RESPONSE OF FUNGI TO DIFFERENT TYPES OF ORGANIC AMENDMENTS IN SOIL CULTIVATED TO AMARANTH

BY

Chinemelum Franklyn UZOWULU

AGR1700342

DEPARTMENT OF SOIL SCIENCE AND LAND MANAGEMENT

FACULTY OF AGRICULTURE

UNIVERSITY OF BENIN

BENIN CITY

MAY, 2024

CERTIFICATION

This is to certify that this research was carried out by **Chinemelum Franklyn UZOWULU** in the Department of soil Science and Land management, Faculty of Agriculture, University of Benin, Benin City.

Dr. I. Ogbemudia Date_____

Project Supervisor

Dr. (MRS) A. O. BAKARE Date_____

Ag. Head of Department

DEDICATION

This work is dedicated to Almighty God for his unquantifiable love and absolute faithfulness, the holy spirit for being my best friend and comfort throughout my studies.

ACKNOWLEDGEMENT

My utmost gratitude goes to God Almighty for seeing me through out the academic journey, I want to acknowledge the dean of faculty of Agriculture, Prof D. N. Izekor, Department of Soil Science and Land Management, The HOD Dr (Mrs) A. O. Bakare, My Project supervisor Dr Ikponmwosa Ogbemudia, thank you so much sir for the support and corrections, Prof. A. I. Ogboghodo, Prof. J. S. Ogeh, Prof. J. U. Chokor, Prof. E. R. Orhue, Prof. Ehigiator, Dr. (Mrs.) F. Okunsebor, Dr. A. Emomu, Dr. H. O. Kadiri, Dr. (Mrs.) V. I. O. Edosa, Mr E. Airueghian and Mr O. Bukola for their positive academic impact.

Sincere appreciation to My parents Mr and Mrs Uzowulu, My lovely siblings, Ekene, Mimi, my aunt Chioma, and my friends Godwin, Michael, DK, Sammy, Godswill. God bless you all for everything.

I will not leave out the esteemed members of SLM class of 2023 (BELLATORES) and my colleagues from other department of the Faculty of Agriculture, The Bellatores FYB committee, as well as AGR Class of 2023.

TABLE OF CONTENT

LIST OF TABLES

LIST OF FIGURES

ABSTRACT

This study investigated the response of soil fungi to various organic amendments; earthworm cast (EC), poultry manure (PM), and inorganic fertiliser (urea) and their impact on the growth of amaranth (Amaranthus cruentus L. The experiment aimed to bridge the knowledge gap regarding the effects of organic amendments on fungal communities and their potential benefits for sustainable amaranth production. The experiment was done using a complete randomised design (CRD) and replicated three times, using soil and a plastic container. Results showed that organic amendments significantly influenced the composition of soil fungal communities compared to the control treatment. Notably, Trichoderma spp., a fungus with high phosphate solubilization potential, was most abundant in urea-amended soil. However, poultry manure (PM) treatment yielded plants with the highest fresh and dry matter weight, likely due to increased nutrient availability and microbial activity stimulated by the breakdown of organic matter. The study showed that PM amendments improved soil health, promoting the growth of beneficial fungi like Trichoderma spp., and enhancing amaranth growth, potentially contributing to sustainable agricultural practices.

CHAPTER ONE

INTRODUCTION

Soil health and productivity are intricately linked to the activity and composition of the soil microbiota, particularly fungi. It is well known that a gram of undisturbed soil contains thousands of individual microbial taxa including fungi, which determine soil fertility and enhance plant growth and health (Vassilev and Mendes, 2024). Fungi facilitates nutrient cycling by breaking down organic matter, making essential elements readily available for plant uptake (Frąc *et al.,* 2018). This is fundamental for sustaining plant growth and maintaining ecosystem function. Fungi participates in nitrogen fixation, hormone production, biological control against root pathogens and protection against drought, and also play an important role in stabilisation of soil organic matter and decomposition of residues (Rashid *et al*., 2016). However, the proliferation of chemical fertilisation in soil–plant systems, has resulted in a decrease in soil natural fertility, optimal plant performance and microbial diversity (Huang *et al*., 2019). In addition to the overload of chemical fertilisers, an increasing number of stress factors, such as salinity, alkalinity/acidity, contamination, nutrient deficiency or drought, soil disturbance due to climate change, and various biotic factors, are affecting the overall soil–crop characteristics, yield and potentially sustainable food security (Vassilev and Mendes, 2024).

Amaranth, (Amaranthus cruentus L.) a pseudo cereal crop, thrives in these environmental conditions, demonstrating tolerance to drought, heat stress, and even marginal soils (Jamalluddin *et al*., 2021). These characteristics make it a promising alternative crop, particularly in the context of a changing climate and increasing demands for sustainable food production. However, optimising amaranth production necessitates a deeper understanding of the intricate interplay between the plant, soil properties, and the resident microbial diversity, with a particular focus on the role of soil fungi (Jing *et al*., 2022). Moreover, with the further

decline in microbial diversity, the prospective outlook of optimal production becomes increasingly slim. The decline in microbial diversity disrupts the ecological balance within the soil, hindering its ability to perform essential functions like nutrient cycling, decomposition, disease suppression and ultimately facilitating optimal plant growth (Bierza *et al*., 2023). This recognition of the detrimental effects of the use of inorganic fertilisers has fueled the rise of organic amendments. Organic farming with the use of organic amendments, such as compost, manure, and biochar, are increasingly employed in sustainable agriculture to improve soil fertility, enhance microbial activity and optimal crop production (Ye *et al*., 2020). These amendments provide a readily available source of carbon energy and nutrients for soil microbes, potentially altering the composition and activity of the fungal population (Reeve *et al*., 2016). Various literature have shown that compost application has the potential to lead to an increase in the abundance of saprotrophic fungi, responsible for decomposing organic matter (Alori *et al*., 2023). Additionally, compost amendments can favour the growth of beneficial AM fungi, potentially enhancing plant growth through improved nutrient acquisition. The best and most logical way to improve soil fertility and increase plant growth is to use and manage plant beneficial microorganisms and so therefore, understanding the specific effects of different organic amendments on the composition, activity and response of soil fungi is crucial for optimising their use in sustainable agriculture (Vassilev and Mendes, 2024). Matching the type of amendment to the desired fungal community composition can unlock the full potential of these practices for improving soil health and yield of crops such as amaranth. While the importance of soil fungi in maintaining healthy and productive soils is well established, the specific effects of various organic amendments on the composition of fungal diversity in an amaranth cropping system remain largely unexplored.

Aim

This research project aims to bridge this knowledge gap by investigating the response of soil fungi to different types of organic amendments with amaranth as the target crop.

Objectives

The objectives are to:

1. Determine the effect of organic amendments on the composition of the soil fungal and its response.
2. Determine the impact of the organic amendments on amaranth

CHAPTER TWO

LITERATURE REVIEW

2.1 Soil Fungi

Soil health, and the closely related terms of soil quality and fertility, is considered as one of the most important characteristics of soil ecosystems. The integrated approach to soil health assumes that soil is a living system and soil health results from the interaction between different processes and properties, with a strong effect on the activity of soil microbiota (Frac *et al.,* 2018). The soil microbiota is critical to plant performance, thus improving the ability of plant-associated soil probiotics is essential for establishing dependable and sustainable crop yields (Wang *et al*., 2023). Part of the soil microbiota exist the fungi, which is a diverse and functionally significant component of the soil microbiome, playing pivotal roles in nutrient cycling, soil formation and plant growth promotion. Their ability to degrade complex organic matter and mobilise essential nutrients makes them indispensable for maintaining soil fertility and ecosystem functioning (Vassilev and Mendes, 2024). Furthermore, Fungi possess the biochemical and ecological capacity to degrade environmental organic chemicals and to decrease the risk associated with metals, metalloids and radionuclides, either by chemical modification or by influencing chemical bioavailability (Harms *et al*., 2011). The trend towards energy- and cost-efficient passive remediation schemes, referred to as monitored natural attenuation, for the reclamation of contaminated land, further enunciates the importance of fungi and its role in the soil (Harms *et al.,* 2011). However, the intensive use of chemical fertilisers and unsustainable agricultural practices has led to a decline in soil microbial diversity, compromising soil health and productivity (Huang *et al.,* 2019). In this context, the exploration of sustainable practices, such as the application of organic amendments, has gained significant attention as a means to promote beneficial soil fungal communities and enhance crop yields (Ye *et al.,* 2020).

Soil fungi exhibit a remarkable diversity, encompassing a wide range of taxonomic groups and functional traits. They can be broadly classified into three main groups based on their lifestyles and interactions with other organisms: saprotrophic fungi, mycorrhizal fungi, and pathogenic fungi (Frąc *et al.,* 2018). Saprotrophic fungi, also known as decomposers, play a crucial role in the decomposition of organic matter, releasing essential nutrients into the soil (Alori *et al.,* 2023). They secrete a variety of extracellular enzymes, including cellulases, lignin-modifying enzymes, and proteases, which enable them to break down complex plant and animal residues (Rashid *et al.,* 2016). This process not only facilitates nutrient cycling but also contributes to the formation and stabilisation of soil organic matter, improving soil structure and water-holding capacity (Bierza *et al.,* 2023).

Mycorrhizal fungi, on the other hand, establish symbiotic associations with plant roots, forming an intricate network of hyphal filaments that extend into the surrounding soil (Bagyaraj & Ashwin, 2017). This symbiotic relationship is mutually beneficial, as the fungi receive carbohydrates from the plant, while the plant benefits from the fungi's ability to enhance nutrient and water acquisition from the soil (Jing *et al.,* 2022). Mycorrhizal fungi can be further divided into ectomycorrhizal and arbuscular mycorrhizal (AM) fungi, with the latter being more prevalent in agricultural systems and forming associations with a wide range of crop plants (Jing *et al.,* 2022).

Pathogenic fungi, while often perceived as detrimental, also play important roles in soil ecosystems. They can act as regulators of plant populations, contributing to plant diversity and ecosystem stability (Rashid *et al.,* 2016). Some pathogenic fungi can also act as biocontrol agents, suppressing the growth of other harmful pathogens or pests (Frąc *et al.,* 2018). The diversity and abundance of these fungal groups in soil are influenced by various biotic and abiotic factors, such as plant species, soil pH, moisture content, and nutrient

availability (Frąc *et al.,* 2015). Maintaining a balanced and diverse fungal community is crucial for optimal soil function and plant growth, as different fungal groups contribute to various processes and interact with plants in unique ways contributing to soil health and productivity (Vassilev & Mendes, 2024).

2.2 Effects of Organic Amendments on Soil Fungi

The application of organic amendments, such as compost, manure, and biochar, has been recognized as a promising strategy for promoting beneficial soil fungal communities and enhancing crop productivity (Ye *et al.,* 2020). These amendments can influence soil fungal communities through several mechanisms, including changes in nutrient availability, introduction of exogenous microorganisms, and alteration of soil physical and chemical properties. Compost, a product of aerobic decomposition of organic matter, is a rich source of organic carbon, nutrients, and diverse microbial communities (Alori *et al.,* 2023). Several studies have reported an increase in the abundance and diversity of saprotrophic fungi following compost application (Reeve *et al.,* 2016; Alori *et al.,* 2023). The readily available carbon and nutrients provided by compost can stimulate the growth and activity of these fungi, which play a crucial role in decomposing organic matter and facilitating nutrient cycling (Alori *et al.,* 2023). Additionally, compost amendments have been shown to promote the growth of beneficial mycorrhizal fungi, potentially enhancing plant growth through improved nutrient acquisition (Reeve *et al.,* 2016). Manure, another commonly used organic amendment, can also influence soil fungal communities. The specific effects of manure on fungal composition and diversity may vary depending on the type of manure (e.g., cattle, poultry, or green manure) and its chemical composition (Hafifah *et al.*, 2016). For instance, cattle manure has been reported to increase the abundance of saprotrophic and AM fungi, while poultry manure tends to favour the growth of saprotrophic fungi (Hafifah *et al.,* 2016).

Green manures, which involve the incorporation of plant residues into the soil, can also shape the fungal community by introducing specific carbon sources and altering soil nutrient profiles (Hafifah *et al.,* 2016). Biochar, a carbon-rich material produced through the pyrolysis of biomass, has gained attention as a soil amendment due to its potential benefits for soil health and carbon sequestration (Yao *et al.,* 2017). The effects of biochar on soil fungal communities have been widely studied, but the results are often variable and dependent on factors such as biochar feedstock, production temperature, and soil properties. Gao *et al* (2021) reported an initial suppression of fungal activity after biochar application, likely due to nutrient immobilisation or the presence of potentially inhibitory compounds. However, over time, biochar can promote fungal growth and activity as microbial communities adapt and utilise the biochar as a carbon source (Bolan *et al*., 2023). It is important to note that the effects of organic amendments on soil fungal communities can be influenced by various factors, including amendment type, application rate, soil properties, and environmental conditions (Reeve *et al.,* 2016; Gao *et al.,* 2021). Additionally, the specific composition of the fungal community can have implications for plant growth and soil health, as different fungal groups contribute to different processes and interactions within the soil ecosystem (Vassilev & Mendes, 2024).

2.3 Organic Amendments and Amaranth Cultivation

Amaranth (Amaranthus cruentus L.) is a highly nutritious pseudocereal crop that has gained increasing attention due to its ability to thrive under extreme soil conditions, drought tolerance, and potential for sustainable cultivation (Jamalluddin *et al.,* 2021). Due to its increasing importance, optimising amaranth production requires a comprehensive understanding of the intricate relationships between the plant, soil properties, and the resident microbial communities, especially fungi (Jing *et al.*, 2022). While the literature on the effects

of organic amendments on soil fungi in amaranth cropping systems is limited, some studies have explored the impacts of these amendments on amaranth growth and yield. For instance, a study by Japakurmar *et al.* (2021) investigated the effects of compost and biochar amendments on the growth and nutrient uptake of amaranth. They found that both amendments improved plant growth, with biochar having a more pronounced effect on nutrient uptake, particularly for phosphorus and potassium. However, the study did not specifically examine the effects on soil fungal communities. In another study, Jamalluddin *et al.* (2021) evaluated the impact of different organic amendments, including vermicompost, poultry manure, and biochar, on the growth and yield of amaranth under water-deficit conditions. Their results showed that vermicompost and poultry manure significantly enhanced amaranth growth, biomass, and grain yield, potentially due to improved soil fertility and water-holding capacity. While the study did not directly assess soil fungal communities, it highlighted the potential benefits of organic amendments for amaranth cultivation under water-stressed conditions. Despite these studies, there is a dearth of comprehensive research specifically investigating the response of soil fungal communities to various organic amendments in amaranth cropping systems. Understanding the effects of these amendments on fungal diversity and composition is crucial, as soil fungi play vital roles in nutrient cycling, plant-microbe interactions, and overall soil health (Vassilev and Mendes, 2024; Jing *et al.,* 2022).

2.4 Organic and Inorganic Amendments

The use of these organic amendments results in higher growth, yield and quality of crops. They contain macronutrients, essential micro nutrients, many vitamins, growth promoting factors like indole acetic acid (IAA), gibberellic acid (GA) and beneficial microorganisms (Sreenivasa *et al.,* 2010). Organic manures used as amendments can improve soil-water-plant relations through modifying bulk density, total porosity, soil water relation and consequently,

increasing plant growth and water use efficiency (Sreenivasa *et al.,* 2010). Nileemas and Sreenivasa (2011) stated that application of liquid organic manure as amendments promotes biological activity in soil and enhances nutrient availability to tomato crop. Awad *et al.,*(2012) stated that organic manure contains high levels of relatively available nutrients elements, which are essentially required for plant growth; moreover it plays an important role for improving soil physical properties. Sustainability in agro ecosystems involves environmentally friendly techniques based on biological and non- chemical methods (Bonato and Ridray, 2010). Organic fertilisers such as poultry manure, cow dung, compost, cattle rumen, food waste, farmyard manure are been used as organic amendments and it's been reported to provide significant quantity of nutrients which are supply to the soil in slow rate over a long period which reduce the nitrate losses in drainage water (Sreenivasa *et al.,* 2010). It improves the activities of microorganism in the soil as well improves water movement and good aeration of the soil among many other advantages but the quantity, transportation, labour and cost still remain as the challenges of commercial use of organic fertilisers.

Inorganic fertilisers are also been used by farmers in the production of crops and vegetables and many researchers have documented reports on the increased yield of various crops through its application (Akanbi, 2012). One of the important factors for better production of tomato is proper use of fertilisers. Nitrogen requirement for the growth of plants is comparatively larger than other elements and it plays a key role in tomato growth alongside potassium when it is applied during growing stage. Nitrogen promotes plant organs development and results in abundant chlorophyll except root growth that is relatively poor (Lincoln *et. al.* 2006) and its deficiency results in stunted growth of the plant, leading to premature flowering and short growth cycle. To achieve maximum yield, timing of fertilisers application and appropriate source is also necessary for improved nitrogen management. It was asserted that inorganic fertilisers can improve crop yields and soil pH, total nutrient

content, and nutrient availability. (Islam *et al*., 2018). Among the commonly used inorganic fertilisers by vegetable farmers are the NPK and Urea but the scarcity and high price of these fertilisers remain a challenge facing crop production (Akanbi, 2012).

2.5 Poultry Manure

Poultry manure is used as soil amendment and contains fairly high nutrient composition particularly nitrogen than other sources of animal manure (Sunassee 2012). Poultry manure promotes and enhances the growth and yield of plants such as vegetables e.g tomatoes, because it is not all macro and micro nutrients that are readily available for the plant uptake, and this could bring about slow growth and poor yield (Ismail *et al.,* 2014). Wilson *et al.,* (2016) and Sharpley *et al.,* (2010) described poultry manure as a mixture of bedding material (wood shaving, sawdust, grain husk, etc.) poultry dropping, feed, water, vaccine, feather. It contains all 13 of the essential plant nutrients that are used by plants. These include nitrogen (N), phosphorous (P), potassium (K), calcium (Ca), magnesium (Mg), sulphur (S), manganese (Mn), copper (Cu), zinc (Zn), chlorine (Cl), boron (B), iron (Fe), and molybdenum (Mo). Plant nutrients originate from the feed, supplements, medications, and water consumed by the birds. Using poultry manure as a fertilizer for plants may provide a portion, or all, of the plant requirements (Griffiths, 2011). The amount of nutrients provided depends on the nutrient content of the manure (l kg of nutrient/t of manure) and the amount of manure applied (t of manure/hectare). The amount of manure applied per acre (called the application rate) is typically based on the nitrogen needs of the plants. However, phosphorous requirement can also be used to determine the application rate (Griffiths, 2011).

According to Tabler and Berry, (2013), they reported that the composition of chicken manure varies according to age of the chicken, moisture content and age of the manure, kind and amount of litter, and storage and handling practices. The only sure way to know the composition is to analyze the material. Conversion of the uric acid and urea to ammonia is

rapid during the first 2 weeks after the addition of manure to a warm moist soil, but conversion of the organic forms of nitrogen to an available form is slow during the first 4 weeks after its addition (Griffiths, 2011). Phosphorus is primarily organic and becomes available as the manure decomposes, but all may not be available until the next cropping or season (Sharpley *et al.,*2010). Other plant nutrients become available during decomposition of chicken manure and, like phosphorus, may not all be available until the next cropping season (Tabler and Berry, 2013).

2.6 Earthworm Cast

Earthworm cast, also known as vermicompost, is a natural and nutrient-rich fertilizer produced by earthworms as they break down organic matter (Sunassee, 2012). It contains a balanced mix of nutrients, including nitrogen, phosphorus, potassium, and other micronutrients essential for plant growth (Ismail et al., 2014). Earthworm cast has been shown to improve soil fertility, structure, and overall health, leading to enhanced plant growth and yield (Wilson et al., 2016). Like poultry manure, earthworm cast is a valuable source of plant nutrients, including nitrogen, phosphorus, potassium, calcium, magnesium, sulfur, manganese, copper, zinc, chlorine, boron, iron, and molybdenum (Sharpley et al., 2010). The nutrient content of earthworm cast can vary depending on factors such as the type of organic matter being broken down, the age of the worms, and the moisture content of the cast (Tabler and Berry, 2013). According to Griffiths (2011), earthworm cast is a slow-release fertilizer, with nutrients becoming available to plants as the cast decomposes. The conversion of organic forms of nitrogen to available forms is slow, taking several weeks to occur. Similarly, phosphorus and other nutrients become available during decomposition, but may not all be available until the next cropping season (Sharpley et al., 2010).Overall, earthworm cast is a valuable natural fertilizer that can improve soil fertility and plant growth, making it a useful alternative to synthetic fertilizers.

Summarily, the application of organic amendments, such as compost, manure, and biochar, has been recognized as a promising strategy for promoting beneficial fungal communities and enhancing crop productivity in sustainable agriculture systems. While several studies have investigated the effects of these amendments on soil fungal communities in various cropping systems, there is a lack of research specifically focused on amaranth cultivation, which is becoming increasingly important for global food security. Given the potential of amaranth as a sustainable and resilient crop, understanding the response of soil fungi to different organic amendments in amaranth cropping systems is of utmost importance. Furthermore, the existing literature reveals variations in the effects of organic amendments on fungal communities, which can be influenced by factors such as amendment type, application rate, soil properties, and environmental conditions. These variations highlight the need for context-specific studies to develop targeted strategies for optimising soil fungal communities and maximising the benefits of organic amendments for amaranth production. To address these research gaps, this study employs a culture-dependent sampling or methodology. It entails cultivating amaranth under the influence of different organic amendments. This approach would not only provide insights into the composition of soil fungal communities but also enable the identification, characterization, and response of specific fungal taxa that may play crucial roles in amaranth growth and soil health. By determining the response of soil fungi to different organic amendments in amaranth cropping systems, this research has the potential to contribute valuable knowledge to the field of sustainable agriculture. The findings can inform the development of targeted amendment strategies to promote beneficial fungal communities, improve soil fertility, and enhance the productivity and resilience of amaranth cultivation. Ultimately, this research may pave the way for more sustainable and environmentally friendly practices in amaranth production, contributing to food security and environmental conservation efforts.

CHAPTER THREE

3.0 MATERIALS AND METHODS

3.1 Study Area

This study was carried out at the Ugbowo campus of the University of Benin, Benin City in Nigeria. The area lies between latitude 6°23’37” and 6°24’26” North and longitude 5°36’25” and 5°38’09” East, with coastal plain sand as the parent material. The natural climate is Humid Tropics. The natural vegetation is rain forest. The rainy season is bimodal with peak in July and September. Average rainfall is between 1500mm-2500mm annually. Mean maximum and minimum temperature are 31°C and 22°C. . The soils are predominantly derived from Coastal plain sands underlaid by sedimentary rocks which were characterized mainly by sands, grits and clay. (Kadiri, 2022).

3.2 Sample Collection Before And After Planting.

The soil used for this research was obtained from the Faculty of Agriculture, University of Benin, Edo state, Nigeria; the area lies on latitude 6°24'05" N and longitude 5°37'31" E. The soil was sampled from ; depth 0-15cm using soil auger at different points and were bulked into a large composite sample. The soil was sieved to remove debris and weighed 15kg into 12 plastic container\perforated bucket (four treatment with three replicate for each sample). Sample was obtained with the use of a sanitized trowel and please in Petri dish for laboratory analysis.

3.3 Fertilizer Application.

Poultry manure wad obtained from the Faculty of Agriculture Animal Farm, University of Benin, Edo state, Nigeria. The cured poultry manure was applied at the rate of 0.5kg into three sampled filled perforated buckets each, one weeks before planting. Earthworm caste was obtained from a reserved forest at Jemikin Okitipupa, Ondo State, Nigeria. Earthworm

caste sample was randomly collected from the top layer of the soil from the forest with coordinates Latitude 6°30'39"N and Longitude 4°45'14"E. Earthworm caste was applied at the rate of 2kg two weeks before transplanting. While inorganic fertilizer was purchased from an Agricultural input dealer at New Benin, Benin city, Edo state; And was applied at the rate of 3grams into threesample filled perforated bucket each, at two weeks after transplanting.

3.4 PLANTING MATERIALS.

Amaranthus hybridus L seeds were bought from the local market at New Benin Market, Benin City. The amaranth was sowed two weeks after organic manure was incorporated into the soil sample. Planting was done with three seeds per bucket due to correction and viability.

3.5 EXPERIMENTAL PROCEDURE

A total of twelve (12) plastic buckets were used, resulting from 3 plastic buckets per replicate of four treatments. The experiment was laid out in a Completely Randomised Design (CRD). The plants were watered daily throughout the period of the experiment. Weeding was exercised every two weeks. Parameter reading was conducted every other week. Thereafter, harvesting was done every other day in a week from the 9th – 14th week.

The treatments were made up of:

1. 3 replicates with zero organic / inorganic amendment applied and this served as Control (labelled as C1, C2 and C3).
2. 3 replicates with cow dung applied, (labelled as N1, N2 and N3).
3. 3 replicates with organic fertiliser (Poultry manure) applied, (labelled as PM1, PM2 and PM3).
4. 3 replicates with earthworm caste applied (labelled EC1, EC2 and EC3).

3.6 PLANT PARAMETERS MEASURED

Plant height

The height was measured from the soil level to the point where the flag leaf emerges from the plant. The height of all the plants per treatment was measured and the mean calculated and the unit expressed in centimetres (cm) at 2 weeks interval.

Number of leaves

The number of leaves per treatment was obtained by counting all the fully open leaves per plant. The mean was calculated and recorded at 2 week interval.

Girth

The girth was measured at the base of the plant from the soil level. The girth of all plants per treatment was measured, the mean was calculated and the unit was expressed in centimetres (cm) at 2-week intervals.

3.7 SOIL LABORATORY ANALYSIS

3.7.1 Available Phosphorus (P) Determination (Bray-1 Method)

Five grams (5g) of soil was weighed into the extractor cup and 30ml of Bray-1 solution was added into the cup. The solution was stirred in a mechanical shaker for 5 minutes. The solution was then filtered into a reagent bottle. 1ml extract (aliquot) was pipetted into a 50ml volumetric flask. 6 ml of distilled water with 2 ml of colour developing reagent was added and mixed well. 1 ml of ascorbic acid solution was then added. The solution was left for about 10 minutes for the colour to develop. The solution was measured at about 650 nm in a visible-range spectrophotometer. A graph of absorbance against parts per million (ppm) standard and ppm P interpolation was done.

Calculated ppm P (µg P/kg soil) = R × 30/5 = R × 6 (Hossner, 1996; Ibitoye, 2008).

3.7.2 Nitrogen Determination (Kjeldahl method)

Digestion

One gram (1g) of air-dried soil of fine tilth was weighed into 250 ml kjeldahl flask. One catalyst tablet (consisting of $CuSO_4$, K_2SO_4 and a pinch of Selenium) was added. 20 ml of concentrated H_2SO_4 was added to the solution and the mixture was heated till it became clear (light green colour). The solution was then allowed to cool after it was removed from heat. About 10 ml of water was added and the content was filtered using Whatman 45 filter paper into 100 ml volumetric flask. It was made up to mark and shaken together for proper mixing (Hossner, 1996; Ibitoye, 2008).

Distillation

10 ml aliquot was transferred into 500 ml Kjeldahl flask and 30 ml of water was added. 15 ml of NaOH (excess base) was added and heat was applied. 2.5 ml distillate was collected in a 5 ml boric acid indicator. The NH_4 – Nitrogen was determined by titrating distillate with 0.01M standard HCl until colour change from green to pink was observed.

% Nitrogen calculated as follows: $T \times M \times 14/1000 \times V1/V2 \times 100/W$

Where;

T = titre value of sample

M = concentration of HCl used for titration in molarity

1000 = constant

V1 = final volume of digest

V2 = volume of aliquot used for distillation

W = weight of sample used.

3.7.3 Organic Carbon Content

1g of soil was weighed into a 250ml conical flask and 10 ml of potassium dichromate was then added to the flask. 20 ml of concentrated H_2SO_4 was added vigorously. It was allowed to

stay for 30 minutes and then 100ml of water was added to the solution. 5 drops of Ferroin indicator was then added and the mixture was titrated with 0.5N ferrous sulphate ($FeSO_4.7H_2O$). A blank titre was also taken but without sample. Percentage organic carbon was calculated as follows:

Where,

B = blank titre value

T = sample titre value

N = concentration of $FeSO_4$ in Normality

F = correction factor (1.33)

Percentage organic carbon was converted to percentage organic matter by multiplying the results by 1.724 and answers were reported in g/Kg by multiplying percentage organic carbon value or percentage organic matter value by 10 (Hossner, 1996; Ibitoye, 2008).

3.7.4 Exchangeable Bases (K, Ca, Mg, Na) Determination

10 g of soil was weighed in a 250ml soil shaking bottle. 100 ml of 1N ammonium acetate was added and was shaken in a mechanical shaker for 1 hour. The soil was filtered using a Whatman 45 filter paper into a 100ml volumetric flask and made up 100ml mark with ammonium acetate solution and stored in a 100ml plastic reagent bottle. Potassium (K) and Sodium (Na) were read in a 'Jennway model' flame photometer and Calcium (Ca) and Magnesium (Mg) were read in a Unican series 969 model Atomic Absorption Spectrophotometer (AAS) (Hossner, 1996; Ibitoye, 2008).

3.7.5 pH determination

20g of soil was weighed into a 50 ml beaker and 20 ml of distilled water was added. The mixture was stirred for 30 minutes intermittently and pH read in a standardised pH metre (Hossner, 1996; Ibitoye, 2008).

3.7.6 Exchange Acidity

5g of air-dried soil was weighed into a shaking bottle and 50mlk of 1M KCl was added and mixture shaken for 1 hour in a mechanical shaker. The soil was filtered into a 100ml volumetric flask and was made up to 100 ml mark. 23ml of soil extract was measured into a 250ml conical flask. Five drops of phenolphthalein indicator were added. Mixture was titrated with 0.05N NaOH to the pink colour end point.

Calculation for E.A on: (Meq/100g)

3.7.6.1 For Exchange Aluminium in soil

5ml of 3N NaF was added to the titrated extract and the mixture was titrated with 0.05N HCl to end point.

Therefore, exchange Al^{3+} = (Meq/100g)

Where,

V = titre value

W = weight of sample used

(Hossner, 1996; Ibitoye, 2008).

3.7.7 Particle size determination (Hydrometer method)

51g of air-dried soil (2mm) in a 250 ml beaker and 50 ml of Calgon (sodium hexametaphosphate + sodium carbonate) were added to the soil sample. 100 ml of distilled water was added and stirred vigorously for 1 minute using a glass rod and allowed to stand for 30 minutes. The suspension was transferred into a sedimentation cylinder and made up to a 1-litre mark with distilled water.

3.8 IDENTIFICATION AND ENUMERATION OF TOTAL VIABLE BACTERIA AND FUNGI

3.8.1 Culture Media Used

➢ Potato-dextrose agar preparation

This medium was prepared from commercially available dehydrated powder. In the preparation, 39g of potato dextrose agar powder was weighed and dissolved in 1000ml distilled water in a conical flask then shaken until the agar dissolved. The flask was then corked with cotton wool and aluminium foil to avoid contamination. The prepared medium was placed in an autoclave for sterilization at 121°C, 15 lbs pressure for 15 minutes. It is used for isolation and enumeration of fungi.

➢ Nutrient agar preparation

This was used to culture non-fastidious organisms and for bacterial heterotrophic plate counts. In the preparation, 28g of nutrient agar powder was weighed on the weighing balance and dissolved in 1000ml distilled water in a conical flask then shaken until the agar dissolved; the flask was then corked with cotton wool and aluminium foil to avoid contamination and placed in the autoclave for sterilization at 121°C, 15 lbs pressure for 15 minutes.

Glassware

- Disposable petri-dishes/plates
- Test tubes and test tubes rack
- Conical flasks and beakers
- MacCartney bottles and Durham's tubes
- Spirit lamps and disinfectant
- Slides and cover slip
- Bijoux bottles

Sterilizing Materials and Measures

• Autoclave

• Bunsen burner

• Oven

All glassware were properly washed with water, detergent, disinfectant and brushes, and were sterilised in an air oven at 160°C for 2 hours to achieve maximum sterilisation.

Other materials include

• Weighing electronic balance

• Cotton wool and aluminium foil paper

• Inoculating loop

• Refrigerator

• Inoculating needle

• Incubator

• Sterile distilled water

• Whatman filter paper

• Micropipette

• Microscope etc.

Reagents

• Methylated spirit

• 70% ethanol

• Crystal violet

• Grams iodine

• Safranin

• Carbon fusion

• Oil immersion

• Lactophenol blue

3.9 SAMPLE COLLECTION

Working in an aseptic environment, the soil samples were obtained with sterile spatula and transferred to sterile polythene bags. 3 samples were collected and were taken to the laboratory in sterile plastic containers for microbiological analysis.

3.9.1 Sample processing procedure

A stock solution of each sample was prepared by weighing 1g of each sample into 9 ml of sterile distilled water to make a 10% stock solution. This solution constitutes the first dilution. Aliquot of the desired dilutions were introduced to the Petri plate with the help of a micropipette.

3.9.2 Microbiological analysis (serial dilution)

0.1ml of each stock solution was dispensed into a sterile test tube containing 9.9ml of sterile distilled water to give a 1/1000 dilution which represents the 3rd dilution with 0.1ml aliquot of the 3^{rd} dilution transferred to another 9.9ml of sterile distilled water to give the 5^{th} dilution of 1/100000 fold.

0.1ml of each of the used dilutions was dispensed into nutrient agar and potato dextrose agar by the pour plate methodology. The aliquot sample of these dilutions was duplicated for confirmation and to check distribution of the cells in the diluents.

The inoculated plates were then incubated at 37°C for 24 hours for nutrient agar and 48-72 hours for potato dextrose agar at 25°C. After incubation, discrete colonies on the nutrient agar plates and potato dextrose agar plates were counted and the unit expressed in cfu/g.

3.9.3 Plate count

The bioload of the microorganisms is expressed in colony forming units (cfu). The agar plates having the culture colonies after incubation were divided into four and two vertically

opposite sections, were counted and multiplied by two. This was multiplied again by the dilution factor and gave the colony forming unit per gram (cfu/g) of the sample.

3.9.4 Identification

Pure culture isolation

Fresh nutrient agar plates, mannitol salt agar plates, MacConkey agar plates, streaked inoculated for pure culture from plates of different colonies that were inoculated at 37°C overnight. This was done in order to identify different isolates of bacterial colonies based on appearance and morphology (form and structure). This permits the selection and transfer of different species from mixed culture and transfer of a single colony to a sterile medium for cultivation of pure culture.

Fungi identification

Characteristics and identification of fungi isolates using gram staining

Procedure

I. A thin film of the test organism was made on a glass slide by heat fixing over a Bunsen burner with the help of an inoculating loop.

II. The glass slide was stained with crystal violet for 60 seconds and flooded with water.

III. Lugol iodine was applied as a mordant for 60 seconds and flooded with water.

IV. Acetone-ethanol mixture was used to decolourise it for 20 seconds and flooded with water.

V. The slide was counterstained with Safranin for 60 seconds and flooded with water.

VI. The slide was allowed to air-dry in a slanting position.

VII. Examination of the slide with an oil immersion was carried out under the microscope..

3.10. STATISTICAL ANALYSIS

Plant and soil parameters measured were subjected to Analysis of Variance (ANOVA) using GENSTAT 8th edition while Duncan's New Multiple Range Test was used to separate the

means at 5% level of probability. Shannon and simpson were used to quantify and identify the diversity of microbial communities

.

CHAPTER FOUR

4.0 RESULTS AND DISCUSSION

Table 1 shows the physical and chemical properties of the soil. Table 2 shows the fungi species and phosphate solubilization abilities in Soils

4.1 PHYSICAL PROPERTIES

4.1.1 Particle Size Distribution

The results (Table 1) of the particle size distribution of the soils incorporated with different organic manure showed a sand fraction of 806.7 g kg-1 in the control experiment (no organic manure), 806.7 g kg-1 in soils fertilised with urea (N), 783.3 g kg-1 for soils fertilised with earthworm cast (EC) and 826.7 g kg-1 for soils fertilised with poultry manure (PM). Silt fractions ranged from 40 g/kg in control to 76.67g/kg in PM. Soils fertilised with poultry manure and earthworm cast (EC) displayed a higher silt content (76.67 g/kg), while soil fertilised with urea had a silt content of 46 g/kg. Clay content decreased, with 153.30 g/kg of sand in the control, 146.70 g/kg in urea, 140 g/kg in Earthworm cast and 106.7 g/kg in poultry manure. These observations are in tandem with similar studies like Ye *et al.,* 2020 and Alori *et al.,* 2023. Based on the values, all treatments resulted in a sandy soil texture (percentage of sand > 70%). There were no significant differences ($p > 0.05$) in particle size distribution between treatments.

4.2 CHEMICAL PROPERTIES

4.2.1 pH

The pH levels of the soil ranged from generally acidic in all treatments, ranging from 4.73 (urea amended soil) to 5.6 (poultry manure amended soil). The pH levels were higher among

organic treatment than inorganic treatment and control. There were no significant differences in pH between the treatment types. The lack of significant pH difference despite using organic amendments could be due to the study's timeframe. Some organic matter decomposition processes can generate organic acids that might temporarily decrease soil pH. However, the buffering capacity of the soil and the specific type of organic amendment can influence the overall effect on pH. Long-term studies (Reeve *et al.,* 2016) suggest that organic amendments may gradually increase soil pH, which was observed minimally in the study. As observed by Bolan *et al.* (2023) organic amendments can stimulate beneficial microbes and contribute to the breakdown of organic matter and the release of nutrients while also consuming protons (positively charged hydrogen ions) during respiration. This can lead to a gradual increase in soil pH over time.

4.2.2 Organic Carbon

Organic carbon was highest in EC (20.28 g/kg) due to the high organic matter content of the soil, and lowest in the control (10.21 g/kg). The organic carbon content of soil fertilised with urea (N) was 11.29 g/kg while PM treatment showed a higher content of 15.31 g/kg. This indicates that organic amendments increased substantially the level of soil organic carbon compared to the control and inorganic treatment. This observation is in concert with Reeve *et al.* (2016) who highlighted the connection between organic amendments and improved soil health, including increased organic matter content.

4.2.3 Total Nitrogen

Total nitrogen content in the soil was highest for the soil treatment with earthworm cast (1.47 g/kg) followed by soil treated with poultry manure (1.11 g/kg), then soil treated with urea fertiliser (0.82 g/kg) and control (0.74 g/kg). This suggests that both organic amendments (PM and EC) increased the total nitrogen content compared to the control and inorganic fertiliser. This observation is in concert with Alori *et al.,* and Hafifah *et al.,* who highlighted

the potential of organic amendments like PM and EC to decompose overtime releasing a high volume of essential nutrients like Nitrogen into the soil. There were no significant differences in total nitrogen between treatments.

4.2.4 Available Phosphorus

Available phosphorus was significantly higher ($p < 0.05$) in the PM treatment (235.17 mg/kg) compared to all other treatments (control: 24.96 mg/kg, N: 26.93 mg/kg, EC: 25.19 g/kg). This suggests that poultry manure was the most effective amendment in increasing available soil phosphorus. This is in line with Wang *et al.* (2023). While Wang *et al*. (2023) focused on beneficial fungi and plant production, their study suggests that some amendments like poultry manure can harbour microorganisms with phosphate-solubilizing capabilities. The presence of such microbes in PM could have facilitated the release of bound phosphorus in the soil, leading to increased availability.

4.2.5 Exchangeable Cations (K, Na, Mg, and Ca) and Acidity (H and Al)

The table (2) displays the concentration of exchangeable cations (Ca, Mg, K, and Na) and exchangeable Al and H in cmol/kg. PM amendment resulted in the highest levels of Ca (3.03 cmol/kg) and Mg (0.91 cmol/kg) compared to other treatments. This was followed by EC with Ca levels of 2.33 cmol/kg. Conversely, control treatment had the highest exchangeable Al (0.18 cmol/kg) and H (0.43 cmol/kg), indicating higher potential soil acidity. Higher levels of Ca and Mg indicate higher fertility status, due to the presence of organic amendments, which facilitate the presence of soil microbiome and nutrient disbursement enabling optimum growth of plants.

4.2.6 Cation Exchange Capacity (CEC)

Cation exchange capacity (CEC) represents the soil's ability to hold onto positively charged ions (cations). The table shows the CEC values for each treatment. PM treatment had the highest CEC (5.46 cmol/kg), followed by EC (3.76 cmol/kg) and control and N (both at 2.74 cmol/kg). This suggests that organic amendments, particularly PM, improved the soil's capacity to retain essential cations, which is paramount for soil microbes and plant development.

4.2.7 Potassium

Available potassium content showed the following trend: PM (0.29 cmol/kg) > EC (0.11 cmol/kg) > control (0.09 cmol/kg) > N (0.07 cmol/kg). While PM and EC had slightly higher values, there were no statistically significant differences ($p > 0.05$) between any treatments for potassium. PM might have contained slightly more available K compared to EC and the other treatments. However, the total K content in the soil and the texture of the soil might have been a limiting factor, leading to the non-significant differences observed. Nonetheless, the study observations are in tandem with previous literature like Reeve *et al.* (2016), and Hafifah *et al.* (2016) which linked the potential of organic amendments to improve soil quality, and have higher content of nutrients such as K present.

Table 1: Some Physical and Chemical Properties of Soil After Experiment

Treatment	Sand	Silt	Clay	pH(1:2)	EC	Organic C	Organic M.	Total N	Avail. P	Ca	Mg	Na	K	Al	H	ECEC	%BS
	g/kg	g/kg	g/kg	H_20	µS/cm	g/kg	g/kg	g/kg	mg/kg	.....cmol/kg…..							

Control	806.70^{ab}	40.00^{c}	153.30^{c}	5.39^{a}	44.80^{b}	10.21^{b}	17.60^{b}	0.74^{b}	24.96^{b}	0.82^{b}	0.35^{b}	0.88^{bc}	0.09^{b}	0.43^{ab}	0.18^{b}	2.74^{c}	77.94^{b}
Urea	806.70^{ab}	46.67^{bc}	146.70^{a}	4.73^{b}	106.30^{b}	11.29^{b}	19.46^{b}	0.82^{b}	26.93^{b}	0.78^{b}	0.43^{b}	1.07^{a}	0.07^{b}	0.73^{a}	0.35^{a}	3.42^{bc}	69.35^{b}
Earthworm Cast	783.30^{c}	76.67^{a}	140.00^{a}	5.43^{a}	73.90^{b}	20.28^{a}	34.98^{a}	1.47^{a}	25.19^{b}	2.33^{a}	0.41^{b}	0.72^{c}	0.11^{b}	0.07^{b}	0.12^{b}	3.76^{b}	94.67^{a}
Poultry Manure	826.70^{a}	76.67^{ab}	106.70^{b}	5.63^{a}	241.70^{a}	15.31^{a}	26.41^{a}	1.11^{a}	235.17^{a}	3.03^{a}	0.91^{a}	0.95^{ab}	0.29^{a}	0.09^{b}	0.35^{b}	5.46^{a}	95.00^{a}
SED	12.47	9.13	9.13	0.17	38.90	0.94	1.62	0.07	17.99	0.23	0.09	0.07	0.04	0.22	0.05	0.39	5.34

Table 2. Fungi Species and Phosphate Solubilization Abilities in Soils

	IAA production	Phospate Solubilization	Colony Diameter	Halozone Diameter	PSI (day1)	PSI (day2)	PSI (3days)
Trichoderma sp.	+	+	5	14	2.80	4.20	6.33
Cladosporium spp.	-	-	-	-	-	-	-
Aspergillus niger	+	+	5	10	2.00	3.00	4.52
Rhodotorula spp.	-	-	-	-	-	-	-
Penicillium sp.	+	+	5	8	1.60	2.40	3.62

Mucor mucedo	-	-	-	-	-	-	-
Aspergillus flavus	-	-	-	-	-	-	-

4.3 Fungi Species and Phosphate Solubilization

Table two (2) indicates the presence of various fungal species in the soil and their potential for phosphate solubilization. Species present include *Trichoderma spp Cladosporium spp, Aspergillus niger, Rhodotorula spp, Penicillium spp, Mucor mucedo, and Aspergillus flavus*. The table suggests that the amendments influence the resident fungal communities with varying abilities to solubilize phosphate, potentially improving plant growth by making phosphorus more bioavailable. This is relevant to studies like Bolan et al. (2023) which discuss the potential of organic amendments to stimulate beneficial microbes. Trichoderma spp. appears to be the most promising candidate for phosphate solubilization because it has the highest PSI (Phosphate Solubilization Index). Phosphate solubilization refers to the ability of the fungus to convert insoluble phosphate forms in the soil into soluble forms that plants can readily absorb. A "+" indicates the fungus can solubilize phosphate, while a "-" indicates that the fungus can't. This finding aligns with studies like Saravanakumar *et al.* (2013) which highlight the phosphate-solubilizing abilities of Trichoderma species. Furthermore, Hafifah *et al.* (2016) emphasised the potential of organic amendments to improve soil quality, potentially creating a more favourable environment for beneficial microbes like phosphate-solubilizing fungi to thrive. Overall, the table provides valuable insights into the potential role of fungal communities in phosphate cycling within the experiment. The presence of

phosphate-solubilizing fungi, particularly Trichoderma sp., suggests a possible mechanism by which amendments could enhance plant growth through improved phosphorus availability.

Plant and Fungi Growth Parameters

Figure 1 illustrates the level of fungi present in soils treated with different organic amendments. N recorded the highest level of fungi present, followed by PM, EC and control. This suggests a high concentration of fungi in N-amended soils. This is as a result of urea's readily available form of nitrogen source for fungi, which potentially facilitated its rapid population growth compared to other treatments.

Figure 2 indicates a potential influence of organic amendments on amaranth fresh weight (grams). PM treatment yielded plants with the highest average fresh weight, followed by N, EC, and control treatments. This is because organic amendments like PM and EC decompose over time, releasing nutrients like nitrogen, phosphorus, and potassium into the soil. These nutrients are essential for plant growth and development. The table showing soil properties (Table 2) show that PM treatment resulted in higher levels of available nutrients compared to other treatments. This could explain the potentially higher fresh weight observed in PM-amended soil. While N amendment provides nitrogen, its availability for immediate plant uptake might be lower compared to organically derived nitrogen from decomposed PM or EC. Furthermore, PM and EC can stimulate the growth and activity of beneficial soil microbes, including bacteria and fungi. These microbes can further contribute to nutrient cycling, breakdown of organic matter, and production of plant growth regulators, indirectly influencing plant growth and potentially leading to variations in fresh weight.

Figure 3 shows the dry matter weight of amaranth plants (g) following application of different organic amendments. Plants grown in PM-amended soil exhibited the highest dry matter weight, followed by EC, N and the control treatment. This suggests that organic

amendments, particularly PM, can positively influence the dry matter weight of amaranth plants significantly.

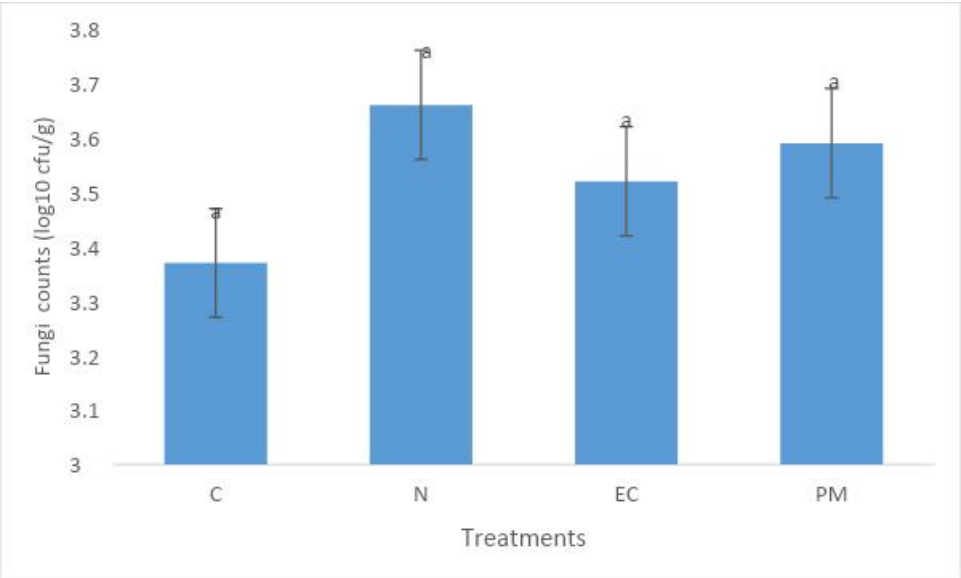

Figure 1. Fungi Count (cfu) in different soil treatment

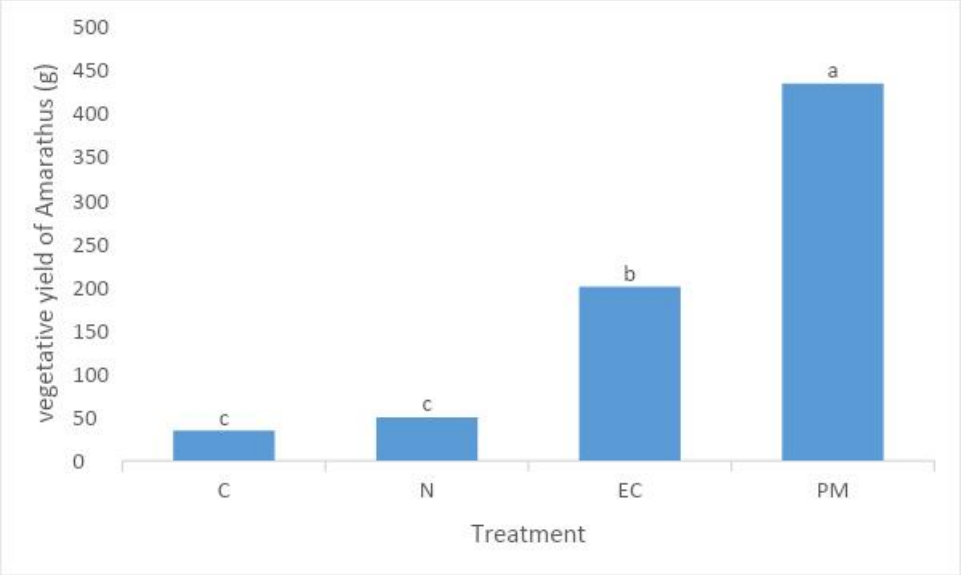

Figure 2. Fresh weight of Amaranth

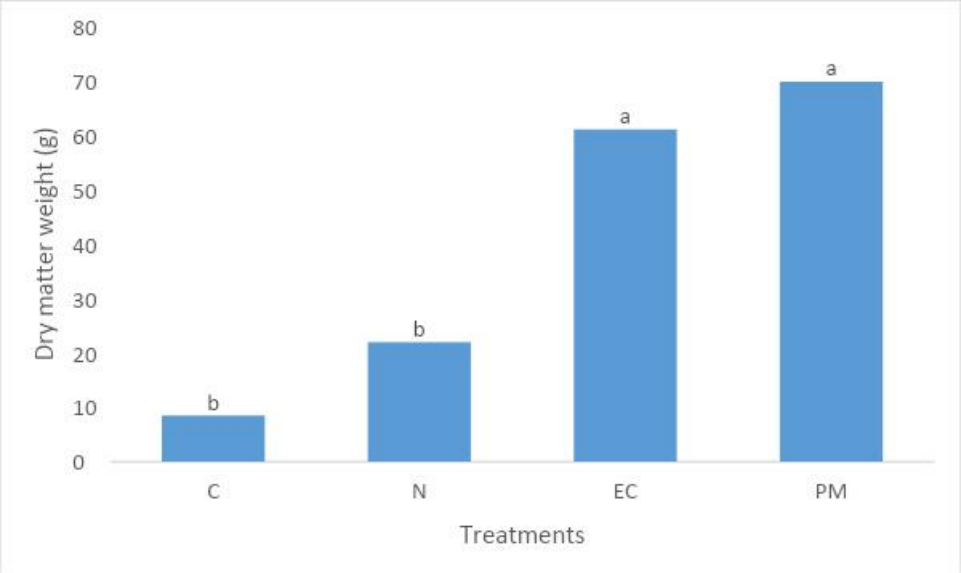

Figure 3. Dry Matter Weight of Amaranth

CHAPTER FIVE

5.1 CONCLUSION

From the study, organic amendments, particularly poultry manure, have shown promise for improving soil properties, microbial activity and plant growth. It was observed that organic amendments (earthworm cast and poultry manure) increased organic carbon, total nitrogen, available phosphorus, and cation exchange capacity compared to the control and inorganic fertiliser (urea). Poultry manure had the highest levels of nitrogen, available phosphorus and potassium in the soil. Trichoderma spp. was the most promising fungi for phosphate solubilization which facilitated nutrient availability and plant performance. However, fungal growth was highest in urea-amended soil. Nonetheless, soil amended with poultry manure

yielded plants with the highest fresh and dry matter weight, likely due to increased nutrient availability and microbial activity.

5.2 RECOMMENDATIONS

For improved microbiome such as fungi in the soil, which helps to stimulate nutrient facilitation, it is recommended that organic amendments, like poultry manure, should be utilised. Poultry manure evidently increased the beneficial microbes like phosphate-solubilizing fungi, and increased levels of several nutrients essential for plant growth compared to inorganic fertiliser (urea) and no amendment (control).

REFERENCES

Alori, E. T., Olaniyan, F. T., Adekiya, A. O., Ayorinde, B. B., Daramola, F. Y., Joseph, A., and Babalola, O. O. (2023). Response of soil microbial community (bacteria and fungi) to organic and inorganic amendments using tomato as a test crop. *Air, Soil and Water Research.*

Bagyaraj, D. J., and Ashwin, R. (2017). Soil biodiversity: role in sustainable horticulture. *Biodivers Hortic Crops*, 5, 1-18.

Barnett, H.L, and Hunter, B.B. (1996) Illustrated genera of Imperfect Fungi. *Burgess Publishing Company*, Third edition.

Bierza, W., Woźniak, G., Kompała-Bąba, A., Magurno, F., Malicka, M., Chmura, D., and Piotrowska-Seget, Z. (2023). The Effect of Plant Diversity and Soil Properties on Soil Microbial Biomass and Activity in a Novel Ecosystem. *Sustainability*, 15(6), 4880.

Bolan, S., Hou, D., Wang, L., Hale, L., Egamberdieva, D., Tammeorg, P., and Bolan, N. (2023). The potential of biochar as a microbial carrier for agricultural and environmental applications. *Science of the Total Environment,* 163968.

Frąc, M., Hannula, S. E., Bełka, M., and Jędryczka, M. (2018). Fungal biodiversity and their role in soil health. *Frontiers in microbiology*, 9, 316246.

Frąc, M., Jezierska-Tys, S., and Yaguchi, T. (2015). Occurrence, detection, and molecular and metabolic characterization of heat-resistant fungi in soils and plants and their risk to human health. *Advances in agronomy*, 132, 161-204.

Gao, W., and Liu, Y. (2021). Different responses of soil bacterial and fungal communities to 3 years of biochar amendment in an alkaline soybean soil. *Frontiers in Microbiology*, 12, 630418.

Griffiths, B. (2011). Soil fertility and fertilizers. In White, R. E. (Ed.), *Soils for sustainable agriculture* (pp. 155-176). CRC Press.

Hafifah, S., Maghfoer, M. D., & Prasetya, B. (2016). The potential of tithonia diversifolia green manure for improving soil quality for cauliflower (Brassica oleracea var. Brotrytis L.). *Journal of Degraded and Mining Lands Management*, 3(2), 499–506.

Harms, H., Schlosser, D., & Wick, L. Y. (2011). Untapped potential: exploiting fungi in bioremediation of hazardous chemicals. *Nature Reviews Microbiology,* 9(3), 177-192.

Hossner, L.R. (1996). Dissolution for total elemental analysis. Methods of soil analysis: part 3 chemical methods, 5, 49-64.

Huang, R., McGrath, S. P., Hirsch, P. R., Clark, I. M., Storkey, J., Wu, L., and Liang, Y. (2019). Plant–microbe networks in soil are weakened by century-long use of inorganic fertilizers. Microbial biotechnology, 12(6), 1464-1475.

Ibitoye, A. A. (2008). Laboratory Manual on Basic Soil Analysis. Department of Crop, Soil and Pest Management FUTA. Foladave Nig. Ltd. Pp. 4-91.

Ismail, S. M., Ozores-Hampton, M., & Sargent, S. A. (2014). Vegetable production using vermicompost. *University of Florida IFAS Extension*.

Jamalluddin, N., Massawe, F. J., Mayes, S., Ho, W. K., Singh, A., and Symonds, R. C.
(2021). Physiological screening for drought tolerance traits in vegetable amaranth (Amaranthus tricolor) germplasm. *Agriculture, 11(10), 994.*

Japakumar, J., Abdullah, R., & Rosli, N. S. M. (2021). Effects of biochar and compost applications on soil properties and growth performance of Amaranthus sp. grown at an urban community garden. AGRIVITA *Journal of Agricultural Science*, 43(3), 441-453.

Jing, J., Cong, W. F., & Bezemer, T. M. (2022). Legacies at work: plant–soil–microbiome interactions underpinning agricultural sustainability. *Trends in Plant Science*

Kadiri, O. H., Agbogun, L., & Edosa, V. I. O. (2022). Characterization and Classification of Soils on Agricultural Landscape in Benin Rainforest Zone, Nigeria. *Nigeria Journal of Soil and Environmental Research, 21, 122-128.*

Rashid, M. I., Mujawar, L. H., Shahzad, T., Almeelbi, T., Ismail, I. M., & Oves, M. (2016). Bacteria and fungi can contribute to nutrients bioavailability and aggregate formation in degraded soils. *Microbiological research, 183, 26-41.*

Reeve, J. R., Hoagland, L. A., Villalba, J. J., Carr, P. M., Atucha, A., Cambardella, C., Davis, D. R., & Delate, K. (2016). Organic farming, soil health, and food quality: Considering possible links. In Sparks, D. L. (Ed.), *Advances in agronomy* Vol. 137, pp. 319–367.

Sharpley, A. N., McDowell, R. W., & Kleinman, P. J. A. (2010). Phosphorus loss from land to water: Integrating agricultural and environmental management. *CRC Press.*

Sunassee, S. N. (2012). Vermicomposting: A sustainable approach to waste management and soil fertility. *Journal of Sustainable Development, 5(1), 1-12.*

Tabler, G. T., & Berry, I. L. (2013). Chicken manure management and utilization. *University of Arkansas Division of Agriculture.*

Vassilev, N., & Mendes, G. D. O. (2024). Soil Fungi in Sustainable Agriculture. *Microorganisms*, 12(1), 163.

Wang, Y., Liu, Z., Hao, X., Wang, Z., Wang, Z., Liu, S., ... & Li, R. (2023). Biodiversity of the beneficial soil-borne fungi steered by Trichoderma-amended biofertilizers stimulates plant production. *Biofilms and Microbiomes, 9(1), 46.*

Wilson, C. G., & Power, I. L. (2016). Vermicomposting: A review of the process and its applications. *Journal of Environmental Science and Health, Part B, 51(1), 1-13.*

Yao, Q., Liu, J., Yu, Z., Li, Y., Jin, J., Liu, X., & Wang, G. (2017). Changes of bacterial community compositions after three years of biochar application in a black soil of northeast China. *Applied Soil Ecology, 113, 11-21.*

Ye, L., Zhao, X., Bao, E., Li, J., Zou, Z., & Cao, K. (2020). Bio-Organic fertilizer with reduced rates of chemical fertilization improves soil fertility and enhances tomato yield and quality. *Scientific Reports, 10, 177.*