrade any aerobically degradable compound in which oxygen and nutrient like nitrogen and phosphorus is injected to the contaminated site.

In this process, the distribution of these nutrients and oxygen is dependent on soil texture. In this process enough oxygen is provided through low air flow rate for microbes.

However in biosparging, air is injected below the ground water under pressure to increase the concentration of oxygen. The oxygen is injected for microbial degradation of pollutant. Biosparging increase the aerobic degradation and volatilization. This process is effective in reducing petroleum products at underground storage tank sites. In case of bioagumentation, microorganisms having specific metabolic capability are introduced to the contaminated site for enhancing the degradation of waste where soil and ground water are contaminated with chlorinated ethers such as tetrachloroethylene and trichloroethylene. In this process all chlorinated etheres are decomposed or broken down into ethylene and chloride which are non – toxic.

In this paper, the waste management technique adopted to remove pollutants is known as Phytoremediation which is also cost effective and affordable technique which utilizes natural plants that are able to bioaccumulate toxins in their tissues which are then harvested for removal of wastes and the kind of plants that were used are called *Phragmites karka* which had shown a high efficiency in the removal of Nitrogen (N), phosphorus (P) and Arsenic in contaminate soil and hence I can admit that marsh plants such as *Phragmites karka* are the most effective and efficient bioaccumulators for toxic removal management for soil contaminants.

This thesis will create awareness to the community about the adverse effects of using the poultry manures in agricultural sector along the shores of water bodies for instance along the shores of Lake Victoria because using these kinds of manures pose several threats to their health and as well in the environment.

Among several impacts of growing vegetables using these kind of manures (poultry manure) is the spread of cancers to the community since these poultry manures carry Arsenic which causes skin cancer (Bolan et al., 1992). Either swallowing water contaminated with excessive nitrates brought by excessive use of poultry manures causes methemoglobinemia (Avery, 1999. &Manassaram, 2010).

Another impact that this article addresses to the community is that excessive use of these poultry manures causes overgrowth of water blooms that cause a reduction of oxygen in water hence death of aquatic organisms and if it happens that a kind of water blooms accumulated are blue – green algae also known as cyanobacteria will result into a release of toxins in water that cause hazards such as neurotoxins, dermatoxins, endotoxins, respiratory toxins as well hepatoxins (Babica, Blaha and Marsalek, 2009).

Poultry manures also contain several other contaminants such as antimicrobials which are released in fecal droppings which cause health problems to various aquatic organisms.

The burning issue of today's environment problem is the release of toxic contaminants from various man made sources resulting in contamination of natural resources of earth and leading to scarcity of clean water and soil contamination. To overcome these drawbacks, a much better perspective is to completely destroy the pollutants, or to transform them into some biodegradable substances. This approach can be achieved by using a technique known as bioremediation which acts as an option to clean and conserve the environment and its resources by destroying various contaminants using natural biological activity. It is considered as safer, cleaner, cost effective and environmental friendly technology.

Phytoremediation is a newly evolving field of science and technology that uses green plants to clean up polluted soil, ground water and waste water. Phytoremediation is defined as the use of green plants including grasses and woody species to remove environmental contaminants as heavy metals, metalloids, trace elements organic compounds and radioactive compounds in soil and water. Phytoremediation takes advantage of the unique and selective uptake capabilities of plant root systems together with the translocation, bioaccumulation, and contaminant storage / degradation abilities of the entire plant body. The mechanisms and efficiency of Phytoremediation depend on the type of contaminant, bioavailability and soil properties and the uptake of contaminants in plants occurs through the root system

CHAPTER TWO

LITERATURE REVIEW

2.1 .GENERAL IMPACTS OF POULTRY LITTER

Poultry industry is one of the largest and fastest growing sector in the world which is motivated by an increasing demand for broiler chicken meat and as well a high demand for eggs as a result there is a shifting in a method of raising chicken where growers raise these chickens in the confinement unit in which a feed is provided by growers in a house where the chickens are confined so that the chickens does not look after the feed in the surroundings on their own (Chalamila, 2007).

This modern technology of growing broiler chickens result in an accumulation of wastes within a broiler chicken house commonly known as broiler chicken litter or poultry litter which is a mixture of feed droppings, fecal droppings as well bedding materials (Bolan et al., 1992).

The major challenge facing this sector of poultry production is the daily accumulation of litter within the growing house which results in daily over loading of wastes due to the fact that most of the feed supplied to the chicken house is not consumed but drop on the ground and then mixed with bedding materials such as wood chavings, cereal straw, husk and paper clippings (Swaim and Sundaram, 2002). Figure 2.1 shows a hips of poultry manure being dumped in a farm along lake Victoria shores for agricultural purposes.

There are three common practices of broiler litter management in the broiler unit house and these include single use litter, partial re-use and multi- use litter. The single – use litter involves the total clean – out of the house after each flock and replacement of the bedding material.

Partial re- use involves the removal of litter from the brooding section for spreading on the grower section of the house and then new bedding material is spread on the brooding section. However the partially spent litter is often composted for a few days to elevate its temperature in order to kill the pathogens and some of the spent litter may be removed after each batch, and after 2 to 5 batches the house is totally cleaned out.

The amount of total solids (day matter) excreted by the birds can be estimated from the dry matter digestibility of the diet. Broiler chickens generally digest about 85% to 90% of the dry matter of the feed (NRC, 1994). Broiler chickens consume

approximately 2.5 to 3.0 kg of dry matter up to 35 days of age and 5 to 6 kg of dry matter up to 49 days of age (FSA, 2007). At a moisture content of 90%, total manure production will be around 4 and 6 kg for 35 and 49 days old birds and it has been estimated that broiler chickens excrete approximately 55% of the total N, 70% of P and 80% of K.

Figure 2.1 A hips of Poultry manures on farms

However poultry litter is economically significant in agricultural sector as it provides poultry manure which adds essential major plant nutrients which include Nitrogen (N), phosphorus (P) and Potassium (K) and studies have indicated that poultry manures contain much amount of Nitrogen and phosphorus (ASABE, 2005; AXTELIRS 1986; Bitzer and Sims, 1988) as seen in table 2.1.

Table 2. 1 . Nutrient content of man (91 kg)dry . (Bolan et al. 1992).

Nutrients	Poultry manure composts	
	Layer	Broiler
Nitrogen	32.8	25.7
Phosphorus	10.8	6.7
Potassium	15.2	10.1

There are four forms of N in organic N, labile organic N, ammonium and nitrate N (Sims and Wolf, 1994; Sharply and Smith, 1995; Diaz et al., 2008). Complex forms of organic N in poultry litter include constituents of feathers, split and undigested feed, and bedding materials. Labile organic N is Uric acid and Urea.

Phosphorus in poultry litter is about two thirds present as so lid – phase organic P and one third as in organic P (Edwards and Daniel, 1992; shapley et al; 2004). Apart from major plant nutrients, there are also trace elements and their concentrations. Since a major portion of the trace elements ingested is excreted in faces and Urine, the concentrations of trace elements in manure by – products depend primarily on their concentrations in the diet (krishnamachari, 1987; Miller et al; 1991). Kunkle et al. (1991) noticed that Cu concentrations in poultry manure – by products were linearly related to Cu added in the diet.

The major problem facing poultry industry is the accumulation of wastes which have environmental impacts in air, water and soil and the major contaminants spread by poultry litter include:-

a. Disposal of excess Nitrogen and phosphorus

b. Disposal of Arsenic

c. Disposal of Pathogens

d. Disposal of Endocrine disrupting compounds

e. Disposal of Antimicrobials

f. Disposal of Ammonia in the atmosphere

g. Disposal of Toxins in water

2.2 .SPECIFIC IMPACTS OF POUTRY LITTER

2.2.1: DISPOSAL OF EXCESS NITROGEN AND PHOSPHORUS TO ENVIRONMENTS

Disposal of excess nitrogen in the form of Nitrates (NO^-_3) causes a defect called methemoglobinemia in infants which is brought about by drinking water having high concentrations of $NO_3^-$by pregnant women.

Methemoglobinemia or sometimes called blue baby syndrome occurs when methemoglobin in the form of hemoglin which iron is oxidized to its ferric state and is unable to deliver oxygen. Methemoglobinemia occurs when amounts of methemoglobin in the blood become high enough to manifest clinical symptoms of cyanosis, usually about 15% of total circulating hemoglobin.

Methemoglobinemia occurs for various reasons including generic abnormalities in hemoglobin that make the protein more susceptible to oxidation and exposure to oxidant drugs and chemicals including nitrate. Infants under 6 months of age are more susceptible to methemoglonemia because they have lower amounts of a key enzyme called NADH – cytochrome $b_5$reductase (methemoglobinreductase) which converts methemoglobin back to hemoglobin.

For over 40 years there has existed a widespread belief that nitrates in drinking water are the primary cause of infantile methemoglobinemia. Hunter comly originally proposed this theory in 1945 in a report in the Journal of the American Medical Association after treating several infantile methemoglobinemia victims exposed to nitrate – contaminated water where comly proposed that because nitrites (NO^-_2) are known to react directly with hemoglobin to form methemoglobin, nitrates (NO^-_3) from drinking water must be converted to nitrites within the gastrointestinal tract of infants. Because many infants did not appear susceptible to methemoglobinemia from nitrate – contaminated water, comly suggested that nitrate – to nitrite conversion might only occur in the presence of a bacterial infection of the upper gastrointestinal tract where such reactions could occur before nitrates are absorbed.

These nitrate – derived nitrites comly then react with hemoglobin to form methemoglobin and in sufficient quantities lead to cyanosis of methemoglobinmia.

Hence it was decided that limiting infant exposure to nitrates was the most prudent approach to protect infants from methemoglobinemia. A survey conducted by American Public Health Association (APHA) to determine a safe level of nitrates in water and a total of 278 cases with 39 deaths were compiled. The results showed that methemoglbinemia incidence correlated with increasing nitrate levels because no infantile methemoglobinemia cases were observed with concentration – 10ppm nitrate – nitrogen concentrations.

Another impact resulted from the disposal of excess Nitrogen and phosphorus nutrients of poultry manure is an overgrowth of algae from called cyanobacterial water blooms which represent a major ecological and human health problem worldwide. This kind of algae forms secrete various kind or toxins in aquatic systems and hence create environmental threat to aquatic organism and studies show that about 40% of lakes and reservoirs in Europe, America as well Asia are now eutrophic and promote favourable conditions for cyanobacteria mass development (Bartram et al; 1999).

Cyanobacteria blooms have severe impacts on ecosystem functioning by disturbing the relationships among organisms, changes of biodiversity light conditions or oxygen concentrations.

Also the occurrence of cyanobacterial mass populations can create a significant water quality problem especially as many cyanobacterial species are conpable of synthesizing a wide range of ordours, noxious compounds or potent toxins (Sivonea Jones, 1999) and it has been estimated that about 25 to 75% of

cyanobacterial blooms are toxic and secrete toxins called cyanotoxins such as hepatoxins, neurotoxins, derma toxins, respiratory toxisns as well endotoxins. The potential impacts of cyanobacteria include health impacts such as gastrointestinal disorders, liver inflammation, cardiac arrhythmia (Babica, Blaha and Marsalek, 2009).

According to their classical structures, cyanotoxins (cyanobacteria toxins) fall into several main groups such as:Peptidy, heterocyclic compounds, cyanobacterial lipopolysaccharides. Hepatotoxic heptapeptides or microcystins are the most prevalent cyanotoxins in the environment and they are present in high amounts in cyanobacterial biomass and studies have shown that this group of cyanobacterial algae blooms/ microcystins brings an acute toxicity to animals and humans several experiments with manuals eg rodents showed significant subchronic and chronic toxicity of orally administered microcystins where harmful effects of microcystins such as increased mortality, liver injury (including histopathological changes, chronic inflammation, degeneration of hepatocytes, increased liver enzyme levels, renal damage. The majority of microcystin producing blooms have also shown to involve in many incidents of fatal animal poisoning in cattle, sheep, chickens, horses, poultry and wild birds, fishes. However the wide range of aquatic organisms is directly exposed to microcystins contained in their food or to microcystins dissolved in water which causes a wide range of effects.

Microcystins and many other cyanotoxins are released into the environment throughout the summer season and normally released into the surrounding water by senescence and lysis of the blooms and any form of toxin present could then come into contact with a wide range of aquatic organism including phytoplankton, phytoplankton grazers, invertebrates, fish or aquatic plants.

Bioaccumulation is an important process through which chemicals can accumulate and affect living organisms such as aquatic organisms. Cyanobacterial hepatotoxins such as microcystins, accumulate in animal tissue but do not cause acute death of animal in environment with the natural concentration of microcystins. Many death losses were mainly caused by neurotoxins which caused inassive death of Pleistocene large manual in the lake basin of Neumark – Nord in Germany 150,000/= years ago.

(Braun and Pfeiffer 2002). In case of lethal dose of microcystins death of vertebrates animals is mostly the consequence of severe liver damage which starts with cytoskeletal disorganization and can include cell blebbing, cellular disruption, lipid peroxidation, loss of membrane integrity, DNA damage, apoptosis, necrosis and ultimately death by hemorrhagic shock.

The target organ for microcystins is mainly liver / heap topancreases, where microcystins enter the meubrane through specific mechanism. Since zooplankton is one of the most important link between primary producers and higher producers such as fishes thus zooplankton may be an important vector of cyanobacterial toxins along the food chain.

2.2.2: DISPOSAL OF ARSENIC IN THE ENVIRONMENTS

Applying poultry litter in the soil may also add a trace element called Arsenic (As) which is a toxic element and this element may be absorbed by crops being grown in the contaminated soil and eventually may get transmitted to the human bodies through eating food containing arsenic (Bolan et al. 1992). Much of this Arsenic in poultry litter comes from commercial broiler operations which use Arsenic as a feed additive and it is normally given to broiler chickens in the form of roxarsone and arsenilic acid as feed additive of conventionally – raised broilers. It is used to control protozoan parasites known as coccidians and enhance weight gain (Morrison, 1969). Arsenic in the soil includes the following forms, arsenious (H_3AsO_3, $H_3AsO_3^-$ and $H_3AsO_3^{2-}$), arsenic acids (H_3AsO_4, $H_3AsO_4^-$, $H_3AsO_4^{2-}$), arsenates, arsenites and methylarsenic acid in which the inorganic forms of arsenic is more toxic than organic forms (Tangahu,2011).

And it has to be noted that Roxarsone is normally added to poultry feed at a rate of 22.7 to 45.5grams per ton or 0.0025 to 0.005 percent.

Most of the roxarsone passes through the birds and is excreted unchanged and each broiler chicken excretes about 150 milligrams of roxarsone during the 42 day growth period in which it is administered. Litter collected following a single flock of birds can contain from 1 to 70 milligrams of arsenic per kilogram of litter with 30 to 50 milligrams per kilogram commonly found or

0.003 to 0.005 percent. Poultry houses are only partially cleaned, following each flock of birds, increasing the concentration of arsenic in the litter.

The movement and toxicity of arsenic is affected by chemical and microbial reactions which readily transform roxarsone into inorganic forms of arsenic. These inorganic forms are then subject to a variety of chemical and biological reactions in the soil. Soil mineralogy, soil moisture, soil pH and microbial reactions all determine arsenic mobility, its uptake by plants, and its toxicity (BC Bellows, 2005). When arsenic is bound to soil minerals it is relatively immobile but when arsenic is dissolved in water it can be taken up by plants and is subject to runoff or leaching. Thus arsenic is more likely to damage the environment, affect crop growth, or endanger animal and human health than is arsenic which is bound to soil particles.

Arsenic is more likely to bind to soil particles in soil that is:-

- Field moist or dry
- Neutral to slightly acidic in its reaction or p^H
- Rich in iron, aluminum, manganese or limestone.

But Arsenic is more likely to be soluble in soil that is:-

- Met or muddy
- Alkaline but without limestone mineralogy.

- Relatively high in concentrations of phosphate or nitrate
- Sandy

In wet soils that have a high (alkaline) p^H, soil chemistry will favour arsenite over arsenate resulting in high arsenic toxicity. If poultry litter containing arsenic is added to upland, arable soils that have loamy or claylike textures, neutral or semi acid p^H, and are not subject to water logging, the arsenic will be relatively stable low toxicity.

In contrast, if poultry litter containing arsenic to soils that are wet, alkaline, or have a sandy texture, the arsenic will have a high toxicity and a high potential for contaminating ground or surface water through leaching or runoff. Plant uptake of arsenic will be greatest on sandy soils with low to moderate levels of organic matter and excessive amounts of phosphorus or nitrate. The greatest risk of contamination from arsenic in poultry litter comes when litter is removed from poultry house but not mixed with soil and this happens when the litter is stacked in piles before spreading or when it is applied to the soil and not mixed in through tillage.

Recent studies have shown that organic compounds tend to displace arsenic bound to iron oxide resulting in the release of dissolved arsenic into the soil and this process not only increase the amount of dissolved arsenic but also its availability and toxicity since the organic matter displaced arsenite more readily than arsenate.

It has to be understood that Arsenic and phosphorus are chemically very similar since both bind to iron and aluminium oxides and as well both are major components of the clay coatings on soil aggregates.

Since phosphorus is much more abundant in agricultural soils than arsenic, it crowds arsenic off binding sites hence increasing the solubility and mobility of arsenic. Because of the chemical similarity of phosphorus and arsenic, plants confuse the two chemicals and then these plants take up arsenic and metabolize it as through it were phosphorus.

Many mychorrhizal fungi facilitate plant uptake of phosphorus and also increase plant up take of Arsenic. In sandy soils, phosphorus additions stimulate plants to take up take of Arsenic. In sandy soils, phosphorus additions stimulate plants to take up additional arsenic.

While soluble or dissolved arsenic poses the greatest risk for environmental contamination and wind or water erosion then transport arsenic as a result contaminating rivers and streams.

Arsenic contamination pose a major threat to health of human beings as it causes cancers (Martinez, 2011) and there is strong body of evidence linking arsenic with a variety of health problems, from acute toxicities to chronic diseases which can take years to develop.

The diseases associated with arsenic contamination include skin lesions, hyper tension, some endemic peripheral vascular disorders, diabetes, severe arteriosclerosis, neuropathies.

According to the international Agency of Research on cancer (IARC) Arsenic has been classified as a class I human carcinogen and there is sufficient evidence of carcinogenicity to humans.

Skin and several types of internal cancers, including bladder, kidney, liver, and lung have been associated with arsenic ingestion. Skin cancer is the most common form of neoplasm associated with arsenic ingestion while lung cancer corresponds to the most deadly form of cancers.

Studies have shown that the most common malignancies found in patients with long-term exposure to arsenic include Bowen's disease, basal cell carcinoma (BCC) and squamous cell carcinoma (sq CC). Arsenic – related skin SqCC can develop from Bowen's disease where as arsenic related BCC develops usually in multiple foci and areas of the body covered from sun exposure in contrast to cases originating from other skin carcinogens such as UV – light.

Arsenic – related Bowen's disease can appear 10 years after arsenic exposure while other types of skin cancer can have a latency period of 20 or 30 years.

Premalignant skin lesions are relatively early manifestations of arsenic toxicity and are often considered precursors to arsenic – induced skin BCC and SqCC tumors.

These lesions include dermal manifestations such as hyper pigmentation "raindrop" pattern of pigmentation and hyperkeratosis characterized by skin thickening mainly at palms and the feet. These lesions are commonly found in chronically exposed populations and are considered a diagnostic criterion of arsenicosis.

In the case of lung cancer, there is a significant relationship between lung cancer and ingested arsenic and this was discovered following a therapeutic application of this metalloid in psoriasis patients treated with fowler's solution. Studies have shown that a high concentrations of arsenic in drinking water pose a significant threat by causing lung cancers (NRS 1999).

Another risk brought by arsenic to human beings is the spread of a disease called Blackfoot disease (BFD) which is an endemic, peripheral arterial disease characterized by severe systemic arteriosclerosis and spontaneous gangrene resulting in amputations and is common to individuals exposed to arsenic.

2.2.3: DISPOSAL OF TOXIC GASES IN THE ATMOSPHERE

Furthermore, another threat posed by poultry litter is air pollution caused by ammonia volatilization which causes the formation of acidic rain (Ritz et al., 2004).

Ammonia is a byproduct from Excretion of Nitrogen (N) which is excreted once excess proteins and amino acids are fed to poultry chickens which are not all digested and then part of undigested proteins is excreted in fecal waste and

approximately 50% of the N content of freshly excreted poultry manure is in the form of uric acid. Then N in Uric acid can be very quickly converted to ammonia (NH_3) by hydrolysis, mineralization and volatilization.

Microbial degradation of Uric acid in the litter is the primary source of NH_3 formation and *Bacillus pasteurii* is one of the primary Uricolytic bacteria that facilitate NH_3 production. For optimum growth, these bacteria require a pH around 8.5 and the decomposition process requires Uric acid, water, and oxygen to react giving off NH_3 and carbon dioxide (CO_2). The process of decomposition of uric acid into NH_3 also involves several enzymes, including uricase and urease. Uricase converts uric acid into allontoin, which is later converted into glyoxylate and urea. With the addition of water (moisture), urease breaks urea down into NH_3 and CO_2.

The formation of NH_3 continues will the microbial breakdown of manure under both aerobic and anaerobic conditions. The water soluble characteristic of NH_3 allows it to be dissolved in the moisture on mucous membranes and eyes, and it is also associated with dust particles. Ammonia does not have ionic charge hence making it readily released into the atmosphere in gaseous form. Protonating NH_3 into non volatile ammonium (NH_4^+) require an acidic environment and factor that contribute to the formation of NH_3 include temperature, moisture, pH and nitrogen content of the litter or manure. Temperature, moisture, pH have direct influence of the litter on the living environment of the microorganisms that facilitate the conversion of uric acid

into ammonia. High house temperatures increase both bacterial activity and ammonia production. The pH has a direct effect on litter moisture. Ammonia production is negligible when manure or litter pH is at levels less than 7, in area sing as pH approaches 7.0 and high when pH approaches 7.0 and high when a pH of 8.0 or greater is reached. Typically, the pH of poultry manure as extracted and the pH of litter are between 7.5 and 8.5. It is estimated that 50 to 89% of N in manure is converted into NH_3. Studies have shown that the levels of NH_3 within a poultry house should be maintained low in order to prevent detrimental effects on health issues and the levels of NH_3 should not exceed 25ppm in poultry houses. However, prolonged exposure to concentrations as low as 20 ppm can be detrimental to bird health.

Broiler feed consumption and feed efficiency has been shown to decrease during exposure to levels of NH_3 ranging from 25 to 125ppm. Symptoms of NH_3 poisoning in poultry include tracheal irritation air sac inflammation, conjunctivitis and dyspnea. Exposure to 20ppm for long periods of time has resulted in a variety of disorders including respiratory tract damage.

Levels of 75 to 100ppm are associated with changes in the respiratory epithelium including loss of cilia and increased number of mucus – secreting cells. While exposure to 25ppm for 42 days resulted in decreased feed efficiency and after 56 days resulted in airsacculits following infections bursal disease exposure.

Exposures to 46 to 102ppm resulted in eye damage in the form of keratoconjuctivitis. After eye damage has occurred birds may experience difficult in finding feed and water sources. Bird performance and health can therefore be affected by both respiratory disease and physical damage due to increased NH_3 concentration. Higher levels of ammonia emissions is detrimental to environmental concern, and once emitted NH_3 can rapidly react with acidic compounds found in the atmosphere such as nitric acid, sulphuric acid and be converted to aerosolized ammonium particles typically as ammonium sulphate and ammonium nitrate.

As aerosols, N compounds can impact ecological balance, biodiversity, and water systems. Deposition back onto the soil, vegetation or water usually occurs within a matter of days and thus in relatively close proximity to the emission source and once deposited, N can impact soil acidity forest productivity, terrestrial ecosystem biodiversity, stream acidity, and coastal productivity. Also high N concentrations in the atmosphere.

Contributes to the formation of acid rain that may damage plant life, cause excessive fertilization of soils, and vegetation, increase algal blooms in surface waters, and damage aquatic life.

In general plant growth globally is limited by N, and deposition of N, therefore can cause increased plant growth. European forests that receive N from atmosphere deposition show an increase in nitrate leaching as much as 30% of

inorganic N deposition (Ritz et al; 2004). Emissions of N compounds can result in N fertilization and species change in natural ecosystems.

A number of fertilization studies have demonstrated that increased N availability promotes the dominance of fast – growing, nutrient – rich plant species to the detriment of shower growing nutrient – poor species.

Aerosolized ammonium contributes to particulate matter (PM) in the atmosphere specifically PM 2.5 or PM with an aerodynamic diameter of 2.5 microns or less.

Such small diameter PM contributes to atmospheric haze and may have a negative impact upon human respiratory health. Due to health and environmental impacts of NH_3 volatilization, strategies for reducing NH_3 volatilization should be directed towards reducing NH_3 formation. Since NH_3 losses immediately once formed immediately, potential strategies for control of NH_3 in poultry production include among others: Ventilation, dietary manipulation, and manure management.

Traditionally, improving air quality in poultry has been largely accomplished through ventilation. Increased ventilation rates reduce NH_3 concentration within the house but translate directly into higher emissions. Ventilation is therefore more of an inhouse air quality control method then a strategy to inhibit the formation and emission of NH_3

Most acidifying agents function similarly to reduce NH_3 volatilization by lowering the P^H of manure or litter and thereby reducing microbial microbial activity. Use of these agents has been shown to improve bird performance and lower the energy usage needed to ventilate poultry houses. Additional benefits with the use of acidifying agents have been documented as evident by reduction in the incidence of ascites, reduction of respiratory lesions, reduction of litter *Escherichia coli*, and reduction of water soluble phosphorus concentrations in litter. The use of acidifying agents has been shown to be effective in controlling NH_3.

2.2.4: DISPOSAL OF ANTIMICROBIALS IN THE ENVIRONMENTS

Another impact brought by poultry litter in the environment is the disposal of antimicrobials in the environment which are discharged through fecal droppings. Antimicrobials are antibiotics which are given to poultry chickens as food additives during growing processes of Broiler chickens and there is a wide range of these antibiotics which include Gentamycin, spectinomycin, bacitracin, bambermycin, chlortetracycline, dihydrostreptomycin, erythromycin, lincomycin, neomycin, tetracycline and tylosin (Bolan et al., 2010).

These antibiotics are used to treat clinical diseases, to prevent and control common infectious diseases and also to promote animal growth. The different applications of antibiotics in food animals have been described as therapeutic use, prophylactic use, and subtherapeutic use. Antibiotics can be used to treat a

single animal with clinical disease or a large group of animals (Landers et al., 2012). Despite the widespread adoption of antibiotic use in food animals, reliable data about the quantity and patter of use are not available.

Quantifying antibiotic use in food animals is challenging due to variations in study objectives as investigations may measure only therapeutic uses, only non therapeutic uses, or a combination depending on their outcome of interest. Although limited, the available data suggest that food animal production is responsible for a significant proportions of antibiotic use. Forinstance, in 1989, the institute of medicine estimated that approximately half of the 31.9 million pounds of antimicrobials consumed in the U.S were for non therapeutic use in animals. More recent estimates by the union of concerned scientists, an advocacy group that supports reduced agricultural antimicrobial use suggest that 24.6 million pounds of antimicrobials are used for non therapeutic purpose in chickens and cattle, compared with just 3.0 million pounds used for human medicine. The twelve classes of antimicrobials being in use include arsenicals, polypeptides, glycolipids, tetracycline, elfamycin, macrolids, lincosamides, polyethers, beta – lactams, quinoxalines, streptogramins, and sulphonamides – may be used at different times in the life cycle of poultry, and cattle. While some of antimicrobiasl used in animals are not currently used in the treatment of infections in humans, but still others such as tetracyclines, penicillins and sulphonamides are also used to treat human disease. However recent studies have revealed that, there is an association between antibiotic use in food animals and antibiotic resistance in humans. Antibiotic – resistant bacteria of

animal origin have been observed in the surrounding farming operations, on meat products available for purchase in retail food stores, and as the cause of clinical infections and subclinical colonization in humans.

Antibiotic use in animals can have direct and indirect effects on human health in which direct effects are those that can be causally linked to contact with antibiotic – resistant bacteria from food animals and indirect effects are those that result from contact with resistant organisms that have been spread to various components of the ecosystems (eg. Water and soil) as a result of antibiotic use in food animals.

2.2.5: DISPOSAL OF PATHOGENS IN THE ENVIRONMENTS

Also poultry litter contamination in the soil adds pathogens (harmful microorganisms) and the most common pathogen found in the poultry litter is called Salmonella typhimurium. Recent studies have indicated that Salmonella typhimurium not only contaminate in the soil but also may be contaminated in the poultry meat during meat processing activities when dressing is not conducted well (Bisgaard et al., 2003). Salmonella gets into the soil when manure is applied onto the soil and conditions exist that does not kill the salmonella.

The process of manure handling, storage and spreading time all play important roles in the life cycle and survival of Salmonella in the soil.

Salmonella is considered to be an enteric or fecal organism because it is normally found in the intestine of birds and mam-mals and when an animal defecates, the salmonella passes out of the body and then if the unprocessed manure is then spread onto the fields, lawns or gardens, the salmonella can now be found in the soil.

Salmonella is one of the leading causes of food borne infections in the world by consuming poultry products including eggs and meat. According to US Food and Drug Administration (2009), 2 to 4 million cases of salmonellosis in humans occur every year only in US. Salmonella causes a wide range of diseases with enteric and typhoid fever, food poisoning, diarrhea and gastro-enteritis (Maqsood, 2012).

Poultry feed is considered to be the main source of transfer of salmonella into poultry flocks including dust, cooling system and feed ingredients can be the major sources of salmonella contamination during the feed milling process. Feed ingredients and environment which harbor *Salmonella* can mix contamination in feed which results in the cross contamination from feed to the animals. In humans, Salmonella causes a wide range of diseases with enteric and typhoid fever, food poisoning, diarrhea and gastro-enteritis while in poultry, it causes a variety of acute and chronic diseases including. Paratyphoid, fowl typhoid, avian arizonosis, enterotoxigenic diarrhoea, plugue and shigellosis.

Salmonella can infect poultry flocks through feed, water, hatching eggs and through environmental factors including birds, insects, rodents and farm workers. The symptoms of disease are acute with prolonged effects of abdominal cramps, fever and mild diarrhea. Salmonella species include S. typhimurium, S. Enteriditis, and S. infantis do not have host specificity and cause diseases in all kinds of animals and human, while in Poultry S. pullorum and S. gallinarium commonly cause Pullorum disease and fowl typhoid and these infections can be ingested through feces, fluff, litter and water. Various studies have revealed that, one of the leading causes of food borne infections in the world is still due to S. enteriditis by consuming poultry products including eggs and meat.

Food poisoning in human beings is closely related by the use of poultry products which are contaminated with salmonella. It has also found that birds contaminated with salmonella may spread contamination to healthy birds due to salmonella present in the environment.

Studies conducted in 1980 on salmonella prevalence found that poultry eggs without proper cooking methods were a major risk factor for the outbreak of *S. enteritis* in the United States. From then the National Salmonella Surveillance system (CDC) was developed in order to collect data of outbreaks from all locations and the results from 1985 to 1995 showed that incidence rate of *S. enteritis* increased from 2.38 to 3.9 per 100,000 population while with a decline of 49% it came down to 1.98 per 100,000 in 1999. The reason for this decline

in infection and outbreaks could not be proved. It was thought that the implementation of prevention and control measures played a major role during the 1990s. These control measures mainly dealt with safe handling methods including proper cooking of eggs, regulations regarding refrigeration, quality assurance programs, educational messages, on farm testing and traceability.

One of the major contributing factor for the spread of salmonella is when animals are given feed contaminated with salmonella and it has been found that in the region where endemic infection is well controlled or absent, Salmonella contaminated feed is a major source for introducing Salmonella in animal food production. Recent studies to investigate the prevalence of Salmonella during milling process found that dust and feed ingredients can be a major source of Salmonella contamination.

Samples from 3 feed mills which were individually producing 100,000 to 400,000 tons of feed every year and the temperature of each sample was recorded. The results showed significantly higher *Enterobacteriaceae* counts in those feed samples which were also positive for Salmonella as compared to the feed samples which were not contaminated with Salmonella. The data showed that maintaining high temperature during pellet making was not sufficient to eliminate Salmonella and the distribution of contamination was also uneven. But 85^{o}C temperature was required during pelleting to eliminate Salmonella completely.

2.2.6: DISPOSAL OF ENDOCRINE DISRUPTORS IN THE ENVIRONMENTS

Other wastes excreted by poultry chickens that are present in the poultry litter are Estrogenic compounds commonly known as Endocrine Disruptors or Endocrine disrupting compounds (EDG), which are defined as compounds which affect the endocrine system. According to the world Health Organization (WHO), an endocrine disruptor is an exogenous substance or mixture that alters function(s) in an intact organism or its progeny, or populations.

However the Environmental Protection Agency of the United States (USEPA) which defines an endocrine disrupting compounds as agents which interfere with the synthesis, secretion, transport, binding action or elimination of natural hormones in the body that are responsible for the maintanance of homeostasis reproduction, development or behaviour.

Endocrine disrupting compounds are chemicals whose structure are very diverse which contain one or more aromatic rings and some are chlorinated. However all these compounds share common mechanisms and biological effects such as, mimicking or antagonizing the effects of hormones, altering the pattern of synthesis and the metabolism of hormones and as well modifying hormone receptor levels.

By their interaction with hormone receptors and various processes such in the endocrine and neuronal system they interfere with the homeostasis of the body.

These endocrine Disrupting compounds might also alter hormone biosynthesis, hormone storage, hormone transport and clearance, hormone receptor recognition or binding, post receptor activation or induce oxidative stress. As a consequence these compounds have the potential to exert detrimental effects on man, plants, animals and eventually whole ecosystems.

Most of the chemicals with endocrine activity described so for are estrogenic and are of three classes namely, Estriol, Estradiol and Estrone and hence are referred to as Estrogenic Disruptors compounds (Mbuthia *et al.,* 2014). Estrogens are key regulators of physiological changes associated with reproduction in both sexes and also regulate important physiological processes including immune function and mineral homeostasis. A wide range of Estrogenic disrupting compounds (EDCs) alter the function(s) of the endocrine system and cause adverse health effects in an intact organism or its progeny. EDCs may act at several levels and the best studied actions are those in which compounds bind to estrogen receptors (ERs) and minic or block normal estrogenic actions.

Estrogenic EDCs are structurally diverse compounds from multiple sources that have estrogenic or anti – estrogenic activities although they may also affect other endocrine systems. Sources of estrogenic EDCs include natural estrogens produced by plants (phytoestrogens), fungi (mycoestrogens) and cyanobacteria,

synthetic therapeutic drugs and numerous synthetic compounds mainly used in industry and agriculture eg. Polychlorinated biphenyls (PCBs), organochlorine pesticides (OCPs). Many EDCs are of anthropogenic origin and have been accumulating in the aquatic environment for decades and their lipophilic and persistent nature means that they bioaccumulate or biomagnify in marine organisms. Aquatic contaminants can compromise reproduction, development, immune response and other physiological processes which can ultimately affect the survival of fish.

In addition to the direct impact of aquatic contaminants on fish population, the ecological importance of fish means that they also indirect affect the environment and when eaten by humans and wild life pose a health risk and negatively impact the economics of fisheries and aquaculture (Estêväo, D., Power, M.& Pinto, S. 2014). The estrogenicity of EDCs have mostly been evaluated in relation to their binding or activation of intracellular ERs, which regulate many of estrogens actions in target cells. The adverse effects of EDCs include induction of hepatic vitellogenin production, reduced gonadal growth, male gonad feminization, alters sex ratios.

However exposure to EDCs to tetrapods is known to affect both Osteoclasts (OSC) and Osteoblasts (OSB) which are responsible for bone formation, thus exposure to EDCs affects both OSB and OCS functions and bone characteristics. Moreover in mammals, exposure to EDCs during the perinatal period may have an impact during adult life.

Recent studies on EDCs suggest that EDCs have an impact on Skeletal development, morphology and anomalies in fish. Other impacts brought by EDCs include, missing of some features in fishes such as eyes, tails, fins, but also they cause body deformation such as deformed head, heart as well abnormalities such as abnormality in body colour (Mbuthia et al., 2014).

2.3: REMEDIATION OF CONTAMINATED SOIL USING MARSH PLANTS

This literature has been conducted a study to investigate on how the contaminants brought about by poultry litter would be eliminated from the contaminated soils along the shores of lake Victoria and this research has learned that although there are several ways which can be used to remove contaminants from contaminated areas but the most commonly used method is called phytoremediation (Seghatoles and Moosavi 2013).

The term Phytoremediation refers to a diverse collection of plant based technologies that use different plants as a containment, destruction or an extraction technique (Sakakibara,2012).

This technology has been receiving attention lately as an innovative, cost – effective to the more established treatment methods used at hazardous waste sites. Phytoremediation is an emerging technology that uses various plants to degrade, extract, contain or immobilize contaminants including metals, pesticides, hydrocarbons and chlorinated solvents from soil and water.

Plants act as solar – driven pumping and filtering systems as they take up contaminants – mainly water soluble through their roots and transport or translocate them through various plant tissues where they can be metabolized, sequestered or volatilized.

Approximately 400 plant species from at least 45 plant families have been so far reported to hyperaccumulate metals, and some of the families are Brassicaceae, Faboceae, Euphorbiaceae, Asterraceae, Lamiaceae and Scrophulariaceae.

Indian mustard (Brassica juncea L.), and Sunflower (Helianthus annus L.) have reportedly shown high uptake and tolerance to heavy metals. The roots of Indian mustard are found to be effective in the removal of Cd, Cr, Cu, Ni, Pb and Zn, while sunflower can remove Pb, U, Cs and Sr from hydroponic solutions. The success of Phytoremediation depends mainly on the choice of plant which must obviously posses the ability to accumulate large amounts of heavy metals (hyperaccumulation).

Hyperaccumulators such as Thlaspicaerulescens or Alyssum bertolonii, producing a relatively low amount of above ground biomass but accumulating high amounts of one or more elements. The specific plant and wild species that are used in this technique are effective at accumulating increasing amounts of toxic heavy metals and they are known as accumulators.

These plants accumulate heavy metals at high concentrations above ground than do non – hyperaccumulators growing in the same conditions without showing any observable symptoms in their tissues.

Phytoremediation can be classified into different applications such as phytofiltration or rhizofiltration, phytostabilization, phytodegradation and phytoextraction.

2.3.1: PHYTOEXTRACTION

Phytoextraction is a technology that uses plants to absorb metals from soil and translocate them to the harvestable shoots where they are accumulated. The roots and shoots are subsequently harvested to remove the contaminants from the soil (Lasat,2000). Jiang et al.(2004) found that Elsholtziasplendes performed better in the remediation of contaminants in which the exchangeable form of Cd was partly removed by the plant uptake that accompanied with the intake of nutrition while the exchangeable form of Cd decreased in the planted soil.

2.3.2: PHYTOSTABILIZATION

Phytostabilization referred to as in – place inactivation and is primarily used for the remediation of soil, sediment and sludges. This technology uses plant roots to limit contaminant mobility and bioavailability in the soil. In phytostabilization, plants are responsible for reducing the percolation of water within the soil matrix, which may create a hazardous leachate inhibiting direct

contact with polluted soil by acting as barrier and interfering with soil erosion, which results in the spread of toxic metals to other sites. Phytostabilization is a suitable technique to remediate Cd, Cu, As, Zn and Cr (Bolan et al.,2011).

2.3.3: RHIZOFILTRATION

Rhizofiltration is primarily used to remediate extracted ground water, surface water and waste water with low contaminant concentrations (Abdullahi,2015). Rhizofiltration involves the use of plants to clean various aquatic environments, which can be used to remove Pb, Cd, Cu, Ni, Zn and Cr, which are primarily retained within the roots. Sunflower, Indian Mustard, tobacco, spinach and Corn have been shown ability to remove lead from water with sunflower having the greatest ability.

2.3.4: PHYTODEGRADATION

Phytodegradation is the use of plants and microorganisms to uptake, metabolize and degrade the organic contaminant. In phytodegradation, plant roots are used in association with microorganisms to detoxify soil contaminated with organic compounds. It is also known as phytotransformation and it remediates organic compounds including herbicides, insecticides, Chlorinated solvents and inorganic contaminants (Newman,2004).

Phytodegradation is the breakdown of organic contaminants within plant tissue in which plants produce enzymes such as dehalogenase and oxygenase that help catalyze degradation. In this technology, both plants and the associated

microbial communities play a significant role in remediating contaminants (Nagan,2011).

2.3.5: PHYTOVOLATILIZATION

Phytovolatilization is the use of green plants to extract volatile contaminants such as Hg and Se from polluted soils and to ascend them into the air from their foliage. Phytovolatilization involves the use of plants to take up contaminants from the soil transforming them into volatile forms and transpiring them into the atmosphere (Sakakibara,2010).

2.4 .RELEVANT MEANS TO REMEDIATE SOILS CONTAMINATED WITH POULTRY LITTER

This literature have examined the impacts of poultry litter on the environment and applied the technology called Phytoremediation on contaminated soils polluted with Poultry litter using macrophytes plants called *Phragmites karka*.

This technology, Phytoremediation has been utilized based on the fact that it is cost effective and environment friendly which utilizes plants in order to clean up the environment contaminated with N, P and As of poultry litter .

This literature provides an option way of cleaning the environment especially on shores of Lake Victoria where farmers have been growing various kinds of crops using poultry manure.

This study have witnessed an accumulation of large amounts of poultry manures along growing sites which farmers accumulate as stocks / reserves for future use. This is a serious problem which needs a relevant measure in order to solve it, and this literature have proposed the use of *Phragmites karka* which will help the environment retain its status. In this Literature marsh plants called *Phragmites karka* were grown in the Laboratory Setting and supplied with poultry litter in order to assess their efficiency in waste removal. In this study, two equal sized – 1m x 2m x 1.5m beds (b_1 and b_2) were constructed in the Laboratory field and cutted pieces of *Phragmites karka* were planted in the two beds by using native soils filled at a depth of 0.5m. In bed_1 *P. karka* plants were grown closely packed while in bed_2 *P. karka* were few and more spaced.

The two beds were periodic supplied with tap water as these macrophytes survives in aquatic environments and after three months of period from the time of plantation, 500g of poultry manures were added in the two beds and then contaminated soil from different points together with the added manures about 5g was collected from each bed in order to determine initial levels of N, P and As . Also samples of leaves measured at about 2g was collected from the grown *P.karka* in order to determine the initial levels of N and P.

After a period of two weeks, soil samples as well samples of leaves of *P. karka* were collected in the similar way as in the previous and then the levels of N, P and As were determined. Soil samples from contaminated farm with poultry manures as well from uncontaminated farm were also analyzed for the levels of

N, P and As. Also samples of leaves of local spinach grown in a contaminated soils with poultry manures as well those grown in uncontaminated soils with poultry manures were analyzed in order to investigate the levels of N, P and As in those local spinaches grown in the two areas.

For soil samples, 1g of soil for each sample was used for analysis while 0.2g of plant tissues (leaves) were used for analysis of contaminants (N, P and As). During sample preparation, the p^H of the soil was measured and it was found that the pH of the soil was pH > 7 for various contaminated soil samples and the pHfor uncontaminated soil samples was about 5.8 ie pH< 7.

The method used for the detection of contaminants levels when pH> 7 was Olsen Sodium bicarbonate method while when pH< 7, the method employed was Bray method.

It has to be noted that the difference in the number of Macrophytes (*P. karka*) in which bed_1 occupied with more closely plants while bed_2 occupied with more spaced plants was relevant in this literature in order to reveal which criteria is more convenient for much uptake of contaminants that can be applied on the ground in affected areas.

The basic principle upon which this vascular plant works is that it takes up the contaminant as a nutrient element from the soil and bioaccumulate it into its tissues

which are stems and leaves (Phytoextraction) and then the nutrient elements are assimilated through degradation by conversion into other useful forms for the utilization of the body by a process described as assimilation and it is achieved by enzymes such as Nitrosedictase, Lactase, dehalogenase and Nitrilase, and this process is called Phytodegradation.

As this plant takes up the contaminants as nutrients and bioaccumulate them into its body the contaminants levels will be decreasing progressively.

Another mechanism on this principle is that once the nutrient has been loaded into its body some may get lost in volatilized form by which the plant convert a contaminant of solid or liquid origin into a gaseous form a process called volatilization. Since this process is carried out by a plant it is described as Phytovolatilization.

It is to be noted that a factor like rainfall may disturb and may also affect the relevance of data collected since in this progress the volume of water was maintained at 10 litre and was replaced upon dryness for wetness occurred in which tap water was used throughout the progress during which the program was conducted from September to

November 2014 as a growing season before data collection and after this period it followed a period of data gathering procedures from December 2014 to May 2015 for a period of Six months.

A factor of rainfall was overcomed by constructing a roofing which comprised transparent sheet material which allows a regulated amount of sunlight to pass through it and hence prevents the entrance of rain water to the plants and this roofing material was supported by four wooden sheets on the four edges and the height of the roof was 4.5m high which was able to accommodate the height *Phragmites karka* throughout growing seasons.

After sample collection procedure, the next procedure that followed was sample analysis which involved sample digestion using a Kjedahl digestion instrument followed by calorimetric titration which involved various relevant reagents for the analysis of Nitrogen (N). Either for analysis of Phosphorus involved digestion procedure followed by formation of blue Phosphomolybdenum complex.

Analysis for Arsenic (As) involved digestion produce followed by determining the concentration of As on the Atomic Absorption Spectrophotometer with vapour Generation Accessory (AAS – VGA).

In this literature, the results were as follows:-

- During the first four months from December 2014 to March 2015, the uptake of contaminants were high and slowed during the period from April to May and this is due to the fact that plants uptake of nutrients is high during younger period than during old period.

- There were a differences in the uptake of contaminants (N, P and As) between bed_1 and bed_2 and the results recorded have shown that, uptake of N, P and As in bed_1 was significant higher than in bed_2,forinstance in December 2014 the results show that in bed_1 Nitrogen(N) in the soil decreased from 3.7% to 2.4% showing a decrease of 1.3% for N and Phosphorus (P) in the soil decreased from 2.5% to 1.3% showing a decrease of P in the soil by 1.2%. But in bed_2 a decrease of N ranged from 3.7% to 2.6% showing a difference of 1.1% while a decrease of P in bed_2 ranged from 2.5% to 1.8% showing a difference of 0.7% for P. Hence plants uptake of contaminants were higher in bed_1 than in bed_2 since bed_1 occupied with much closely plants while bed_2 occupied with few plants.

- Contamination by Arsenic were not so alarming in poultry litter investigated, and only few samples showed the prevalence of Arsenic in a very minimal amounts for instance in those samples that indicated the prevalence of Arsenic showed Arsenic levels of 0.32 ppm/0.00032%, 0.25ppm/0.000025%.

  Most samples of poultry litter showed no prevalence of Arsenic ie<0.001ppm.

  In some incidence, the results showed that the levels of Arsenic (As) after treatment became <0.001ppm while the initial levels of Arsenic (As) was 0.1ppm. This incidence is due to the process called Phytovolatilization which involves a conversion of a contaminant when in a solid form or liquid form into a gaseous form by a macrophyte plant and due to the absence of Arsenic (As) after treatment,

this suggests that the plants used *Phragmites karka* concentrated these contaminants (As) into their bodies then converted them into a volatile form.

- Also this literature have shown that the levels of Nitrogen (N) and Phosphorus (P) in the leaves of *P,karka* grown in the laboratory field were higher after contamination with poultry litter.

Furthermore, the results revealed that the levels of Nitrogen (N) and Phosphorus (P) in contaminated soil farms were higher while in uncontaminated soil farms the levels of N and P were low. But on both farms-contaminated soil farms and uncontaminated soil farms ,there were no contamination of Arsenic (As). Hence there were no significance difference in the levels of Arsenic on the two farms.

2.5: DEFINING KEY WORDS

In this literature, the key words used are as list below:-

1. Phytoremediation – The use of plants to eliminate pollutants.
2. *Phragmites karka* /common reed – A macrophyte plant that grows in a wet/aquatic environment.
3. Bioremediation – The use of living organism such as plants, animals or microorganisms to eliminate pollutants

4. Bioventing – The use of a combination of oxygen, nutrients and microbes to degrade pollutants

5. Biosparging – The use of a combination of microbes and oxygen to degrade pollutants.

6. Bioagumentation – The use of a microbes having specific metabolic capability to degrade pollutants.

7. Macrophyte – A vascular plant with extensive below ground biomass and above ground biomass

8. Cyanobacteria – The blue green algae which secretes harmful chemicals in the environment.

9. Litter – A waste produced by poultry chickens such as broiler chickens or layers.

10. Poultry – Any domesticated bird.

11. Methemoglobinemia – Tendency by which Iron (Fe) in hemoglobin is more susceptible to oxidation when exposed to oxidants such as nitrites.

2.6: THEORETICAL / CONCEPTUAL FRAMEWORK

2.6.1 .Challenges Facing Poultry Industry

In recent years cities in various countries have expanded due to an increase of human population which in turn has increased the demand for food worldwide,

as a result producers in various agricultural sectors have been forced to increase the quantity of various food products and Poultry industry is one among the fast growing sector worldwide due to a high demand for meat and eggs.

The main challenge facing this sector of poultry industry is the large quantities of wastes being produced by broiler chickens called poultry litter which is a mixture of fecal droppings, feathers bedding materials such as saw dust.

Due to a rapid expansion of poultry industry there is a large quantity of poultry litter being produced and only a few quantity of this poultry litter is used for agricultural activities as manure, but most of this poultry litter is unused and hence pose an environmental threat since in some cases poultry litter is carried by runoff down stream or rivers and enters ground water or surface water which causes contamination in the environments. The release of toxic contaminants to the environment has led to the scarcity of clean water, loss of soil fertility as well a loss of biodiversity. The biodiversity of plants and animal species play an important role in the development of healthy and productive ecosystems and thus play an important role of economic benefits to man and environment.

2.6.2: Means to Overcome Challenges of Poultry Industry

To overcome these drawbacks, a much better perspective is to completely destroy the pollutants or to transform them into some biodegradable substances. This approach can be achieved by using a technique known as bioremediation which acts as an option to clean and safe environment and its resources by destroying various contaminants using natural biological activity. It is

considered as safer, cleaner, cost effective and environment friendly technology which generally have a public acceptance and can often be carried out at any site.

This technology called bioremediation, is defined as the process by means of various biological agents primarily microorganisms to degrade the environmental contaminants into less toxic forms.

The first patent for a biological remediation agent was registered in 1974 using a strain of Pseudomonas putida to degrade Petroleum. In 1991, about 70 microbial genera were reported to degrade petroleum compounds.

U.S. EPA has defined bioremediation agents as microbiological cultures, enzymes and nutrient additives that significantly increase the rate of bioremediation to mitigate the effect of various pollutants.

The main advantages of bioremediation: low cost, high efficiency, minimization of chemical and biological sludge, selectivity to specific metals, no additional nutrient requirement, regeneration of biosorbent and the possibility of metal recovery.

Bioremediation can occur on its nature or can be spurred through addition of fertilizers for the enhancement of bioavailability within the medium (biostimulation), Bioventing, bioleaching, bioreactor, bioaugmentation, composting, biostimulation, land farming,Phytoremediation and rhizofiltration – are all examples of bioremediation technologies. On the basis of removal and

transportation of wastes, bioremediation technology can be classified as in situ and ex situ. In situ bioremediation involves treatment of contaminated material at the same site, while ex situ involves complete removal of contaminated material from one site and transfer it to another site, where it is treated using biological agents. Both in situ and ex situ depend essentially on microbial – metabolism, however so far in situ methods are preferred over ex situ for ecological restoration of contaminated soil, water and environment. Phytoremediation is an emerging technology that uses various plants to degrade, extract, contain or immobilize contaminants from soil and water. This technology has been receiving attention lately as an innovative, cost-effective alternative to the more established treatment methods used at hazardous waste sites

2.6.2.1: Mechanisms of Phytoremediation

Phytoremediation is a newly evolving field of science and technology that uses plants to clean up polluted soil, ground water, and waste water. Phytoremediation uses green

plants including grasses, and woody species to remove, contain or render harmless environmental contaminants such as heavy metals, metalloids, trace elements, organic compounds, and radioactive compounds in the soil or water. This definition includes all plant – influenced biological, Chemical and Physical processes that aid in the uptake, sequestration, degradation and

metabolism of contaminants either by plants, soil microbes or plant and microbial interactions.

Phytoremediation takes advantage of the unique and selective uptake capabilities of plant roots systems, together with the translocation, bioaccumulation, and contaminant storage[degradation abilities of the entire plant body. Plant – based soil remediation systems can be viewed as biological treatment systems with extensive, self – extending uptake network that enhances the below – ground ecosystem for subsequent productive use. Phytoremediation avoids excavation and transport of polluted media thus reducing the risk of spreading the contamination and has the potential to treat sites polluted with more than one type of pollutant.

Some drawbacks associated with phytoremediation are:- dependency on the growing conditions required by the plant (ie, climate, geology, altitude and temperature), large scale operations requires access to agricultural equipment and knowledge, tolerance of the plant to the pollutant affect the success for remediation, contaminants collected in senescing tissues may be released back into the environment in certain seasons, time taken to remediate sites far exceeds that of the other technologies and contaminant

solubility may be increased leading to greater environmental damage and the possibility of leaching.

The mechanisms and efficiency of phytoremediation depend on the type of contaminant, bioavailability and soil properties. There are several ways by which plants clean up or remediate contaminated sites. The uptake of contaminants in plants occurs through the root system in which the principal mechanisms for preventing toxicity are found.

The root system provides an enormous surface area that absorbs and accumulates water and nutrients essential for growth along with other non – essential contaminants.

Phytoremediation involves the following mechanisms-

Phytoextraction. This is also called Phytoaccumulation and it refers to the uptake of contaminants in the soil by plant roots into the above ground portions of the plants.

Phytoextraction is primarily used for the treatment of contaminated soils. This technique uses plants to absorb, concentrate and precipitate toxic materials from contaminated soils. There are several advantages of Phytoextraction for instance it is fairly low cost, the contaminant is permanently removed from the soil, the amount of waste material is substantially decreased during remediation, the contaminant can be recycled from the contaminated plant biomass.

Phytoextraction is a technology that uses plants to absorb metals from soil and translocate them to the harvestable shoots where they are accumulated. The

roots and shoots are subsequently harvested to remove the contaminants from the soil.

Studied conducted by Jiang et al.(2004) found that Elsholtziasplendes performed better in the remediation of contaminants in which the exchangeable form of Cd was partly removed by the plant uptake that accompanied with the intake of nutrition while the exchangeable form of Cd decreased in the planted soil.

However there are several factors limiting the extent of contaminant Phytoextraction including:-

- Contaminant bioavalability within the rhizosphere.
- Rate of contaminant uptake by roots
- Rate of xylem loading / translocation to shoots
- Cellular tolerance to toxic material

Phytostabilization referred to as in – place inactivation and is primarily used for the remediation of soil, sediment and sludges. This technology uses plant roots to limit contaminant mobility and bioavailability in the soil. In phytostabilization, plants are responsible for reducing the percolation of water within the soil matrix, which may create a hazardous leachate inhibiting direct contact with polluted soil by acting as barrier and interfering with soil erosion, which results in the spread of toxic metals to other sites. Phytostabilization is a

suitable technique to remediate Cd, Cu, As, Zn and Cr. Rhizofiltration is primarily used to remediate extracted ground water, surface water and waste water with low contaminant concentrations.

Rhizofiltration involves the use of plants to clean various aquatic environments, which can be used to remove Pb, Cd, Cu, Ni, Zn and Cr, which are primarily retained within the roots. Sunflower, Indian Mustard, tobacco, spinach and Corn have been shown ability to remove lead from water with sunflower having the greatest ability.

Phytodegradation is the use of plants and microorganisms to uptake, metabolize and degrade the organic contaminant. In phytodegradation, plant roots are used in association with microorganisms to detoxify soil contaminated with organic compounds.

It is also known as phytotransformation and it remediates organic compounds including herbicides, insecticides, Chlorinated solvents and inorganic contaminants. Phytodegradation is the breakdown of organic contaminants within plant tissue in which plants produce enzymes such as dehalogenase and oxygenase that help catalyze degradation. In this technology, both plants and the associated microbial communities play a significant role in remediating contaminants.

Phytovolatilization is the use of green plants to extract volatile contaminants such as Hg and Se from polluted soils and to ascend them into the air from their

foliage. Phytovolatilization involves the use of plants to take up contaminants from the soil transforming them into volatile forms and transpiring them into the atmosphere.

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2.6.3: Adaptation of Macrophytes (P.karka) to their functions

Macrophytes(P.Karka) have several intrinsic properties that make them more adaptable to their role in contaminants removal. The most important functions of the machrophytes in relation to the treatment of waste water are the physical effects brought about by the presence of the plants. These macrophytes stabilize the surface of the soil, provide good conditions for physical filtration, prevent vertical flow systems from clogging, insulate against frost during winter, and provide a huge surface area for attached microbial growth. Macrophytes mediate transfer of oxygen to the rhizosphere by leakage from roots increases aerobic degradation of organic matter and nitrification.

In well drained soil, the pore spaces are filled with a relatively high content of oxygen. Microorganisms living in the soil and roots of plants growing in the soil therefore are able to obtain oxygen directly from their surroundings.

Macrophytes are morphologically adapted to growing in water – saturated sediment by virtue of large internal air spaces for transportation of oxygen to roots and rhizomes. The internal oxygen movement down the plant serves not only the respiratory demands of the tissues, but also supplies the rhizosphere with oxygen by leakage from the roots. This oxygen leakage from roots creates oxidized condition and stimulates both aerobic decomposition of organic matter and growth of nitrifying bacteria.

Generally, the roles of macrophytes can be divided into the following categories:-

(a) Physical

Macrophytes stabilize the surface of the plant beds, provide good conditions for physical filtration, and provide a huge surface area for attached microbial growth.

(b) Soil hydraulic conductivity

Soil hydraulic conductivity is improved in an emergent macrophyte plant system. Turnover of root mass creates macropores in the bed soil system allowing for greater percolation of water thus increasing effluent/ plant intaractions.

(c) Organic compound release

Emergent macrophytes have been shown to release a wide variety of organic compounds through their root systems, at a rate up to 25% of the total Photosynthetically fixed carbon. This carbon release may act as a source of food for deniitrifying microbes. Decomposing plant biomass also provides a durable, readily available carbon source for the microbial population.

(d) Microbial growth

Macrophytes have above and below ground biomass to provide a large surface area for growth of microbial biofilms. These biofilms are responsible for a majority of the microbial processes in a bed system including Nitrogen and Phosphorus removal.

Plants create and maintain the littler / hums layer that may be likned to a thin biofilm. As plants grow and die, leaves and stems falling to the surface water in a bed creates multiple layers of organic debris (the litter / humus component), and this accumulation of partially decomposed biomass creates highly porous substrate layers that provide a substantial amount of attachment surface for microbial organisms.

(e) Creation of aerobic soils

Macrophytes mediate transfer of oxygen through the hollow plant tissue and leakage from root systems to the rhizospere where aerobic degradation of organic matter and nitrification will take place.

Macrophytes have adaptations in the lignified layers in the hypodermis and outer cortex to minimize the rate of oxygen leakage. The high Nitrogen and Phosphorus removal of *Phragmites karka Phragmites karka* (common reed) is most likely attributed to the characteristics of its root growth. Phragmites allocates 50% of plant biomass to root and rhizome systems. Increased root biomass allows for greater oxygen, transport into the substrate creating a more aerobic environment favouring nitrification reactions.

Nitrification requires a minimum of 2mg O_2/L to proceed at a maximum rate. These marsh plants /Macrophytes preferred because they have a rapid and relatively constant growth rate in a tropical system, these kinds of plants have a higher growth rate and are easily propagated by means of runners and by bits of

mats breaking off and drifting to new areas.The principal Pollutant removal by macrophytes include biological processes such as microbial metabolic activity and plant uptake as well as physico – chemical processes, adsorption and filtration.

Microbial degradation plays a dominant role in the removal of solute / colloidal biodegradable organic matter in the contaminated areas. Biodegradation occurs when dissolved organic matter is carried into the biofilms that attached on plant stems, root systems and surrounding soil by diffusion process.

2.6.4: Pollutant Removal Mechanisms

A pollutant may be removed as a result of more than one process such as:

(a) Nitrogen removal mechanisms.

Nitrogen removal is through nitrification / Denitrification processes. Nitrogen exists in various forms, namely Ammoniacal Nitrogen (NH_3 and NH_4^+), organic Nitrogen and oxidized Nitrogen (NO_2 and NO_3^-). The removal occurs through either plant up take or denitrification,volatilization of ammonia(NH_3),storage in detritus and sediment,uptake by macrophytes and storage in plant biomass. A majority of Nitrogen removal occurs through either plant uptake or denitrification process. Nitrogen uptake is significant if plants are harvested and biomass is removed from the system.

At the root- soil interface, atmospheric oxygen diffuses into the rhizosphere through the leaves, stems, rhizomes and roots of the macrophytes, thus creating an aerobic layer in the bed where the macrophyte have been grown.

Nitrogen transformation takes place in the oxidized and reduced layers while Ammonification takes place where organic N is mineralized to NH_4^+-N in both oxidized and reduced layers.

The oxidized layer and the submerged portions of plants are important sites for nitrification in which Ammoniacal Nitrogen (AN) is converted to nitrite N (NO_2 – N) by Nitrosomonas bacteria which is either taken up by the plants or diffuses into the reduced zone where it is converted to N_2 and N_2O by the denitrification process. Denitrification is the permanent removal of Nitrogen from the system however the process is limited by the number of factors such as temperature, pH; redox potential, carbon availability and nitrate availability. The extent of Nitrogen removal depends on the amounts of Nitrogen in the polluted site. If the nitrogen content is low, the macrophytes will completely directly with nitrifying and denitrifying bacteria for NH_4^+ and NO^-_3 removal, while in high Nitrogen content, particularly Ammonia, this will stimulates nitrifying and denitrifying activity.

(b) Phosphorus removal mechanisms

Phosphorus is present in wastewaters as orthophosphate, dehydrated orthophosphate (polyphosphate) and organic Phosphorus. The conversion of

most of Phosphorus to the orthophosphate forms ($H_2PO_4^{=}$, $PO_4^{=}$, $PO_4^{3=}$) is caused by biological oxidation.

Most of the phosphorus component may fix within the soil media where phosphate removal is achieved by Physicochemical processes, by adsorption, complexation and precipitation reactions involving calcium (Ca), Iron (Fe) and Aluminium (Al). However the removal of Phosphorous is more dependent on biomass uptake by the macrophyte / marsh plant system with subsequent harvesting.

NB. Nitrogen uptake by the macrophyte is taken up in a mineralized state and incorporated it into plant biomass. Accumulated Nitrogen is released into the system during a die-back period. Plant uptake is not a measure of net removal and this is because dead plant biomass will decompose to detritus and litter in the life cycle and some of this Nitrogen will leach and be released into the sediment.

Recent studies show that only 26 – 55% of annual N and P uptake is retained in above – ground tissue while the balance is lost to leaching and litter fall.

(c) Metals removal mechanism

Metals such zinc, copper, Arsenic, cobalt etc are removed from the system through processes such adsorption, precipitation, complexation, sedimentation, erosion and diffusion through Physical – Chemical pathway.

Metals are also removed by direct uptake by macrophytes.

(d) Pathogens removal mechanism

Pathogens such as bacteria are removed mainly by sedimentation, filtration and adsorption by biomass and by natural die-off and predation.

(e) Other pollutant removal mechanisms- Evapotranspiration is one of the mechanisms for pollutant removal.

Atmospheric water losses that occur from a contaminated water and soil is termed as evaporation and from emergent portion of plants is termed as transpiration.

The combination of both process is called evapotranspiration. Daily transpiration is positively related to mineral adsorption and daily transpiration could be as an index of the water purification capability of plants.

NB. Nitrogen removal from contaminated soil / water involves - three pathways

Mineralization

Mineralization is the biological transformation of organically combined nitrogen to ammonium nitrogen during organic matter degradation. And this can be both aerobic and anaerobic process and is often referred to as ammonification. Mineralization of organically combined nitrogen releases

inorganic nitrogen as nitrate, nitrites, ammonia and ammonium making it available for plants, fungi and bacteria.

Nitrification

Nitrification is the biological conversion of organic and inorganic compounds from a reduced state to a more oxidized state and it involves a conversion of ammonia (NH_3) into Nitrate (NO^-_3)

It involves the following stages:-

1. Conversion of NH_3 into NH_4^+

This happens when ammonia (NH_3) combines with water.

$NH_3 + H_2O \longrightarrow NH_4^+ + OH^-$

Upon formation of NH_4^+ it can be absorbed by plants and algae and converted back into organic matter or the ammonium ion can be electrostatically held on negatively charged surfaces of soil particles.

This NH_4^+ ion under aerobic conditions reacts with oxygen to form nitrite ($NO_2^=$) and then nitrite ($NO_2^=$) is converted into nitrate (NO_3^-).

$2NH_4^+ + 3O_2 \longrightarrow 4H^+ + 2H_2O + 2NO_2^=$

Then: $2NO_2^= + 0_2 \longrightarrow 2NO_3^=$

There are two bacteria which facilitates this conversion. The first is called Nitrosomonas sp. Which oxidizes ammonium to Nitrite andNitrobactersp.Oxidizes nitrite (NO_2^-) to nitrate ($NO_3^=$).

Dinitrification

This is the biological conversion of Nitrate (NO_3-) to Nitrogen (N) and this process is facilitated by a kind of bacteria called achrobacter and bacillus.

This process is sometimes called Volatilization.

The general equation for such conversion is:-

$$NO_3^- \longrightarrow 2NO^- \longrightarrow NO \longrightarrow N_2O \longrightarrow N_2$$

NB. It has to be noted that phosphorus removal in the bed is done by two means.

The first means is by Chemical effect in which the macrophyte aerate the contaminated environment providing aerobic environments to the soil particles. This aerobic condition enhances a process called precipitation where oxides of Ca, Al and Fe under aerobic conditions combines with phosphates of the contaminated soil particles to form, Al – Phosphate, Ca – phosphate and Fe – phosphate, these phosphates are then precipitated onto the sediments. Phosphorus can also be removed by macrophytes through their roots which uptake the contaminants (adsorption) and concentrate them into their bodies and since these macrophytes have extensive growth in the below ground (roots)

and above ground (stems and leaves) thus facilitate in huge accumulation of biomass in their tissues which makes these macrophytes to be more preferable for contaminants removal.

CHAPTER THREE

METHODOLOGY

3.1. STUDY AREA

This research was conducted in Mwanza Region, Tanzania at Nyasaka Islamic High School where *phragmites karka* plant species were grown and through this laboratory working site, samples of soils contaminated with poultry litter as well leaves were collected from the two beds where *Phragmites karka* were grown as seen in Figure 3.1.

Figure 2: Showing *Phragmites karka* Grown in Two Beds

It is to be noted that a factor of rainfall which may also affect the relevance of data collected since in this progress the volume of water was maintained at 10 litre and was replaced upon dryness for wetness occurred in which tap water was used throughout the progress during which the program was conducted from September to November 2014 as a growing season before data collection and after this period it followed a period of data gathering procedures from December 2014 to May 2015 for a period of Six months. A factor of rainfall was overcame by constructing a roofing which comprised transparent sheet material which allows a regulated amount of sunlight to pass through it and hence prevents the entrance of rain water to the plants and this roofing material was supported by four wooden sheets on the four edges and the height of the roof was 4.5m high which was able to accommodate the height *Phragmites karka* throughout growing seasons.

Samples of soils contaminated with poultry litter was also collected from the shores of lake Victoria in order to analyze the levels of contamination in farming areas where local farmers have been engaging in agricultural activities using poultry litter in order to increase output of their crops . Figure 3.2 shows a location of agricultural activities along the shore of lake Victoria.

3.2. THE DESIGN OF THE RESEARCH

The design of the research involved the following procedures:-

i. Along the shores of lake Victoria, samples of soils were collected from a farm which uses poultry manures and another farm on which crops were grown without the use of poultry manures.

ii. In the laboratory field, two beds b_1 and b_2 each of size of 1mx2mx1.5m were constructed and whose roof on four poles was made of transparent sheet placed at a height of 0.5m above ground. This transparent roof material used in order to prevent entrance of rainwater in the two beds (b_1 and b_2).

In the two beds b_1 and b_2, native soils was added and cutted pieces of *Phragmites karka* were grown in the two beds and supplied with tap water. Upon three months of growth of *Phragmites karka* plants from September to November 2014, 500g of poultry litter was added in the two beds (b_1 and b_2) and this kind of Poultry litter was collected from various Poultry chickens growers.

3.3. SAMPLING PROCEDURES

Samples of soils as well leaves of *P.karka* were collected by using a sampling procedure called random sampling.

In this sampling design, every item of the universe has an equal chance of inclusion in the sample.

Random sampling ensures the law of statistical regularity which states that if on an average the sample chosen is random one, the sample will have the same

composition and characteristics as the universe (Kothari, 2004). In brief, the implications of random sampling are:-

- It gives each element in the population an equal probability of getting into the sample and all choices are independent of one another.
- It gives each possible sample combination an equal probability of being chosen.

3.3.1 Sampling procedures for soil samples from contaminated and uncontaminated soil farms.

Soil samples from contaminated soil and uncontaminated soil farm was collected from ten (10) different points and put in plastic bags which were free from contamination.

3.3.2 Sampling procedure for samples of soil from the laboratory setting

Soil samples from five (5) different points was collected from each bed (b_1 and b_2),and then composed to form a single unit and put in plastic bags free from contaminants.

3.3.3 Sampling procedures for samples of leaves of P. karka from laboratory field.

Five (5) g of leaves of *P.karka* was collected from each bed (b_1 and b_2) for analysis and were put in plastic bags free from contaminants.

3.4. SAMPLE ANALYSIS FOR SOIL SAMPLES

3.4.1. SAMPLE ANALYSIS FOR NITROGEN (N) IN SOIL SAMPLES

After samples of soil have been collected from cropland areas as well from the Laboratory field, then 1g of soil sample was first air dried and once the sample dried, the next step involved dilution of the sample and the pH of the soil was measured.

The pH of the soil form the Laboratory field contaminated with poultry litter was greater than 7 ie pH > 7 and the method applied when pH greater than seven was Olsen method, while the pH of the soil in the cropland area was also greater than 7 ie pH> 7. But the pH from uncontaminated crop land area was less than 7 ie pH< 7, the method for soil samples whose pH < 7 was Bray Method.

After pH measurement of soil samples the next step involves a process called Digestion where by a soil sample in a hard glass tube was placed in a Kjedahl digestion apparatus. In the digestion apparatus, 1ml of 0.5M concentrated Sulphuric acid was added in the tube containing an analyte then followed by heating the mixture forming ammonium sulphate, $(NH_4^+)_2\ SO_4$ as analyte of interest.

Figure 3.3: Kjedahl Digestion Apparatus

The next step after digestion, was Kjedahl distillation, where the analyte of interest was transferred into a Kjedahl distillation apparatus followed by addition of sodium Hydroxide (NaOH) where the analyte $(NH_4)_2SO_4$ reacts with NaOH forming gaseous ammonia (NH_3) but due to the presence of a condenser Unit within the apparatus, gaseous ammonia (NH_3) was condensed into a liquid ammonia. In the Kjedahl distillation Apparatus, a weak acid called Boric acid H_3BO_3 was added which reacted with liquid ammonium to form ammonium Borate as seen in figure 3.4.

Figure 3.4 Kjedahl Distillation Apparatus

Then this weak salt- ammonium borate is titrated with a standard solution of sulphuric acid using a mixture of indicators of methyl red and Bromothymol blue and at equivalent point, the resultant solution was green in which the concentration of Nitrogen in mg/kg is determined using a calorimetric curve as a Kjedahl – N. The following is a summary of equations showing reactions involved during reactions.

$$NH_4^+ + H_2SO_4 \longrightarrow (NH^+_4)_2SO_4 + 2H^+$$

$$(NH^+_4)_2SO_4 + 2NaOH \longrightarrow 2NH_3 + 2H_20 + Na_2SO_4$$

$$H_3BO_3 + 3\,NH_3 \longrightarrow BH_{12}N_3O_3$$

3.4.2. SAMPLE ANALYSIS FOR PHOSPHORUS (P) IN SOIL SAMPLES

After sample collection, 1g of soil sample was air dried followed by measurement of soil p^H when the pH> 7, Olsen sodium bicarbonate method was used and when the pH < 7, Bray method was used.

After measurement of soil pH, the step that followed was a digestion of soil sample by a wet acidic digestion procedure in which a dilute orthophosphate solution and ammonium molybdate reacts under acid condition to form a heteropoly acid, molybdophosphoric acid in the presence of vanadium to form a yellow vanadomolybdophosphoric acid. The concentration of P is measured as the blue or yellow colour and determined spectrophotometrically using a spectrophotometer measured in mg/kg.

3.5. SAMPLE ANALYSIS FOR LEAF SAMPLES

Analysis procedures for leaf samples started with drying of samples to an oven overnight at 70^oC. The next step that followed involved grinding of leaf samples to powdered form.

3.5.1. SAMPLE ANALYSIS FOR NITROGEN

The powdered form of leaf sample was digested followed with Kjedahl distillation, and then last with titration procedure as in the as indicated by the following equations:-

$$NH_4^+ + H_2SO_4 \longrightarrow (NH^+_4)_2SO_4 + 2H^+$$

$$(NH^+_4)_2SO_4 + 2NaOH \longrightarrow 2NH_3 + 2H_20 + Na_2SO_4$$

$H_3BO_3 + 3\ NH_3 \longrightarrow BH_{12}N_3O_3$

3.5.2. SAMPLE ANALYSIS FOR PHOSPHORUS

After sample collection, 2g of soil sample was air dried followed by measurement of soil pH when the pH> 7, Olsen sodium bicarbonate method was used and when the pH< 7, Bray method was used. After measurement of soil pH, the step that followed was a digestion of soil sample by a wet acidic digestion procedure in which a dilute orthophosphate solution and ammonium molybdate reacts under acid condition to form a heteropoly acid, molybdophosphoric acid in the presence of vanadium to form a yellow vanadomolybdophosphoric acid. The concentration of P is measured as the blue or yellow colour and determined spectrophotometrically using a spectrophotometer measured in mg/kg.

3.6. SAMPLE ANALYSIS FOR ARSENIC IN SOIL AND LEAF SAMPLES

After sample collection and sample digestion, then Arsenic (As) was analysed by using Atomic Absorption Spectrophotometer with Vapour Generation Accessory (AAS – VGA). AAS – VGA) for analysis of total Arsenic (AS (III) + As (V) is very crucial and it requires a reduction of As (V) for correct analysis. As is reduced to AsH_3vapours and finally for free As atoms which are responsible for absorption signal in AAS.

To accomplish this, vapour generation assembly attached to AAS has acid channel filled with 10M HCl and the reduction channel with sodium borohydrid and the concentration of Arsenic was given in mg/kg.

CHAPTER FOUR

RESULTS AND DISCUSSIONS

4.1 To Determine the Levels of N, P and As in Soil Contaminated With poultry litter

Results have shown a decrease in the levels of N, As and P for soil samples but however there was a little decrease in the removal efficiency by Phragmites karka in march 2015 due to plant maturity as seen in table 4 .1 for values of N and P. The results in table 4.1 has shown a decrease in the levels of both Phosphorus and Nitrogen in a horizontal basis: In February 2015, the levels of P before remediation was 2.2 % and after was 1.7% , giving a decrease of 0.5%. While for N, the levels before remediation was 2.8% and after was 2.0% giving a decrease of 0.8%

4.1.1 The Levels of Phosphorus (P) in the Soil Before and After Remediation

Results in figure 4.1 have shown that the amounts of pollution by phosphorus before remediation using Phragmites karka was higher than after remediation indicating the efficiency of this kind of plant in removing contaminants. However the amount of phosphorus removed from march was significant low and this is because this kind of phytoremediator have a life time of one year and during this time it had a life time of seven months which caused a decrease of its removal efficiency due to maturity.

Table 4.1: The levels of N and P in the contaminated soil before and after remediation in bed 1 by P.karka.

		Levels of contaminants in the soil sample			
		Before treatment		After treatment	
Month	Sample	Available phosphorus (P) (%)	Total Nitrogen (N) (%)	Available phosphorus (P) (%)	Total Nitrogen (N) (%)
Dec 2014	1	2.5	3.7	1.3	2.4
	2	2.3	3.5	1.5	2.6
Jan 2015	1	1.8	2.4	1.2	1.8
	2	1.5	2.0	1.0	1.6
Feb 2015	1	2.2	2.8	1.7	2.0
	2	2.5	3.2	2.42	3.15
March 2015	1	1.4	2.6	1.34	2.52
	2	1.6	2.9	1.55	2.86
April 2015	1	3.1	3.5	3.07	3.44
	2	2.9	3.2	2.85	3.16
MAY 2015	1	2.5	3.8	2.46	3.78
	2	2.2	3.6	2.17	3.56

During a young life time from December to January there was a significant high removal efficient of contaminants since during this period many physiological processes are needed by a plant such as root developments as well development of a bud and stem growth and these results are similar to Jia et al.(2016) who revealed that Rye plants played a significant role in the removal of Zinc, Lead and Organic compounds for instance,removal rates for Zinc in some instance was 29.5%. However results were presented using a line graph which showed that there was an increase in the rates of removal of contaminants, which meant a decrease in contaminant levels from contaminated soil, and this efficiency shown by Rye plant is a similar result from the efficiency shown by Phragmites karkon from the observation

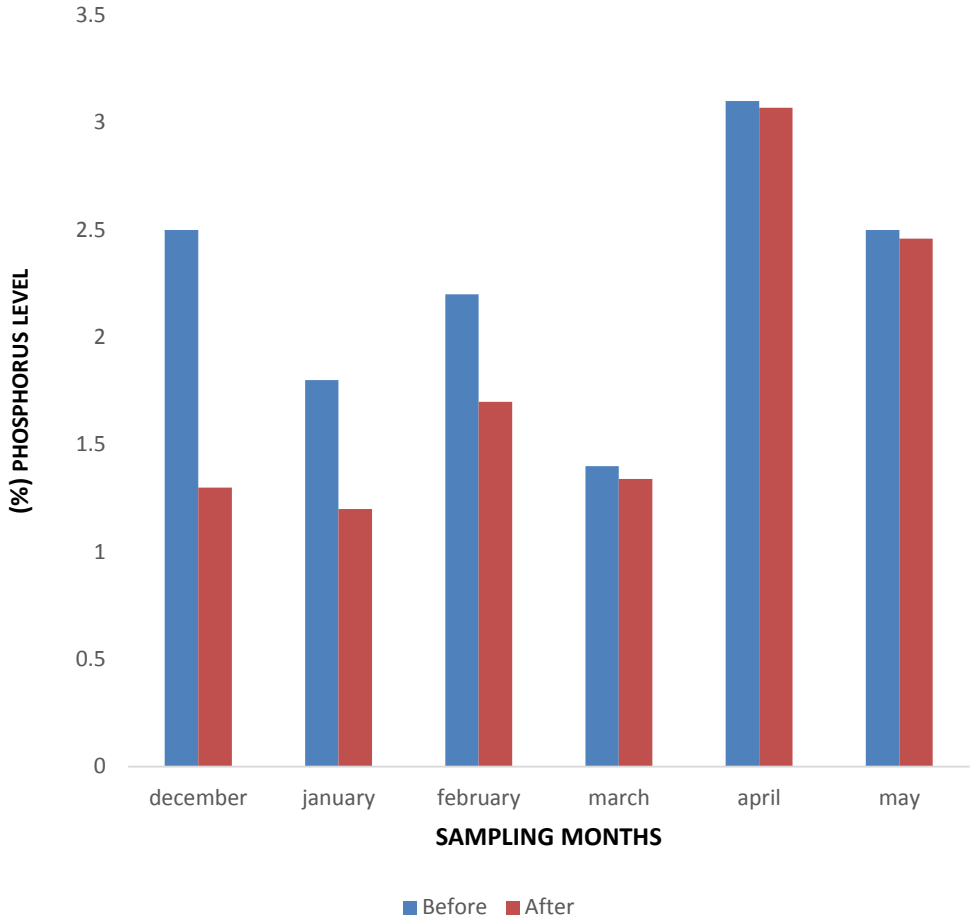

Figure 4.1: Phosphorous Level in the Contaminated Soil before and After Remediation

4.1.2 The Levels of N in Soil Samples Before and After remediation

Results shows that there were a decrease in the levels of Nitrogen which means that the levels of Nitrogen before remediation was high while after remediation was low indicating a decrease in the concentrations of Nitrogen.

In December2015, First Week: The levels of N before remediation was 3.7% and after remediation was 2.4% showing a decrease of 1.3% as seen in table 4.1.

Results also shows that there were a linear decrease of Nitrogen when verifying the results using a linear graph produced a linear graph as seen in figure 4.2

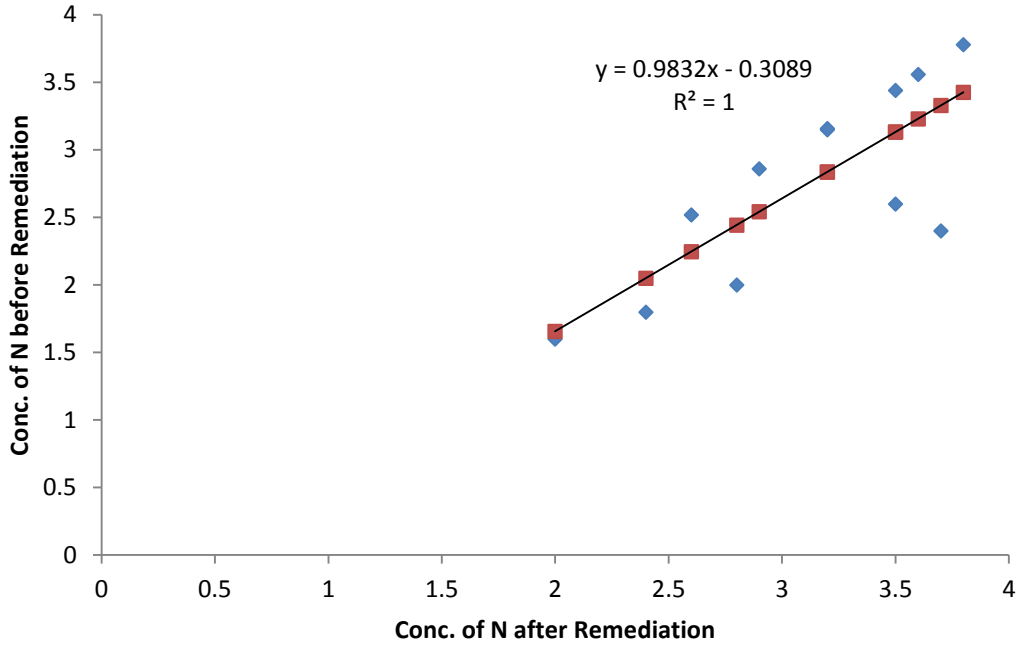

Figure 4.5 : Graph showing Nitrogen Linear Regression

Results from figure 4.2 shows that there were a linear decrease for values of Nitrogen when values of N before remediation were plotted against those values after remediation.

4.1.3 The Levels of Arsenic in Soil Samples Before and After Remediation

Table 4.2: The Levels of Arsenic (As) In a Contaminated Soil by Poultry Litter in a Laboratory Setting Before and After Treatment by *P. karka*

SAMPLE	TOTAL As BEFORE TREATMENT (ppm)	TOTAL As AFTER TREATMENT (ppm)
1	0.32	0.21
2	0.25	0.16
3	<0.001	<0.001
4	<0.001	<0.001
5	0.10	<0.001
6	<0.001	<0.001
7	<0.001	<0.001
8	<0.001	<0.001
9	<0.001	<0.001
10	<0.001	<0.001

Table 4.2 shows a decrease in the levels of Arsenic after remediation in a laboratory setting where *Phragmites karkon* plant species were grown: In sample 01, the levels of Arsenic before remediation was 0.32 X 10^{-5} % and after remediation was 0.21 X 10^{-5} % indicating a decrease of 0.11 X 10^{-5} %. But some samples had small values of Total Arsenic as seen in sample 5 in which Total Arsenic before remediation was 0.1 X 10^{-5} % and after remediation was 0.001 X 10^{-5} % which is beyond detection limit of Atomic Absorption Spectrophotometer.

Discussions by using Hypotheses Testing

Hypothesis simply means a mere assumption or some supposition to be proved or disproved. For a researcher, hypothesis is a formal question that he intends to resolve. Thus hypothesis may be defined as a proposition or a set of proposition set forth as an explanation for the occurrence of some specified group of phenomena either asserted merely as a provisional conjecture to guide some investigation or accepted as highly probable in the light of established facts.There are two types of Hypotheses which are Null Hypotheses H_O and Alternative Hypotheses H_a.The Null Hypotheses Ho is usually the one which one wishes to prove and the Alternative Hypotheses Ha is the one which some one wishes to disprove. In this discussion using Hypotheses testing. A T- test was used for testing the relevance of data results at a level of significance of 5%.In case 1, discussions were focused on the understanding of the impacts of *P. karka* on the contaminated soil by Poultry litter and asses the efficiency of this plant species on the remediation process in order to clean the environments.

I. The levels of Phosphorus (P)

This part tried to evaluate whether there was a decrease or not in the levels of Phosphorus in the contaminated soil when *P. karka* were used in the Laboratory Setting. This involved the use of Hypotheses as follows:-

Null Hypothesis (Ho): There was no significant decrease in the levels of P after treatment in contaminated soil in bed $_1$.**Alternative Hypothesis (Ha**): The level of Phosphorus in contaminated soil decreased significantly in bed 1 after treatment.

Table 4.3: Testing Hypotheses-1

Sample	P after treatment Xi	P before treatment Yi	Difference (Di = Xi – Yi)	Difference squired Di^2
1	1.3	2.5	-1.2	1.44
2	1.5	2.3	-0.8	0.64
3	1.2	1.8	-0.6	0.36
4	1.0	1.5	-0.5	0.25
5	1.7	2.2	-0.5	0.25
6	2.42	2.5	-0.08	0.0064
7	1.34	1.4	-0.06	0.0036
8	1.55	1.6	-0.05	0.0025
9	3.07	3.1	-0.03	0.0009
10	2.85	2.9	-0.05	0.0025
11	2.46	2.5	-0.04	0.0016
12	2.17	2.2	-0.03	0.0009
N=12			$\Sigma Di = -3.94$	$\Sigma Di^2 = 2.9584$

Calculated t value = -1.828

Critical level: $t_{(n-1),\ \alpha} = t_{(12-1),0.05}$

$t_{(11)}, 0.05 = 1.796 = -1.796$ on one tailed test=left

Note: When the calculated value is higher than the tabulated value (critical value) then the null hypothesis (H_o) is rejected but when the critical value is less than the calculated value then the null Hypothesis is accepted.The results show that the

calculated value is -1.828 which is higher than the tabulated value then the null hypothesis (H_o) is Rejected and the alternative hypothesis is accepted. This means that there was a significant decrease in the levels of Phosphorus after treatments in bed_1 using *Phragmites karka*. And hence *P. karka* proved to be a relevant macrophyte for contaminants removal in soils contaminated with poultry litter.

II. The levels of Nitrogen

This part tried to evaluate whether there was a decrease or not in the levels of Nitrogen in the contaminated soil when *P. karka* were used in the Laboratory Setting.This involved the use of Hypotheses as follows:-Null Hypothesis (Ho): The levels of Nitrogen in contaminated soil decreased significantly in bed $_1$ after treatment.

Alternative Hypothesis (Ha): There was no significant decrease in the levels of Nitrogen after treatment in contaminated soil in bed.

Table 4.4: Testing Hypotheses -2

Sample	Levels of N after treatment Xi	Levels of N before treatment Yi	Difference (Di = Xi – Yi)	Difference squired Di^2
1	2.4	3.7	-1.3	1.69
2	2.6	3.5	-0.9	0.81
3	1.8	2.4	-0.6	0.36
4	1.6	2.0	-0.4	0.16
5	2.0	2.8	-0.8	0.64
6	3.15	3.2	-0.05	0.0025
7	2.52	2.6	-0.08	0.0064
8	2.86	2.9	-0.04	0.0016
9	3.44	3.5	-0.06	0.006
10	3.16	3.2	-0.04	0.0016
11	3.78	3.8	-0.02	0.0004
12	3.56	3.6	-0.04	0.0016
N=12			$\Sigma Di = -4.33$	$\Sigma Di^2 = 3.6801$

From the table above, by applying the relevant formula for Mean of Difference and Standard deviation of difference at 5%

The calculated t value was -1.7165

The critical t value was -1.796

Since the observation value / calculated value lies in the Acceptance region and it is less than the Critical value then the null hypothesis (Ho) is Accepted this means that, the levels of Nitrogen (N) decreased significantly in bed $_1$ after treatment with *P. karka* .

III. The levels of Arsenic (As)

HYPOTHESES TESTING

Ho: There was a significant decrease in the levels of Arsenic after remediation

Ha: There was no significant decrease in the levels of Arsenic after remediation .Level of significance was 5% At a level of significant of 5%, observed t value was -1.7331 whereas the critical value of t was -6.314, and since the observed t value was less than the critical value hence,

These results justify that there was a significant decrease in the levels of Arsenic in soil samples after remediation.Some samples of contaminated soils in the Laboratory setting have shown Prevalence of Total Arsenic in a very low concentrations while most samples had no Arsenic contaminations, hence this

might be a justifications that there is a possibility that these local growers of Poultry chicken do not use growing bousters rich in Arsenic .Generally speaking, the levels of Arsenic after remediation was very low and in some cases indicated below detection limit < 0.001

4.2. To determine the Efficiency of Phragmites karka Species

Table 4.5: The levels of N and P in the leaf samples of *P. karka* in bed1

Month	Sample	Levels of contaminants in the leaf samples			
		Initial levels		Final levels	
		Available (P) (%)	Total (N) (%)	Available (P) (%)	Total (N) (%)
	1	0.2	1.6	0.8	2.3
Dec 2014	2	0.8	2.3	1.4	2.8
	1	1.4	2.8	1.9	3.3
Jan 2015	2	1.9	3.3	2.2	3.5
	1	2.2	3.5	2.5	3.8
Feb 2015	2	2.5	3.8	2.54	3.83
March	1	2.54	3.83	2.58	3.87
2015	2	2.58	3.87	2.61	4.00
	1	2.61	4.00	2.63	4.04
April 2015	2	2.63	4.04	2.65	4.06
	1	2.65	4.06	2.66	4.07
May2015	2	2.66	4.07	2.68	4.09

Results in table 4.5 shows an increase of the levels of P and N : In Jan 2015 , the levels of P in leaf samples changed from 1.9% to 2.2% showing an increase of 0.3%. The levels of N in leaf samples changed from 3.3% to 3.5% showing an increase of 0.3%.

But there were a small change in the remove of N and P from Feb 2015,which shows the levels of P in leaf samples changed from 2.58% to 2.61% showing an increase of 0.04% while the levels of N changed from 3.87% to 4.00% showing an increase of 0.03%. This small change in the remove of N and P is mainly due to plant maturity which makes it decrease in consumption of nutrients

4.2.1 The Levels of P in Leaf Samples of Phragmites karka in Laboratory Setting.

Results in **figure 4.3** have shown that there was an increase of the levels of Phosphorus (P) in leaf samples of *Phragmites karkon* with an increase of time of exposure to contaminants and this result is analogous to a study by Woranan et al (2016) : A Case Study of *Gynura Pseudochina (L)* DC. on "Heavy Metals" which have indicated that after the leaves of the plant species were dried and analyzed for Zinc and Cadmium, results indicated an increased in the concentrations of both Zinc and Cadmium.

Results have shown a decrease in the bioaccumulation of both Phosphorus and with an increasing time of maturity of the Phragmites karkon due to decrease of physiological processes as a plant gets matured.

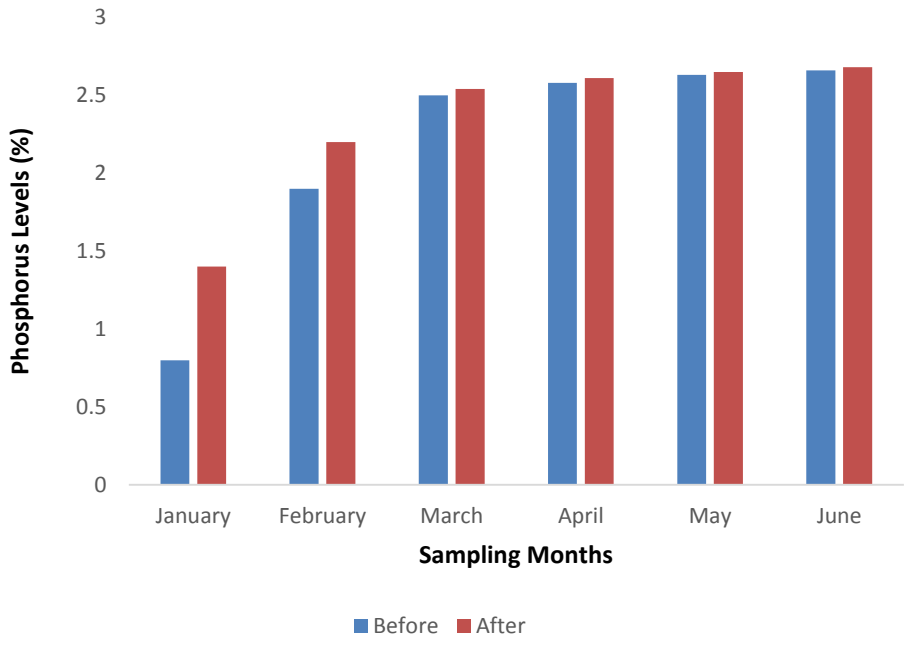

Figure 4.6: Phosphorous Level in the Leaf Sample of *P. karka* Before and after Remediation

4.2.2 The Levels of N in Leaf Samples of *Phragmites karka* Before and After Remediation in Laboratory Setting.

Results in figure 4.4 sh**ows** an increase in the levels of Nitrogen (N) for samples of leaves of *Phragmites karkon* indicating that *Phragmites karkon* have an ability to remove soil Nitrogen from a contaminated soil with poultry litter

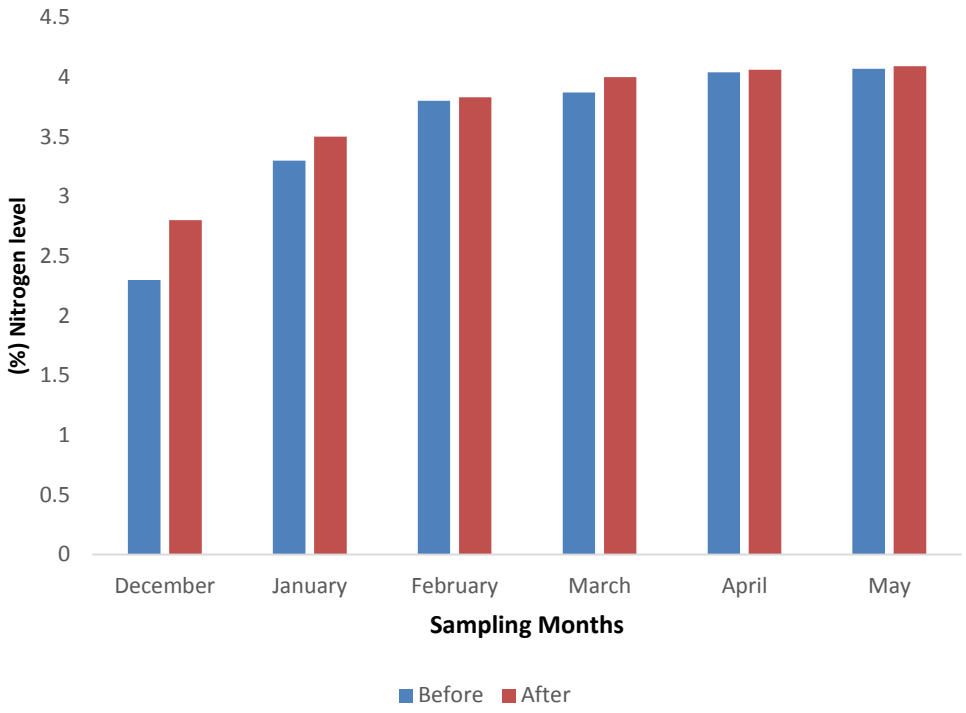

Figure 4.7: Nitrogen Level in the Leaf Sample of *P. karka* Before and after Remediation

Discussions by using Linear Regression

This method was used in order to justify if there was a correlation between the increase in the concentrations of contaminants in leaf samples of *P. karka* and the quantity of manures added in a prolonged period of time.This part tried to assess the impacts of *P. karka* on the contaminated soil through reviewing the weight build up in the Leaf samples of *P. karka* .In this study. *P. karka* were considered to be relevant plants for Bioremediation when a plot of graph gave a linear shape and hence justified that there were an increase of contaminants in the leaves of *P. karka* .

Table 4.6: Regression Observation of Phosphorus

Observation	*Predicted Y*	*Residuals*
1	2.186374194	-0.886374194
2	1.976289032	-0.476289032
3	1.451076129	-0.251076129
4	1.135948387	-0.135948387
5	1.871246452	-0.171246452
6	2.186374194	0.233625806
7	1.030905806	0.309094194
8	1.240990968	0.309009032
9	2.816629677	0.253370323
10	2.606544516	0.243455484
11	2.186374194	0.273625806

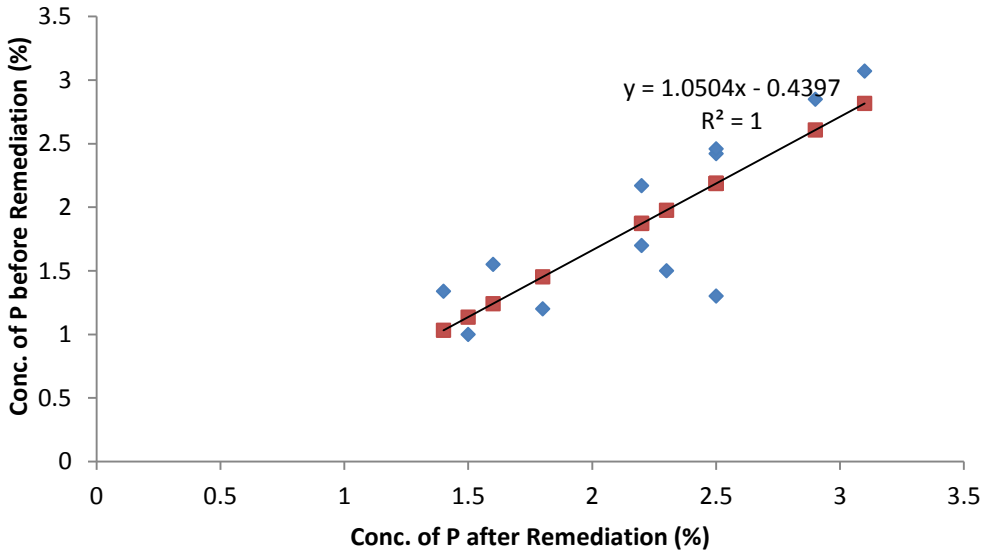

Figure 4.8: A Graph Showing Phosphorus Remediation Linear Regression

Results from figure 4.5 shows that, a linear graph which has been formed suggests that there were a linear correlation between the amounts of manures added in the beds and the decrease in the concentrations of the contaminants, which shows a uniform decrease in concentrations of contaminants for every quantity of manures added. This suggests a significant removal or decrease by certain amounts of the contaminant. Secondly, a deviations from a linear pattern of some points for a bluish dots suggests that there were some random variations among some samples in their concentrations of contaminants in which some samples did not differs more in their concentrations while others differs more in their concentrations.

Table 4.7: Table showing Nitrogen Linear Regression

Observation	Predicted Y	Residuals
1	3.32911	-0.92911
2	3.132462	-0.53246
3	2.0509	-0.2509
4	1.657604	-0.0576
5	2.444195	-0.4442
6	2.837491	0.312509
7	2.247547	0.272453
8	2.542519	0.317481
9	3.132462	0.307538
10	2.837491	0.322509
11	3.427434	0.352566

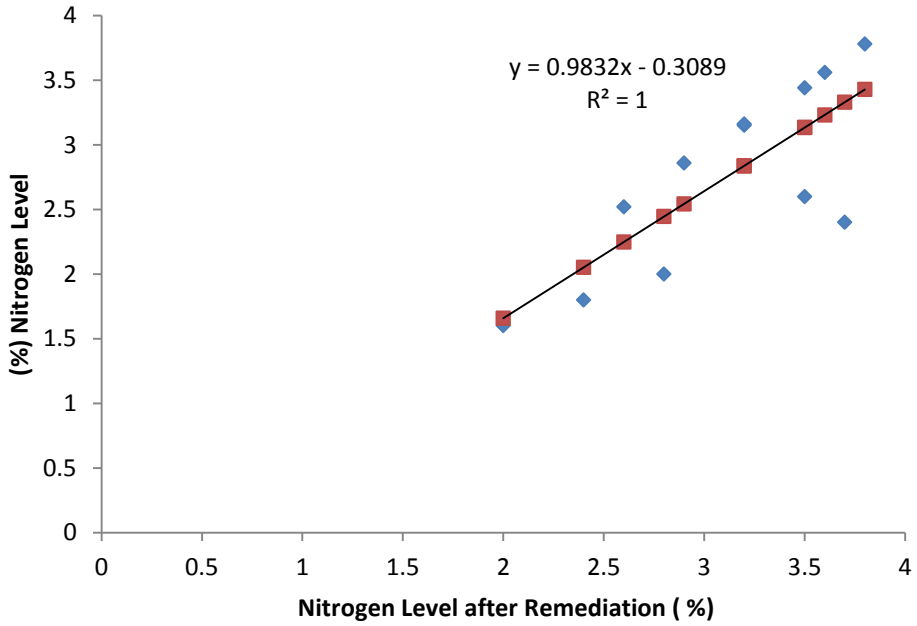

Figure 4.9: A graph showing Nitrogen Linear Regression

Results in figure 4.6 shows that, a linear graph which has been formed suggests that there were a linear correlation between the amounts of manures added in the beds and the decrease in the concentrations of the contaminants, which means that there were a uniform decrease for every quantity of manures added and this shows a significant removal or decrease by certain amounts of the contaminant. Secondly, a deviations from a linear pattern of some points for a bluish dots suggests that there were some random variations among some samples in their concentrations of contaminants in which some samples did not differs more in their concentrations while others differs more in their concentrations.

Poultry litter and asses the efficiency of this plant species on the remediation process in order to clean the environments. In this case, an increase in the biomass

of *Phragmites karka* was considered to be a measure of the efficiency of wastes removal and this involved the following methods;

4.3. The Levels of N, P or As in Contaminated and Uncontaminated Soil Areas along Lake Victoria Shores

This part presents results of farms whose soil has been contaminated with poultry litter and a farm whose soil has not been contaminated with poultry litter. Results showed high amounts of Nitrogen and Phosphorus but with no Arsenic contamination in a farm whose soil has been contaminated with poultry litter.

4.3.1: Contaminated Soil Farm with Poultry Litter

4.3.1.1. The Amounts of P in Contaminated Soil Farm

Results in table 4.8 showed that in a contaminated soil farm with poultry litter there was a high amounts of P. Sample 01 of a contaminated soil farm in table 4.8 showed that the amounts of P was 0.50% which is beyond standard levels of 0 to 0.0025% low, 0.0025 to 0.005% medium and + 0.005% as high (Jove, 2018).

Table 4.8. Showing Amounts P in Contaminated Soil Farm with Poultry Litter

Sample	Total N (%)
O1	0.5
02	1.8
03	1.3
04	1.7
05	0.5
06	1.9
07	1.4
08	1.2
09	0.8
10	1.6

4.3.1.2: The Amounts of N in Contaminated Soil Farm

The amounts of N in table 4.9 of a contaminated soil farm with poultry litter had high amounts of N. Sample 02 of table 4.9, showed amounts of N of 2.7% which is beyond standard levels: 0 to 0.00 15% low, 0.0015 to 0.003% medium and + 0.003% high (Jove**,**2018**).**

Table 4.9: Showing Amounts of N in Contaminated Soil Farm

Sample	Available P (%)
01	2.3
02	2.7
03	3.0
04	2.8
05	2.6
06	3.4
07	2.1
08	2.3
09	2.7
10	2.0

4.3.1.3. The Amounts of Arsenic in Contaminated Soil Farm

Results showed that there were no Arsenic contamination in a contaminated soil farm as seen in table 4.9.1 in which all values were beyond detection limit of $<1x10^{-7}$ % equivalent to < 0.001mg/kg

Table 10: Showing Amounts of Arsenic in Contaminated Soil Farm

Sample	Total Arsenic (%)
01	$< 1x10^{-7}$
02	$< 1x10^{-7}$
03	$< 1x10^{-7}$
04	$< 1x10^{-7}$
05	$< 1x10^{-7}$
06	$< 1x10^{-7}$
07	$< 1x10^{-7}$
08	$< 1x10^{-7}$
09	$< 1x10^{-7}$
10	$< 1x10^{-7}$

4.3.2: Uncontaminated Soil Farm with Poultry Litter

4.3.2.1: The Amounts of P in Uncontaminated Soil Farm

The amounts of Phosphorus in uncontaminated soil farm in table 4.9.2 was low and did not exceed the standard levels known: Sample 08 of table 4.3.4 shows that the amounts of P was 0.0046% which lies within the range of standard limits of: 0 to 0.0025% low, 0.0025% to 0.005% medium and + 0.005% high (Jove, 2018).

Table 4.11: Showing Amounts of P in Uncontaminated Soil Farm

Sample	Available P (%)
01	0.0043
02	0.0036
03	0.0040
04	0,0048
05	0.0035
06	0.0037
07	0.0034
08	0.0046
09	0.0042
10	0.0039

4.3.2.2 : The Amounts N in Uncontaminated Soil Farm

The amounts of Total N in uncontaminated soil farm in table 4.9.3 was low and did not exceed the standard levels known: Sample 04 of table 4.3,5 shows that the amounts of N was 0.0023% which lies within the range of standard limits of: 0 to 0.0015% low, 0.0015% to 0.003% medium and + 0.003% high (Jove, 2018).

Table 4.12: Showing Amounts of N in Uncontaminated Soil Farm

Sample	Total N (%)
01	0.0016
02	0.0022
03	0.0020
04	0.0023
05	0.0021
06	0.0015
07	0.0019
08	0.0017
09	0.0024
10	0.0018

4.3.2.3: The Amounts of Total Arsenic in Uncontaminated Soil Farm

The amounts of Total Arsenic was beyond detection limit by a Spectrophotometer which means that there was no Arsenic contamination in soils which was not contaminated with poultry litter as seen in results recorded in table 4.9.4 in which all the values shows a value of $< 1x10^{-7}$ % equivalent to < 0.001mg/kg.

Table 4.13: Showing Total Arsenic in Uncontaminated Soil Farm

Sample	Total As in %
01	$< 1x10^{-7}$
02	$< 1x10^{-7}$

03	$<1x10^{-7}$
04	$<1x10^{-7}$
05	$<1x10^{-7}$
06	$<1x10^{-7}$
07	$<1x10^{-7}$
08	$<1x10^{-7}$
09	$<1x10^{-7}$
10	$<1x10^{-7}$

4.4: Percent Removal Efficiency of *Phragmites karka* in Bed_1 and Bed_2

Results have shown that the removal efficiency of Bed 1 was significant higher than for Bed 2 as seen in table 4.9.5: In Dec 2014, initial levels of P in the leaves of *Phragmites karkon* in Bed 1was 0.2% while final levels of P was 0.8 % and this indicates an increase of 0.6 % (part per hundred unit) and in percentage the value is 60 percent. But the initial levels of P in Bed 2 was 0.1% and the final levels was 0.5%, this indicates an increase of 0.4% which is equivalent to 40 percent, This indicates a difference of 20 percent high in Bed 1. The initial levels of N for Bed 1 in Dec 2014 indicates a value of 1.6 % (part per hundred) while the final value was 2.3% givingj a difference of 0.7% which is equivalent to 70 percent but the initial levels of N in Bed 2 was 1.2% while the final level was 1.7% giving a difference of 0.5 % which is equivalent to 50 percent. This indicates a difference of 20 percent in high in Bed 1.

Table 4.14: Percent Increase of N and P in Leaf Samples of *Phragmites karka* for Bed$_1$ and Bed $_2$

		Initial levels in Bed 1		Final levels in Bed 1		Percent increase in Bed 1		Initial levels in Bed 2		Final levels in Bed 2		Percent increase in Bed 2	
Month	Sample	P (%)	N (%)	P (%)	N (%)	P (%)	N (%)	P (%)	N (%)	P (%)	N (%)	P (%)	N (%)
Dec 2014	1	0.2	1.6	0.8	2.3	0.6	0.7	0.1	1.2	0.5	1.7	0.4	0.5
	2	0.8	2.3	1.4	2.8	0.6	0.5	0.5	1.7	0.9	2.0	0.4	0.3
Jan 2015	1	1.4	2.8	1.9	3.3	0.5	0.5	0.9	2.0	1.2	2.4	0.3	0.4
	2	1.9	3.3	2.2	3.5	03	03	1.2	2.4	1.4	2.5	0.2	0.1
Feb 2015	1	2.2	3.5	2.5	3.8	0.3	0.3	1.4	2.5	1.5	2.7	0.1	0.2
	2	2.5	3.8	2.54	3.83	0.04	0.04	1.5	2.7	1.52	2.72	0.02	0.02
March	1	2.54	3.83	2.58	3.87	0.04	0.04	1.52	2.72	1.54	2.73	0.02	0.01
2015	2	2.58	3.87	2.61	4.00	0.03	0.13	1.54	2.73	1.55	2.75	0.01	0.02
April 2015	1	2.61	4.00	2.63	4.04	0.02	0.04	1.55	2.75	1.554	2.78	0.004	0.03
	2	2.63	4.04	2.65	4.06	0.02	0.02	1.554	2.78	1.556	2.79	0.002	0.01
May 2015	1	2.65	4.06	2.66	4.07	0.01	0.01	1.556	2.79	1.561	2.796	0.005	0.006
	2	2.66	4.07	2.68	4.09	0.02	0.02	1.561	2.796	1.568	2.801	0.007	0.005

These results have shown that the removal efficiency of *Phragmites karkon* plant species is higher when the plant species are grown in abundant quantities and this is analogous to findings by Mojiri et al.(2013) who found that Typha domingensis plant species were able to remove heavy metals from a contaminated area in Urban waste leachate and the contaminants levels decreased with an increase in number of plant species and with an increase of time.

CHAPTER FIVE

CONCLUSION AND RECOMENDATIONS

Macrophytes (*Phragmites karka)* is so relevant for the remediation of contaminated soils polluted with poultry litter. This study have shown their high efficiency in the removal of contaminants (N, P and As) in contaminated soils caused by Poultry litter in the laboratory setting and all the results have shown that the removal efficiency by these marsh plants were significant high. Also, hence based on these findings, the community needs to be well addressed about the impacts of growing crops along the shores of the lake and also along the banks of other water sources using poultry manures as these have adverse effects to the environments as revealed in the study's experimental results as well in literature review.

Secondly, the Government needs to reinforce the relevant environmental officials to adopt the use of these Marsh plants (*Phragmites karka*) in the remediation of contaminated areas because their performance was significant high in this study.

However this literature have investigated on the actual causes of the prevalence of higher values of Nitrogen (N) and Phosphorus (P) in poultry feeds through investigating on milling machines on how the feed is prepared.

This study discovered that poultry feeds commonly used by local farmers include the following ingredients:=

- Sun flower hulls
- Sardine
- Snails shells
- Maize husks
- Rice husks

(a) Sunflower hulls

Sunflower hulls are the by product of the dehulling of sunflower seeds before they are used for oil extraction.

Sunflower seed contain about 20.30% hulls that are often removed before oil extraction due to their deterious effects on oil presses and because they reduce the quality of both oil and meal. A well manageddehulling process yields seeds with 8-12% hulls remaining on the kemels.

Hulls provide energy or other purposes such as composting, bedding material, or as a low-quality roughage for livestock. Sunflower hulls contains a large quantity of proteins and hence most broiler chickens growers prefer as relevant feeds for broiler chickens and as well for layers.

(b) Sardine

Sardine is a common name that refers to a small, oily fish within the herring family of clupeidase. Typically, sardines are caught with encircling nets

particularly purse seines, and many modifications of encircling nets are used including traps or weirs. Weirs are usually stationary enclosures composed of stakes into which sardines are diverted as they swim.

The chief use of sardines is for human consumption, but fish meal is used as animal feed, while sardine oil has many uses including manufacture of paint, varnish and linoleum.

Broiler chicken growers as well layers growers use sardines as source of proteins for their chickens.

(c) Snails shells

Snails shells are the remains of dead snails which growers of domestic broiler and layers chickens use as a component of broiler chickens and layers feeds, as a source of nutrients such as Nitrogen (N) from proteins extracted in the shells of snails.

(d) Maize and rice husks

Husks are the outer coats enclosing the seeds of grains such as maize and Rice and most growers of chickens prefer for their higher rich in energies.

In this study it has found that chickens feed is a composition of several components that have been mentioned earlier ie sunflower hulls, sardine, snails shells, husks of maize and rice seeds and this composition of several ingredients has a high amounts of Nitrogen (N) and Phosphorus (P).

Based on these findings, this research article has concluded that poultry feeds used by most local poultry chickens growers is mainly composed of substances which are common in our environments and not industrially made feed additives such as roxarsone which is most common in western countries which is used as feed additive in order to boust growth (growth bouster) but causes adverse effects to our health by causing skin and lung cancer.

Researchers on future studies might find the following:-

- As to why there were no significant differences in the contamination by Arsenic between the contaminated soil farms and uncontaminated soil farms.
- As to why the levels of Arsenic in poultry houses were very low and not alarming.
- As to why very few samples indicated the prevalence of Arsenic but most of samples indicated no prevalence of Arsenic despite the information we have from several research papers which revealed the prevalence of Arsenic in high concentrations in almost all poultry litter samples and the most common feed additives to boust growth for broiler chickens is called Roxarsone.

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