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EVALUATION AND OPTIMIZATION OF MUNICIPAL SEWAGE SLUDGE VERMICOMPOST PROCESS AS AN EFFECTIVE OPTION FOR ORGANIC WASTE RECYCLING

By

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A thesis submitted to the African Centre of Excellence for Water Management (ACEWM) in partial fulfillment of the requirement for a Master's Degree in Water Management (Water Supply& Sanitation) of Addis Ababa University

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CERTIFICATE OF APPROVAL

This is to certify that we the undersigned, have examined the Master of Science (MSc) thesis entitled "**EVALUATION AND OPTIMIZATION OF MUNICIPAL SEWAGE SLUDGE VERMICOMPOST PROCESS AS AN EFFECTIVE OPTION FOR ORGANIC WASTE RECYCLING**" and that in our opinion it is fully adequate, in scope and quality, as an MSc thesis for the Degree of Master of Science in Water Management (Water Supply and Sanitation).

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DECLARATION

I, Beauty Banda declare that this thesis is a result of my effort and hard work and to the best of my knowledge the findings of this work have never been submitted in any form for another degree, diploma, or any award to Addis Ababa University or any other University or Tertiary institution. Where assistance was sought from information taken from published and unpublished work of others, it has been accordingly acknowledged in the text and a list of references is given.

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DEDICATION

This thesis is dedicated to my parents; Mrs. Beauty Kalala Banda and Mr. Douglas Banda. To my siblings Ntangu and Mayeso Banda and lastly to my cheerleader Mr. Kasongo Kalaba. Thank you for your care, encouragement, love, and support.

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ABSTRACT

Population increases and unplanned urbanization in urban cities like Addis Ababa have led to an increase in wastewater generation and sludge production. The management of sludge is among the most costly and challenging processes of any WWTP bearing the fact that disposal options should protect public health and the environment. This study, therefore, employed an ecological cheap, and natural way of organic waste recycling for agricultural practices using the vermicomposting approach. Samples of wet and dry municipal sludges were obtained from the outlet of the USAB and sludge drying beds of Kaliti WWTP to characterize the municipal sludges. Vermicompost was produced from the dry sludge samples amended with cow dung and coffee husks to balance the carbon to nitrogen ratios of the organic waste for faster degradation. Optimization of the process parameters was carried out using Design Expert Version 10.0.7 under an RSM and Box-Behnken Design. The three parameters and their ranges namely moisture content (60-90%), turning frequency (1-3 turnings/week), and mixing ratio of substrates (50:50 to 80: 20 wt.%) of municipal sewage sludge to amendments, respectively were chosen. The experimental data on the quantity of N, P, and K were fitted into a quadratic polynomial model using multiple regression analysis. The regression coefficients of 99.91%, 89.65%, and 99.46% for NPK observed between the predicted and actual values were obvious that the developed regression models fitted the experimental data well. A regression of coefficient of $>$ 0.75 is an indication of the aptness or suitability of the model. The experimental results show that the moisture content of 71.95%, mixing ratio of sludge to amendments (72.3: 27.7% wt.), and a turning frequency of 2 turnings per week were the best operating conditions. Under these conditions, the maximum calculated yield of N, P, and K was 2.76%, 1.80%, and 1.88% respectively. A plastic pot experiment was, also, set up to evaluate the efficiency of the vermicompost to enhance plant growth using a germination index value. The study was conducted through the effect of increasing concentration of vermicompost (T_1- 0% VC, T_2 - 50% VC, and T_3 -100% VC w/w) in the target germination rate or index. Results showed that the germination index produced from T_3 was > 33 times T_2 and > 66 times T_1. It was inferred that vermicomposting of municipal sewage sludge is an effective option for recycling nutrients in organic waste.

Keywords: Optimization, Vermicomposting, Municipal sewage sludge, *Eisenia fetida*, Coffee husks, and Cow dung.

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ABBREVIATIONS

EC	Electrical conductivity
C/N	Carbon to Nitrogen ratio
MC	Moisture Content
MR	Mixing Ratios
TF	Turning Frequency
NPK	Nitrogen, Phosphorus, and Potassium
CH	Coffee Husk
MSS	Municipal Sewage Sludge
SS	Sewage sludge
TN	Total Kjeldahl Nitrogen
OM	Organic matter
APPRC	Ambo Plant Protection Research Centre
ECDSWC	Ethiopian Construction Design & Supervision Works Corporation
DoE	Design of Experiment
CW	Cow Dung
VC	Vermicompost
RSM	Response Surface Methodology
RSCL	Receiving soil contaminant limits
BBD	Box Behnken Design
SDG	Sustainable Development Goals
TOC	Total Organic Carbon
ANOVA	Analysis of Variance
WWS	Wastewater Sludge
WWTP	Wastewater Treatment Plant
BW	Biowaste
FC	Fecal Coliforms
TC	Total Coliforms

1.0 INTRODUCTION

1.1 Background

It is becoming a challenge to manage human waste in proper accordance with public health and environmental laws. The United Nations in its Sustainable Development Goals (SDG) number 6, has put a target goal to improve water quality by reducing pollution, eliminating dumping, and minimizing the release of hazardous chemicals and materials by halving the proportion of untreated wastewater by the year 2030 (Mara & Evans, 2018). While the management of human waste remains a challenge, it offers an opportunity that can benefit millions of poor farmers. Wastewater reuse and waste composting have a long tradition that can be traced back to centuries (Olufunke Cofie, 2016).

Converting human waste into a resource can contribute positively to efforts to sustainably intensify agricultural production. This is among the solutions that can be up-scaled to achieve SDG 2, which has a target to end hunger, achieve food security, promote sustainable agricultural systems, and improve nutrition by the year 2030 (Gil et al., 2019). Current estimates show that nearly a 690million people are hungry or 8.9% of the world's population goes without food each day, everywhere. In the year 2019, nearly one in ten people around the world, a figure close to, 750million people globally were exposed to severe levels of food insecurity (Egal, 2019). The Covid-19 pandemic could now double that estimate increasing the number of people affected by hunger globally.

Urbanization and rapid population growth are factors that have led to an increase in the provision of water supply which has led to an increase in wastewater generation and the production of sludge. This sludge usually requires a high cost for proper handling, disposal, and treatment (Abdel Wahaab et al., 2020). Globally the main disposal methods for sewage sludge are; landfilling, sea disposal, and incineration. Land application is the most common end use of sewage sludge method being practiced in most developing countries. The treated sludge is spread on land as a soil conditioner, organic fertilizer, or as a biofertilizer (Ødegaard et al., 2002).

Human excreta and urine are rich in nitrogen, phosphorous, and, potassium which are important nutrients needed for the growth of plants. On average 500 liters of human urine contains; 5.6 kg of nitrogen, 0.4 kg of phosphorus, and 1.0 kg of potassium. Human feces, on the other hand, contains; 0.09 kg of nitrogen, 0.19 kg of phosphorus, and 0.17 kg of potassium. Therefore, a total mass of; 5.7 kg of nitrogen, 0.7 kg of phosphorus, and 1.2 kg of potassium (NPK) can be obtained from human excreta, an equivalent mass of nutrients needed to grow 250 kgs of cereals annually (Sakthivel, 2011).

One of the biggest setbacks in the use of excreta or sludge in food production is the diverse contaminants it contains. Among them are; pathogens, trace elements, pesticides, pharmaceuticals, among others. These contaminants bring negative environmental effects to the soil once applied directly without treatment and may create odor and hygiene concerns.

One successful natural biological treatment technology that has been used for decades to recover nutrients from sewage sludge is composting. Composting is a process that is aimed at destructing pathogens, stabilizing organic matter, and producing material or compost that can be used in crop production (Kosobucki et al., 2000). Common practiced composting technologies are; aerated static heap or pile, windrow composting, in-vessel composting, and vermicomposting.

Unlike ordinary composting technologies, vermicomposting is relatively quick and produces finely divided rich stable castings or humus. Worm composting method, that utilizes worms or associated microbes to degrade organic waste and nutrients such as nitrogen, phosphorous, potassium, and calcium present in the feed material into forms that are much soluble and available to plants than those in the parent substrate (Ludibeth et al., 2012).

Composting of sewage sludge and any other organic material promotes nutrient recycling in the ecosystem and provides sustainable ways of waste management. Important parameters that affect the composting process include; pH, moisture content, the particle size of substrates, the mixing ratio of substrates, carbon-nitrogen ratio, and the nature of the bulky agent used.

Resource recovery of sludge or organic waste in Ethiopia is not carried out at a larger scale just as, is the case in many African countries. Sludge production, collection, treatment, and disposal practices are not well-known. Findings from a survey carried out in South Africa, report that only 86 of the 950 wastewater treatment plants in the country compost their municipal sludge

(Spinosa, 2015). Sludge had either accumulated on-site or disposed of or simply just airdried. In countries like Ghana, particularly in Accra wastewater reuse has been practiced over the years without much legal control, wastewater from drains and sludge has been used to grow a wide range of vegetables without proper treatment. In Burkina Faso, the reuse of wastewater has been restricted only for some selected vegetables (Nikiema et al., 2011).

Municipal sewage sludge has high moisture content and has a low carbon to nitrogen ratio, for this reason, it is usually co -composted with other organic materials. Common organic materials used for co-composting sewage sludge include; organic fractions of municipal solid wastes, sawdust, wood chips, and many other agricultural wastes (Uçaroğlu & Alkan, 2016).

1.2 Statement of the problem

In most developing countries including Ethiopia, owing to technical deficiency, limited reinforcement of environmental laws, there has been a lack of capability of handling municipal sewage sludge generated which results in serious environmental pollution. Sludge is disposed of without any treatment and just air-dried in sludge drying beds and most cases are used as soil conditioners or organic fertilizers without any treatment. In other cases, this sludge is dumped in dumpsites or river bodies where it causes eutrophication of water bodies and produces offensive odors. This inadequate treatment and disposal of sewage sludge have caused serious environmental pollution, as water bodies passing through the city of Addis such as the Akiki River have been polluted with fecal matter from such practices.

The absence of strict regulatory standards in the use of organic amendments as fertilizers or soil conditioners has led to the use of biosolids and effluents from wastewater treatment plants without any prior treatment. An example, are farmers that live close to the Kaliti WWTP. These farmers irrigate and enrich their crops with the sludge without any prior treatment (AAWSA, 2014). This puts the health of the public at risk because it causes the egression of human pathogens to the water, soil, and accumulates in the food chain, which can cause diseases (Zhou et al., 2020).

Figure 1.1: Vegetables irrigated using Sludge at Kaliti WWTP

On one hand, coffee husks are massively produced in Ethiopia and becoming difficult to manage and dispose of because they are bulky and have a low commercial value. For a single kilogram of coffee beans produced, about 1kilogram of husks are generated (Degefe et al., 2012). Coffee husk was used in the vermicomposting process to boost the composting process through facilitating aeration. Coffee husk is also known to be a good absorbent for heavy metals such as Chromium and Lead (Geleti Misganu, 2019). The coffee husk also contains proteins, sugar, and minerals which are vital for the growth of micro-organisms. Thus, it is expected that coffee husks and cow dung can boost the stabilization of the municipal sewage sludge during the aerobic vermicompost process.

Even though vermicomposting is an effective natural process for the stabilization of organic waste, it has its limiting factors. Among the limiting factors are; Temperature, moisture content, carbon to nitrogen ratio, mixing ratios, and the particle size of the feedstock. The effect of C/N, worm stocking density, and moisture content on vermicomposting has been majorly studied by several researchers however, the effect of mixing ratios of substrates particularly in the vermicomposting of sewage sludge has been fairy ignored. There are wide uncertainties regarding optimal mixtures, what to mix, and in which proportions. The optimal tuning frequency of the feedstock is another factor that has been fairly ignored and taken for granted. This study, therefore employed a pilot-scale setup to optimize vermicomposting parameters ranging from a set of values obtained from the literature. The process parameters that were optimized in this study were the moisture content, the mixing ratios, and the turning frequencies of the feedstock.

Research questions

- What are the characteristics of the sewage sludge obtained from the Kaliti Wastewater Treatment plant?
- What patterns of chemical and physical change occur during the vermicomposting process?
- What are the optimal operating conditions resulting in good quality of compost?
- How much time duration is required to mature the compost?

1.3 Objectives

Main objective

The general objective of this study was to evaluate and optimize the vermicomposting process parameters to produce high-quality compost from municipal sewage sludge through the co-composting agent of; cow dung, and coffee husk using *Eisenia fetida* earthworms.

Specific objectives

i. To characterize the physicochemical and biological properties of municipal sewage sludge obtained from the Kaliti wastewater treatment plant.

ii. To produce good quality vermicompost from the dry sludge and optimize vermicompost process parameters (*moisture content, mixing ratio, and turning frequency).*

iii. To characterize the vermicompost for its physicochemical and microbial properties in the use as an organic fertilizer.

iv. To evaluate the performance of the vermicompost as an organic fertilizer using germination index values.

1.4 Significance of the research

In most developing countries there is limited knowledge of sludge characterization, handling, and resource recovery. Sewage sludge is still seen as a waste rather than a resource. This research aimed at generating knowledge on the characteristics of the Municipal sewage sludge produced at the Kaliti Wastewater Treatment Plant and inform Policymakers on the safe potential reuse of the Municipal sewage sludge in Agricultural practices.

1.5 Limitations of the study

i. The nutrient and pathogen composition of municipal sewage sludge is highly variable from one treatment plant to another making results difficult to compare or validate.
ii. The sludge composition is dependent upon the origin of the sludge and the treatment efficiency of the treatment plant.

2.0 LITERATURE REVIEW

2.1 Sludge treatment methods

Municipal Sludge produced from secondary and primary units of a wastewater treatment often has a lot of water as a result, processes such as thickening, dewatering, conditioning, and drying are used to remove the water for easier handling and transportation. Sludge stabilization and disinfection techniques are carried out to reduce the volume of sludge, reduce odor emissions, and avoid vector attractions. Sludge stabilization techniques involve a biological process such as Anaerobic digestion, aerobic digestion, and composting. Chemical stabilization involves lime and nitrate treatments. Thermal drying and pasteurization of sewage sludge are also carried out as physical stabilization processes.

2.1.2 Anaerobic digestion

Stabilization of sewage sludge using anaerobic digestion yields biogas and biomass. Biogas is a mixture of gases usually methane, and carbon dioxide. The biogas that is produced can be used to produce electricity on the plant. The biomass is a solid-like residue that is rich in nitrate and can be used as a fertilizer. The advantage of this treatment or stabilization method is that raw sewage sludge can be used in the production of biogas and biomass. Raw sewage sludge is difficult to handle, dispose of, and utilize.

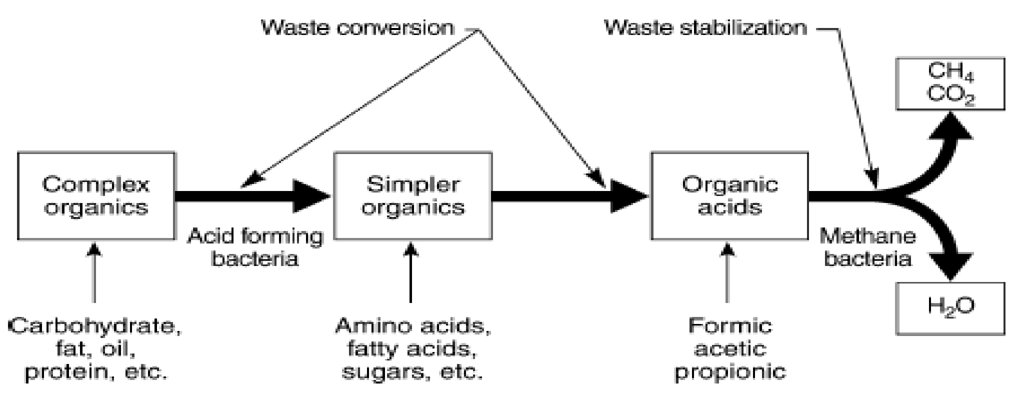


Figure 2.1: General anaerobic biological reactions adapted from (Tobergte & Curtis, 2007)

Anaerobic digestion is widely established in developed countries. Countries like Germany produce 3,517 Gwh/y of electricity from their 1,258 sewerage plants (Hanum et al., 2019). Anaerobic digestion at a large scale requires a high level of investment in large tanks and other process vessels.

2.1.3 Aerobic digestion

Aerobic digestion is a biological process of stabilizing sewage sludge from activated sludge systems, trickling filters, primary sludges, or any mixtures of the same in the presence of oxygen. It can be done in open or closed systems where oxygen is supplied by aerators or pumps. The main principle used in the aerobic digestion process is that when food has depleted the micro-organisms in the sludge metabolize their cellular mass.

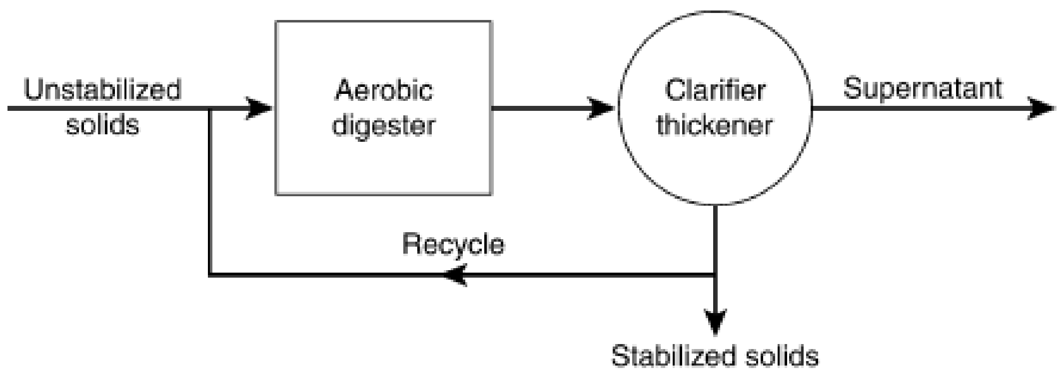


Figure 2.2: Conventional aerobic digestion process adapted from (Tobergte & Curtis, 2007)

Aerobic digestion is easier to handle compared to anaerobic digestion and provides a better reduction in volatile solids. Because of the presence of oxygen, it does not generate odor. The end product is stable humus that can be used in agriculture and land remediation practices. This digested sludge however has very poor mechanical dewatering characteristics. The main disadvantage of this stabilization process is that some contaminants such as hormones, heavy metals are not removed (Tobergte & Curtis, 2007). This method requires high energy costs for the supply of aerators and pumps.

2.1.4 Composting

The sewage sludge composting method is the most preferred method of sludge stabilization as it requires fewer investments, capital cots, and skilled labor (Kosobucki et al., 2000). Composting stabilizes the organic matter found in sewage sludge by making the nutrients found in sludge in a more soluble form that can easily be taken up by plants. The rich organic fertilizers produced from the composting process can be used in agriculture to replace chemical fertilizers. Chemical fertilizers promote the pollution of water bodies and cause acidification of soils (Savci, 2012).

2.1.5 Other stabilization methods

Other sewage sludge stabilization process involves the use of lime to adjust the pH of sludge and prevent odors.

2.2 Composting of municipal sewage sludge

Composting of sewage sludge is the most common method of treating sewage sludge carried out in most developing countries because it does not require huge investments and provides a wide range of benefits if done properly (Kosobucki et al., 2000). To increase aeration during composting of sewage sludge different bulky agents are mixed. Common bulky agents that have been employed in various studies include; sawdust, food waste, municipal solid waste, and many agricultural and food wastes (Uçaroğlu & Alkan, 2016). An analysis carried out by Yuvaraj et al., (2021), in which the centrality role of cattle solid wastes was evaluated, reports that cattle dung plays a central role in the mineralization, nutrient recovery, and microbial activity of the earthworm. In an experiment carried out by Manaf et al., (1974), to determine which bedding material either newspaper or sawdust is more suitable for vermicomposting by which biological parameters such as cocoon production, growth rate, number of worms, and worm biomass were used as indicators results reveal that different types of bedding material will influence the growth of the worm. The newspaper bedding was more influential in worm biomass production and growth rate while sawdust bedding produced better cocoons and many worms. This shows that any agricultural or domestic waste can be used as a bulking agent in vermicomposting so long it's rich in carbon to provide energy for microbes and is low in salt content and ammonia.

2.3 Methods of composting

In broad terms, there are two main types of composting systems; the OPEN WINDROW SYSTEM such as the static piles, and the CLOSED IN VESSEL SYSTEM which can be static or movable closed structures where aeration and moisture are controlled by mechanical means. The identification of the best-suited option for compositing depends on the scale of composting, input materials, investments among other reasons (Olufunke Cofie, 2016).

Table 2.1: Common composting technologies and their descriptions (Olufunke Cofie, 2016)

Composting Technology	Merit	Demerit
Static pile	• The simplest form of composting. • Requires minimum management and equipment.	• Takes a longer duration to produce the mature compost.
Trench and pit composting	• Requires low-capital investment. • Suitable for dry areas.	• Leaching control is difficult. • Labor intensive.
Aerated static pit/heap	• The land requirement is low • Allows for capturing and treating the air to reduce odor generation.	• A compost pile can dry out quickly • Requires regular monitoring. • Capital intensive investment may be required.
Windrow	• Suitable for outdoor composting. • Methane emissions are lower compared to passively aerated piles.	• Anaerobic conditions may occur resulting in odor emissions
In-vessel composting	• Less land requirement • Shorter composting period	• Very costly method
Vermicomposting	• Stable compost produced.	• Periodic monitoring

2.4 Factors affecting composting and co-composting process

They are many factors that affect the composting process such as the proportions of the mixture, the aeration rate, oxygen consumption rates, compost recycling, moisture content, pH, and carbon to nitrogen ratio (Kaur, 2018).

A study was carried out by Tena et al., (2020), on the evaluation of co-composting methods using effective microorganisms to improve the quality of co-composting of fecal sludge with organic solid waste for mobile Eco San toilets and organic solid waste in Bahir Dar city. Parameters such as pH, temperature, and moisture content were monitored. The authors found that the optimum pH for co-composting ranged from 5.40 to 7.75 and the pH value directly affects the microbial activity of the organic matter. The temperature was seen to raise during the first 15 days of co-composting and reached a maximum value of 40.25 ℃ and slowed down in the last few days of the co-composting process showing an indication of reaching the curing stage. The maximum moisture content in his study was recorded on the 3rd day of co-composting. This shows that pH, temperature, and moisture content changes during co-composting, and are important parameters to be considered during the design of the composting process.

In another research that was carried out by Lu et al., (2009), to study the characteristics of co-composting of municipal solid waste and sewage sludge in which four main factors were investigated; aeration pattern, the proportion of municipal solid wastes and sewage sludge, the aeration rate, and mature compost were investigated through changes of temperature, oxygen consumption rate, organic matter, moisture content, carbon to nitrogen ratio, nitrogen loss, sulfur, and hydrogen. Results reveal that a continuous aeration pattern was superior during composting and a mixture of 3:1 (w: w) of municipal solid waste and sewage sludge was beneficial to composting as it maintained the highest temperature for the longest duration achieved the fastest organic matter degradation and the highest nutrient in the final composting product.

Uçaroğlu & Alkan (2016), carried out a study to investigate the compostability of wastewater treatment sludge containing different bulking agents, to determine the most efficient bulk agent. In his study, compost trials of mixtures of wheat straw, plane leaf, corncob, and sunflower with wastewater sludge were performed in laboratory reactions, and in all the experiments carried there was an equal ratio of the mixture of 60 % wastewater sludge and 40 % bulk agent. Parameters such as temperature, dry matter, organic matter, pH, electrical conductivity, and carbon to nitrogen ratio were monitored. Results showed that the highest parameter of organic matter degradation, loss of dry matter, and the temperature was achieved for wastewater sludge and corncob mixtures. This shows that the selection of the best bulk agent is also an important parameter that affects the composting process.

2.5 The general concept of vermicomposting

Vermicomposting is a modern inexpensive and eco-friendly biotechnology in which earthworms are employed as natural bioreactors to decompose the organic matter. The earthworms in vermiculture are introduced to organic waste in piles of elongated rows that are covered with protective layers to prevent waterlogging (Olunfunke Cofie et al., 2016).

Vermicomposting is a non-thermophilic, bio oxidative process that uses earthworms and microbes to transform organic waste into rich humus called a Verm cast (Amare, 2015). This process is also called worm composting. It is altogether a natural system in which the earthworms play their major roles in degrading the organic portion of the waste. The use of earthworms in solid waste management is called vermicomposting.

Municipal sewage sludge vermicomposting has been successful in earlier studies, as indicated by (Sinha et al., 2010). In his study, conducted on the vermi- stabilization of sewage sludge: converting a potential biohazard into a safe biofertilizer. The study consisted of five treatments or experimental designs: Treatment 1 consisted of only sludge acting as a control, Treatment 2 consisted of sludge and earthworms, Treatment 3 consisted of the sludge, earthworms, and cow dung, Treatment 4 consisted of sludge and cow dung and lastly Treatment 5 consisted of sludge and organic garden soil. From their findings, the authors observed that the treatment with earthworms: Treatment 2 and Treatment 3 respectively, showed the most significant and rapid changes during the experimental period. This shows the significant roles, earthworms have in the degradation of organic waste. Other studies have evaluated the vermicomposting of municipal sludge and have been successful (Boruszko.,2020; Suleiman et al.,2017).

2.5.1 Brief description of the earthworm

There are many earthworm species known to man of which mostly belong to the phylum *Annelida* and class *Oligochaeta* and family *Lumbricate*, that comprises *Apporrectodea, Bimastos, Dendrobaena, Eisenia,* and many more (Águila-Juárez et al., 2011).

Not all worms are good composters or decomposers. *Epigeal* and *Anecic* species are the two known to produce humus. *Epigeal* are surface feeders while *Anecic* are burrow feeders. Earthworms are hermaphrodites meaning they consist of both male and female and are bisexual organisms. An adult earthworm consists of a reproductive structure called a clitellum, where cocoons are produced. Earthworms feed on a variety of organic matter processing them with the help of other microbe decomposers such as; fungi, nematodes, bacteria among others. An adult earthworm, which is usually 8-10 cm in length on average, can consume 25 % -35 % of its body weight each day.

Figure 2.3: *Eisenia andrei* (top) and *Eisenia fetida* (bottom) adapted from (Dominguez, 2018)

An adult earthworm processes organic matter by first sucking it to its tract, grinding it, mixing it with other body fluids modify the levels of acidity in the process, inoculating it with other micro-organisms, and lasting excreting organic matter called castings. For this reason, earthworms are known to be good bio-remediators of toxic compounds such as heavy metals (Sinha et al., 2010). The most common earthworms to be used in vermicomposting processes are the; *Dendrobaena hortensis*, *Eisenia fetida*, *Eudrilus eugeniae,* and *Perionyx excavatus* (Dominguez, 2018).

Suleiman et al.,(2017) conducted a comparative study to assess the effectiveness of vermicomposting process applied to 3 different sewage sludge using 3 different earthworm species: *Eisenia fetida*, *Eisenia andrei,* and *Dendrobaena veneta.* The authors found that *Eisenia fetida* species exhibited the highest ability to survive in harsh environments and accumulate heavy metals. Other comparative studies that have assessed the survival and adaptability of different species of earthworms are (Loehr et al., 1985).

Table 2.2: Common earthworm species used in vermicomposting process

Scientific name	***Common name***	***Application***	***Reference***
Eisenia fetida	Red wrigglers, tiger worm, or redworm	Most widely used. Determine total mass reduction and stabilization of waste	Suleiman et al, (2017).
Eudrilus eugeniae	Nightcrawlers	Second widely used for vermicomposting. native to equatorial West Africa	Boruszko, (2020).
Perionyx excavates	-	Common species in India in the Eastern Himalayas.	Amare, (2015).

2.6 Factors affecting vermicomposting

The major factors that affect vermicomposting and have been studied by researchers include; pH, temperature range, carbon to nitrogen ratio, nutrients, aeration, and moisture content. According to Sinha et al. (2010), optimum pH values of the survival of worms are in the range of 4.5 and 9, and a temperature range of 20 ℃-25 ℃ with a moisture content of 60-70 %.

As compared to other earthworm species *Eisenia fetida* has a low mortality rate since it can balance and model its energy expenditure priorities. This attribute enables it to survive in extremely toxic and environmental conditions (Sinha et al., 2010).

2.6.1 Aeration

Aeration is important during the composting period because it provides oxygen for bacteria preventing anaerobic activity. In aerobic composting of the heap and pile methods, aeration is achieved by frequently turning the pile or heap. In vermicomposting, the worms maintain aerobic conditions in the wastes by ingesting solids and converting a portion of the organics to worm biomass and respiration products and expel partially stabilized matter as discrete castings. Aeration during vermicomposting is also related to particle size. Although larger particles favor airing, they take a long time to degrade. For this reason, the recommended particle size is 5 cm (Águila-Juárez et al., 2011).

2.6.2 Moisture content

Moisture content is important to support the metabolic activity of micro-organisms. When the moisture content is too low, metabolic activity occurs slowly, and when the moisture content is too high anaerobic conditions occur. Worms only thrive in aerobic conditions and die in anaerobic conditions. The optimum moisture level which prevents the worm from losing weight and dehydrating is 60 % and 80 % (Kaur, 2020).

2.6.3 Carbon to nitrogen ratio

The carbon to nitrogen ratio of the raw materials is important as a main component of the required nutrient. The optimal C/N ratio depends on the type of feed substrate, the species of earthworm, and the desired final product. The optimum carbon to nitrogen ratio of raw materials is between 25: 1 and 30: 1 (Teklebrahan, 2014). Higher carbon to nitrogen ratios results in the limitation of the growth of the microorganisms resulting in a larger composting time while a lower carbon to nitrogen ratio leads to the underutilization of Nitrogen. It is therefore important to ensure that the raw materials used in the vermicomposting process have the right ratio.

2.6.4 Temperature

Verm stabilization of sewage sludge has been reported to proceed more rapidly at temperatures of 13℃- 22℃. Temperatures at 50℃ are considered to be stressed conditions that result in the death of the worms. For the growth of the *Eisenia fetida* worm, optimum temperature ranges from 15 ℃ *and* 25℃.

2.6.5 pH

The habitat of the worm requires a pH from 6.8 to 9.0 to reproduce and grow, although slight variations are tolerable by the earthworm. The pH during the process of vermicomposting can either increase or decrease depending on the environmental conditions and feedstock substrate. When there is seen to be a reduction in pH mainly attributed to the production of carbon dioxide and organic acids, the worm secretes calcium carbonate to neutralize acidity and avoid mortality (Águila-Juárez et al., 2011). Changes in pH are also caused by the mineralization of nitrogen.

Gurav & Pathade., (2011) carried out a study on the production of Vermicompost from Temple Waste (Nirmalya): A case study. In which temple wastes mainly of flowers, leaves, fruits, sugar, and jaggery were used for eco-friendly treatment methods like bio methanation and vermicomposting. The study employed effluent from the biogas digester run on Ganesh temple waste admixed with temple solids and cattle dung. Optimization of parameters such as moisture content, particle size, pH, and temperature of the vermicompost using *Eudrilus eugenia* earthworm species was the main objective of the study. Results found that treatments of the temperature of 25℃, a pH of 8.0, the particle size of 1-2 mm, and 80 % moisture content were optimum parameters for vermicomposting.

Few studies have been carried out to evaluate the treatment of municipal sewage sludge using the vermicomposting method especially on investigating the effect of operating parameters and how these parameters affect the final vermicompost product. This research, therefore, intended to broaden the knowledge by investigating the effect of operating parameters such as; turning frequency, mixing ratios, and moisture content on the quality of vermicompost.

Table 2.3: Summarized research performed on vermicomposting and its results

Author	Objective	Parameters	Results
Belmeskine et al., (2020)	Assessing earthworm's acclimatization in the sewage sludge.	Weight and Length of the worms.	An increase in the average weight of the worms; *10.62*, *23.89*, and *35.72%* after 7, 14, and 21 days.
Hanc et al., (2020)	Investigating the feasibility of vermicomposting malting sludge and its mixtures with straw pellets.	The ratio of the mixtures in the proportions of *100%*, *75%*, *50%*, and *25%* of malt sludge.	Treatments with *100%* and *75%* malt sludge caused the death of the worms.
Kendie., (2009)	The effects of carbon to nitrogen ratio vermicomposting of Rice husk and cow dung with fresh biosolid.	Variation of carbon to nitrogen ratio in the proportion of *15:1,25:1,35:1,45:1and 55:1* of different treatments was used.	The ratio of *25:1* created the optimum conditions for the survival and growth of earthworms.
Degefe et al., (2012)	Evaluate the performance of epigeic earthworms; *E. Andrei*.	Moisture content, temperature, and equal ratio of mixtures.	An increment of *NPK* and a reduction in the carbon to nitrogen ratio.
Teklebrahan., (2014)	Optimization of Municipal Solid Waste Vermicomposting Parameters.	MC in the range of *(60-90%).* C/N in the range *(20-30).* Worm Stocking density in the range of *(0.8-2.0kg/m^2).*	Worm stocking density was found to be the most important parameter influencing the vermicomposting process of municipal solid waste.

3.0 METHODOLOGY

3.1 Study area

Addis Ababa is one of the fastest-growing cities in sub-Saharan Africa. It is home to 25% of Ethiopia's urban population. The city lies on a high-altitude plateau of 2100-2800 m and sloping to the South on the Akaki River. The city of Addis has an area of 527 km^2 and an estimated population of 5,005,524 a number close to 4% of the entire Ethiopian population. The city has an annual growth rate of 4.37 % (UNICEF, 2019).

Addis Ababa has twelve wastewater treatment plants. The Kaliti wastewater treatment plant is among the biggest in terms of size. This WWTP is suited in the industrial area of Addis Ababa, sloping down to the little Akaki river. The Kaliti WWTP was recently expanded and commissioned in the year 2018 and has a design treatment capacity of $100\,000\,m^3/day$ (AAWSA, 2019). About 40% of households in Addis have a wastewater collection system either through piped sewer lines or vacuum trucks, while the rest use on-site sanitation technologies.

Kaliti WWTP has six treatment units. The six treatment units are; grit and grease removal unit, the up-flow anaerobic system blanket, trickling filters, and sedimentation tanks, disinfection, and sludge drying bed unit. The UASB (up-flow anaerobic sludge blanket reactor) at Kaliti WWTP comprises four units which are; primary clarifier, biological reactor, secondary clarifier, and sludge digester. Non- biodegradable suspended solids are removed from the primary clarifier. The biological reactor removes biodegradable compounds by converting them to methane while the Sludge digester improves the dewatering characteristics of the Sludge. One sludge drying bed has a typical dimension of 6 m wide, 6-30 m length with a sand layer ranging from 230-300 mm depth. The sludge drying beds have been designed based on a sludge layer of 30 cm for UASB sludge and 40 cm for latrine sludge. The sludge amount in the reactor is managed through regular discharge of sludge to the sludge drying beds.

The sludge is discharged through values in the storage tank, this sludge takes approximately 15 days to dry in the sludge drying beds. The treatment plant also receives about 5,600 m^3 of sewage from on-site sanitation systems such as septic tanks and pit latrines per week and is discharged into the latrine sludge drying beds which take a longer time to dry (AAWSA, 2014).

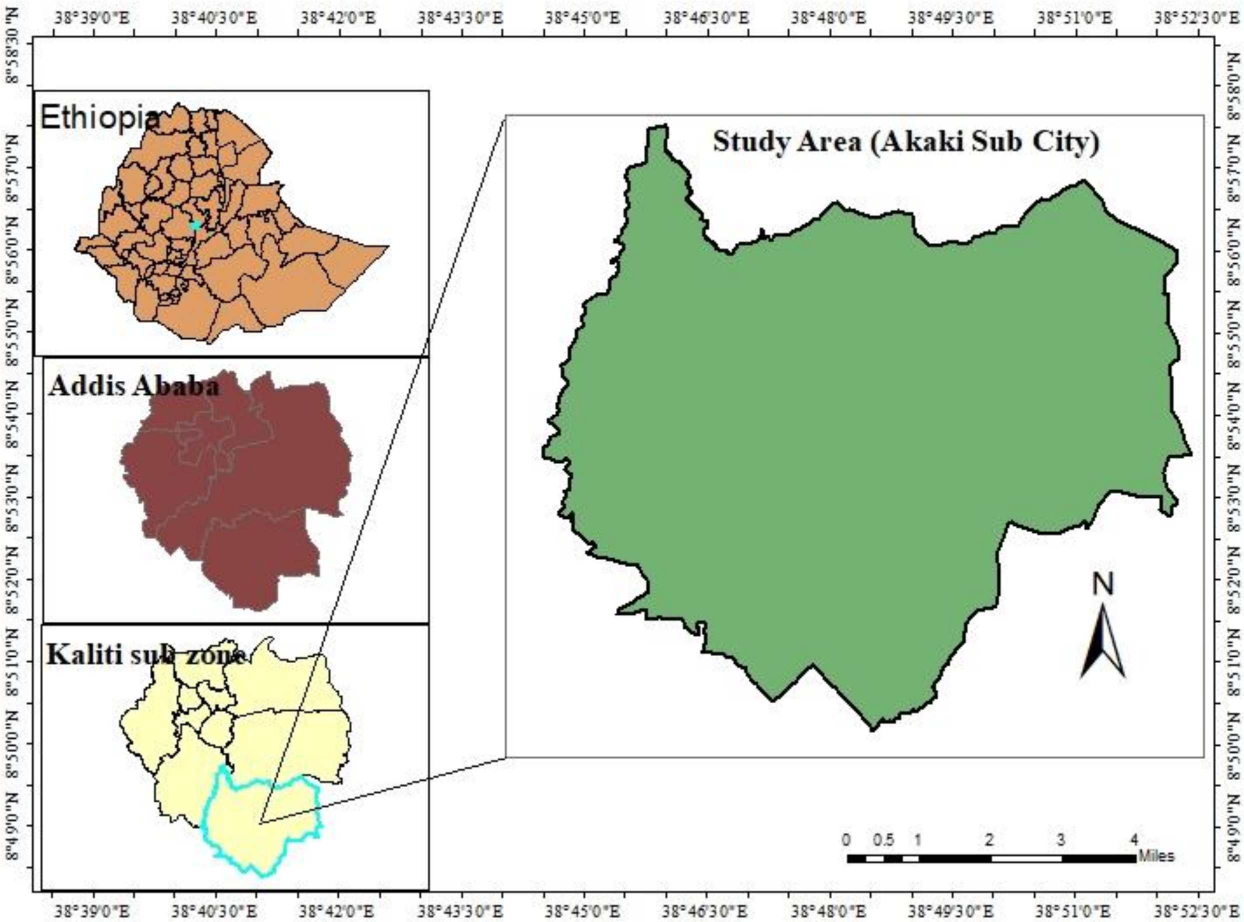


Figure 3.1: Study map of Kaliti WWTP in Akaki Sub-city (8.925083 N, 38.754910 E)

3.2 Sewage sludge sampling

Standard sampling procedures from EPA POTW Sludge Sampling and Analysis Guidance were followed in this study (EPA, 1989). Secondary sludge after treatment from the Up-flow Anaerobic Sludge Blanket Reactor (UASB) was used in this study. To get a good representative sample of the sewage sludge, grab samples of wet sewage sludges were taken at four different locations in the sludge drying beds and were mixed to provide for the final vermicomposting process. Sludge drying beds that were recently loaded were used to collect the samples of the sludge.

Figure 3.2: Sludge sampling from the drying beds

A minimum of 7 samples was collected from each drying bed in 1-liter capacity sampling bottles and transported in a cooler box kept below 4℃ to the laboratory for analysis. Liquid Sludge was also sampled in this study, to see the variation in physical and chemical characteristics between the liquid and dry sludge. The liquid sludge was sampled from the outlet of the UASB reactor. To get a good representative sample, equivalent volumes were collected in a 3mins time interval at all 5 depths of the outlet of the UASB.

3.3 Materials used in the vermicomposting process

Human waste is relatively rich in nitrogen and has a high moisture content, for this reason, it is highly dense and has a low carbon to nitrogen ratio. To balance this ratio and fasten the rate of decomposition or degradation, other carbon-rich materials were added to the vermicomposting process in this study. Cow dung is a material that is used as an amendment in most vermicomposting processes because it plays an important role in mineralization, nutrient recovery, and microbial and earthworm activity during vermicomposting (Yuvaraj et al., 2021). Cow dung is rich in nitrogen and easily biodegradable, a total quantity of 10 kg pre-composted cow dung was used in this study. To fasten the rate of decomposition and balance the carbon to nitrogen ratio of materials as well as the moisture contents, a highly carbonaceous material such as coffee husks was used in this study. A quantity of 20 kg shredded coffee husks was used in this study.

Nitrogen content during composting is important for the maintenance and formation of micro-organism's body cells. Coffee husks are rich in carbon, which is an important source of energy in metabolic processes which accelerate the rate of decomposition. In this study, red wrigglers worms or *Eisenia fetida* was used. These worms are the most widely used in most vermicomposting studies (Amare, 2015; Ludibeth et al., 2012; Degefe et al., 2012). These worms were collected from the Ethiopia Agricultural Research Ambo plant protection research center.

Figure 3.3: Materials used in Vermicomposting process

3.4 Characterization of sewage sludge samples

3.4.1 Laboratory preparation of sewage sludge samples

The characterization of sewage sludge for its physical, chemical, and biological characteristics was based on Standard Methods for the Examination of Water and Wastewater (APHA, 2002), and ASTM Standard Test Methods. Samples were air-dried and grounded to pass through a 2 mm screen before analysis. All analyses were done in triplicates and results were averaged. All instruments were calibrated before use and results were read to the nearest 0.01unit.

Parameter analysis

Analysis of pH was based on Standard method 9045 D soil and waste. Samples of sludge that were air-dried were measured to obtain a mass of 20 g. Then 100 ml of distilled water was added to a 250 ml beaker to obtain a supernatant suspension of 1: 5 (wt. /v) ratios of samples to the water mixture. The suspension was continuously stirred for 5mins using a glass rod and allowed to stand for 1 hour. Electrical conductivity and total dissolved solids were measured from the suspension made for pH using a conductivity meter of model 145.

Moisture content and total solid of sewage sludge:

A well-mixed sludge sample was weighed using an electronic mass balance to obtain a mass of 10 g of sewage sludge. The 10 g of sewage sludge was put in a crucible dish with known weight and dried in the oven for 24 hours at a temperature of 105℃. A desiccator was used to cool the sample to room temperature.

The moisture content of sewage sludge was calculated using the formula below;

$$MC = \frac{A - B}{B} \times 100 \dots \dots \dots \dots \dots \dots \dots .. (3.1)$$

Where;

A = mass before oven drying

B= mass after oven drying.

And the Total Solid was calculated using the formula below;

$$Total\ Solid(\%) = 100 - MC \dots \dots \dots \dots \dots ... (3.2)$$

Determination of ash content:

Determination of ash and organic matter was based on ASTM D 2974-87 Standard Test Methods. The oven-dried test specimens from the moisture content were used to determine the ash content of the sludge. The mass of the dish and specimen was determined before placing it in the furnace at temperatures of 550℃ for 2 hours. The sample was later cooled in a desiccator. The ash content of the sludge was determined using equation 3.3:

$$Ash\ content = \frac{C}{D} \times 100 \ldots . \ldots (3.3)$$

Where;

C = Mass of crucible dish + specimen before combustion

D = Mass after combustion

Determination of volatile solids:

The amount of organic matter or volatile solids in the sludge samples was determined by Gravimetric determination. The weight loss after a sample is ignited to dryness at 550℃

$$\%Total\ Volatile\ solids\ as\ \%TS = \frac{A - B}{A - D} \times 100 \ldots \ldots \ldots (3.4)$$

Where;

A = the final weight of the dried residue + crucible dish at 105℃

B = the final 550℃ $weight$ of the inorganic ash + crucible dish

D = the crucible dish weight

Determination of carbon content:

Volatile solids largely (carbon, oxygen, and nitrogen) are the components that burn off an already dry sample in a laboratory furnace at 550℃ leaving only ash and other mineral elements that do not oxidize. Most biological materials have a carbon content between 45 to 60 % of the volatile solids fraction (Adams et al., 1951). For this reason, the carbon content was calculated from volatile solid data, using a constant factor for the conversion from volatile solids to carbon by an empirical equation as reported by (Norbu, 2002) and the formula is:

$$\%\ Carbon = \frac{\%VS}{1.8} \ldots \ldots \ldots \ldots \ldots (3.5)$$

Determination of total nitrogen Kjeldahl:

A compost sample of 0.5 g was accurately weighed and transferred into a digestion tube. A catalyst mixture of 2 g catalyst was added and a few carborundum boiling stones were later mixed and rinsed with a little water to moisten the mixture. Concentrated sulphuric acid of 7 ml was added and mixed by swirling. The digestion tube stand with the samples was placed beside the block digester and an exhaust manifold was fitted on top of it. The tubes with the rack and exhaust manifold were placed on the digestion block preheated in the fume-hood. Digestion was for 3 hours until the digest was white or pale yellow on a block digester pre-heated to 300℃. The digest was allowed to cool and 50 ml of distilled water was added cautiously then allowed to cool again. The acid digest was transferred quantitatively to the macro-Kjeldahl flasks and rinsed using distilled water. Then 20 mls of the boric acid solution was measured from a dispense into a receiver Erlenmeyer flask corresponding to the number of samples. An indicator solution of 2 drops was added and placed under the condenser. The end of the condenser was immersed in the boric acid solution to prevent any loss of ammonia.

Calculation:

$$\text{TKN}(\%) = \frac{(a-b)}{s} \times N \times 0.014 \times 100 \times mcf \dots\dots\dots\dots\dots (3.6)$$

Where;

a= ml of H_2SO_4 required for titration of a sample

b= ml of H_2SO_4 required for titration of blank

s= air-dry sample weight in grams

N= Normality of H_2SO_4 (0.1 N)

100= ml of the solution

0.014= meq weight of nitrogen in g

mcf= moisture correction factor

Determination of available phosphorus:

A compost of 5 g was weighed into 250 ml polythene while shaking, including two blanks and a reference sample. The sample was shaken for 30 minutes on a mechanical shaker and was filtered through a Whatman No. 42 filter paper. Until the filtrate was clear, if not clear, 1 spoon P- free charcoal was added and shaken, and filtered again. Then pipetted in short test tubes of 3 mls of standard series of the blanks and the sample extract. Then 3mls of the mixed reagents were slowly added by pipette and swirl (CO_2 evolution). Then the solution was allowed to stand for at least 1 hour for the blue color to develop to the maximum. The absorbance was measured on a spectrophotometer at 882 or 720 nm (Sertsu & Taye, 2000).

Calculation;

$$P_{(PPm)} = (a - b) \times \frac{100}{s} \times mcf \ldots\ldots\ldots\ldots\ldots (3.7)$$

Where;

a= mg/l of P in sample extract

b= mg/l of P in blank

s= sample weight in gram (5 g)

100= ml of extracting solution

mcf= moisture correction factor

Determination of exchangeable potassium:

Potassium in the vermicompost samples was determined using dry ashing at $650 - 700$℃ and dissolving in concentrated hydrochloric acid. This was done by taking 5 grams of the sample and placing it in a crucible dish. The sample was made to ignite to ash at temperatures of $650 - 700$℃ in a furnace. The sample was later cooled and dissolved in a volume of 5 mls of concentrated hydrochloric acid and later transferred to a 250 ml beaker where it was heated. A

100 ml volumetric flask was later used in which the sample volume was marked. The solution was later filtered and the filtrate was later diluted with some distilled water. This was done to ensure the concentration of potassium in the sample remained in the ranges of $0\ to\ 20\ ppm$ if required. Exchangeable potassium was determined using a flame photometer using the k-filter. The different concentration of potassium of the standard solution in the flame photometer was read and the standard curve was prepared by plotting the reading against the different concentration of k (Sertsu & Taye, 2000).

Calculation;

$$k(\%) = R \times 20 \times dilute \ldots\ldots\ldots\ldots\ldots\ldots (3.8)$$

Where;

R =*ppm of K* in the sample solution (obtained by extra plotting from the standard curve)

Determination of organic carbon:

A sample of mass 10 g that had previously been oven-dried in an oven at 105℃ for 24 hours was placed in a pre-weighed crucible dish and the sample was made to ignite to temperatures of 650-700℃ for 6-8 hours in a furnace. Then later cooled to room temperature in a desiccator for 12 hours. The contents of the crucible dish were later weighed and recorded.

Calculation;

Total organic carbon (TOC) was later calculated using an empirical formula following (Adams et al., 1951)

$$TOM(\%) = \frac{intial_{wt} - final_{wt}}{intial_{wt}} \times 100 \ldots\ldots\ldots\ldots\ldots (3.9)$$

$$TOC(\%) = \frac{TOM}{1.8} \ldots\ldots\ldots (3.10)$$

Where;

TOM = Total organic matter

TOC = Total organic carbon

Determination of carbon to nitrogen ratio:

The carbon to nitrogen ratio of the municipal sewage sludge was estimated using the formula below;

$$R = \frac{Q_{1\,C_1}\ (100 - M_1) +\ Q_2 C_2\ (\ 100 - M_2\ ) + \cdots +\ Q_n C_n\,(100 - M_n)}{Q_1 N_1(\ 100 - M_1) + \cdots Q_n N_n\,(100 - M_n)} \ldots\ldots\ldots (3.11)$$

Where;

R = C/N ratio of compost mixture

Q_n = mass of material n (wet weight basis)

Cn = carbon (%) of material n

Nn = nitrogen (%) of material n

Mn = is moisture content (%) of material n.

When the above formula is simplified, it gives;

$$Carbon\ to\ nitrogen\ ratio(R) = \frac{Carbon\,\%\ of\ the\ sludge}{Total\ nitrogen\,\%} \ldots\ldots\ldots\ldots (3.12)$$

Microbial analysis of sewage sludge sample

Determination of total coliforms, E. coli, and fecal coliforms:

Appropriate municipal sewage sludge was collected using sterilized clean bottles from the Kaliti Wastewater Treatment plant and transported to the Bioengineering laboratory for total coliform analysis. A 10 g of sewage sludge was diluted into 100 ml sterilized distilled water and this volume (100 ml) of the diluted sample was filtered through a 47 mm, 0.45μm pore size cellulose ester membrane filter that retains the bacteria present in the sample. The filter was placed on a 5 ml plate of Mackonkey agar or an absorbent pad saturated with 2-3 ml Mackonkey broth, and the plate was incubated at 37°C for up to 24 hours. The total coliform bacterial colonies that grew on the plate were inspected for the presence of blue, red, and white color, and the colonies were counted using a colony counter (APHA, 2002).

Determination of helminth eggs count:

The Helminth eggs were done by using the $ZnSO_4$ (1.3 specific gravity) suspension method. A mass of 1 g of the sample was added into a 15 ml test tube and a few milliliters of 0.1% was added until it became 10 ml. The mixed samples were centrifuged at 3000 rpm for 3 to 5 minutes and then the supernatant was discarded. The decanted was collected and placed into other test tubes. The collected deposits were re-suspended in a few milliliters of $ZnSO_4$ until the test tubes were almost full and the vortex was well mixed. The tubes were again centrifuged at 2000 rpm for 3 to 5 minutes. The supernatant was then transferred into another test tube using a clean pipette. The test tubes were filled with distilled water to reduce the specific gravity of $ZnSO_4$ so as not to damage the eggs and also to allow them to deposit upon centrifugation. The test tubes were centrifuged again at 2500 rpm for 3 minutes. The supernatants were removed and deposits were combined into one test tube using water to recover all the deposits and were centrifuged again at 2500 rpm for 3 minutes to get the final deposit. The final deposit was removed from the test tube using a pipette and was placed onto one or more microscopic slides. The coverslips were placed over slides and were examined under a microscope of a 40× objective lens. The helminth ova were reported as eggs per gram of compost or sludge (Moodley et al., 2008).

Heavy metal analysis:

To determine the health and environmental hazard of the sludge and compost. Heavy metal analysis of 5 trace elements was conducted using flame atomic absorption spectrometry. These 5 heavy metals included; Cadmium, Lead, Chromium, Zinc, and Copper which are among the heavy metals of primary concern in the environment based on their high levels of toxicity following previous studies (Agoro et al., 2020; Águila-Juárez et al., 2011).

Heavy metal analysis was carried out at Ethiopian Construction Design and Supervision Works Corporation Laboratory. Sample preparation of the sludge involved air-drying the sludge in natural light. Then afterward, 2 g of the measured already grounded sewage sludge was placed in a conical flask containing 20 ml of concentrated HNO_3 (55%). This sample was heated at 90℃ for 45 mins. The temperature was then later increased to 150℃ held for 10 mins. HNO_3 of volume 10 mls was periodically added to the sample mixture to avoid or prevent dryness. Later the sample was allowed to cool to room temperature and filtration was followed using a Whatman filter paper. The digested sample was later transferred into a 100 ml standard flask.

The heavy metal contents in the sludge and compost were analyzed using a flame atomic absorption spectrophotometer (model novAA 400P) with Aspect LS 1.5.4.0 novAA 400P Tech: Flame software.

[A] [B] [C]

[D] [E] [F]

Figure 3.4: Laboratory characterization of sludge samples. A- measuring the weight of sample B- an oven used determining moisture content C- desiccator with silica gel as a drying agent D- sludge sample in crucible dish before determining ash content E- furnace F-crucible dish after being placed in the furnace.

3.5 Experimental setup and description

The 3 potential parameters that were varied in this experiment were moisture content (*MC*), turning frequency (*TF*), and Mixing ratio (*MR*) of substrates. As seen from the literature review other factors affect the vermicomposting process, in this experiment these other factors were held constant at their optimum levels because of different constraints. The levels of the selected factors were determined from literature research.

A small-scale vermicomposting experiment was set up. The experimental setup consisted of plastic bins having a capacity of 10 L. The plastic bins were used as vermi-reactors because they are easy to maintain and are easily constructed. The plastic bins were perforated in the bottom sides and the cap of the bin for aeration and drainage. The vermicomposting process was carried out in a greenhouse with ambient average temperatures of 37 ℃ at Addis Ababa University, School of Natural and Computational Sciences at Arati-kilo campus. Each treatment unit had the sewage sludge substrate, cow dung, coffee husks, and earthworms as per their proportions. The control experiments had no earthworm in the mixture of the substrate but were also maintained to compare the effect of earthworms in the conversion of substrates. A general simple schematic description of a composting process is presented in figure 3.5.

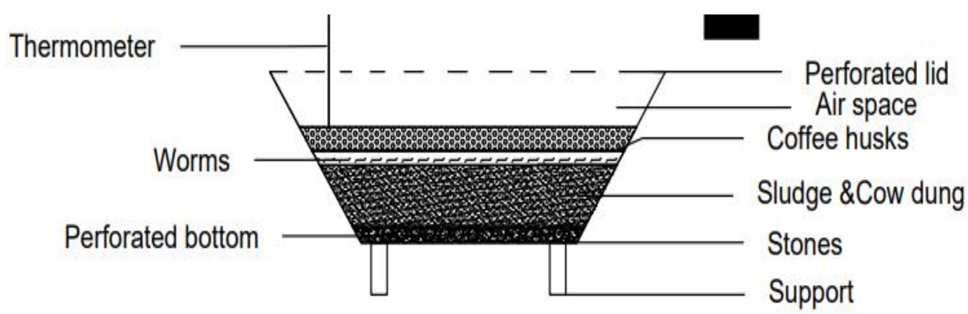


Figure 3.5: General sketch of a vermicompost bin

3.6 Design of experiment

The vermicomposting experiment was conducted for a maximum period of 50 days, from the 10th March 2021 to the 27th April 2021 (Biruntha et al., 2020). The experiment was laid out in a randomized design. *Design expert software version 10.0.7* was used to randomize the runs and design the experiment. Three factors having three levels each were considered in this experiment. The levels of the selected factors in this study were determined from literature research as presented in Table 3.1:

Table 3.1: Level and code of variables for Box-Behnken Design

Factors	Unit	Coded symbol	Level 1	Level 2	Level 3
MC	(%)	A	60	75	90
Mixing ratios	*w /w*	B	50:50	70: 30	80:20
Turning frequency	Turnings/ week	C	1	2	3

The experimental design used was a Box-Behnken under response surface methodology. In a Box-Behnken design, each numeric factor is set to 3 levels (-1, 0, +1). This design was chosen because it takes a less sample size hence saves time and it is more efficient and easier to arrange and interpret than other designs. In an ordinary multilevel categoric design or a general factorial design, the experiment having 3 factors at 3 levels and 2 duplications would have 54 experimental runs. In Box-Behnken design the same experiment would have a less sample size.

Box-Behnken design required 17 runs for modeling a response surface. Details of the experimental runs with input parameters are given in Table 3.2. Randomization of the experiment is important for concluding the experiment in a manner that the conditions in one run neither depend on the conditions of the previous runs. Five center points were used in this design to estimate experimental error and ensure that statistical analyses such as ANOVA are analyzed.

Table 3.2: Box-Behnken Design and combination of process variables

Run order	A: MC	B: MR	C: TF
1	60	50	2
2	60	65	1
3	75	80	1
4	60	80	2
5	60	65	3
6	75	65	2
7	75	50	1
8	90	65	1
9	90	50	2
10	75	65	2
11	75	65	2
12	75	65	2
13	75	50	3
14	90	65	3
15	75	65	2
16	75	80	3
17	90	80	2

3.7 Experimental procedure

The coffee husks were first shredded to reduce the particle size to 1-2 mm which is the optimum value (Gurav & Pathade, 2011). The sewage sludge was mixed with cow dung and coffee husks to give the optimal conditions of C/N ratios under different mixing ratios of substrates. A worm stocking ratio of 60 adult earthworms was stocked in each bin having a capacity of 5 to 6 kg of biowaste. Following recommended stocking density of 12 worms for every 1 kg of biowaste (Kendie, 2009).

Eisenia fetida earthworms were used in this study. The bins were initially filled to a 2 cm height with 12 mm nominal size chips of stone(aggregates) and shredded newspaper as bedding material to ensure proper circulation of air and water. The bins were perforated in the bottom and sides to increase aeration and avoid anaerobic activity. A mixture of municipal sewage sludge, cow dung, and coffee husks in different mixing ratios of substrates and moisture content was used above the bedding material to provide a natural habitat to the earthworms.

Although the reproduction of the earthworm depends on several important parameters, the most important of all parameters are temperature, moisture, and pH (Norbu, 2002). Temperature readings were taken twice a week throughout vermicomposting and weekly averaged values were considered. The response variables considered in this experiment were nutrient parameters such as NPK.

To evaluate the performance of the vermicompost in enhancing plant growth, 3 different treatments of increasing vermicompost (w/w) were employed. Treatment 1 which acted as a control had no vermicompost in it, Treatment 2 had 50 % vermicompost and Treatment 3 had 100% vermicompost. The 3 different treatments were set up in plastic pots of 7 Liter capacity at Addis Ababa University, School of Public Health Seferem Salam Campus premises. The plastic pots set up were placed in a shed of ambient temperatures of 25℃. In each of the 3 Treatments, 150 Lettuce seedlings (*Lactuca sativa)* were planted beneath a soil depth of $\frac{1}{8}$ inch deep. Watering of the seedlings was done at a 3days interval. The performance evaluation of the vermicompost was then calculated using germination index values (GI) given in equation 3.13;

$$GI\% = \frac{Number\ of\ emerged\ plants}{Number\ of\ seeds\ sown} \times 100 \ldots\ldots\ldots\ldots\ldots..(3.13)$$

3.8 Parameter analysis

To investigate the patterns of chemical and physical changes that occur during the vermicomposting process. Samples of the compost were analyzed during the period of vermicomposting. For each analysis, samples of the compost were drawn from 3 locations of each bin evenly spaced on top, middle, and bottoms then thoroughly mixed. About 20 g of the homogenized wet sample (free from earthworms, hatchlings, and cocoons) was drawn from each bin immediately for analysis.

3.9 Statistical analysis

Design-expert 10.0.7 (State-Ease) was the software used for designing the experiment, statistical analysis (analysis of variance), and response surface methodology studies. Design-expert was used to generate all graphs as well. Box-Behnken under Response surface methodology experimental method was used to optimize municipal sewage sludge vermicomposting parameters under 3 operating conditions. The response variables were total nitrogen, available phosphorus, and exchangeable potassium. The significance of the result was set from the analysis of variance (ANOVA).

4.0 RESULTS AND DISCUSSION

4.1 Characterization of the Liquid and Dry Wastewater Sludge

For the efficient end-use of sludge or disposal options, characterization of the physical, chemical, and biological composition of the sewage sludge samples was performed. The results for selected physical, chemical, and biological characteristics of liquid wastewater sludge obtained after final clarification from the outlet of the USAB digester are presented in Table 4.1. Results were given as mean values with their standard deviation of the three replicates.

Table 4.1: Selected physio-chemical and biological characteristics of Liquid Sludge

Parameters	**Quantity / Units**
pH	6.8 ± 0.03
Electrical Conductivity	$2.05\ mS/cm \pm 0.03$
Total dissolved solids	$995\ mg/l \pm 34.56$
Moisture content	$96\% \pm 0.82$
Dry residue/ Total solid	$4\ mg/l \pm 0.82$
Heavy metals	
Cadmium	$Below\ Detection\ limit$
Lead	$0.01 \pm 0.0008\ mg/l$
Chromium	$0.91 \pm 0.001\ mg/L$
Zinc	$21.69 \pm 0.698\ mg/L$
Copper	$2.55 \pm 0.236\ mg/L$
Bacteriological Parameters	
Total coliforms	5.24×10^6 CFU/ml
E. coli	2.38×10^6 CFU/ml
Qualitative parameters (1: weak -3: strong)	
smell	2
viscosity	2
color	2

The characteristics of the sewage sludge vary significantly between two wastewater treatment facilities and its composition is highly changeable during the process of treatment (Rorat et al., 2019). This statement is in harmony with the findings of a study that was carried out in Poland to determine the physical and chemical properties of the sewage sludge sampled from two municipal WWTPs. The authors observed a significant difference between the composition of nutrients and metals from one treatment plant to the other (Kiper et al., 2019). The variability in the composition of sewage sludge may also be attributed to the different materials used for anal cleaning, climatic factors, and the source area of the influent (Schoebitz et al., 2014).

As seen in Table 4.1, the composition of sewage sludge is highly variable. The variability in the composition of sewage sludge in this study may have been caused by the following reasons;

- The origin or the source of the incoming wastewater
- The composition of the incoming wastewater volume
- The type of wastewater treatment technology employed
- The treatment efficiency of the WWTP.

In this study, the mean pH value of the aqueous sewage sludge was found to be 6.8 ± 0.03. A slightly acidic to neutral pH values were observed in the sewage sludge samples obtained from Kaliti WWTP. This can be attributed to the presence of ions in the sludge samples (Romanos et al., 2019). The variations in sewage sludge parameters were also noted by several other researchers (Tilahun, 2009).

The pH value is an important parameter of both the aqueous and dried sewage sludge samples. In cases where the value is low indicating acidity, it can cause corrosiveness of metallic pipes. If the sludge were to be used for agriculture purposes it would gradually affect the $pH$of the soil which can later affect nutrient uptake. The pH value of the aqueous sludge in this study was close to neutral which is the safe and optimum value for its use in agriculture.

To vary the moisture content between the two sludge samples, samples of the dry sludge were obtained from the sludge drying beds of the WWTP. Other selected physical, chemical, and biological characteristics of the dry sewage sludge were analyzed and results are presented in Table 4.2.

Table 4.2: Selected physio-chemical and biological characteristics of dry sludge

Parameter	Quantity/ Unit
pH	7.42 ± 0.06
Electrical conductivity	$3.56\, mS/cm \pm 0.02$
Moisture content	$71.78\% \pm 0.03$
Total solids or Dry Residue	$28.22\% \pm 0.03$
Ash content	$13.01\% \pm 0.02$
Total Organic matter	$86.99\ \% \pm 0.03$
Total Dissolved solid	$1005\, mg/l \pm 1.24$
Volatile solids	$53.61\% \pm 0.01$
Carbon content	$29.78\ \% \pm 0.01$
Total nitrogen	$9.46\ \pm 0.15\, mg/kg$
Total nitrogen	$0.94\ (wt\%)$
Carbon to nitrogen ratio	3.1
Bacteriological Parameters	
Total coliforms	4.34×10^{6} **CFU/ml**
Total fecal coliforms	1.66×10^{6}CFU/**ml**
Helminth eggs	$70\ /g \pm 2\ of\ sludge.$

Table 4.2 shows a significant variation in moisture content between the liquid and dry sewage sludges. The moisture content of liquid sludge was reported to be 96% $\pm$ 0.82 while the dried sewage sludge samples have a moisture content of 71.78% $\pm$ 0.03. The liquid sewage sludge as seen in Table 4.1 has a moisture content close to 100 %, this is an indication that it is almost entirely a liquid. Liquids are heavy and, in most cases, difficult to transport and handle. For this reason, dewatering of the sludge is usually employed. The method of dewatering liquid sludge determines the overall moisture variation between the liquid and dry sludge samples.

It is also seen in Table 4.2 that the dry sewage sludge samples have a higher electrical conductivity than the liquid sludge samples by a mean value of $1.51 mS/cm \pm 0.02$. The increase in EC values of the dry sludge may have been attributed to the increase in total dissolved solids (TDS) in the sludge drying beds. Kaliti WWTP employs the conventional sand drying beds to dewater the sludge. The sludge is loaded into the convention sand drying bed. Consequently, there is an increase in TDS from the ions in the sand. This may have caused an increase in EC values in the dry sludge. The mineralization and release of different salts such as nitrates and potassium from the decomposition of easily biodegradable matter in the sludge drying beds may have also attributed to this variation. Despite having higher EC values in the dry sewage sludge samples, the values obtained in both samples were still lower than the maximum tolerable limits of plants $4.0 mS/cm$ (Belmeskine et al., 2020). Plants and soils are negatively affected by high EC values. A high EC value in a sample is an indication of the high salt concentration. An increase in salinity negatively affects crop access to soil water by increasing the osmotic strength of the soil solution. Even though the dry sewage sludge samples recorded high EC and TDS values, these species of earthworms (*Eisenia fetida*) can survive and adapt to extreme environmental conditions (Bhat et al., 2018).

To successfully use sludge as an organic fertilizer, the OM content of the sludge is an important parameter. The mean values of OM of dry sludge samples obtained from the characterization study were $86.99\% \pm 0.03$. This value indicates that sludge is rich in organic matter nutrients. The C/N ratios obtained in this study were very low as presented in Table 4.2. This was mainly due to the high nitrogen values observed in sludge and lower carbon values. As suggested by many authors the optimal C/N ratios required for composting are in the ranges of (20:1 to 30:1) parts of carbon to nitrogen (Ndegwa & Thompson, 2000).

Results obtained from proximate analysis such as; ash content and volatile solids of sludge were $13.01\% \pm 0.02$ and $53.61\% \pm 0.01$ respectively. Similar results were obtained by Yassin, (2019). In his study, based on the characterization result of the ash content of the sewage sludge obtained from Kaliti WWTP, his findings reveal that sewage sludge ash can also be used as supplementary cementitious material for concrete. The value for ash obtained from the dry sludge samples in this study, reveals that sewage sludge is rich biomass with much appreciable mineral or inorganic content, hence resource recovery can also be explored in other streams such as in the construction industry.

Heavy metal analysis of Sewage Sludge Samples:

Among the factors that restrict the use of municipal sewage sludge in agricultural practices are pathogens and heavy metals. Kaliti WWTP only treats domestic sewage from households, therefore the presence of heavy metals in sewage sludge samples may be attributed to corrosion within the sewerage network and also from the process of galvanizing Zinc. Some of these trace elements may not be treated during the treatment process which results in them being discharged into the environment and may create toxic hazards (Tilahun, 2009).

To determine the health and environmental hazard of the municipal sludge from the Kaliti Wastewater Treatment plant. Heavy metal analysis of 5 trace elements was conducted using flame atomic absorption spectrometry. These 5 heavy metals included; Cadmium, Lead, Chromium, Zinc, and Copper which are among the heavy metals of primary concern in the environment based on their high levels of toxicity following previous studies (Agoro et al., 2020 ; Águila-Juárez et al., 2011). Results for heavy metal analysis of the sewage sludge samples are presented in Table 4.3 and a more descriptive presentation is given in figure 4.1

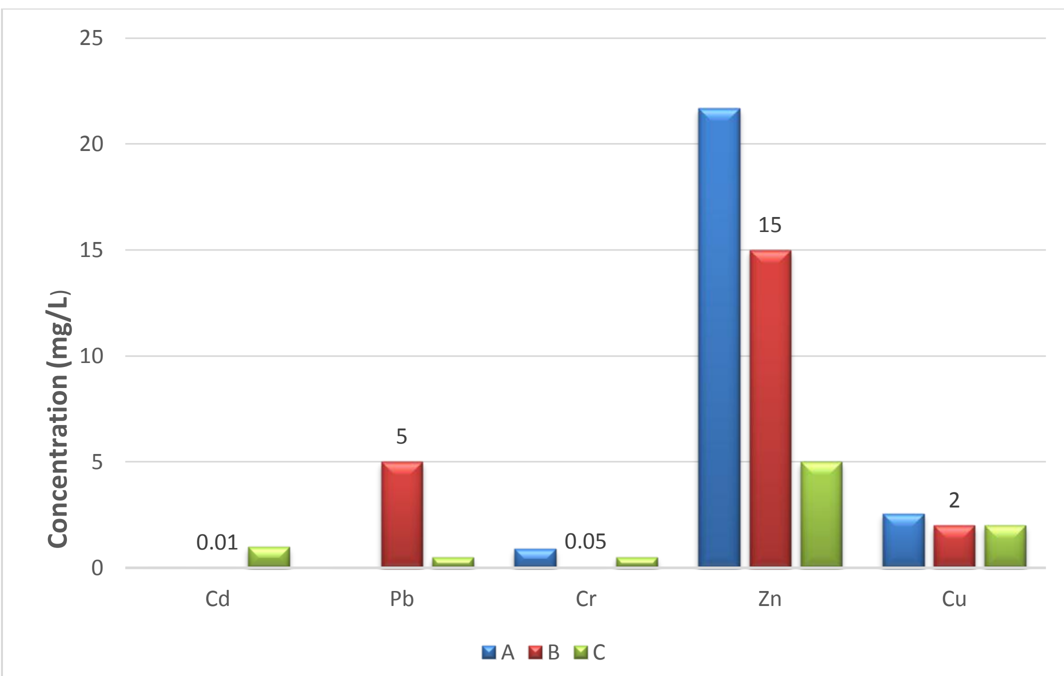


Figure 4.1: Metal concentrations in Sewage Sludge. *A- Results from this study *B- WHO (2006) Guidelines for the safe use of WW and Excreta *C- Controlled Application of Effluents to Land (Addis Ababa City Municipality,2003)

As seen from figure 4.1 and Table 4.3, the concentration of heavy metals in sewage sludge samples varied from being below the detection limit for Cadmium to 21.69 $\pm$ 0.698mg/L for Zinc elements. Similar results were also obtained by Tilahun.,(2009), in his Optimization study of Anaerobic Co-digestion of sewage sludge from Kaliti WWTP with brewery yeast waste from Gonder Beer.

It can also be seen in figure 4.1 that the trace element Lead (Pb) was below the emission limit value set by The Environmental Protection Authority of Ethiopia. Trace elements such as Chromium (Cr) and Copper (Cu) were slightly above both regulatory standards. According to the WHO regulatory standard, (Cr) and (Cu) were slightly above hazardous levels by $> 0.86mg/L$ and $> 0.55mg/L$ respectively. The trace element Zinc exhibited the highest concentration and exceeded all regulatory permissible limits. Similar results were also obtained by (Zhang et al., 2017).

Table 4.3: Threshold levels of Heavy Elements for crop production

Element	Recommended Max conc.(mg/L)	Remarks***(WHO, 2006)***
1. Cadmium (Cd)	0.01	• Above threshold levels may be toxic to crops such as beans, beets, and turnips.
2. Chromium (Cr)	0.05	• This element is not essential for plant growth.
3. Lead (Pb)	5	• At high concentrations, Lead inhibits plant cell growth.
4. Copper (Cu)	2	• Toxic to several plants at concentrations as low as 0.1 to 1.0mg/L nutrient solution.
5. Zinc (Zn)	15	• Toxic to a wide range of plants at varying conc.

As seen from results obtained from heavy metal analysis of the municipal sewage sludge. To avoid the accumulation of heavy metals such as Chromium, Zinc, and Copper in plants and soils and eventually humans it is important to treat the sewage sludge before being applied in agriculture practices. Heavy metals in human beings have severe endocrine-disrupting properties (Q. Lu et al., 2012).

Microbial Analysis of Sewage Sludge:

Pathogens such as bacteria and helminthic infections are mainly transmitted through the fecal-oral route. When a person eats food or fluids that have been contaminated by fecal matter either by flies, fingers, or crops irrigated by untreated sewage. Microbial analysis of the municipal sewage sludge samples was carried out in this study to determine the safe use of the municipal sewage sludge in agricultural practices. Results for the microbial analysis of the municipal sewage sludge are presented in Table 4.4:

Table 4.4: Microbial evaluation of sewage sludge

Indicators of fecal contamination	**Range in this study (CFU/ml).**	**Other studies*(Tilahun, 2009).* (MPN/100 ML)**	**[c]Recommended microbiological quality guidelines *(WHO, 2006).***
Liquid SS			
1. Total coliforms	5.24×10^6	35×10^4	$\leq 1000 CFU/ML$
2. E. coli	2.38×10^6	Not determined	$\leq 1000 CFU/ML$
Dry SS			
1. Helminths eggs	$70/g$	$15{,}000 - 45{,}000 no/L$	$< 10^A$ $< 35^B$
2. Total fecal coliforms	1.66×10^6	17×10^4	$\leq 1000 CFU/g$
3. Total coliforms	4.34×10^6	Not determined	$\leq 1000 CFU/g$

Note - (A-biosolid of excellent quality, B- biosolid of good quality according to (WHO, 2006) Standards. c- Recommended microbiological quality guidelines are merely dependent upon the reuse condition and are categorized into three groups (A, B, and C).

A- Crops to be eaten uncooked B- irrigation of cereals C- Localized irrigation of crops.

As presented in table 4.4, the microbial population (coliforms, bacteria, helminth eggs) in the sewage sludge samples is very high relating to poor sanitation practices (Tilahun.,2009; Mengistu et al., 2017). To facilitate a further reduction in pathogens other mechanisms such as; predation, starvation, temperature, ultraviolet radiation, changes in pH and time may be employed. The microbial analysis of sewage sludge samples indicates the unsafe use of untreated sewage sludge in agricultural practices based on recommended guidelines for the safe use of wastewater, excreta, and greywater in agricultural practices (WHO, 2006)**.**

Indicators of fecal contamination such as *E. coli* and helminth eggs (*Ascaris*, *Ancylostoma*, *Necator*, *Hymenolepis*, *Strongyloides*, *Toxocara*, *Trichuris*, *Taenia* spp) are among the most major risk of using untreated sludge and wastewater in agricultural practices. These eggs which are mainly associated with poor sanitation can survive in the environment for a very long time. The use of untreated municipal sewage sludge and wastewater in agricultural practices may present a great health hazard to field workers and crop consumers who may be at great risk of suffering from helminthic infections and bacterial infections such as typhoid and cholera. Apart from the public health concern, the use of raw sewage sludge negatively affects the environment causing eutrophication of water bodies and algae blooms.

4.2 Study on Sewage Sludge Vermicompost process

4.2.1 Physiochemical changes during vermicomposting

The degradation of organic waste and interaction of earthworms and micro-organisms brings about significant physical-chemical changes in the compost. This was seen in the results presented in Table 4.5. According to Norbu, (2002), the stabilization and mineralization of organic matter by earthworms is sorely dependent upon three important parameters which are; temperature, moisture, and pH. Additional parameters such as EC, ash content, and variation in the organic matter were analyzed in this study during the process of vermicomposting.

Table 4.5: Physical-Chemical changes during the vermicomposting process of sewage sludge

Parameters	**0-days**	**7th Day**	**20th Day**	**40th Day**	**50th Day**
pH	7.42 ± 0.06	7.57 ± 0.12	8.68 ± 0.18	8.00 ± 0.12	7.34 ± 0.05
$Ec(mS/cm)$	3.56 ± 0.02	3.15 ± 0.03	2.76 ± 0.12	2.17 ± 0.03	1.349 ± 2.17
$MC(\%)$	71.78 ± 0.03	67.86 ± 2.1	54 ± 4	57.9 ± 2	61.4 ± 0.1
$Ash(\%)$	13.01 ± 0.02	18.98 ± 1.45	28.05 ± 3.67	29.85 ± 5.17	16.5 ± 0.46
$OM(\%)$	86.99 ± 0.02	81.02 ± 1.45	71.95 ± 3.67	70.15 ± 5.17	83.5 ± 0.46
$TDS(mg/L)$	1005 ± 1.24	1580 ± 1.5	1380 ± 1.8	1080 ± 2.0	496 ± 16.77
$DM(\%)$	28.22 ± 0.03	32.14 ± 2.1	46 ± 4	42.1 ± 2	38.6 ± 0.1
$Carbon(\%)$	$29.78\% \pm 0.01$	45.01 ± 1.45	39.97 ± 3.67	38.97 ± 5.17	46.39 ± 0.46

Mean value and SD of the three replicates.

As seen in Table 4.5, there were significant changes in physical and chemical parameters during the vermicomposting process. This was also observed by several other researchers (Amouei et al., 2017). There was a general increase in the pH during the first 20 to 40 days. The variation in the pH value may have been caused by the alkalination of the biowaste from the release of ammonia gas during the degradation of the organic waste. Towards the 50th day of vermicomposting the pH value of the compost dropped to neutral, indicating the stability and maturity of the compost. Humus has a pH buffering capacity. The results of variation in pH during the vermicomposting period are in agreement with other studies(Uçaroğlu & Alkan, 2016).

Concerning EC there was a decreasing trend throughout the vermicomposting period. A decrease in EC values may be due to the stabilization of final mixtures and the decrease in ions after forming complex humus. The mineralization of the organic matter increasing inorganic matter content may also have been one of the reasons for a significant reduction in EC values.

The decomposition of OM is directly related to microbial degradation which usually results in the loss of OM. This was observed during the 7th to 40th day of composting. This was in agreement with Tena et al., (2020) who reported that a decrease in OM is an indication of the rapid decomposition of the biowaste due to the action of the micro-organisms.

The volatile matter or the organic matter of the biowaste gradually reduced. On the other hand, an increase in Ash content was observed during the period of vermicomposting. This increase corresponded to the recommended values ($< 35\%$) of ash according to Tena et al., (2020). Figure 4.2 presents a more graphical presentation of the physical-chemical changes during the period of vermicomposting

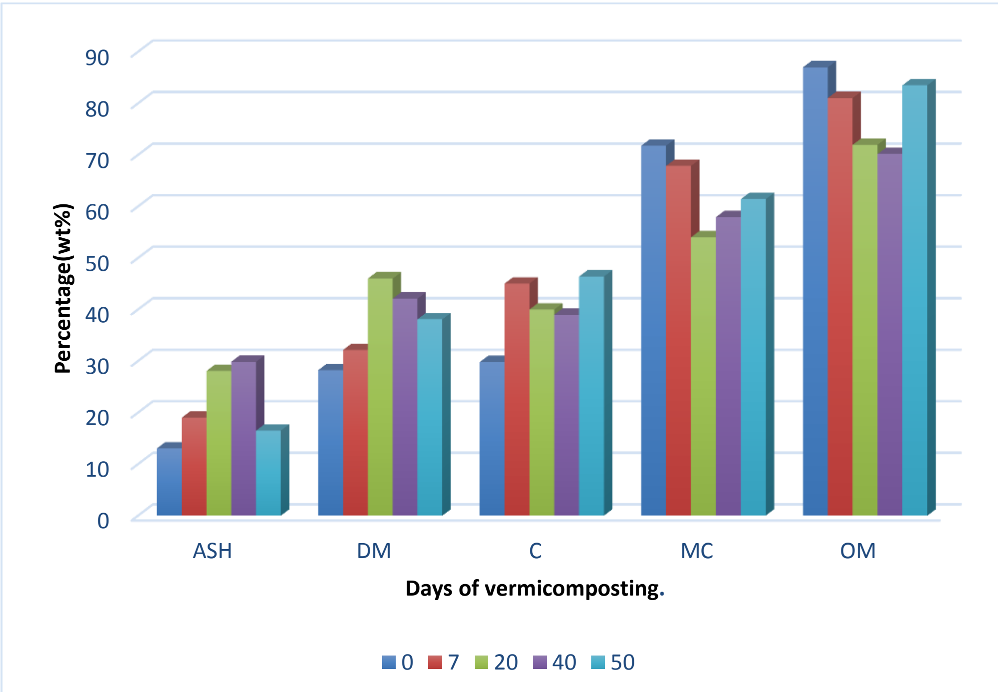


Figure 4.2: Variation in physio-chemical parameters during vermicomposting. *Note- parameters were analyzed on the 0, 7, 20, 40, and 50 days of vermicomposting.

Temperature variation:

Ordinary composting and co-composting technologies involve the thermophilic, mesophilic, and maturation phases during the biodegradation of organic waste. Unlike these technologies, vermicomposting is a non-thermophilic composting technology (Amare, 2015). As Ansari & Ori (2020) mention, the temperature for the survival of the *Eisenia fetida* earthworms is in the range of $0°C\ to\ 35°C\ (25°C\ optimum)$. These earthworms have a wide range of temperature tolerance levels, unlike other earthworm species. According to Chin et al., (2018), *Eisenia fetida* earthworms can survive up to temperature ranges of $40°C$ in the mesophilic range. During the process of vermicomposting, temperature readings were taken twice a week from all the 17runs and weekly averaged results were considered. Figure 4.3 shows the temperature profile change for the first 5runs in the 6weeks of composting. Similar patterns in temperature profile during vermicomposting for other runs are seen from Appendix 3.

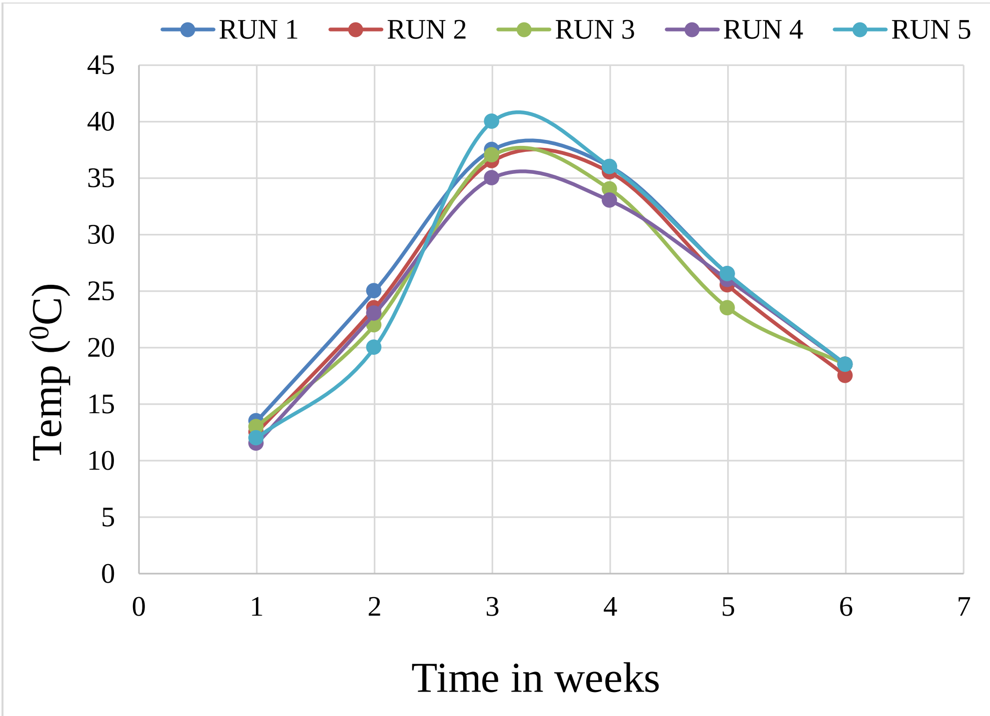


Figure 4.3: Variations in temperature during vermicomposting

As shown in figure 4.3, the temperature of the vermi-reactors or runs were kept within the optimum ranges required for vermicomposting. The minimum temperature value of the vermi-reactors recorded during the process of vermicomposting was 11.5℃ and the maximum value was 40℃. Like in any other composting technology, the temperature of the biowaste has a great influence on the microbial population and is an indication of the activities occurring during the decomposition process. At the minimum temperature, which was 11.5℃ during the first week of vermicomposting the earthworms were immobile and were not reproducing not until the temperature of the vermi-reactors had reached optimum values. Rapid cocoon production was observed in the second to the third week of vermicomposting. As seen in figure 4.3, a parabolic profile was observed during the composting period.

The microbiology of the activity of the earthworms and the micro-organisms may have caused the variation of temperature. As the natural process proceeded, there was an increase in the microbial activity of the earthworms and the microorganisms, leading to an increase in the temperature of the vermicompost. The maturity phase of the vermicompost showed a reduction in temperature readings. This shows that temperature variation is a good indicator of the microbial activity of the vermicompost. Temperature is also an important factor that affects the activity, metabolism, growth, and reproduction of the earthworm (Ansari & Ori 2020).

4.3 Effect of *Eisenia fetida* earthworms on vermicomposting

The degradation mechanism of the sewage sludge depends on the joint action of the earthworms and micro-organisms. In the gut of these earthworms are valuable degradative enzymes that stabilize different components of organic waste material. The earthworms later release rich castings which contain nutrients that are more soluble and available to plants than those in the parent substrate. Another important role earthworms play during the degradation of organic matter is increasing aeration through their borrowing actions. This reduces bad odor. Odor reduction was observed in this study, as the process of vermicomposting proceeded, the reactors that had earthworm in them lost their foul smell and developed an earthly smell like that of soil while on the other hand, the control experiment with no earthworm in it produced a bad foul smell. As Sinha et al., (2010) mention, earthworms have the potential to increase the rate of aerobic decomposition through their burrowing actions and at the same time have the ability to stabilize organic residues in the sludge by reducing harmful pathogens through the release of antibacterial coelomic fluid.

4.4 Maturation period of vermicompost

As seen from this study, 5 to 8 kg of biowaste can be degraded by 60 adults *Eisenia fetida* earthworms in a 10 L capacity vermi-reactor in a period of 5 to 6 weeks of the vermicomposting period. From observation of this study, during the first week of the vermicomposting, the earthworms were immobile with a low feeding rate. This was because there were trying to be acclimatized to their new environment. During the third to fourth week of vermicomposting, there was seen to be a rapid production of cocoons indicating rapid reproduction of the earthworms. Towards the latter part of the vermicomposting period, in the 5^{th} to 6^{th} weeks of vermicomposting, there was seen to be a change in the physical appearance of the compost as seen in figure 4.4. The vermicompost of municipal sewage sludge amended with cow dung and coffee husks developed a dark black fine color with finely divided and homogenized crushed crumbs an indication of mature castings. Similar results were obtained by Kendie, (2009), based on the author's findings, 60-70 adult *Eisenia fetida* earthworms were able to degrade organic waste in a 10 L capacity vermi-reactor provided optimum operating conditions in as little as 50-60 days of vermicomposting.

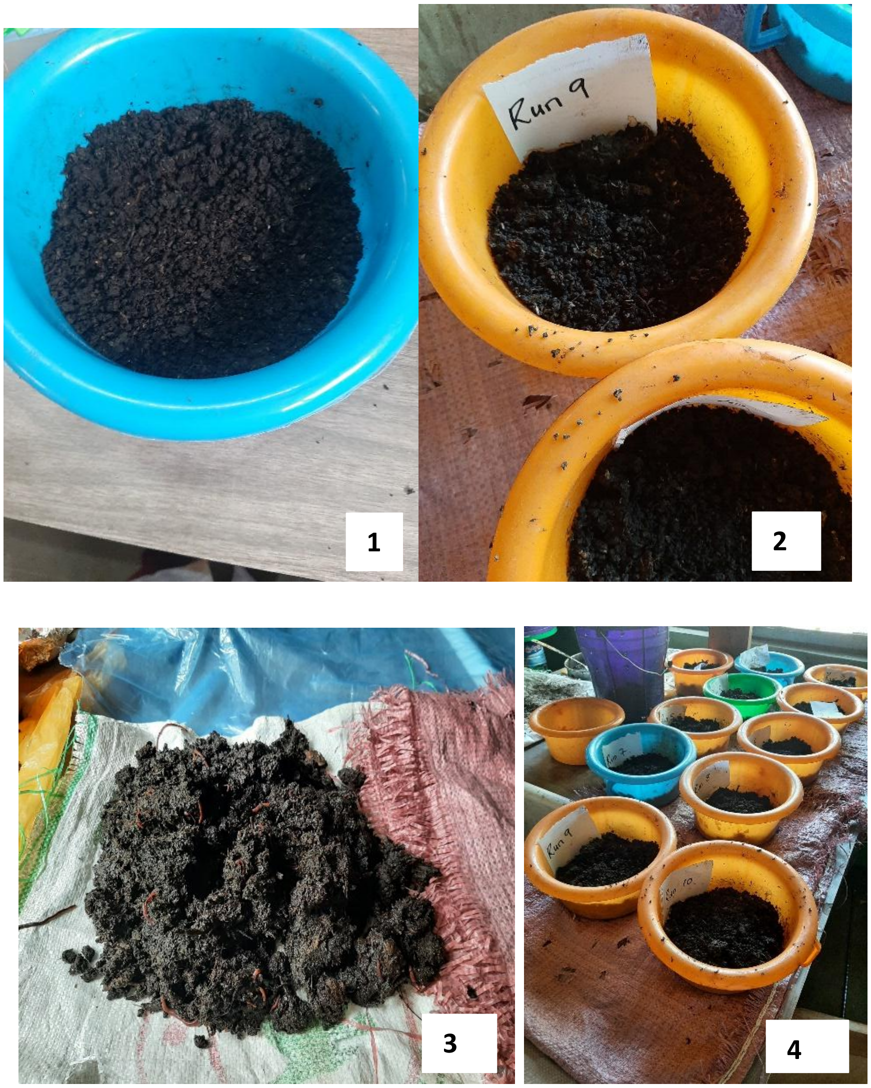


Figure 4.4: Harvesting of the vermicompost: 1 and 2-black gold castings. 3-separating worms from castings. 4- individual experimental bins

4.5 Characterization of the vermicompost

Physic-chemical and biological characteristics of the vermicompost obtained from optimum operating conditions (*moisture content 60 to 90%, mixing ratios 50 to 80 w /w, and turning frequency 1 to 3 turnings per week*) are presented in Table 4.6. The values are given as mean values and SD of the three replicates.

Table 4.6: Physicochemical and biological characteristics of the vermicompost against recommended guidelines

Parameter	Quantity/ unit (*This study*)	Guidelines for Assessing Compost Quality for Safe and Effective Utilization in Vegetable Production- (Ozores-Hampton, 2017)
pH	7.34 ± 0.05	$Ranges\ from\ 5-8 (optimum\ is\ 6-7.5)$
EC	$1.349\ mS/cm \pm 2.17$	$< 4.0\ mS/cm$
Organic matter	$83.5\% \pm 0.46$	$40-60\%$
Moisture content	$61.4\% \pm 0.1$	$(< 30\ dry)\ \&(> 60\ wet)$
C/N	$16:1$	$10-25$
Organic carbon (%)	17.11 ± 0.54	$-$
Ash content	$16.5\% \pm 0.46$	$< 35\%$
Total Carbon (%)	46.39 ± 0.46	$-$
Nutrients (NPK)		
Total nitrogen (%)	2.84 ± 0.07	$(0.5-6)\%$
Phosphorus (%)	1.22 ± 0.03	$(0.2-3.0)\%$
Potassium (%)	1.57 ± 0.04	$(0.10-3.5)\%$
Micronutrients(mg/kg-soil)		
Copper (Cu)	10.58 ± 1.64	
Zinc (Zn)	260.09 ± 1.41	-
Manganese (Mn)	241.40 ± 1.18	
Iron (Fe)	415.70 ± 1.58	
Bacteriological parameters (WHO, 2006).		
E. coli	$Not\ detected$	$\leq 1000 CFU/ml$
Helminth eggs	$51/g \pm 2$	$< 10^a$ $< 35^b$
Color	$Dark\ black$	$Dark\ black$
Odor	$No\ foul\ odor$	$odorless$

*Note- (a-biosolid of excellent quality, b- biosolid of good quality), ND-Note detected.

Results from qualitative parameters such as the color and odor of the vermicompost indicated that the decomposition of municipal sewage sludge was successful. The composite vermicompost obtained from all 17runs had a dark black fine color and was finely divided and homogenized. At the end of vermicomposting, there was no foul odor in the runs an indication that all parameters required for vermicomposting were present in optimum condition except for in the control which shows the importance of the earthworms during vermicomposting. The stabilization of final mixtures and decrease in ions after forming complex humus and mineralization of organic matter increasing the inorganic matter content may have resulted in low EC values towards the end of the composting period which were still in the range of the recommended values. At the end of the vermicomposting period, a carbon to nitrogen ratio of 16: 1 was recorded. A carbon to nitrogen in the range of 10-25, is an indication of an advanced degree stabilization of organic matter and reflects a satisfactory degree of maturity of the organic waste (Ozores-Hampton, 2017). The compost vermicompost obtained in this study contained *2.84%* of Nitrogen, *1.22%* of available phosphorus, and *1.57%* of potassium respectively as the major three important elements. Vermicast fertilizer characteristics nutrients vary with earthworm feed type. Typical nutrient analysis of the vermicast is *carbon to nitrogen ratio of 12-15*, *N* range of *(1.5-2.5%*), *K* range of *(1.25-2.25*), and *P* range of *(1-2%)* following guidelines of (Chaoui, 2010).

EPA Victoria, (2004) classifies biosolids in two independent factors based on contaminant concentrations and microbiological quality post-treatment. From the initial characterization of the sludge samples, a contaminant grade 2 of the sludge samples were met because contaminant levels were excessive. Despite the initial biosolid material being a contaminant grade 2 after the 50 days of vermicomposting a contaminant grade 1 was achieved. A contaminant grade 1 is the highest quality biosolid with relatively low contaminant levels.

Table 4.7: Contaminant limits for classifying biosolids based on EPA Victoria, (2004) guidelines

Contaminant	**Results in this study**	**Contaminant grade 1 & RSCL**	**Contaminant grade 2**
Zinc	260.09	300	2500
Copper	10.58	150	2000

Note: Values are in mg/kg dry weight *RSCL – Receiving soil contaminant limits

As seen in Table 4.7, contaminant levels for Zinc and Copper obtained in this study was that of contaminant grade 1. Contaminant grade 1 limits are also applicable for receiving soil contaminant limits set by EPA Victoria, (2004). These limits have been set up to ensure the application of biosolids does not cause chemical contamination of receiving soils and are established based on the most conservative endpoint (from human health, environment, and food safety).

The selected metals analyzed from composite samples of the vermicompost were within the acceptable limit except for iron which was above the acceptable standard for human consumption. Metals are naturally occurring in the earth’s crust. In this experiment has earlier outlined, the presence of trace elements may have been attributed to galvanizing of zinc or corrosion within the sewerage network. Among the metals that were analyzed; zinc, copper, manganese, and iron (Table 4.8) all were within the acceptable levels of the FAO/ WHO standards except for the Zinc element (FAO/ WHO, 2011).

Table 4.8: Metal analysis for composite vermicompost samples

Parameter	Results from this study	Acceptable limit (FAO/ WHO)
Zinc	260.09	99.4
Copper	10.58	73.3
Manganese	241.40	500
Iron	415.70	425.5

Note: Values are in mg/kg dry weight

Results of nutrients and contaminant levels obtained in this study may vary from those obtained in other studies due to several reasons. Some possible reasons for variations may be attributed to the following reasons;

- The different substrates used in the vermicomposting process.
- Municipal sewage sludge is highly variable as stated earlier, hence its nutrient or contaminant composition is highly dependent upon the origin of the wastewater and the treatment efficiency of the WWTP.

The reduction in pathogens levels in the vermicompost as compared to the initial sewage sludge samples in this study may have been attributed to the dilution of sewage sludge with other amendments such as cow dung and coffee husks that had reduced the toxicity levels and as well as the changes in pH during the vermicomposting process to slightly alkaline. At high alkaline levels, pathogens become inactive this is among the reasons why lime is a chemical stabilizer of sludge. Earthworm and bacteria degradation mechanisms may also have led to the reduction of pathogens in the vermicompost as they release antibacterial coelomic fluid (Sinha et al., 2010). Vermicomposting is a mesophilic process as it does not generate any heat, the vermicast was low in pathogen because earthworms consume fungi and some of the aerobic bacteria releasing low pathogen level castings. The significant reduction of pathogens in the vermicompost enables it to be called a Class A biosolid. Class A biosolids products can be used on home lawns and gardens, parks and golf courses, and other places where public contact is likely.

4.6 Optimization of Vermicompost with Response Surface Method

The optimization of key process parameters (*MC, MR, and TF*) for the production of organic fertilizer from municipal sewage sludge was demonstrated using a response surface methodology under a Box Behnken design. The *MC, MR, and TF* were independent variables while *NPK* were considered as the main response factors as seen in Table 4.9;

Table 4.9: Experimental results of N, P, & K obtained from optimal operating conditions

Run order	Moisture Content(A)	Mixing Ratios(B)	Turning Frequency(C)	N %	P %	K %
1	60	50	2	1.54 ±1.76	1.02 ± 0.54	1.24 ±0.63
2	60	65	1	1.58 ±0.06	1.25±0.08	1.36 ±0.05
3	75	80	1	2.58 ±1.29	1.74 ±0.03	1.8 ±0.03
4	60	80	2	1.75 ±1.23	1.54 ±0.03	1.28 ±0.04
5	60	65	3	1.60 ±0.92	1.35 ±0.02	1.25 ±0.02
6	75	65	2	2.73 ±0.62	1.52 ±0.18	1.87 ±0.08
7	75	50	1	2.27 ±1.98	1.11 ±0.92	1.8 ±0.09
8	90	65	1	0.85 ±0.04	1.28 ±0.63	0.6 ±0.07
9	90	50	2	0.83 ±0.03	1.25 ±0.06	0.59 ±0.04
10	75	65	2	2.78 ±1.42	1.82 ±0.18	1.85 ±0.18
11	75	65	2	2.73 ± .06	1.78 ±0.09	1.86 ±0.54
12	75	65	2	2.75 ±0.08	1.79 ±0.54	1.83 ±0.04
13	75	50	3	2.34 ±1.41	1.34 ±1.58	1.45 ± .08
14	90	65	3	0.82 ±0.03	0.24 ±0.62	0.53±0.54
15	75	65	2	2.7±0.08	1.7 ±0.12	1.79 ±0.58
16	75	80	3	2.34 ±0.58	1.54 ±0.04	1.69 ±0.92
17	90	80	2	0.82 ±0.54	0.54 ±0.06	0.54±0.12

The data represents the mean ± standard deviation of three replicates

4.6.1 Development of the empirical model

The data gathered in Table 4.9 was subjected to a regression analysis utilizing RSM of *Design expert version 10.0.7*. This was done to examine if there is a relationship between the dependent and independent variables. Second-order polynomial equations for responses *NPK* were therefore developed and are presented in terms of coded factors as seen from equations *4.1,4.2 and 4.3.*

$$N = +2.74 - (0.39 * A) + (0.068 * B) - (0.023 * C) - (0.047 * AB) - (0.013 * AC) - (0.077 * BC) - (1.33 * A^2) - (0.16 * B^2) - (0.19 * C^2) \dots\dots\dots\dots (4.1)$$

In terms of nitrogen as seen from the second-order polynomial 4.1, the independent terms *A*, *B*, and the quadratic terms A^2, B^2, and C^2 and the interactive term *AB* were all significant ($p < 0.05$). These model terms had significant effects on the yields of nitrogen. The quadratic regression model equation for the response of phosphorus is given by equation 4.2:

$$P = +1.72 - (0.23 * A) + (0.080 * B) + (0.11 * C) - (0.31 * AB) - (0.28 * AC) - (0.11 * BC) - (0.51 * A^2) - (0.12 * B^2) - (0.17 * C^2) \dots\dots\dots\dots (4.2)$$

The most significant model terms on the response of phosphorus were independent terms *A* and *B*, interactive terms *AC* and quadratic terms A^2 all significant at ($p < 0.05$). In the quadratic regression model equation for the response of potassium equation 4.3, the most significant model terms were independent terms *A* and *C* and quadratic terms A^2, $B^{2,}$ and C^2 which were all significant ($p < 0.05$).

$$K = +1.84 - (0.36 * A) + (0.029 * B) + (0.080 * C) - (0.023 * AB) + (1.000E - 022 * AC) - (0.060 * BC) - (0.84 * A^2) - (0.089 * B^2) - (0.066 * C^2) \dots\dots\dots\dots\dots\dots\dots\dots\dots\dots\dots\dots (4.3)$$

Model fitting and ANOVA analysis:

The fit summary tab in tables 4.10, 4.11, and 4.12 shows that the quadratic models were best suited for all the 3 responses (NPK). The quadratic model was suitable for all the 3 responses because it had a high Adjusted R^2 and predicted R^2. The quadratic model had the smallest $p - value$s ($p < 0.05$), showing its significance as compared to other models. The lack of fit value in all quadratic models for NPK responses was not significant ($p > 0.05$) which is a clear indication that independent variables affected the dependent variables. All three models were significant ($p < 0.05$). This can be seen from the fit summary tab presented in Tables 4.10, 4.11, and 4.12.

Table 4.10: Fit summary tab for Nitrogen response

Source	Sequential p-value	Lack of fit p-value	Adjusted R-Squared	Predicted R-Squared	
Linear	0.5826	< 0.0001	-0.0649	-0.4846	
2FI	0.9976	< 0.0001	-0.3786	-2.0693	
Quadratic	$\mathbf{< 0.0001}$	**0.25488**	**0.9979**	**0.9904**	***Suggested***
Cubic	0.2548		0.9985		*Aliased*

Table 4.11: Fit summary tab for phosphorus response

Source	Sequential p-value	Lack of fit p-value	Adjusted R-Squared	Predicted R-Squared	
Linear	0.4222	0.0069	0.0007	-0.4494	
2FI	0.2885	0.0069	0.0931	-0.8724	
Quadratic	**0.0056**	**0.0635**	**0.7635**	**-0.3709**	***Suggested***
Cubic	0.0635		0.9212		*Aliased*

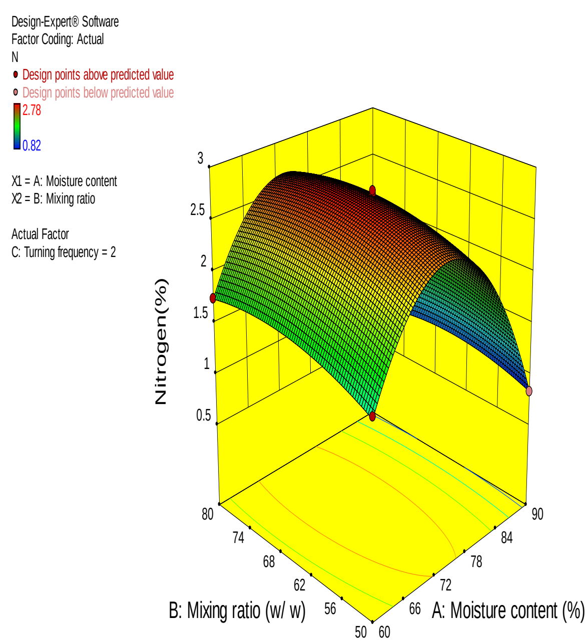
Table 4.12: Fit summary for potassium tab response

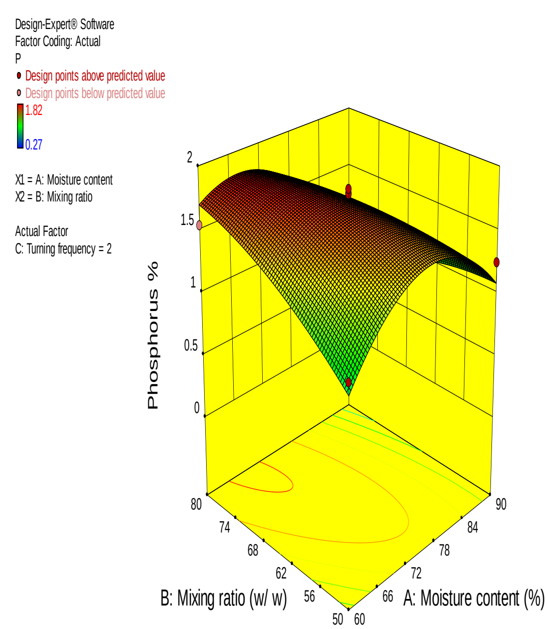
Source	Sequential p-value	Lack of fit p-value	Adjusted R-Squared	Predicted R-Squared	
Linear	0.2602	< 0.0001	0.0862	-0.3035	
2FI	0.9965	< 0.0001	-0.1816	-1.7521	
Quadratic	< 0.0001	**0.0550**	**0.9878**	**0.9279**	***Suggested***
Cubic	0.0550		0.9962		*Aliased*

The adequacy of the developed models was checked using Analysis of Variance (ANOVA) and by plotting diagnostic plots. This was done to ensure the models provided an adequate approximation for application in the real system. This is presented in Appendix 4 (ANOVA and diagnostic plots of *NPK).* The statistical properties of the developed models were further checked using; residual analysis, normal probability plots, graphs or residuals, residuals vs runs, and plots of predicted vs actual values which are all attached in Appendix 5. The adequacy check of the models revealed that the model was adequate and all statistical properties were considered.

4.6.2 Main parametric and interaction effects

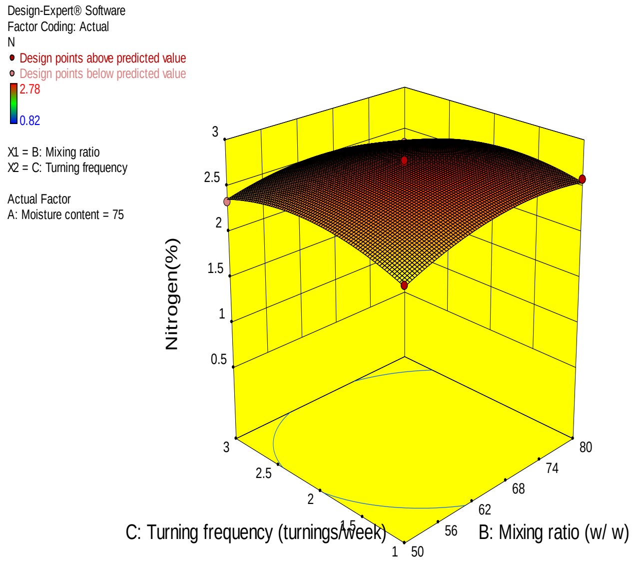
3D graphs also known as response surface graphs generated by the models were plotted to investigate the interactive relationships between the independent and dependent variables. These response surface plots were generated by the software (*Design expert version 10.0.7*) by keeping one factor at the central point and varying the other two factors within the experimental range.





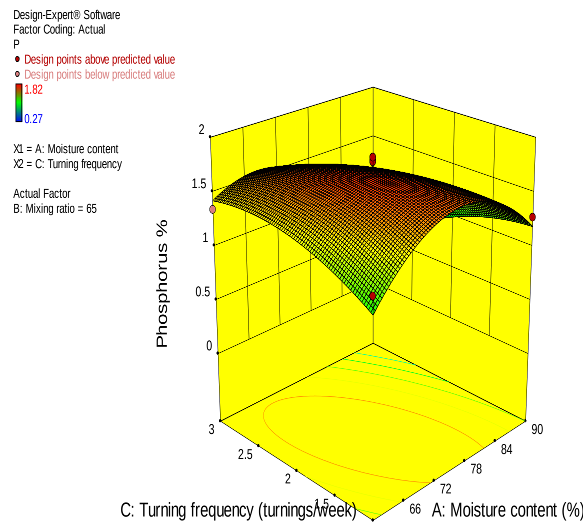
N (%): MR (wt):MC (%) *P(%): MR (wt): MC (%)*

[A] ***[B]***



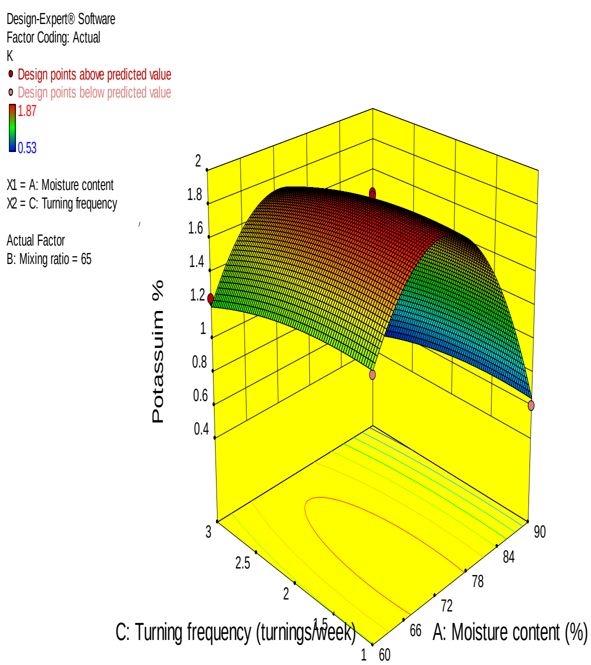
N(%): TF : MR(wt)

[C]



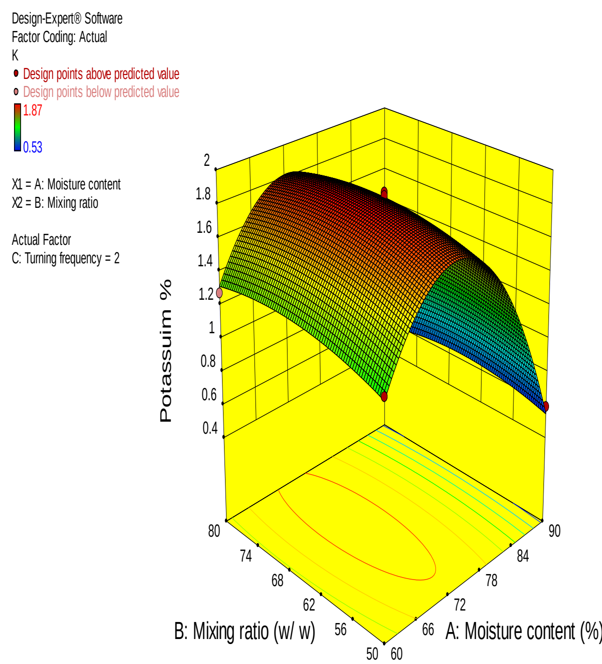
P(%): TF : MC(%)

[D]



K (%): TF : MC (%)

[E]



K (%) : MR (wt) : MC (%)

[F]

Figure 4.5: Response surface plots for NPK responses

Effect on nitrogen:

The model terms that had a significant effect on the yields of nitrogen during the vermicomposting process were: *A, B, AB, BC,* A^2, B^2, and C^2 which were all significant at $p < 0.05$ (*ANOVA tables Appendix 4*). As seen from figure 4.5A, from the interactive effects of process parameters moisture content (A) and mixing ratio(B). An increase in mixing ratios of the sewage sludge directly increased the quantity of nitrogen % in the vermicompost. On the other hand, increasing the moisture content of the feedstock during vermicomposting increased the quantity of nitrogen*%* only up to an optimal value.

An increase in moisture content beyond the optimal point resulted in to decrease in nitrogen*%*. These interactive effects can be attributed to the fact that municipal sewage sludge is rich in nitrogen, so increasing its proportion in the vermicomposting process positively increases the nitrogen content of the vermicompost. Similar results were obtained by other researchers such as Ludibeth et al., (2012). Findings by the authors indicated that an increase in the mixing ratio of the sewage sludge would increase the available nitrogen content in the vermicompost. An increase of up to *90%* of sewage sludge could not facilitate the survival of the earthworm due to the dense nature of the sludge. The decrease in nitrogen quantities after optimal points were achieved in this study is in harmony with the findings of other researchers that suggest that optimal vermicomposting is between the range of *70 to 80%* of the moisture content(Askun et al., 2018). Because excess moisture in the bins creates anaerobic conditions which destroys much of the goodness of the vermicompost. Above *90%* moisture content of the feedstock the pore spaces get displaced by excess moisture creating anaerobic conditions and less oxygen transport, as a result, the microbes in the compost consume oxygen in the moisture and this affects the activity of the earthworms (Gurav & Pathade., 2011).

Increasing the turning frequency affected the quantity of nitrogen until optimal points were reached as seen from interactive effects in figure 4.5C. Frequently turning the compost results in ammonia loss and subsequential low values of nitrogen. The significant effect of process parameters; *mixing ratio, turning frequency and moisture content* on responses *NPK* obtained in this study are in harmony with prior findings. Soto-Paz et al., (2020) carried out a pilot-scale study to determine the influence of *mixing ratio* and *turning frequency* on the co-composting of biowaste and sugarcane filter cake. Findings from the authors suggest that the application of these two strategies simultaneously has a significant effect on the process parameters *($p \leq 0.05$)*. From their findings a mixture of *80: 20* of the biowaste and sugarcane filter cane and a turning frequency of *2* turnings each week had significantly increased the quantity of *TN (2.4%)* and *TP (1.6%)* at the composting process.

Effect on phosphorous:

The model terms that had a significant effect on the yields of phosphorus during the vermicomposting process were: *A, B, AB, AC, and* A^2 which were all significant at $p < 0.05$ (*ANOVA tables Appendix 4*). As seen from Figures 4.5*B and D*, from the interactive effects of process parameters of *moisture content (A*) and *mixing ratio(B)*, a similar trend from the effect on nitrogen was observed. From the 3D graphs, it was observed that process parameters such as *moisture content* and *turning frequency* increased phosphorus (%) in the vermicompost until an optimal point, beyond this point an increase resulted in a decrease in phosphorus content. Similarly, TF has an increase in phosphorus only up to optimal values. In the case of mixing ratios, the phosphorus content was sharply increased as the mixing ratios of sludge were increased. This phenomenon can be attributed to the fact that the sewage sludge is rich in NPK. So, an increase in its ratio directly increases phosphorus in the materials. Although sewage sludge is rich in *NPK* and a direct increase in its proportional results in an increase of *NPK* nutrients, prior studies suggest that earthworms cannot survive in treatments having a mixing ratio of sewage sludge greater than *95%.* Treatments having a mixing ratio of sewage sludge *(< 80%)* exhibit the highest survival and reproduction of the earthworm (Ludibeth et al., 2012).

Effect on potassium:

The model terms that had a significant effect on the yields of phosphorus during the vermicomposting process were: *A, C,* $A,^2$, $B^{2,}$ and C^2 which were all significant at $p < 0.05$ (*ANOVA tables Appendix 4*). As seen from Figures *4.5 E and F*, independent terms *moisture content and turning frequency* have a significant effect on the yields of potassium. An increase in the *moisture content* and *turning frequency* increases the yields of potassium only up to an optimum point. A pilot study by Teklebrahan, (2014), suggests that *moisture content of 74.62%* of the feedstock is optimal to produce the maximum quantity of NPK nutrients.

As seen from this study a strong relationship exists between process parameters such as; *moisture content, turning frequency, and the proportion of mixing rations* on the quality of the vermicompost. Though an increase in the mixing ratio of sewage sludge facilitates an increase in *NPK* it does not support the survival and reproduction of the earthworm

Process optimization of the vermicomposting process

In this research work, the objective function of the response factors was to maximize the yields of *NPK* respectively. The accompanying constraints: *moisture content (60-90%), mixing ratios(50-80%wt*), and *TF (1 -3 turnings/week)* were kept in the range, to provide the ideal outcomes of the developed models. Response optimizer software that is available in the design expert software *version 10.0.7* under numerical optimization was utilized with a desirability of 1. This was done to provide the ideal outcomes of the response and the independent factors. The ideal conditions for obtaining the maximum yield of *NPK* from the vermicomposting of municipal sewage sludge were a; the *moisture content of 71.95%, mixing ratios* of up to *72.352wt%,* and a *turning frequency* of *2 per week*. These conditions would result in maximum *nitrogen of 2.76%, phosphorus of 1.80%*, and *potassium of 1.88%* respectively. The standard error models from the optimization results were; *1.56% for nitrogen, 9% for phosphorus, and 2.5% for potassium*. This demonstrated that the models developed are quite accurate as the percentages of prediction error were in good agreement.

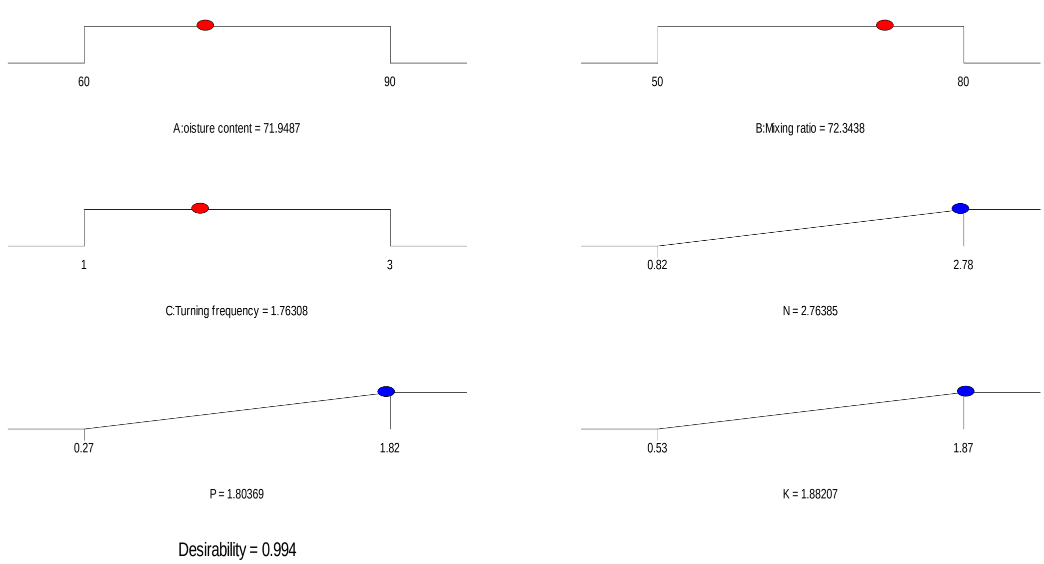


Figure 4.6: Ramp plot of optimization solution for the responses

4.7 Performance evaluation of vermicompost to enhance plant growth

Evaluation of the vermicompost to enhance plant growth was carried out by cultivating 150 Lettuce seedlings (*Lactuca sativa)* in 3 different plastic containers having 3 different treatments.

Three different treatments employed were:

$$T_1 = 0\% Vermicompost + 100\% soil (control\ experiment).$$

$$T_2 = 50\% Vermicompost + 50\% soil\ (wt/wt)$$

$$T_3 = 100\% Vermicompost + 0\% soil$$

Performance evaluation of the vermicompost was determined by the germination index value:

$$GI\% = \frac{Number\ of\ emerged\ plants}{Number\ of\ seeds\ sown} \times 100$$

Table 4.13: Performance evaluation of the vermicompost to support plant growth

Treatments	No. of seeds sown	No. of plants emerged	$GI\%$
T_1	150	30	20
T_2	150	80	53
T_3	150	130	86
$T_2 - T_1$	-	50	33
$T_3 - T_2$	-	50	33
$T_3 - T_1$	-	100	66

Table 4.13 describes the effect of the vermicompost of municipal sewage sludge on the germination of vegetable salad planted in two weeks. As seen from Table 4.13, an increase in the vermicompost shows an increase in the germination index. This is because the vermicompost contains plant growth promoters and other micronutrients that enhance plant growth. From figures 4.7 and 4.8, it is observed that the number of plants that emerged or the germination rate produced using *100%* vermicompost was *33%* greater than the germination index of using *50%* vermicompost and *66%* greater than the (control - 0%vermicompost). A clear indication that vermicompost prepared from municipal sewage sludge enhances plant growth. Improved seed germination in several other plants by the vermicompost was also observed by (Lazcano & Domínguez, 2014).

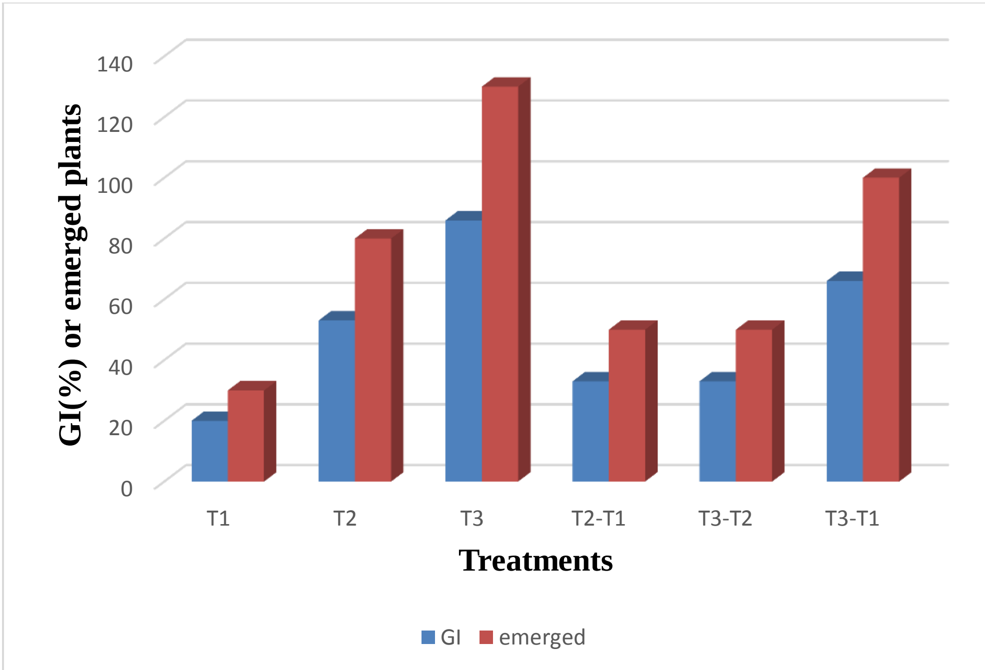


Figure 4.7: Histogram showing performance evaluation of the vermicompost

Therefore, the study showed that the growth and germination of the salad seeds grown in two weeks were positively affected by the increasing dose of the vermicompost which indicates the good performance of the vermicompost as seen in figure 4.8.

Figure 4.8: Experimental treatments for evaluation of vermicompost to enhance growth (*from left to right*); Treatment 3 (100% VC), Treatment 2 (50% VC), Treatment 1 (0% VC)

5.0 CONCLUSION AND RECOMMENDATION

The result in the present study indicates that vermicomposting is among the natural effective options for recycling organic waste, particularly municipal sewage sludge. Municipal sewage sludge which is a potential biohazard can be converted into safe organic fertilizer via vermicomposting which can be applied on farms. Vermicomposting is therefore an ecological natural method that can be employed to stabilize and naturally treat the municipal sewage sludge. This effective organic waste recycling can benefit millions of poor farmers globally and help achieve SDG number 2 which has a target to end hunger, achieve food security and promote sustainable agricultural systems by the year 2030. Among other reasons for promoting organic waste recycling in the use of fertilizers is the reduction of over-dependency on chemical fertilizers which are produced from vanishing earth resources.

According to the characterization results obtained from this study, there is a potential of exploring resource recovery in other streams among construction. A pot experimental setup in this study reveals that vermicompost can enhance plant growth twice compared to plants grown in ordinary soils. Optimization of process parameters using the RSM a Box Behnken Design reveals that moisture content of *71.95%* and a mixing ratio of *72.3%* of the municipal sewage sludge with a turning frequency of 2 times a week produced the maximum quantity of *N-2.76%, P-1.80%,* and *K-1.88%* respectively. Overall, vermicomposting of municipal sewage sludge amended with cow dung and coffee husk is, therefore, a promising approach for converting this potential biohazard into safe organic fertilizer as earthworms play an important role in the stabilization and degradation of organic waste. To strengthen this paper, the following recommendations should be considered;

- To facilitate a further reduction in pathogen levels in the vermicompost, other treatment methods such as thermophilic composting should be carried out before the vermicomposting process.
- More studies or research should be employed to find efficient ways of removing contaminants such as heavy metals among others to facilitate resource recovery from sewage sludge.
- Efforts should be made by WWTP to exhibit a better removal capacity of the trace elements at safe levels for both aquatic life and humans.

- As the majority of the population in Africa is dependent upon on-site sanitation systems, studies to characterize the composition of fecal sludge should be taken up to increase resource recovery from human waste.
- Strict regulatory guidelines should be put in place and enforced in the use of organic amendments as fertilizers.
- Awareness campaigns should be encouraged to overcome the negative public perception of the reuse of sewage sludge in agriculture which is among the biggest hindrance to ecological sanitation.
- Business model studies should be carried out to estimate cost recovery and anticipate potential markets for vermicompost products.

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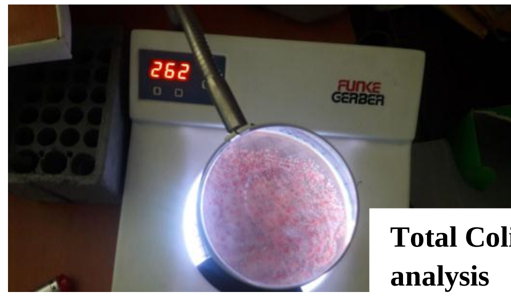
APPENDICES

Appendix 1:

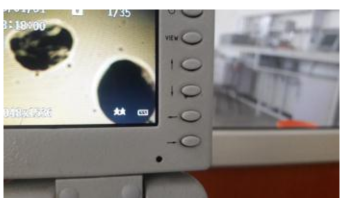
Vital pictures during the research work

Sampling at KWWTP

E.fetida



Total Coliform analysis



Helminth eggs analysis

Vermi-composting setup in a green house.

AAS cookbook machine at ECDSWC Laboratory.

A handful of *E.fetida* earthworm

Adult *Eisenia fetida* earthworm and cocoons

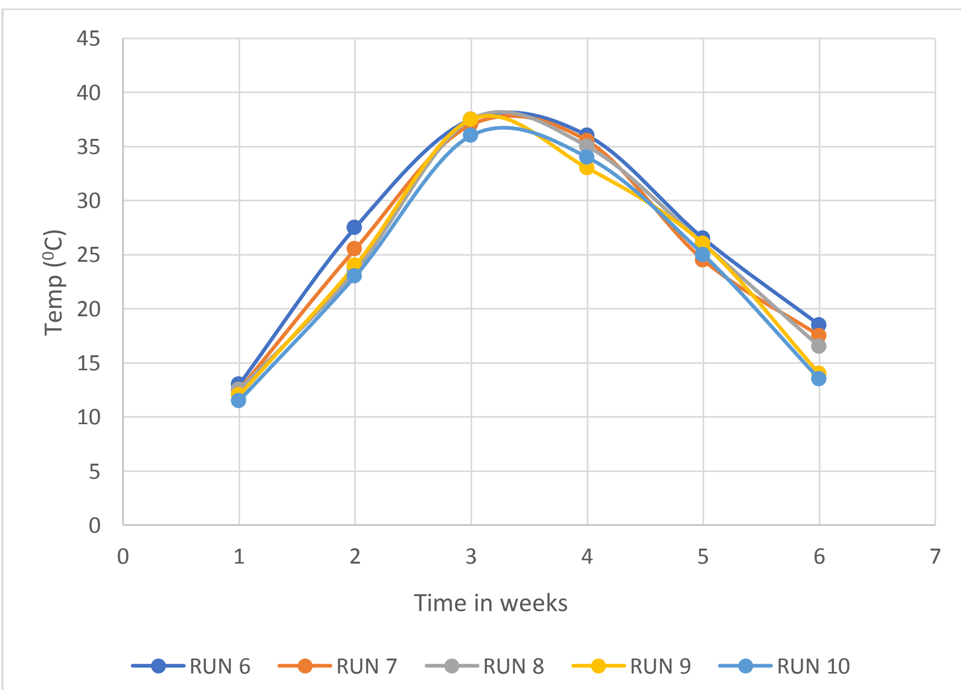
Appendix 2:

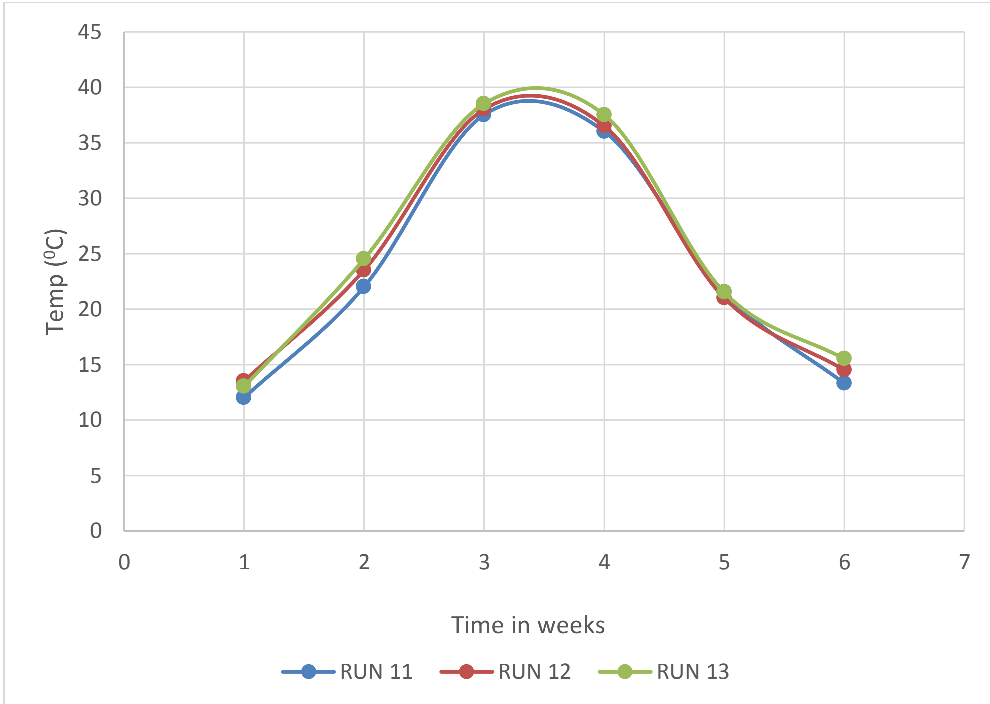
Table 1: Characteristics of materials used as feedstock in the experiment

Parameter	Coffee husks	Cow dung
pH	5.32	7.8
Moisture content (%)	13.0	64.3
Organic carbon (%)	54.5	23.0
Total Nitrogen (%)	1.83	1.82
C/N ratio (%)	29.8	12.6

Appendix 3:

Temperature profile change during vermicomposting





45
40
35
30
25
20
15
10
5
0
Temp (⁰C)
0
1
2
3
4
5
6
7
Time in weeks
RUN 11
RUN 12
RUN 13

Appendix 4:

ANOVA results of the quadratic regression models of NPK

Table 2: ANOVA results of the quadratic regression model for Nitrogen

Source	**Sum of squares**	**df**	**Mean square**	**F Value**	**p-value**	
Model	9.32	9	1.04	830.44	< 0.0001	**significant**
A-MC	1.22	1	1.22	975.67	< 0.0001	
B-MR	0.036	1	0.036	29.23	0.0010	
C-TF	4.050E-003	1	4.050E-003	3.25	0.1145	
AB	9.025E-003	1	9.025E-003	7.24	0.0311	
AC	6.250E-004	1	6.250E-004	0.50	0.5019	
BC	0.024	1	0.024	19.26	0.0032	
A^2	7.48	1	7.48	5996.77	< 0.0001	
B^2	0.11	1	0.11	89.43	< 0.0001	
C^2	0.16	1	0.16	125.43	< 0.0001	
Residual	8.730E-003	7	1.247E-003			
Lack of fit	5.250E-003	3	1.750E-003	2.01	0.2548	**Not significant**
Pure Error	3.480E-003	4	8.700E-004			
Cor Total	9.33	16				

Table 3: ANOVA results of the quadratic regression model for Phosphorus

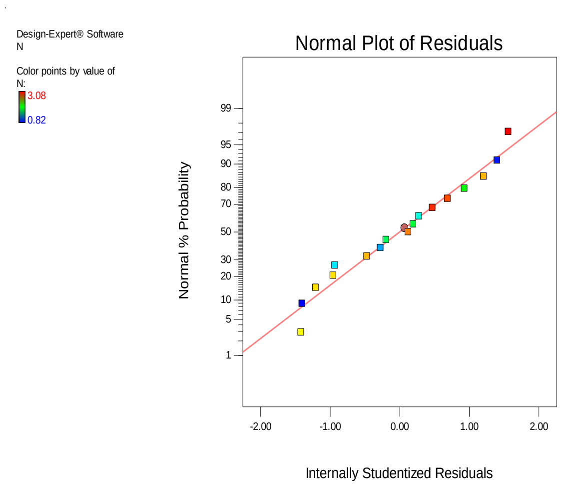
Source	Sum of squares	df	Mean square	F Value	p-value	
Model	2.68	9	0.30	6.74	0.0099	**significant**
A-MC	0.41	1	0.41	9.37	0.0183	
B-MR	0.051	1	0.051	1.16	0.3173	
C-TF	0.097	1	0.097	2.19	0.1823	
AB	0.38	1	0.38	8.56	0.0221	
AC	0.31	1	0.31	6.97	0.0334	
BC	0.046	1	0.046	1.05	0.3403	
A^2	1.12	1	1.12	25.26	0.0015	
B^2	0.060	1	0.060	1.37	0.2806	
C^2	0.12	1	0.12	2.75	0.1414	
Residual	0.31	7	0.044			
Lack of fit	0.25	3	0.083	5.67	0.0635	**Not significant**
Pure Error	0.059	4	0.015			
Cor Total	2.99	16				

Table 4: ANOVA results of the quadratic regression model for Potassium

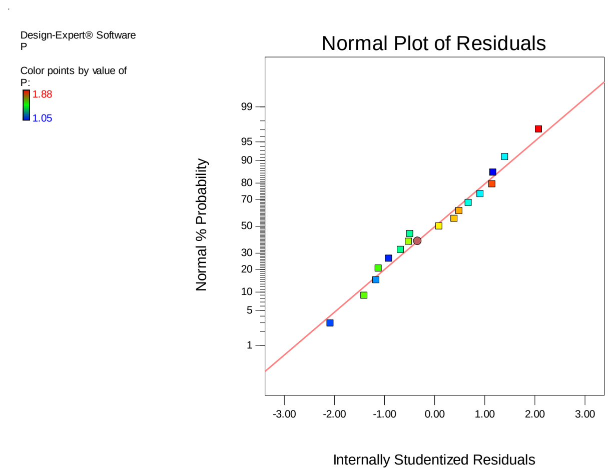
Source	Sum of squares	df	Mean square	F Value	p-value	
Model	4.20	9	0.47	144.37	< 0.0001	**significant**
A-MC	1.03	1	1.03	318.55	< 0.0001	
B-MR	6.613E-003	1	6.613E-003	2.05	0.1957	
C-TF	0.051	1	0.051	15.84	0.0053	
AB	2.025E-003	1	2.025E-003	0.63	0.4546	
AC	4.000E-004	1	4.000E-004	0.12	0.7353	
BC	0.014	1	0.014	4.46	0.0727	
A^2	2.96	1	2.96	916.45	< 0.0001	
B^2	0.033	1	0.033	10.26	0.0150	
C^2	0.018	1	0.018	5.72	0.0481	
Residual	0.023	7	3.232E-003			
Lack of fit	0.019	3	6.208E-003	6.21	0.0550	**Not significant**
Pure Error	4.000E-003	4	1.000E-003			
Cor Total	4.22	16				

Appendix 5:

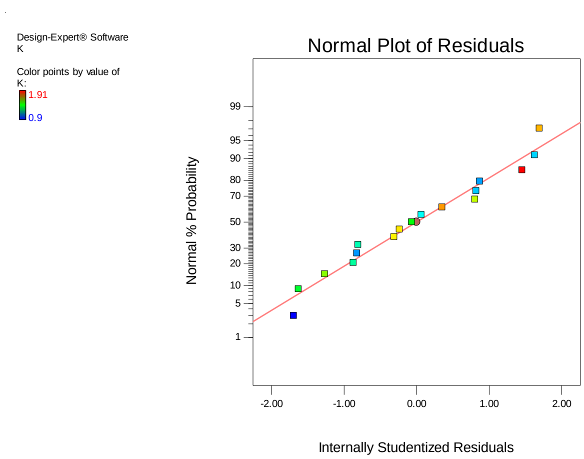
The plot of Residuals from RSM Models



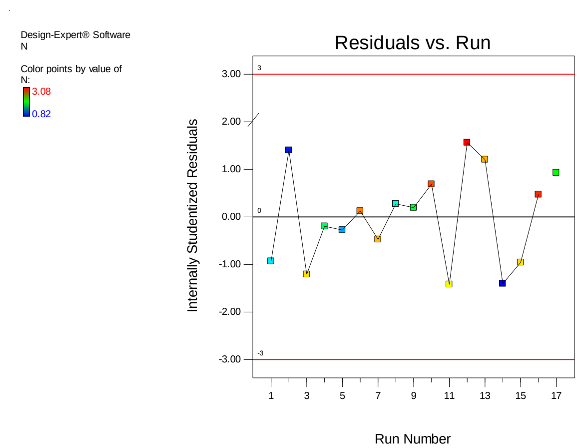
(N)



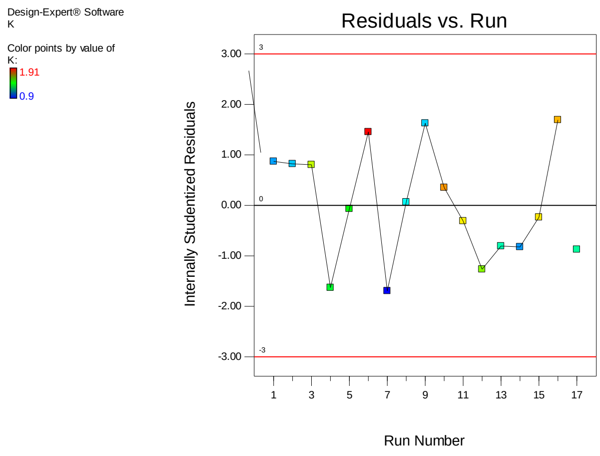
(P)

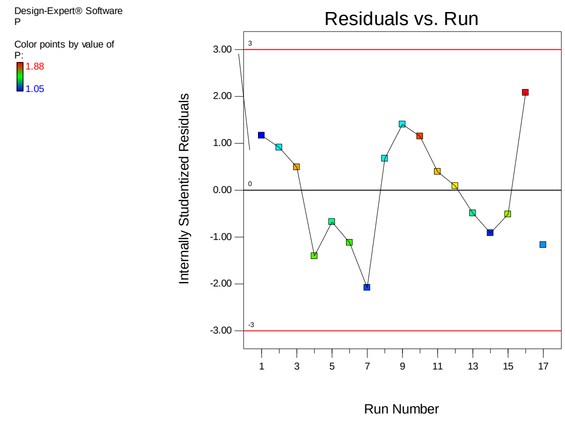


(K)



(N)


Design-Expert® Software
K
Color points by value of
K:
1.91
0.9
Residuals vs. Run
Internally Studentized Residuals
Run Number


Design-Expert® Software
P
Color points by value of
P:
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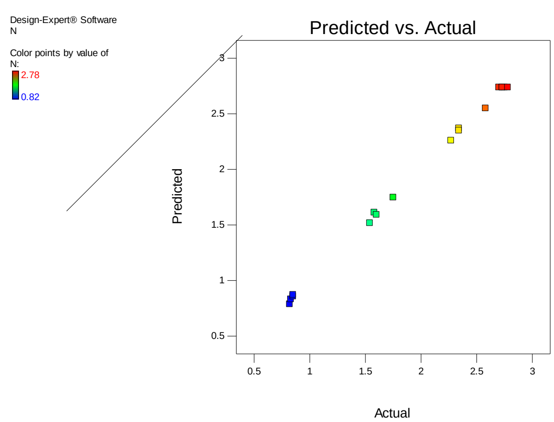
1.05
Residuals vs. Run
Internally Studentized Residuals
Run Number

(P) **(K)**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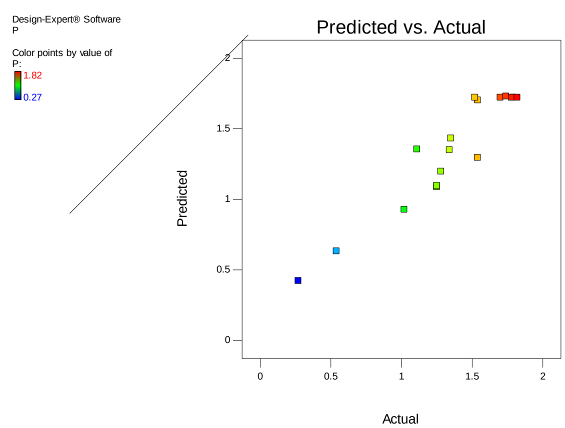
Color points by value of
N:
2.78
0.82
Predicted vs. Actual
Predicted
Actual


Design-Expert® Software
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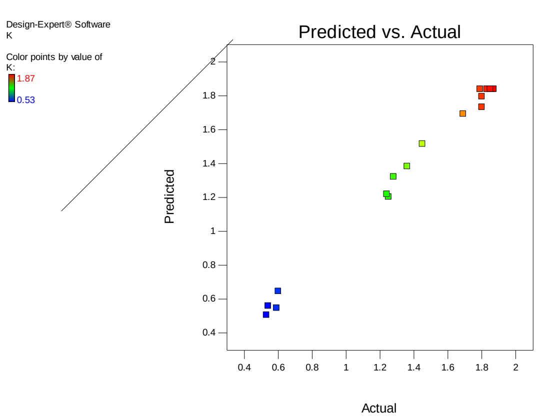
Color points by value of
P:
1.82
0.27
Predicted vs. Actual
Predicted
Actual

(N) **(P)**


Design-Expert® Software
K
Color points by value of
K:
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0.53
Predicted vs. Actual
Predicted
Actual

(K)