CHARACTERIZATION OF BLOODMEAL AND VERMICOMPOST BLENDED BIOFERTILIZERS: CHEMICAL COMPOSITION OF THE OUTPUT, MICROBIAL BIOMASS, AND CHEMICAL COMPOSITION OF DIFFERENT SOURCES OF BLOODMEAL.

A dissertation submitted in partial fulfilment of the requirements for the Master of Science Degree in Food Security and Sustainable Agricultural (Production)

Bindura University of Science Education

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DECLARATION

I hereby declare that the research project entitled "**Characterization of blood meal and vermicompost blended bio fertilizer: chemical composition of output, microbial biomass and chemical composition of different sources of blood meal**." submitted to Bindura University of Science Education, Department of Agricultural Economics, Education and Extension is a record of an original work done by me under the guidance and supervision of **Dr L Musemwa** and this work is submitted in partial fulfilment of the requirements for the award of a Master of Science Degree in Food Security and Sustainable Agriculture. The results embodied in this thesis have not been submitted to any University or Institute for the award of any degree of diploma.

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DEDICATION

I dedicate this work to my beloved and amazing spouse Reginald whose constant support, understanding and encouragement have been my strength throughout this journey. My cherished off springs Tadiwa, Wesley and Ashley, your love, patience, understanding and supportive have given me strength to endure the journey to success. I hope this accomplishment serves as a testament to the power of hard work and determination. Furthermore, I extend my gratitude to my wonderful friends Letwin and Tendai for their sacrifices, unwavering love and support during both highs and lows. Your belief in me is truly invaluable. This achievement is a reflection of your love and support. Indeed, you are true friends!

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ABSTRACT

This study characterized the chemical and microbial properties of vermicompost–blood meal biofertilizer blends under controlled laboratory conditions. It responds to the growing need for alternatives to synthetic fertilizers, which pose environmental and economic challenges in Zimbabwe, especially for smallholder farmers. The research employed a Completely Randomized Design (CRD) and analytical methods including ANOVA and microbial biomass assays to assess key parameters across different blending ratios. Four treatments were tested: Control (100% vermicompost), 1:3, 1:4 and 1:5, pig blood meal–vermicompost blends. Fermentation stabilized pH across all treatments, with values ranging from 9.05 to 9.12. Organic matter increased sharply from 1.50% in the control to 22.8% in the 1:3 blend, while the carbon-to-nitrogen ratio rose from 12.0 to 24.8. Available nitrogen peaked at 3.64% in the 1:3 blend, compared to 1.50% in the control, and available phosphorus reached 0.223% in the 1:4 blend. Microbial assessments revealed that total viable counts (TVC) dropped by 70% in the 1:3 blend versus the control (from 204,000 CFU/g to 62,000 CFU/g), with complete elimination of detectable molds. Analysis of blood meal sources showed cattle blood with the highest nitrogen content (12.04%), followed by pig (11.20%) and chicken (10.05%). These findings demonstrate that a 25:75 blood meal–vermicompost blend optimizes nutrient stabilization, enhances organic matter, and suppresses unwanted microbes, offering a scalable and sustainable alternative to chemical fertilizers for smallholder farmers. It calls for strategic blending of blood meal with vermicompost which can result in a nutrient-enriched, microbially stable, and ecologically sound alternative to chemical fertilizers, with direct implications for crop productivity and soil health management in organic and resource-limited farming systems.

Keywords: blood meal and vermicompost blended bio fertilizers, microbial biomass, chemical composition.

LIST OF ACRONYMS

ANOVA – Analysis of Variance

CFU – Colony Forming Unit

CRD – Completely Randomized Design

NPK – Nitrogen, Phosphorus, and Potassium

pH – Potential of Hydrogen

PLFA – Phospholipid Fatty Acid Analysis.

qPCR – Quantitative Polymerase Chain Reaction

SIR – Substrate Induced Respiration

TKN – Total Kjeldahl Nitrogen

TVC – Total Viable Count

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CHAPTER 1
INTRODUCTION

1.0 INTRODUCTION

With the growing emphasis on sustainable farming practices, organic fertilizers are gaining attention as ecofriendly substitutes for chemical fertilizers. Blood meal, a high nitrogen organic material, and vermicompost, a nutrient rich extract teeming with beneficial microorganisms, offer great potential as bio fertilizer input. However, the performance of their blended formulation, particularly its nutrient content, microbial bio mass, chemical composition of blood meal from different animals requires further investigations. By evaluating these factors, the research seeks to optimize the formulation ensuring maximum efficacy for crop growth while supporting environmentally friendly agriculture. The results will provide valuable insights for improving organic nutrient management systems.

1.1 BACKGROUND OF THE STUDY

Conventional agriculture leads to soil erosion which leads to land degradation, water pollution through the use of synthetic fertilizers and pesticides posing risk to aquatic life and human health, loss of biodiversity due to reduction of beneficial insects and finally contribute to climate change. Though chemical fertilizers release nutrients quickly, they can cause soil degradation over time, whereas bio fertilizers release nutrients gradually thus promoting long-term soil health and plant growth in a sustainable way through improvement of microbial diversity, organic matter content.

Studies indicate that bio fertilizers support beneficial microbial communities that contribute to soil aggregation and porosity, which in turn improves water retention and aeration (Mäder et al., 2020). Furthermore, a study by Meena et al. (2019) found that long-term application of bio fertilizers improved soil pH balance and enhanced enzymatic activity, thereby creating a more favorable environment for plant growth. According to Kumar et al. (2021), bio fertilizers promote root development and nutrient uptake in cereals such as wheat and rice, resulting in increased biomass and grain yield. Furthermore, bio fertilizers have been found to foster stress tolerance in crops by improving their resistance to moisture stress and disease. For example, studies indicate that bio fertilizer-treated plants exhibit higher chlorophyll

content and photosynthetic efficiency, leading to better growth and productivity (Ali et al., 2022).

According to Kuzyakov and Blagodatskaya, (2022), microbial consortia, including fungi and bacteria, co-operate in degrading lignin, cellulose, and other recalcitrant compounds. Fungi such as arbuscular mycorrhizal fungi (AMF) and phosphate-solubilizing bacteria (PSB) like *Pseudomonas* and *Bacillus* are vital in releasing phosphorus from insoluble compounds (Smith and Read, 2022). Recent metagenomics analyses have shown that AMF interactions with rhizobacteria further enhance nutrient cycling and pathogen suppression (Gupta et al., 2023).

The increasing need for sustainable agricultural practices has led to the exploration of organic fertilizers with bio fertilizers gaining prominence due to their ability to enhance soil health, boost crop yields, and support environmental sustainability. The organic amendments include compost which have lower nitrogen content, animal manure being rich in organic matter but should be properly composted to avoid risks of disease-causing organisms and lastly bio char which has limited nutrient availability though it improves carbon sequestration as well as soil structure. Among different organic amendments, blood meal and vermi compost are noble for their rich nutrient profiles as well as microbial activities.

Vermicomposting is a worm mediated bio degradation process using live epigamic worms such as *Eisenia ferida.* Worms plays an important role in the process of nutrient dissolution and bio remediation by enhancing the population of beneficial bacterial (Goswami et al., 2021). The digestive system of worms contains many beneficial microorganisms, nitrogen fixing bacteria and enzymes (Wang et al., 2021). Although microorganisms are responsible for the bio-chemical degradation of organic matter, worms decompose and condition the substrate, increasing the surface area for microorganism activity (Yururdurmaz, 2022). Worms mineralize organic matter through intestinal transit, digest it in the foregut and mid gut and then excrete it through the hind gut, interacting directly with microorganisms (Dume et al., 2022). Sharma et al. (2021) demonstrated that vermin compost application in tomato cultivation resulted in a 30% higher fruit yield compared to conventional compost.

Blood meal, a byproduct from animal processing, which contains 12-15% nitrogen, (Barker and Pilbeam, 2015) making it an effective nutrient source for crops. Hernandez and Lopez, (2023), noted that blood meal application in organic vegetable farming led to a 20% increase in yield compared to untreated soils. On the other hand, Jones et al, (2021) warned that

excessive application of blood meal could lead to nitrate leaching, posing a risk to groundwater quality. However only 3% of nitrogen in blood meal is in soluble form whereas 9% nitrogen is in insoluble form.

On the other hand, vermicomposting produced through the breakdown of organic matter by earthworms is reach in essential micronutrients, beneficial microbes and organic matter. Martinez et al, (2021) suggested that combining blood meal with vermi compost could create a synergistic effect, balancing immediate nitrogen availability with long term soil health benefits. On the other hand, Moyo et al. (2023) found that Zimbabwe lacks a clear and specific regulatory framework for bio fertilizers, creating uncertainty for manufacturers and importers. To add on, Chikukwa and Dube, (2022), highlighted that many bio fertilizer products are not adequately tested before market entry, leading to concerns about efficacy and quality control.

Despite challenges regarding regulation of bio fertilizers in Zimbabwe, Mugabe et al, (2020) conducted a comparative cost analysis and found out that bio fertilizers reduce input costs by 30-40% compared to synthetic fertilizers. However, Chigumira et al, (2021) found that initial adoption costs such as training and product awareness discouraged smallholder farmers from switching to bio fertilizers.

Thus, future research should focus on the potential of farmer training programs as well as regulatory frameworks specific to bio fertilizer production and use. Also, the combination of blood meal with vermicomposting could potentially create a balanced bio fertilizer that promotes nutrient availability and supports microbial biodiversity. It is upon this background that the study aims to investigate blending ratios of blood meal with vermi compost and their effects on chemical composition of the output, microbial biomass and chemical composition of bloodmeal from different livestock.

1.2 PROBLEM STATEMENT

Due to population growth and high demand of food, most farmers use synthetic fertilizer to boost yield in order to meet food demand. The increasing reliance on synthetic fertilizers in agriculture has led to significant environmental and economic challenges, particularly for marginalized communities that often find these inputs unaffordable. The high cost of synthetic fertilizers has made it difficult for many farmers to maintain soil fertility and crop yield, exacerbating food insecurity. Furthermore, the overuse of these chemical fertilizers contributes to climate change through greenhouse gas emissions and leads to land

degradation, as they disrupt soil ecosystems and reduce biodiversity. In this context, there is a pressing need to explore sustainable alternatives, such as bio fertilizers derived from blood meal and vermicompost. These organic amendments not only enhance soil health and improve nutrient dynamics but also promote microbial biodiversity.

However, limited research exists on the optimal blending ratios of these materials and their effects on chemical composition, nutrient content, microbial biomass, and shelf life. This study aims to address these gaps, providing insights that could help mitigate the adverse impacts of synthetic fertilizers while supporting sustainable agricultural practices. These chemical fertilizers thus destroy soil health and pollute the environment whist negatively affecting the natural ecosystem functions leading to land degradation. Studies done by Wang et al. (2022) indicated that chemical fertilizers alter soil microbial diversity by creating imbalances in soil pH and nutrient availability. Prolonged use of nitrogen-based fertilizers reduces soil microbial diversity up to 40%, which extremely limits nutrient cycling and organic matter decomposition, (Wang et al., 2022). Thus continuous use of inorganic fertilizers negatively affects microbial biomass and enzymatic activities especially in conventional agriculture systems. Continuous use of chemical fertilizers negatively affects soil fauna such as earthworms which helps in the maintenance of soil structure and fertility, (Bardgett et al, 2022).

Furthermore, use of chemical fertilizer has led to a 30% decline in earthworm populations in monoculture farming systems, (Bardgett et al., 2022). Besides affecting microbial biomass, chemical fertilizers also contribute to nitrate leaching into ground water which poses health risks and has led to 20-30% of global aquifers exceeding the safe nitrate limit of 10mg/L (set by World Health Organization), (Velthof et al., 2021). Thus, in order to increasing demand for sustainable agricultural practices necessitates the development of effective bio fertilizers that enhance soil fertility and plant growth while minimizing environmental impact. Blood meal, a high nitrogen byproduct of meat industry containing 3% water soluble N and 9% insoluble N, thus vermin compost having high microbial activity, can have the potential to unlock the 9% insoluble N in the blood meal through blending so that it can be released for plant uptake.

However, the optimal blending ratios, chemical interactions and their combined effects on microbial biomass and overall bio fertilizer efficacy remain underexplored. There is lack of comprehensive understanding regarding the chemical composition of bloodmeal from

different livestock especially the nitrogen levels and how these blends can be formulated to maximize nutrient availability, improve microbial activity. Thus addressing this gap is critical for developing a bio fertilizer that not only meets agricultural needs but also promotes sustainable practices in soil management.

1.3 OBJECTIVES

1.3.1 MAIN OBJECTIVE

To investigate the chemical composition of the output, microbial biomass, and the chemical composition of different sources of blood meal.

1.3.2 SPECIFIC OBJECTIVES

1) To analyze the chemical composition of various blood meal and vermicompost blended output.
2) To assess the impact of blending on microbial biomass and diversity within the bio fertilizer.
3) To determine the chemical composition of blood meal from different sources (chicken, cattle and pig).

1.4 RESEARCH QUESTIONS

1) What is the chemical composition of bio fertilizers derived from blending blood meal and vermicompost?
2) What is the impact of blood meal and vermicompost blended bio-fertilizer on microbial biomass and density?
3) What is the chemical composition of blood meal from chicken, cattle and pigs?

1.5 HYPOTHESIS

i) The chemical composition of bio fertilizers derived from blending blood meal and vermicompost vary depending on the specific ratios of each component used in the blend.

ii) Blood meal and vermicompost blended bio fertilizers will significantly enhance microbial biomass and density more effectively than either component used individually.

iii) There is significant difference on the chemical composition of blood meal from chicken, cattle and pig.

1.6 SIGNIFICANCE OF STUDY

This research will provide vital information on the importance of bio fertilizers as a sustainable alternative to chemical fertilizers, particularly in the context of climate change. The findings will benefit poor and marginal farmers in Zimbabwe who struggle with infertile soils, enabling them to formulate bio fertilizers that supply essential nutrients to plants over extended periods. Insights into microbial biomass can help these farmers enhance soil fertility, leading to improved crop yields and quality.

Furthermore, the study's findings on chemical composition of blood meal from different livestock's especially nitrogen will be valuable to various stakeholders in agriculture, including policymakers, research institutions, NGOs, and bio fertilizer manufacturers so that correct blending ratios can be used depending on the source of blood meal used. By providing evidence-based recommendations, the research can facilitate the practical application of bio fertilizers across diverse agricultural contexts, enhancing their usability and effectiveness.

Additionally, the involvement of government and regulatory authorities, such as the Zimbabwe Standards Association, will ensure that bio fertilizer products meet quality standards and regulatory requirements, promoting consumer confidence and safety. Ultimately, the development of cost-effective bio fertilizers can reduce production costs for farmers, thereby enhancing the overall productivity of agricultural systems in Zimbabwe and contributing to food security and sustainable agricultural practices.

1.7 DELIMITATIONS AND LIMITATIONS OF STUDY

1.7.1 DELIMITATIONS

- The study concentrated on specific blending ratios of blood meal and vermicompost, limiting the exploration of other potential combinations that could yield different results.
- The analysis primarily focused on major nutrients (N, P, and K) and specific microbial indicators, excluding other potentially beneficial compounds and microbial functions.
- The chemical composition of blood meal studied includes chicken, cattle and pigs excluding other domesticated animals such as goats, sheep and ducks.

- When it comes to chemical parameters analyzed, only nitrogen content is of major concern leaving behind other major and minor plant nutrients,
- The study was carried out at Fambidzanai permaculture Centre only which may limit the generalizability of the findings to other regions with different soil types and climatic conditions.
- The experiment was conducted under controlled laboratory conditions, which may not account for the natural complexities of natural ecosystems and agricultural practices.

1.7.2 LIMITATIONS

- The chemical composition of blood meal and vermicompost can vary significantly based on source, processing methods and environmental conditions, potentially affecting the consistency of bio fertilizer produced. The limitations were overcomed through the use of uniform raw materials.
- Microbial communities present in both blood meal and vermicompost may differ, which would influence the overall microbial biomass and activity in the blended bio fertilizer and this was addressed through the use of different ratios between vermicompost and blood meal to identify effective combinations that increases microbial bio mass hence facilitating nutrient release.
- Blood composition can be affected by processing method, animal breed, age and diet being given as well as the healthy status of the animals and the limitation was overcomed by using uniform drying process that is sun drying and sourcing blood from commercial abattoirs.
- The results might not apply to blood meal from other sources such as goats and sheep and the researcher overcome this by comparing results with already published literature on chemical composition of sheep and goats.
- The results obtained in controlled laboratory settings might not fully replicate field conditions, limiting the applicability of findings to real-world agricultural settings.

1.8 SUMMARY

The introductory chapter has outlined the scientific and practical relevance of characterizing bloodmeal and vermicompost blended biofertilizers in the context of sustainable agriculture. With increasing concerns over the long-term impact of synthetic fertilizers on soil health and environmental integrity, organic alternatives such as bloodmeal and vermicompost have

gained attention for their potential to supply essential nutrients and enhance microbial activity. It also established the rationale for the study by highlighting the nutrient richness of bloodmeal particularly its high nitrogen content and the biological and structural benefits of vermicompost. However, variability in bloodmeal composition across sources and a lack of standardized blending protocols warrant thorough chemical and microbiological evaluation of the final products. Therefore, this study aims to fill that gap by assessing the nutrient composition of various bloodmeal sources, quantifying microbial biomass in blended biofertilizers, and determining the overall chemical quality of the final output.

CHAPTER 2
LITERATURE REVIEW

2.0 INTRODUCTION

The development and use of organic bio fertilizers have gained significant attention in sustainable agriculture due to their potential to improve soil fertility, enhance plant growth, and reduce reliance on synthetic fertilizers. Among these, the blending of blood meal and vermicompost offers a promising solution, combining the rapid-release nutrient profile of blood meal with the slow-release, organic matter-enriched properties of vermicompost. This combination not only provides essential plant nutrients but also supports soil microbial activity and long-term soil health. Research on the characterization of blood meal and vermicompost blended bio fertilizers focuses on understanding their chemical composition and microbial activity. These factors are critical for determining the bio fertilizer's effectiveness in different agricultural settings. However, the performance of such blends can vary significantly under different storage conditions, with factors such as temperature, humidity, and aeration influencing nutrient stability, microbial biomass, and overall chemical properties. This review explores existing studies on the subject, specifically addressing how the chemical composition of the bio fertilizer and microbial dynamics are impacted by blending. By synthesizing current knowledge, the review aims to identify gaps in research and provide insights into the potential applications and limitations of blood meal and vermicompost blends as sustainable bio fertilizers.

2.1 BACKGROUND OF BIO FERTILIZERS

Bio fertilizers have gained significant attention in recent years as sustainable alternatives to chemical fertilizers. They consist of beneficial microorganisms that enhance soil fertility and plant growth by fixing atmospheric nitrogen, solubilizing phosphorus, and producing growth-promoting substances. According to Kumar et al. (2021), bio fertilizers are critical for addressing global challenges such as soil degradation, climate change, and food security. Furthermore, Kumar, (2021) noted that bio fertilizers represent a sustainable shift from chemical fertilizers, utilizing beneficial microorganisms to enhance soil productivity. Supporting this, Singh, (2019) documented significant crop yield improvements ranging from 10-30% through bio fertilizer applications. Zhang, (2022) further elaborated that bio fertilizers facilitate crucial processes like nitrogen fixation and phosphate solubilization

Recent studies by Meena et al. (2020) highlight the role of nitrogen-fixing bacteria such as Rhizobium, Azotobacter and Azospirillum in improving nitrogen availability in soils. These microorganisms form symbiotic relationships with plants, particularly legumes, and reduce the need for synthetic nitrogen fertilizers. Research by Sharma et al. (2019) emphasizes the importance of phosphate-solubilizing microorganisms (PSMs) like Pseudomonas and Bacillus in converting insoluble phosphates into plant-available forms. This is particularly crucial in soils lacking phosphorus. Recent work by Gouda et al. (2018) demonstrates that Plant Growth-Promoting Rhizobacteria enhance plant growth by producing phytohormones, improving nutrient uptake, and protecting plants from pathogens through antibiosis and induced systemic resistance. Studies by Rillig et al. (2019) show that arbuscular mycorrhizal fungi (AMF) improve phosphorus uptake and enhance plant tolerance to abiotic stresses like drought and salinity. AMF also contribute to soil carbon sequestration, making them valuable in climate-smart agriculture.

2.2 IMPORTANCE OF BLOOD MEAL AND VERMI COMPOST

Blood meal, a byproduct of the meat processing industry, remains a valuable organic fertilizer due to its high nitrogen content and ability to improve soil fertility. Blood meal contains approximately 12-15% nitrogen, making it an excellent organic fertilizer for nitrogen-deficient soils. According to Zhang et al. (2022), blood meal releases nitrogen slowly, reducing the risk of leaching and groundwater contamination. Recent studies by Li et al. (2021) indicate that blood meal enhances microbial activity in the soil, promoting the decomposition of organic matter and nutrient cycling. This improves soil health and fertility over time. Blood meal has been reported to act as a natural pest deterrent due to its strong odor, which repels herbivores like deer and rabbits (Wang et al., 2020). The use of blood meal as an organic fertilizer supports sustainable agriculture by recycling animal byproducts and reducing reliance on synthetic fertilizers (Gupta et al., 2023).

Vermicompost is rich in essential macro- and micronutrients, including nitrogen, phosphorus, potassium, calcium, and magnesium. According to Yadav et al. (2021), these nutrients are present in plant-available forms, making vermicompost highly effective for improving soil fertility. To add on, it also improves soil aggregation, porosity, and water-holding capacity. A study by Singh et al. (2022) found that vermicompost application significantly enhances soil structure, particularly in degraded or sandy soils. Research by Gómez-Brandon et al. (2021)

demonstrates that vermicompost increases the abundance of beneficial microorganisms, such as nitrogen-fixing bacteria and mycorrhizal fungi. Recent studies by Atiyeh et al. (2023) show that vermicompost application increases plant growth, yield, and resistance to pests and diseases. This is attributed to its rich nutrient content and bioactive compounds. Vermicomposting reduces organic waste in landfills and minimizes greenhouse gas emissions. A study by Dominguez et al. (2023) highlights its role in promoting circular economy practices and sustainable waste management.

2.3 CHEMICAL COMPOSITION OF BLOOD MEAL AND VERMI COMPOST

2.3.1 Overview of blood meal

Blood meal is a byproduct of the meat processing industry, derived from the blood of slaughtered animals. It is widely used as an organic fertilizer due to its high nitrogen content and ability to improve soil fertility. Recent studies highlight its role in sustainable agriculture, particularly in organic farming systems. According to Zhang et al. (2022), blood meal has gained popularity as a sustainable alternative to synthetic nitrogen fertilizers, reducing environmental pollution and promoting soil health. It is also recognized for its ability to enhance soil microbial activity and improve nutrient cycling (Li et al., 2021). Anderson, (2018) described blood meal as a sustainable fertilizer derived from processed animal blood, noting its high nitrogen concentration. Liu, (2022) emphasized blood meal's effectiveness as a slow-release fertilizer, maintaining nutrient availability for 3-4 months.

2.3.2 Nutritional profile of blood meal

Blood meal contains approximately 12-14% nitrogen, phosphorus levels 1-2%, iron content 2000-3000 ppm and protein concentration of 80-85%, (Garcia-Lopez., 2020). The potassium content in blood meal is relatively low, typically less than 1%, but it still contributes to overall plant health and stress resistance (Yadav et al., 2021). Blood meal provides trace amounts of essential micronutrients such as iron, zinc, and copper, which are vital for enzymatic processes and chlorophyll synthesis in plants (Singh et al., 2022).

2.3.3 Chemical composition of blood meal

The chemical composition of blood meal is primarily determined by its protein content, which is derived from the blood proteins of animals. Recent work by Patel, (2023) characterized blood meal's chemical attributes as near-neutral pH (6.8-7.2), carbon content: 40-45%, carbon-to-nitrogen ratio: 3:1 which promotes rapid decomposition and nutrient

release in the soil (Edwards et al., 2007). Blood meal is composed of approximately 80-90% protein, primarily in the form of globulins and albumins. These proteins break down into amino acids, which serve as a slow-release nitrogen source for plants (Gouda et al., 2018). Blood meal contains heme iron, a highly bioavailable form of iron that is essential for chlorophyll synthesis and oxygen transport in plants (Rillig et al., 2019). The moisture content of blood meal is typically low (less than 10%), which enhances its shelf life and reduces the risk of microbial contamination. The ash content, which consists of inorganic minerals, is usually around 3-5% (Barker, 2010).

2.3.4 Overview of Vermicompost

Vermicomposting utilizes earthworms to convert organic waste into nutrient-rich fertilizer through bio oxidation and stabilization processes (Singh et al., 2020) .According to several studies, vermicomposting effectively transforms organic waste into nutrient-rich humus-like material through the joint action of earthworms and microorganisms (Kale and Karmegam, 2016; Edwards et al., 2020).The process involves mechanical fragmentation of waste materials, biochemical degradation and microbial enrichment (Kumar et al., 2019). The process is eco-friendly, cost-effective, and significantly reduces the volume of biodegradable waste, making it a valuable tool in addressing global waste management challenges (Pathma and Sakthivel, 2015; Sharma et al., 2022). According to Lim et al. (2018), vermicomposting reduces greenhouse gas emissions and landfill waste while producing a stable organic fertilizer. Recent research (Yadav & Garg, 2021; Sharma et al., 2023) emphasizes its role in sustainable agriculture, particularly in organic farming and urban waste management. Primary species used are *Eisenia fetida, Eudrilus eugeniae* and *Perionyx excavatus* due to their high reproduction rates and waste processing efficiency (Bhat et al., 2017). These species efficiently decompose organic matter and convert it into a stabilized product rich in nutrients (Dominguez et al., 2019). Optimal conditions include 20-30°C temperature, 60-80% moisture, and pH 6.5-7.5 (Devi and Sumathi, 2016).

2.3.5 Nutritional Profile of Vermicompost

The nutritional composition of vermicompost is a key factor influencing its role as an organic fertilizer. Vermicompost is rich in essential plant nutrients such as nitrogen (N), phosphorus (P), and potassium (K), as well as secondary and micronutrients like calcium (Ca), magnesium (Mg), zinc (Zn), and iron (Fe) (Gajalakshmi & Abbasi, 2018). Nitrogen (1.0-2.2%), phosphorus (0.8-1.6%), potassium (0.8-1.5%) (Sharma & Garg, 2018). The nitrogen

content in vermicompost is predominantly in the form of ammonium and nitrate, making it readily available to plants (Bhat et al., 2016). Phosphorus in vermicompost is converted into soluble forms through microbial activity, enhancing its availability for plant uptake (Sharma & Garg, 2020). Vermicompost contains essential micronutrients that is iron (0.2-1.7%), Zinc (0.01-0.05%), Copper (0.01-0.03%) (Adhikary, 2016). To add on, it is rich in beneficial microorganisms that is bacteria ($2x10^8$ CFU/g), fungi ($1x10^4$ CFU/g) (Pathma & Sakthivel, 2015). Research by Pathma & Sakthivel, (2020) indicates that vermicompost contains growth-promoting substances such as humic acids, auxins, and cytokinin, enhancing plant growth and stress resistance. Additionally, Kumar et al. (2022) found that vermicompost improves soil enzyme activity, further increasing nutrient availability for crops.

2.3.6 Chemical Properties of Vermicompost

Vermicompost possesses unique chemical properties that contribute to its effectiveness as a soil amendment. It is characterized by a neutral to slightly alkaline pH (6.5–7.5), which helps in neutralizing acidic soils (Edwards et al., 2019). Similarly studies by (Dominguez et al., 2016; Hussain et al., 2018) show that vermicompost typically has a near-neutral pH (6.5–7.5), making it suitable for most crops. Organic carbon content: 9.5-17.98% (Chatterjee et al., 2016), C: N ratio typically ranges from 12:1 to 17:1 (Kumar et al., 2022), Humic acids: 10.6-28.8 mg/g (Martinez-Balmori et al., 2019), Fulvic acids: 2.8-5.7 mg/g (Goswami et al., 2017), Water-holding capacity: 40-60% (Rajasekar et al., 2023) and Cation Exchange Capacity: 50-70 meq/100g (Lim et al., 2016). Its high CEC (Bhat et al., 2020) enhances nutrient retention in soils. Furthermore, research by (Goswami et al., 2021; Singh et al., 2024) confirms that vermicompost has lower heavy metal concentrations compared to raw organic waste, ensuring safer agricultural use.

2.3.7 Ratios and formulations of blood meal and vermicompost

Research by Bhat et al. (2017) investigated the co-composting of blood meal with organic waste during vermicomposting and found that a ratio of 1:3 (blood meal to vermicompost) mitigates the risk of ammonia toxicity while allowing sufficient nitrogen release. The study also observed that higher blood meal proportions (e.g., 1:1) led to the accumulation of ammonia and nitrites, which could inhibit microbial activity and plant growth. Similar research indicates that a 1:3 to 1:5 ratios (blood meal: vermicompost) optimizes nutrient release while minimizing nitrogen loss (Gómez-Brandon et al., 2016) On the other hand, Gupta et al. (2020) emphasized that the addition of blood meal in a 1:4 ratios with

vermicompost provides a steady nitrogen release, which is advantageous for plant growth. In contrast to the previous authors, Zhang et al. (2018) investigated the chemical interactions when blood meal was incorporated into vermicompost at ratios of 1:5, 1:10, and 1:20 and the findings revealed that the 1:10 ratio optimized nitrogen mineralization while minimizing ammonia volatilization. Zhang et al. (2020) explored a wider range of formulations and concluded that a 1:5 ratios (blood meal to vermicompost) is ideal for sandy soils, where nutrient retention is a challenge and the formulation improves the cation exchange capacity and nutrient-holding ability of the soil. Garg & Kaur, (2019) found that a 1:3 ratios (blood meal: vermicompost) improved nitrogen retention while reducing ammonia volatilization. Lim et al. (2015) tested different formulations and reported that 20% blood meal + 80% vermicompost enhanced microbial activity and nutrient mineralization. Atiyeh et al. (2017), observed that higher vermicompost ratios (70–80%) buffered blood meal's rapid nitrogen release, improving long-term soil fertility.

2.3.8 Chemical interactions of blood meal and vermicompost blend

Chen et al. (2019) highlighted that blood meal, being a protein-rich material, undergoes rapid decomposition when mixed with vermicompost and the humic acids and microbial enzymes in vermicompost facilitate the breakdown of proteins into amino acids and ammonium ions, which are then gradually converted into plant-available forms of nitrogen, such as nitrates. Rodríguez et al. (2021) further elaborated on the role of microbial communities in vermicompost, stating that the addition of blood meal increases microbial activity, leading to faster degradation of organic matter and however warned that excessive blood meal could create anaerobic conditions and produce toxic byproducts such as ammonia and nitrites. Vermicompost helps counteract this by enhancing aeration and microbial diversity. In another study, Kaur et al. (2018) demonstrated that the chemical interaction between the carbon-to-nitrogen (C: N) ratio of vermicompost and the high nitrogen content of blood meal results in a balanced nutrient profile and the interaction reduces the risk of nitrogen leaching and increases the availability of micronutrients such as iron and zinc through chelation with humic substances. Gómez-Brandon et al. (2016), studied nitrogen dynamics in blended organic fertilizers and found that vermicompost's microbial community helps stabilize nitrogen from blood meal. Zhang et al. (2020), demonstrated that vermicompost enhances phosphorus availability when mixed with blood meal due to phosphate-solubilizing bacteria. Saha et al. (2018) noted that blending reduces phytotoxicity-pure blood meal can inhibit seed germination, but vermicompost mitigates this effect.

2.4 NUTRIENT DYNAMICS OF BLENDED FERTILIZERS

2.4.1 Nutrient release patterns from blending blood meal with vermicompost

A synthesis of laboratory and field studies reveals that blending blood meal with vermicompost produces a two-phase nutrient-release profile an early pulse of mineral nitrogen followed by a prolonged, slow-release phase that optimizes both immediate crop nutrition and long-term soil fertility.

Ehiomogue, (2019) incubated soil mixtures amended with vermicompost alone, blood meal alone, and a 1:1 blood meal–vermicompost blend. Pure vermicompost released 34% of its total N by Day 7 and reached 59% by Day 21; pure blood meal released 70% of its N by Day 7 but plateaued at 75% by Day 21. In contrast, the 1:1 blend released 33% of its N by Day 7 and 64% by Day 21 demonstrating that vermicompost moderates the rapid N mineralization from blood meal into a more sustained release (Ehiomogue, 2019). Phosphorus release followed a similar pattern, with blends achieving intermediate cumulative P release compared to the two single amendments.

In an anaerobic incubation study, Hoque et al. (2022) found that soil texture modulates this kinetics. When a 1:1 blood meal vermicompost blend was incubated in floodplain versus terrace soils, cumulative N release at Day 7 was approximately 40% in floodplain soil and 30% in terrace soil; by Day 21 these values rose to 62% and 48%, respectively. This underscores the role of soil properties in governing microbial decomposition and nutrient availability when organic amendments are used in combination (Hoque et al., 2022).

Field trials by Mishra, Singh, and Datta, (2013) further confirmed these findings in situ. Their integrated treatment (blood meal + vermicompost + partial NPK) increased soil total N from 260 to 281 kg ha^{-1}, available P from 18.8 to 20.2 kg ha^{-1}, and K from 281 to 296 kg ha^{-1}, while raising organic matter by 0.17% and stabilizing pH around 7.7 (versus 6.8 in the unfertilized control). These data indicate that blood meal vermicompost blends can deliver an early N pulse for rapid growth and concurrently build long-term soil fertility through enhanced organic matter and balanced nutrient release (Mishra et al., 2013).

2.4.2 Macronutrient (NPK) Release Dynamics

Recent studies have explored the synergistic effects of blood meal and vermicompost on nutrient mineralization. Gómez-Brandon et al. (2016) found that blending blood meal with vermi compost accelerated nitrogen (N) release due to microbial activity, reducing ammonia

volatilization by 20–30% compared to blood meal alone. Similarly, Atiyeh et al. (2018) reported that blood meal-vermi compost mixtures enhanced phosphorus and potassium availability by improving microbial-mediated organic matter decomposition. Zhang et al. (2020) observed that blood meal-vermicompost blends exhibited a slow-release nitrogen pattern, with peak mineralization occurring at 4–6 weeks, aligning better with crop demand than synthetic urea. Kaur et al. (2021) noted that vermicompost's humic acids facilitated phosphorus solubilization from blood meal, increasing plant-available P by 15–25% in alkaline soils. Blood meal, a high-nitrogen by-product, provides a rapid release of nitrogen, which is crucial for early plant growth stages. However, its release can sometimes be too rapid, leading to potential nitrogen losses through volatilization or leaching (Smith et al., 2018). Vermicompost, on the other hand, is known for its slow-release properties and ability to stabilize nutrients in the soil. According to Kumar et al. (2021), when blended, the gradual mineralization of vermicompost complements the rapid nutrient release from blood meal, creating a balanced nutrient supply over time. A study by Wang et al. (2019) demonstrated that vermin compost's microbial population facilitates the conversion of organic nitrogen into plant-available forms while minimizing gaseous losses. Phosphorus availability in these blends is also enhanced due to the organic acids and microbial activity in vermicompost, which solubilize phosphorus compounds. Patel et al. (2020) found that adding vermicompost to blood meal increased phosphorus availability by up to 30% compared to using blood meal alone. Similarly, potassium release is moderated by vermin compost's ability to retain potassium ions, preventing leaching.

2.4.3 Micronutrient Release and Bioavailability

Arancon et al. (2017) demonstrated that blood meal- vermicompost mixtures significantly increased iron and zinc uptake in leafy greens due to enhanced chelation by organic acids in vermi compost. Vermicompost contains chelating agents such as humic and fulvic acids, which enhance the bioavailability of these micronutrients (Chaudhary et al., 2022). When combined with blood meal, the microbial activity within vermicompost accelerates the breakdown of organic matter, releasing trace elements into the soil solution. Studies by Ahmed and Roy, (2023) revealed that the blend improved the uptake of micronutrients by plants, particularly in nutrient-deficient soils. Dominguez et al. (2019) found that vermicompost's microbial consortia reduced iron fixation in calcareous soils, improving its solubility by 35–40%. Yadav et al. (2022) highlighted that blood meal- vermi compost blends released copper and manganese more steadily than inorganic salts, reducing toxicity

risks while ensuring long-term availability. Liu et al. (2023) confirmed that humic substances in vermi compost formed stable complexes with micronutrients, preventing leaching in sandy soils.

2.4.4 Factors Influencing Nutrient Release from Blended Blood Meal and Vermicompost

Biochemical composition and microbial activity influence the decomposition and nutrient release dynamics of organic fertilizers. Blood meal, a high-nitrogen organic material, releases nutrients rapidly due to its high solubility and a low carbon-to-nitrogen (C: N) ratio (Hossain & Anamul, 2018). Furthermore, blood meal undergoes mineralization facilitated by proteolytic microorganisms (Gómez-Brandon et al., 2019). This is in line with Wang et al. (2020) who documented those beneficial microorganisms in vermicompost enhanced nitrogen mineralization from blood meal by up to 45% under controlled conditions. Vermicompost enhances this process by introducing diverse microbial communities that accelerate organic matter breakdown (Yadav & Garg, 2019). On the other hand, vermicompost acts as a slow-release fertilizer due to its stabilized organic matter and humic substances that form complexes with nutrients (Singh et al., 2020). Blending these materials creates a synergistic effect, where vermin compost's microbial activity promotes faster N mineralization from blood meal (Atiyeh et al., 2018). Phosphorus (P) and potassium (K) release from vermicompost is also improved due to humic acid formation, which chelates cations, enhancing their availability (Arancon et al., 2017). Rodriguez-Garcia et al. (2018) reported that pH levels between 6.0-7.5 optimized nutrient availability from blood meal-vermicompost blends, with phosphorus availability particularly sensitive to pH variations. More so, when blended, the interaction between the two materials can modulate nutrient release rates by balancing rapid nitrogen mineralization from blood meal with the slow nutrient availability of vermicompost (Chen et al., 2021). Studies indicate that blending blood meal with vermicompost in a 1:2 ratio optimizes nutrient release, as vermin compost's humic substances reduce N volatilization while improving phosphorus solubility (Dominguez et al., 2023). Field trials demonstrated that blended applications increase crop nitrogen uptake by 20–30% compared to sole applications (Kumar et al., 2021). However, excessive blood meal (>30% of blend) can cause temporary N immobilization due to high C: N ratios during initial decomposition (Singh & Kaur, 2022).

2.4.5 Storage Conditions and Nutrient Stability

Nutrient retention in blended organic fertilizers can be affected by storage conditions. High moisture content (>60%) can lead to anaerobic conditions, reducing N availability through volatilization (NH_3 loss) and denitrification (Chen et al., 2020). Conversely, dry storage (<30% moisture) preserves N but may slow microbial activity, delaying mineralization (Wang et al., 2021). Temperature fluctuations during storage also impact nutrient stability; elevated temperatures (>35°C) accelerate microbial decomposition but may lead to excessive N loss (Eghball et al., 2016). Kumar et al. (2019) demonstrated that storage temperatures between 15-25°C maintained optimal nutrient content in blood meal-vermicompost blends, with nitrogen retention rates 30% higher compared to storage at higher temperatures. Zhang and Lee, (2021) found that moisture levels of 40-60% were optimal for nutrient preservation in blended organic fertilizers, while levels exceeding 65% accelerated nutrient loss through leaching. Optimal storage in airtight containers at 20–25°C with moderate moisture (40–50%) is recommended to balance nutrient preservation and microbial activity (Zhang et al., 2022). Vermicompost, due to its stable organic matter content, buffers nutrient losses during storage by maintaining a balanced microbial ecosystem (Kaur et al., 2019). Blended formulations are more stable under controlled conditions such as low temperatures and moderate humidity levels, which help minimize nutrient losses (Das et al., 2022).

2.4.6 Environmental Factors Affecting Nutrient Release

Environmental conditions such as soil temperature, moisture, and pH modulate nutrient release from blood meal-vermicompost blends. Warmer soils (25–30°C) enhance microbial activity, increasing N mineralization rates (Gopal et al., 2017). Excessive rainfall or irrigation can leach soluble nutrients (NO_3^-, K^+), reducing fertilizer efficiency (Sharma et al., 2018). Soil pH also plays a critical role; neutral to slightly alkaline conditions (pH 6.5–7.5) favor Nitrogen mineralization, while acidic soils (pH < 6) may immobilize Phosphorus (Bhattacharya et al., 2020). For instance, acidic soil conditions can enhance the availability of micronutrients, while neutral to slightly alkaline conditions favor nitrogen release (Ali et al., 2018). Additionally, soil texture influences nutrient retention, with clayey soils reducing leaching but potentially slowing mineralization due to limited aeration (Saha et al., 2016). Soil moisture is critical for nutrient solubilization and microbial activity, with optimal release observed under moderate moisture conditions (Rahman et al., 2020). Temperature fluctuations also affect microbial-mediated decomposition processes, with higher temperatures enhancing the mineralization of organic compounds (Zhang et al., 2023).

Additionally, the presence of a diverse microbial community in vermicompost enhances nutrient cycling and reduces the risk of nutrient leaching (Patil et al., 2021).

2.4.7 Temporal Changes in Nutrient Availability from Blood Meal and Vermicompost blended bio fertilizer

2.4.7.1 Short-Term Nutrient Release Dynamics (0–30 Days)

The initial phase of nutrient release from blood meal and vermicompost blends is characterized by rapid nitrogen (N) mineralization due to microbial decomposition. Blood meal, with its high protein content (~12–14% N), undergoes proteolysis by soil microbes, releasing ammonium (NH_4^+) within 7–14 days (Gómez-Brandón et al., 2019). Vermicompost enhances this process by introducing ammonifying bacteria (e.g., Bacillus spp.), which accelerate N mineralization (Yadav & Garg, 2019). However, excessive BM (>30% of blend ratio) can temporarily immobilize N due to a high C: N ratio, delaying availability (Singh & Kaur, 2022). Phosphorus (P) and potassium (K) show faster release from vermicompost due to humic acid chelation, with significant availability within 10–20 days (Arancon et al., 2017). Blending BM with VC in a 1:2 ratio optimizes early nutrient release while reducing N volatilization (Dominguez et al., 2023).

2.4.7.2 Medium-Term Nutrient Release (30–90 Days)

During this phase, microbial activity stabilizes, and nitrification converts NH_4^+ to NO_3^-, improving plant-available N (Kumar et al., 2021). Studies report a 20–30% increase in N availability in blood meal- vermicompost blends compared to sole applications (Atiyeh et al., 2018). Phosphorus availability peaks at 30–60 days due to continued microbial solubilization of organic P (Gopal et al., 2017). However, environmental factors such as soil moisture and temperature significantly influence release rates. In warm, moist soils (25–30°C), N mineralization is 30% faster than in cooler conditions (Sharma et al., 2018). The temporal dynamics of nutrient release were further explored by Zhao et al. (2018), who observed that the release of nitrogen from a blood meal-vermicompost blend followed a two-phase pattern. An initial rapid release of nitrogen occurred due to blood meal decomposition, followed by a prolonged phase of nutrient release regulated by the microbial processes in the vermicompost. The study highlighted that the blend significantly reduced the risk of nitrogen leaching, especially in sandy soils.

2.4.7.3 Long-Term Nutrient Retention (90–180 Days)

After 90 days, nutrient release slows, humic substances in vermicompost reduce leaching losses, sustaining NO_3^- and K^+ availability (Zhang et al., 2022). Field trials show that blood meal - vermicompost blends maintain 15–20% higher soil nitrogen than synthetic fertilizers after 6 months (Saha et al., 2016). However, excessive rainfall or poor storage can lead to nutrient loss. Blood meal – vermi compost blends stored at >60% moisture lose 10–15% N via NH_3 volatilization (Chen et al., 2020). The study reported that the blend allowed for a more sustained release of nutrients, maintaining availability in the soil for up to 120 days after application. Similarly, Patel et al. (2020) investigated the availability of phosphorus (P) and potassium (K) when using blended amendments. Their findings revealed that vermicompost enhanced the solubilization of phosphorus from blood meal, likely due to increased microbial activity and organic acid production. Potassium availability, while not as directly affected by the blood meal, was slowly released from the vermicompost over the course of 90-150 days.

2.5 MICROBIAL BIOMASS AND DIVERSITY IN BLENDED BIO FERTILIZER

2.5.1 Importance of microbial biomass

Microbial biomass act as a determinant of soil fertility and nutrient cycling capacity in organic amendment systems and ecosystem productivity due to its sensitivity to changes in environmental conditions, agricultural practices, and soil management strategies (Joergensen and Wichern, 2018). Moreover, microbial biomass acts as an early indicator of soil quality, reflecting the short-term impacts of land-use changes and amendments (Luo et al., 2018). Studies have emphasized that microbial biomass serves as an indicator of soil fertility. Studies demonstrate that vermicompost contains 2-3 times higher microbial biomass compared to conventional compost due to earthworm-induced microbial enrichment (Aira et al., 2016). When blended with blood meal, this biomass becomes pivotal in nitrogen mineralization: proteolytic microbes (e.g. Bacillus and Pseudomonas) convert blood meal's protein nitrogen to plant-available NH_4^+ within 7-14 days (Gómez-Brandón et al., 2019). To add on, microbial biomass carbon in vermicompost- blood meal blends increases by 18-25%, enhancing soil organic matter stabilization (Zhang et al., 2022). Wang et al. (2019,) demonstrated that microbial biomass serves as both a source and sink of nutrients, contributing 10-40% of total soil organic matter. They found microbial biomass carbon directly correlates with soil fertility and nutrient cycling efficiency. Zheng and Martinez, (2022) established that microbial biomass acts as an early indicator of soil quality changes,

responding more rapidly than chemical or physical parameters. Their research showed a 25% increase in crop yield when microbial biomass exceeded 400 mg/kg soil.

2.5.2 Assessment methods of microbial biomass

Several methods have been developed to assess microbial biomass, with fumigation-extraction and substrate-induced respiration (SIR) being the most commonly used techniques. The fumigation-extraction method quantifies microbial carbon (Cmic) and nitrogen (Nmic) by measuring the release of carbon dioxide after microbial cells are lysed using chloroform fumigation (Vance et al., 1987). On the other hand, SIR measures microbial activity by monitoring carbon dioxide evolution after the addition of readily available carbon substrates (Anderson & Domsch, 1978). Advanced molecular techniques, such as DNA-based quantification (qPCR) and phospholipid fatty acid (PLFA) analysis, have further improved the precision of microbial biomass assessment, providing insights into microbial community composition and functional diversity (Fierer et al., 2020).

Table 1: Methods of microbial biomass

Method	Application	Key findings	References
Chloroform fumigation-extraction(CFE)	Measures microbial biomass carbon and microbial biomass nitrogen	Vermi compost-blood meal blends shows 30% higher microbial carbon than control soil.	Vance et al. (2017)
PLFA analysis	Profiles microbial community structure	Blood meal increases Gram +bacteria; vermicompost Enhances fungi	Buyer et al. (2021)
Qpcr OF 16s rRNA	Quantifies specific microbial taxa	Nitrosomonas spp. Dominate nitrification phase.	Kumar et al. (2020)

2.6 BLENDING ON MICROBIAL DIVERSITY

Blending organic materials, such as compost and animal-based products, has been shown to influence microbial diversity significantly. For example, the incorporation of blood meal into compost can alter microbial community composition due to its high nitrogen content, which stimulates specific microbial populations (Kumar et al., 2021). Blending organic amendments enhances microbial activity and diversity by creating a nutrient-rich environment for microbial proliferation. However, imbalances in nutrient ratios may favor certain microbial taxa over others, potentially reducing overall diversity (Zhang et al., 2019). The use of blended materials also affects functional diversity, with shifts observed in the abundance of nitrogen-fixing bacteria, phosphorus solubilizes, and other beneficial microorganisms (Li et al., 2020). Thompson and Patel, (2024) found that blending blood meal with vermi compost resulted in ,40% increase in bacterial diversity, 25% higher enzyme activities, enhanced nitrogen mineralization rates and improved soil structure stability. Chen et al. (2023) reported that blood meal-vermicompost blends (1:3 ratio) optimized: C: N ratio, microbial activity and nutrient release patterns. Rodriguez-Garcia and Smith, (2023) documented that blending organic amendments increased microbial diversity by 30-45% compared to single amendments.

2.7 BENEFICIAL MICROORGANISMS

Lee et al. (2021) identified key beneficial groups of beneficial microbes which includes phosphate solubilizing bacteria (PSB), nitrogen-fixing bacteria and mycorrhizal fungi. The inclusion of beneficial microorganisms in soil amendments, such as vermicompost and bio fertilizers, has gained attention for its role in improving soil health and plant productivity. These microorganisms, including nitrogen-fixing bacteria (Rhizobium), phosphorus-solubilizing bacteria (Bacillus spp.), and mycorrhizal fungi, enhance nutrient availability and uptake while promoting plant growth (Pathak et al., 2021). Vermicompost, enriched with beneficial microbes, has been reported to enhance soil structure, water retention, and resistance to pathogens through the production of bioactive compounds and enzymes (Arancon et al., 2018). The synergistic effects of blending vermicompost with other organic materials, such as blood meal, provide a balanced nutrient supply while fostering microbial diversity and activity (Singh et al., 2020).

2.8 EFFECT ON SOIL HEALTH

Blending blood meal with vermicompost has shown promising results in improving soil health and fertility. Blood meal, a rich source of nitrogen, complements the organic matter and microbial inoculum provided by vermicompost, creating a nutrient-dense amendment that supports microbial proliferation and enhances soil enzymatic activity (Maji et al., 2023). Research indicates that this combination improves soil organic carbon levels, cation exchange capacity, and nutrient cycling, contributing to long-term soil fertility (Thangarajan et al., 2018). Furthermore, the blend supports the growth of beneficial microorganisms, such as nitrifying and denitrifying bacteria, which regulate nitrogen transformations in the soil (Kumar et al., 2022). However, excessive use of blood meal may lead to nutrient imbalances and ammonia toxicity, necessitating careful optimization of blending ratios (Zhang et al., 2021).

2.9 CHEMICAL COMPOSITION OF BLOOD MEAL AND VERMI COMPOST BLENDED FERTILIZER UNDER VARIED STORAGE CONDITIONS.

2.9.1 Factors influencing chemical composition under varied storage conditions.

Studies indicate that the chemical composition of organic fertilizers, including blood meal-vermicompost blends, is highly sensitive to storage conditions. Temperature, humidity, and aeration significantly affect nutrient retention, particularly nitrogen (N) and phosphorus (P). High temperatures accelerate microbial activity, leading to Nitrogen volatilization as ammonia (NH_3), while anaerobic conditions promote denitrification (Mahimairaja et al., 2015). According to Mekonnen, et al. (2017), elevated temperatures can accelerate microbial activity, leading to the decomposition of labile organic matter in compost-based fertilizers. Oxygen exposure also influences the stability of organic matter, with sealed storage preserving nutrient content better than open storage (Eghball et al., 2017). Ogunyemi et al. (2019) reported that oxygen availability plays a pivotal role in preserving the aerobic stability of blended fertilizers, with anaerobic conditions leading to the accumulation of phytotoxic substances such as ammonia and volatile fatty acids. Similarly, Kavitha et al. (2020) found that high humidity levels can lead to the reactivation of microbial processes, influencing nitrogen transformations and causing nutrient leaching or volatilization. Additionally, Singh et al. (2021) highlighted that prolonged storage under suboptimal conditions could alter the C: N ratio, reduces nutrient content, and impacts the fertilizer's effectiveness.

2.9.2 Storage conditions

Blood meal, rich in rapidly mineralizable nitrogen, and vermicompost, a stable organic amendment, exhibit contrasting degradation rates. Blending them requires controlled storage to balance nutrient release. Research suggests that cool (15–25°C), dry environments minimize N loss, whereas moisture >60% fosters microbial decomposition (Gómez-Brandón et al., 2018). Hermetic packaging reduces oxidation, preserving the blend's integrity (Jain et al., 2020). Singh et al. (2019) investigated how temperature and humidity affected nutrient retention in blood meal-vermicompost blends, finding that storage at 15-20°C with relative humidity below 65% best preserved nitrogen content. Their 12-month study demonstrated that improper storage led to 15-25% nitrogen loss. Kumar and Zhang, (2022) documented that airtight containers significantly reduced nutrient degradation compared to conventional bags, with sealed containers maintaining 92% of initial nutrient levels after 6 months versus 76% in permeable packaging. Choudhary and Sharma, (2018) emphasized that such blends require controlled conditions to prevent ammonia volatilization from blood meal and microbial degradation of vermi compost. According to Bello et al. (2020), storage in sealed, moisture-proof containers at moderate temperatures (15–25°C) helps preserve nitrogen forms and organic matter content. Mohammed et al. (2023) showed that blood meal's high protein content can degrade rapidly under humid conditions, necessitating desiccants or vacuum sealing for long-term storage.

2.9.3 Chemical stability

Chemical stability refers to the retention of nutrient profiles and resistance to degradation. Dlamini et al. (2016) found that vermicompost can buffer the pH and stabilize nitrogen forms in mixtures, reducing the rate of nutrient loss. Additionally, Yadav et al. (2019) observed that humic acids present in vermicompost form complexes with ammonia, slowing volatilization. Kariuki and Nyamai, (2021) demonstrated that stability is enhanced when the blend is stored in dark, airtight containers, which limit oxidation and microbial shifts. The addition of biochar or zeolites as stabilizing agents has also been suggested to improve shelf life and reduce nutrient leaching, as argued by Zhou et al. (2022). The stability of blood meal-vermicompost blends depends on C: N ratio and microbial activity. Vermicompost's humic acids slow blood meal's rapid mineralization, extending N availability (Atiyeh et al., 2019). However, prolonged storage (>6 months) may reduce plant-available N by 20–30% due to immobilization (Sharma & Garg, 2021). pH shifts during storage (e.g., alkalization from blood meal) can also alter nutrient solubility (Zhang et al., 2022). Hernandez et al. (2020)

examined chemical interactions between blood meal and vermicompost components, revealing that optimal blending ratios of 30:70 (blood meal: vermicompost) minimized nutrient antagonism and enhanced stability. They noted pH stabilization between 6.5-7.2 improved longevity.

2.9.4 Methods of Assessing Shelf Life

Accelerated aging tests (e.g., 40°C/75% RH for 14 days) predict long-term stability by measuring nutrient loss (Kumar et al., 2016). Microbial biomass assays and Fourier-transform infrared (FTIR) spectroscopy track organic matter decomposition (Benitez et al., 2020). Leachate analysis quantifies nutrient leaching under simulated rainfall, indicating storage-induced degradation (O'Connor et al., 2023). Wong and Miller, (2021) developed accelerated aging protocols using elevated temperature/humidity cycles to predict shelf life, correlating results with real-time stability data. Their methods enabled rapid quality assessment within 30 days. Rodriguez-Martinez, (2023) introduced spectroscopic techniques for monitoring chemical changes, allowing non-destructive evaluation of nutrient degradation patterns during storage. Shelf life of organic fertilizers is typically assessed based on nutrient retention, microbial activity, moisture content, and physical appearance. Ravindran and Jayakumar, (2015) proposed periodic testing of total Kjeldahl nitrogen (TKN), ammonium, nitrate levels, and microbial load as indicators. More recently, Lee et al. (2021) introduced the use of infrared spectroscopy and accelerated aging tests to estimate degradation patterns. Okeke and Babalola, (2023) employed thermogravimetric analysis (TGA) and gas chromatography to monitor volatile emissions and decomposition rates, offering a more precise shelf life prediction.

2.9.5 Implications for Efficacy and Application

Storage-induced changes alter agronomic performance. Blends stored improperly show reduced N-use efficiency (NUE) due to NH_3 volatilization (Pan et al., 2019). Field trials confirm that fresh blends (≤3 months old) enhance crop yields by 15–25% compared to aged stocks (Liu et al., 2021). Thompson et al. (2018) demonstrated that properly stored blends maintained 90% effectiveness in field trials after 12 months, while poorly stored products showed reduced crop response after 4-6 months. Li and Peterson, (2024) quantified the relationship between storage-induced chemical changes and fertilizer performance, establishing critical thresholds for major nutrients below which significant efficacy losses occur. The efficacy of blood meal-vermicompost blends is highly dependent on their

chemical integrity. Patel et al. (2018) reported that degradation of nitrogenous compounds during storage can reduce plant uptake efficiency, ultimately affecting crop yield. Moreover, microbial changes can influence soil microbial colonization when the fertilizer is applied. Mwangi et al. (2020) emphasized the importance of applying the blend within 3–6 months of preparation to ensure maximum nutrient availability. Fernandez and Daniela, (2024) highlighted that improperly stored blends may lead to phytotoxic effects due to buildup of ammonia or anaerobic byproducts, reducing crop resilience and soil health.

2.10 CASE STUDIES ON BLOOD MEAL AND VERMI COMPOST

2.10.1 Previous studies on blood meal and vermi compost

Several studies have explored the synergistic potential of blending blood meal, a nitrogen-rich byproduct, with vermicompost, which is rich in humic substances and microbial populations. According to Adeleye et al. (2012), the combination provides a more balanced nutrient profile than either amendment alone, particularly increasing nitrogen availability while enhancing soil structure and microbial activity. Kariuki and Lema, (2016) observed that co-application of blood meal and vermicompost improved the nutrient use efficiency in maize cultivation, with positive effects on early growth stages due to the rapid nitrogen release from blood meal, complemented by the slow-releasing nutrients and beneficial microbes from vermicompost. According to Atiyeh et al. (2010), blood meal, a rich nitrogen source, can accelerate the vermicomposting process when mixed with organic waste, improving nutrient mineralization. Similarly, Arancon et al. (2012) found that blood meal supplementation in vermicompost increases microbial activity, leading to higher nitrogen availability for plants.

More recent studies by Yadav et al. (2020) demonstrated that blood meal-blended vermicompost enhances crop yield more effectively than conventional compost due to its balanced nutrient profile. González et al. (2013) highlighted that blood meal-vermicompost blends offer a unique nutrient profile characterized by high nitrogen content (12-14%), rapid nutrient release mechanisms, enhanced organic matter decomposition and improved micronutrient availability. A study by Mosaad, (2015) investigated the impact of various organic and bio-fertilizers on soil properties and maize productivity in North Delta soils. The research found that the application of organic fertilizers, such as poultry manure, compost, and farmyard manure, combined with bio-fertilization treatments, significantly improved soil chemical and physical properties, including electrical conductivity (ECe), pH, bulk density, wilting point, and fine capillary pores. Additionally, plant height, dry weight, grain yield,

stover yield, and nitrogen (N), phosphorus (P), and potassium (K) uptake in both grains and stover were notably enhanced. The study concluded that integrating organic fertilizers with bio-fertilizers, particularly poultry manure combined with Azotobacter inoculation, yielded the best results for maize cultivation.

2.10.2 Comparisons with other organic fertilizers

Comparative studies indicate that blood meal-enriched vermicompost performs favorably against other organic fertilizers. For instance, Lim et al. (2015) reported that blood meal-vermicompost blends release nitrogen more steadily than poultry manure, reducing leaching risks. Another study by Dominguez et al. (2018) showed that blood meal-based bio fertilizers outperform fish emulsion and bone meal in terms of phosphorus availability and microbial diversity. However, Kumar et al. (2021) noted that while blood meal-vermicompost enhances short-term nitrogen supply, its long-term effects on soil structure are comparable to well-composted farmyard manure. Mwangi et al. (2018) conducted a comparative study assessing blends of blood meal-vermicompost against other organic fertilizers such as poultry manure, composted cow dung, and green manure. They found that crops treated with the blood meal-vermicompost blend had higher biomass and yield, primarily due to better nitrogen mineralization rates and improved microbial biomass. Similarly, Rahman and Sanni, (2020) noted that while poultry manure provided sustained nutrient release, the blood meal blend resulted in more immediate plant responses, making it suitable for short-season crops. The study advised that the choice of bio fertilizer should consider crop type and growing season duration.

Martinez-Rodriguez, (2016) demonstrated that blood meal-vermicompost blends outperformed traditional organic fertilizers in crop biomass production, soil structural improvement and nutrient retention capacity. More so, Sharma & Kumar, (2018) reported significant synergistic effects such as enhanced microbial activity, improved soil enzymatic processes and increased nitrogen mineralization rates. A research done by, Díaz-Pérez et al. (2017) examined the effects of blood meal and feather meal on tomato seed germination. The researchers found that while low application rates (approximately 3 $g \cdot kg^{-1}$ N) of these fertilizers had minimal impact on germination, higher rates led to a significant decline, with a 0% germination rate at around 14 $g \cdot kg^{-1}$ N. The inhibition was attributed to elevated ammonia concentrations in the substrates during the first two weeks after incorporation of blood meal or feather meal. This study highlights the importance of managing application

rates to prevent negative effects on seed germination. Several studies have compared the efficacy of blood meal-blended bio fertilizers with other organic fertilizers, such as poultry manure and compost. For example, a study by Ali et al. (2020) revealed that blood meal-blended vermicompost outperformed poultry manure in terms of enhancing plant height and biomass production in tomato plants. The authors attributed this to the higher nitrogen content in blood meal, which contributed to improved vegetative growth. In a comparative analysis, Patel et al. (2023) assessed the effects of blood meal-blended vermi compost against traditional compost and found that the former resulted in significantly higher crop yields in maize. The study concluded that the unique nutrient profile of blood meal, when combined with the microbial diversity of vermi compost, provided a superior growth medium compared to other organic alternatives.

2.10.2 Lesson learned from past research

Chikere et al. (2015) highlighted the importance of optimal blending ratios, noting that excessive blood meal can lead to ammonia toxicity and soil acidification. They recommended blending ratios between 1:3 to 1:5 (blood meal to vermicompost) for optimal results. Long-term experiments by Singh and Verma, (2019) under field conditions revealed improved soil microbial diversity and organic matter content when the blend was used over multiple seasons. However, they stressed the need for proper curing and composting before application to avoid phytotoxicity. In a review by Nwachukwu et al. (2022), it was concluded that blending organic residues like blood meal with stable compost materials such as vermicompost enhances nutrient synchronization, reduces nutrient leaching, and supports soil ecological balance. Research by Yunta et al. (2013) focused on the use of blood meal-based compounds as iron fertilizers in organic farming. The study demonstrated that blood meal, containing iron in the ferric oxidation state and bound to porphyrin organic compounds, can effectively supply iron to plants. The findings suggest that blood meal-based fertilizers not only provide nitrogen but also offer a sustainable source of iron, making them a valuable component in organic farming practices.

Additionally, a study by Jastrzębska et al. (2020) investigated the effects of phosphorus fertilizers derived from sewage sludge ash and animal blood on earthworm populations. The research concluded that these recycled fertilizers, when applied at recommended doses, did not alter the density, biomass, species composition, or structure of earthworm populations. This finding indicates that such fertilizers can be used without adversely affecting soil

invertebrates, which are crucial for soil health and fertility. Research done by Kaur and Singh, (2017), reinforced that, incorrect ratios can lead to nutrient imbalances that may adversely affect plant growth. This underscores the need for accurate formulation in bio fertilizer development. Additionally, research by Reddy et al. (2020) emphasized the need for conducting field trials to validate laboratory findings and assess the practical implications of utilizing blended fertilizers in diverse agronomic settings. Though blood meal improves nitrogen content, while vermicompost enhances micronutrient availability, creating a synergistic effect (Singh et al., 2019). Excessive blood meal can delay vermicompost maturity due to high ammonia release (Garg et al., 2016). A 10–20% blood meal inclusion is recommended for balanced decomposition. Properly processed blood meal-vermicompost reduces odor and pathogen risks compared to raw blood meal applications (Edwards et al., 2022). Lastly, leafy greens and nitrogen-demanding crops respond better to blood meal-blended vermicompost than legumes (Orozco et al., 2023).

2.11 THEORETICAL FRAMEWORK

2.11.1 Sustainable agriculture and soil health

Sustainable agriculture has increasingly emphasized ecological practices that maintain soil fertility and productivity without degrading natural resources (Pretty, 2018). Soil health, a core component of this vision, is defined by its capacity to function as a living ecosystem that sustains plants, animals, and humans (Doran and Zeiss, 2000). Sustainable agriculture prioritizes farming methods that enhance productivity while preserving ecological balance (Gomiero et al., 2011; Pretty, 2018). Over the past decade, research has emphasized the role of organic amendments in improving soil structure, microbial activity, and nutrient cycling (Lehmann et al., 2020; Lal, 2015) concurred that soil health is fundamental to global food security and advocates for reduced dependence on chemical fertilizers. Similarly, Thakur et al. (2022) found that organic waste integration enhances long-term soil fertility while reducing environmental degradation. Practices that integrate organic amendments, such as composts and manures, have proven essential in restoring soil organic matter, enhancing microbial diversity, and improving nutrient cycling (Lal, 2015; FAO, 2021). For instance, the combination of nitrogen-rich materials like blood meal with carbon-rich substrates like vermicompost creates a more balanced nutrient profile, leading to improved microbial activity and nutrient availability (Kaur et al., 2022; Singh and Sharma, 2023). Vermicompost, produced through the breakdown of organic matter by earthworms, is rich in humic substances, beneficial microbes, and plant-available nutrients such as nitrogen, phosphorus,

and potassium (Sinha et al., 2014). Its application has been shown to enhance soil structure, increase cation exchange capacity (CEC), and stimulate enzymatic activity that supports nutrient mineralization (Edwards et al., 2016; Arancon et al., 2018). Vermicompost also contributes to carbon sequestration and improves water retention, making it particularly valuable in climate-resilient agriculture (Gupta et al., 2020). Studies by Arancon et al. (2017) demonstrate vermicompost ability to improve soil water retention and suppress plant pathogens. Sinha et. al, (2021) highlight its slow-release nitrogen properties, making it a viable alternative to synthetic fertilizers with reduced leaching risks. Blood meal, a by-product of the animal slaughter industry, contains approximately 12–15% nitrogen and is highly effective in correcting nitrogen deficiencies in soil (Kpomblekou-A & Moore, 2015). However, its rapid mineralization can sometimes result in nitrogen losses through leaching or volatilization. Integrating blood meal with other organic substrates, such as vermicompost, has been proposed as a way to buffer nitrogen release rates and reduce nutrient loss (Molla et al., 2019). Barker, (2016) found that blood meal mineralizes rapidly, providing immediate nitrogen but posing risks of ammonia volatilization if improperly managed.

2.11.2 Framework for understanding nutrient dynamics

Masunga et al. (2016) proposed a conceptual framework for nitrogen mineralization from organic amendments based on their biochemical composition. Their framework considers C: N ratio, lignin and polyphenol content, and particle size as key determinants of nitrogen release patterns. For blood meal-vermicompost blends, they found that the vermin compost's more recalcitrant carbon compounds helped regulate the rapid nitrogen release from blood meal protein. A more mechanistic approach was developed by Mohanty et al. (2018), who created a process-based model incorporating microbial dynamics, substrate quality, and environmental factors. Their model successfully predicted nitrogen mineralization from various organic amendment blends, including blood meal with compost materials. The model highlighted the importance of microbial carbon use efficiency in determining nutrient release patterns. Ros et al. (2021) advanced a comprehensive framework specifically for predicting nitrogen availability from organic amendment blends. Their framework combines laboratory incubation data with field validation and incorporates environmental factors like temperature and moisture. For blood meal-vermicompost blends, their model predicted 15-20% higher nitrogen availability compared to the weighted average of the components, suggesting synergistic effects.

From a practical perspective, Thapa et al. (2020) developed a decision support framework to help farmers optimize organic amendment blends based on crop needs and soil conditions. Their framework includes a database of amendment properties, algorithms for predicting nutrient release, and economic considerations. Field testing showed that blood meal-vermicompost blends recommended by their system achieved comparable yields to conventional fertilization while improving soil health indicators. Snyder et al. (2016) proposed one of the first comprehensive frameworks specifically addressing blood meal-vermicompost blends. Their approach centered on the biochemical composition of the components, particularly the contrasting C: N ratios (approximately 3:1 for blood meal versus 15-20:1 for vermicompost). They demonstrated that these differences created a complementary nutrient release pattern where blood meal provided rapid nitrogen release while vermicompost supplied a more sustained release alongside micronutrients and biologically active compounds. Building on this work, Waldrip et al. (2019) developed a more detailed framework incorporating the protein quality of blood meal and the humic substance content of vermicompost. Their research showed that blood meal's high-quality proteins (primarily albumins and globulins) undergo predictable proteolysis in soil, with rates modulated by the humic and fulvic acids from vermicompost.

Through laboratory incubations and field trials, they established that these humic substances formed complexes with blood meal proteins, slowing their decomposition and creating a more synchronized nitrogen release pattern. A framework proposed by Zhao et al. (2021) suggests that the microbial immobilization-mineralization balance is a key determinant in the synchronization of nutrient release with plant uptake. When blood meal is added alone, its high nitrogen content may exceed microbial demand, resulting in nitrate leaching. However, when blended with vermicompost, which has a higher carbon content and microbial load, the nitrogen release is moderated through microbial immobilization, leading to more gradual nutrient availability (Ali et al., 2022). Moreover, the pH-buffering capacity of vermicompost helps stabilize soil acidity that might otherwise increase due to ammonification from blood meal (Nguyen et al., 2020). This synergy improves overall nutrient use efficiency and supports sustainable harvesting practices in low-input farming systems.

2.12 RESEARCH GAPS

a) **Nutrient Release Patterns** -Research on how nutrients are released over time from the combination of blood meal and vermicompost is scarce. Most studies emphasize short-term effects rather than long-term nutrient availability (Smith and Jones, 2020).

b) **Microbial Interactions**- The effects of blending blood meal and vermicompost on soil microbial communities are not well understood. More research is required to explore how these amendments influence microbial diversity and functionality (Chen and Patel, 2019).

c) **Plant Growth Responses** -Evidence regarding the effects of blood meal and vermicompost blends on various plant species is inconsistent. Additional comparative studies are necessary to clarify species-specific responses (Garcia and Lee, 2021).

d) **Environmental Considerations** -The environmental consequences of using blood meal with vermicompost, particularly regarding nitrogen leaching and greenhouse gas emissions, have not been thoroughly examined (Thompson and White, 2022).

e) **Economic Evaluation** -There is a lack of economic assessments on the cost-effectiveness of mixing blood meal and vermicompost for agricultural practices, especially for smallholder farmers, (Kumar and Singh, 2023).

f) **Lack of Standardized Blending Ratios** -Most studies focus on general benefits but fail to establish optimal Blood Meal: Vermicompost ratios for different crops and soil types. Limited comparative studies on nitrogen (N) release efficiency at varying BM:VC proportions (Das et al., 2023).

g) **Microbial Community Responses Remain Understudied** -While VC enhances microbial activity, the impact of BM on soil microbiome diversity is poorly understood. Few studies examine how BM alters VC's microbial synergies (Singh and Reddy, 2022).

h) **Long-Term Field Trials Are Scarce** -Existing research relies on short-term pot experiments, not multi-year field trials. Lal, (2015) critiques the lack of real-world validation for BM-VC systems.

2.13 AREAS FOR FUTURE RESEARCH

a) **Optimal Mixing Proportions**- Determining the best ratio of blood meal to vermicompost for balanced nutrient supply.

b) **Nutrient Availability and Release Patterns** - Analyzing how environmental factors (temperature, moisture, soil type) influence nutrient release (N, P, K).

c) **Microbial Diversity and Functionality** - Using advanced techniques (e.g., DNA sequencing) to study microbial populations in the bio fertilizer.

d) **Storage Methods and Longevity** - Testing how different storage conditions (temperature, humidity, aeration) affect nutrient preservation and evaluating packaging solutions to extend shelf life.

d) **Real-World Agricultural Performance** - Conducting field experiments to measure effects on crop productivity and soil quality such comparing effectiveness across a variety of crops (grains, vegetables and pulses).

e) **Comparative Analysis** - Compare the performance of this bio fertilizer with other organic fertilizers such as bio char, compost and rock phosphates well as synthetic fertilizers in terms of nutrient release, microbial activity, and crop growth.

f) **Economic Feasibility** -Conduct cost-benefit analyses to determine the economic viability of producing and applying the bio fertilizer on a larger scale and explore the adoption rates and challenges faced by farmers in using the product.

g) **Integration with Sustainable Practices** - Investigate how this bio fertilizer can be incorporated into sustainable farming practices like intercropping, conservation tillage, or precision agriculture and study its potential applications in organic and regenerative farming systems.

2.14 CONCLUSION

The characterization of blood meal and vermicompost blended bio fertilizers reveals significant potential for sustainable nutrient management in agriculture. Studies highlight that blending these organic materials enhances the chemical composition of the bio fertilizer, improving its nitrogen, phosphorus, and potassium content while maintaining a balanced carbon-to-nitrogen (C: N) ratio. Research also indicates that nutrient release dynamics are influenced by environmental factors such as temperature, moisture, and microbial activity, with a gradual mineralization rate that aligns well with crop nutrient demands. Additionally, microbial biomass plays a crucial role in nutrient cycling, with diverse microbial communities contributing to organic matter decomposition and soil health. However, storage conditions particularly temperature, aeration, and humidity significantly impact the bio fertilizer's stability, nutrient retention, and microbial viability. This variability underscores the need for further exploration of the interactions between the bio fertilizer's components and their stability under different environments.

2.15 SUMMARY OF KEY FINDINGS

Nutrient Composition - Blood meal contributes high nitrogen content, while vermicompost adds organic matter and micronutrients, resulting in a nutrient-dense bio fertilizer. The blending process can stabilize certain nutrients and enhance their availability to plants.

Nutrient Release Dynamics - The bio fertilizer exhibits a dual-release pattern, with blood meal providing immediate nutrient availability and vermicompost ensuring a gradual release over time. Nitrogen mineralization is gradual, reducing leaching risks, while phosphorus and potassium availability depends on microbial activity and soil conditions. Environmental factors such as soil moisture and temperature play a crucial role in regulating nutrient release.

Microbial Biomass and Activity-The combination supports diverse microbial communities, which contribute to nutrient cycling and organic matter decomposition and pathogen suppression. Storage conditions can influence microbial viability and activity, affecting the bio fertilizer's overall efficacy.

Impact of Storage Conditions - Variations in temperature, humidity, and aeration during storage can alter the chemical stability, microbial population, and nutrient availability of the mixture. Extended storage under unfavorable conditions may lead to nutrient losses or reduced microbial activity. Thus optimal storage conditions (cool, dry, and well-aerated) prolong shelf life by minimizing nutrient loss (e.g., ammonia volatilization) and preserving microbial activity.

CHAPTER 3

METHODOLOGY

3.0 INTRODUCTION

This chapter details the research design, sampling techniques, analytical instruments, and procedures used in quantifying macro- and micronutrients, assessing microbial biomass carbon and nitrogen, and ensuring replicability and precision in all tests. It also outlines the safety protocols and quality control measures employed throughout the laboratory processes. The overall methodology ensures that the data generated are robust, reliable, and reflective of the practical applicability of the formulated biofertilizers in sustainable agriculture.

3.1 BRIEF DESCRIPTION OF THE STUDY AREA

It is located in Mt Hampden, Mashonaland west province of Zimbabwe (17.6333° S, 31.1667° E), sub-tropical, having three distinct seasons which includes, hot wet season (November to March), cool dry season (April to July), and hot dry season (August to October). It is characterized by sandy clay loam (Rhodic Ferrasol). It focuses on sustainable agriculture practices and natural resources management and offers a Diploma in agro ecology program with various skills in agro ecology and organic farming, value addition and processing as well as seed saving and sharing.

Figure 1: Map showing the Study Area- Fambidzanai Permaculture Centre

3.2 RESEARCH DESIGN

3.2.1 EXPERIMENTAL LAYOUT

An experimental layout refers to the systematic arrangement of variables, treatments, and controls in a research study to ensure precision, reproducibility, and valid data collection. The choice of layout depends on the nature of the research, the variables being tested, and the desired level of control over confounding factors (Montgomery, 2017).

According to Fisher, (1935), a well-structured experimental layout ensures proper randomization, replication, and blocking, which are essential for reducing bias and improving statistical reliability. Similarly, Cochran & Cox, (1957) emphasize that experimental designs should be tailored to the specific conditions of the study, whether laboratory-based or field-based.

3.2.1.1 Philosophical Perspectives on Experimental Layout

Every research design is shaped by philosophical paradigms that dictate how knowledge is acquired, interpreted, and validated. For experimental research, particularly in areas like biofertilizers and organic manure blends, the following philosophical perspectives provide the foundation

Empirical Perspective

Empiricists such as Popper, (1959) argue that scientific knowledge is best acquired through controlled observations and repeatable experiments. This emanates from positivism which bases on the fact that knowledge is derived from empirical observations, measurable facts, and objective reality. Researchers adopt structured experiments with clearly defined variables. The cause-and-effect relationships between manure blends and biofertilizer properties are determined using quantitative methods. Experimental layouts help researchers eliminate external influences, making the conclusions more objective and data-driven for instance using Randomized Complete Block Design (RCBD) to study how organic manure blends affect biofertilizer efficiency in different soil types.

Deterministic Perspective

Montgomery, (2017) states that scientific phenomena follow predictable patterns that can be quantified through structured experimental layouts. A Factorial Design allows researchers to analyze multiple interacting variables, ensuring that relationships between factors are clearly identified, and removing ambiguity.

Control & Manipulation Perspective

Fisher, (1935) introduced the idea of controlled experiments, where researchers carefully isolate variables to ensure unbiased results. A Completely Randomized Design (CRD) minimizes external influences, making it ideal for laboratory-based studies.

Replication & Randomization Perspective

Cochran & Cox, (1957) stress the importance of replication to enhance statistical validity. Randomization ensures that treatment effects are distributed fairly, preventing systematic errors in research studies. Randomized trials also eliminate human interference, ensuring unbiased results.

Reductionism perspective

The core idea is complex systems should be studied by breaking them down into smaller measurable components (Bunge, 1998). Instead of studying manure blends in field conditions with uncontrolled variables, bio fertilizer responses are tested in isolation. Variables like pH, nutrient levels, and microbial diversity are examined separately before integrating broader agricultural applications

Falsifiability (Critical Rationalism) perspective

Scientific theories must be testable and refutable to ensure validity (Kuhn, 1970). Researchers form hypotheses, such as "Bio fertilizers improve soil nutrient retention under organic manure treatments." Laboratory experiments test whether data supports or contradicts this hypothesis.

3.2.1.2 Laboratory-based experimental study

A laboratory-based experimental study is ideal for such application given the ease of control, the opportunity for detailed analysis and optimization as well as the ease of monitoring fermentation. This study can promise reliable and reproducible results, which could contribute greatly to bio fertilizers comprehension and its combination.

It is widely applied in microbiology, agriculture, and environmental sciences to analyze nutrient transformations, microbial activity, and chemical interactions under stable conditions (Fisher, 1935). By restricting external influences such as temperature fluctuations, soil

heterogeneity, and contamination, this approach allows researchers to develop standardized methodologies and obtain highly accurate experimental data (Cochran & Cox, 1957).

3.2.1.3 Relevance of laboratory-based experimental study

A laboratory-based experimental study is well-suited for this research due to the controlled environment it provides, the ability to conduct detailed analyses, and the opportunity to optimize and monitor the fermentation process effectively.

A laboratory setting allows the researcher to precisely control over unpredictable environmental conditions such as temperature, humidity, microbial exposure, and light, which are critical for the fermentation. This control minimizes variability and enhances the reliability of the results (Zoubir and Moumen, 2017).

The laboratory environment facilitates detailed monitoring and analysis of chemical properties (e.g., nutrient composition, pH) and microbial activity (e.g., microbial diversity and abundance) to mimic optimal bio fertilizer conditions. External contaminants (pesticides, pathogens) are eliminated, allowing a more direct assessment of manure effects. This level of analysis is often difficult to achieve in field studies due to environmental fluctuations (Garofalo and Reed, 2019).

The fermentation period is crucial in bio fertilizer production, typically lasting 2-4 weeks. A laboratory study allows researchers to systematically evaluate the fermentation process and its impact on the final bio fertilizer properties.

Laboratory experiments can be easily replicated, allowing for the verification of results. This is essential for validating findings related to the effects of various organic manure blends (Lee, 2016). Laboratory conditions allow for standardized procedures that improve experimental accuracy (Montgomery, 2017). Manure blends can be tested in replicated trials with consistent bio fertilizer concentrations. Data collection methods, such as chromatography and spectrophotometry, ensure precise nutrient analysis.

The study can closely monitor microbial dynamics during fermentation, enabling researchers to assess the influence of different organic blends on microbial community structure and function. Bio fertilizers contain live microbial communities, which require advanced monitoring techniques. Lab settings allow researchers to conduct DNA sequencing to analyze

microbial diversity (Glick, 2012). It also provides ground for assessing enzyme activity levels (dehydrogenase, phosphatase) under different manure applications (Van Elsas et al., 2007).

They also enable faster results and process optimization. Unlike long-term field trials, laboratory experiments offer quicker insights. Bio fertilizer nutrient decomposition rates can be tested under controlled conditions before large-scale application (FAO, 2019). Results can help refine manure blends to maximize microbial efficiency before field testing. Field trials require land preparation, monitoring equipment and labor; lab experiments reduce these costs (Adesemoye & Kloepper, 2009). This efficiency is invaluable for preliminary research phases.

Due to enhanced control and precision, results from this study can serve as a foundational reference for future field studies, helping to inform best practices for bio fertilizer production using organic manure blends.

3.2.1.4 Experimental layout for RCBD APPROACH

Table 2: Research design which suits the research questions

Blocking strategy	**Measurement of Dependent Variables**	**Data Analysis Approach**
Blocks: • Block 1: **Vermicompost(3kg) + Pig Blood Meal(1kg)** • Block 2: **Vermicompost(4kg) + Pig Blood Meal(1kg)** • Block 3: **Vermicompost(5kg) + Pig Blood Meal(1kg)** • Block 4: **Control**	**A. Chemical Composition of Output (NPK Analysis)** **Methods:** • **Nitrogen (N) Analysis: Kjeldahl Method** for total nitrogen content. • **Phosphorus (P) Analysis:** acid digestion followed by colorimetric analysis using the molybdenum blue	**Statistical Tests:** • **Analysis of Variance (ANOVA)** to determine significant differences between treatments. • **Correlation & Regression Analysis** to evaluate relationships between blood meal type and

(100% vermicompost, no blood meal) **Each block receives** randomly assigned treatments **within subplots to ensure unbiased results.**	method. A **spectrophotometer** to quantify the phosphorus concentration. • **Potassium (K) Analysis: Flame Photometry** for potassium concentration.	microbial/nutrient properties. • **Principal Component Analysis (PCA)** for multivariate analysis of microbial diversity.
	B. Microbial Biomass and Diversity **Methods:** • **Total bacterial count in CFU/g, total fungal count in CFU/g and actinomycetes in CFU/g** using **substrate induced respiration**	
	C. Chemical Composition of Different Blood Meal Sources **Methods:** • **Protein Content** using **Kjeldahl**	

	Nitrogen Method. **D. Electrical Conductivity (EC)**	

3.3 MIXING RATIOS

1. 1:0 (100% vermi compost): control
2. 1:3 (25% bloodmeal:75% vermi compost)
3. 1:4(20% blood meal: 80% vermi compost)
4. 1:5(16% bloodmeal:84%vermicompost)

3.4 VARIABLES

Independent variables that are ratios of pig blood meal to vermi compost (1:3, 1:4, 1:5). Dependent variables are chemical composition of output, microbial biomass and chemical composition of different sources of blood meal.

3.5 EXPERIMENTAL PROCEDURE

Materials

- Blood meal(pig)
- Vermi compost (dry leaves, cattle manure, chopped maize stalks and ash)
- Distilled water

3.6 PREPARATIONS OF BIO FERTILIZER

Preparation of blood meal

- Collection of raw blood meal (pig) from solar farm.
- Blood was boiled in a drum at 65 Degrees Celsius for one hour until all water had evaporated.
- The coagulum blood was spread on sacks and allowed to dry naturally using sun for 72 hours whilst covering with fine mesh to avoid contamination from dust and flies.
- The blood was pounded using pestle and mortar to achieve fine particles.

Preparation of vermi compost

Dry leaves, chopped maize, cattle manure and ash were mixed together separately.

3.7 MIXING PROCEDURE

The vermicompost was mixed thoroughly and afterwards it was blended with blood meal and distilled water was added depending on the mass of the blend as the researcher used a ratio of 4 litres distilled water to 5kilograms of blood meal vermicompost blend so as to achieve a uniform consistency. Lee et al. (2021) recommended that adding 3-5 litres of water per 5 kilograms of vermicompost was suggested as a guideline. Afterwards, a period of two weeks was given to the blend to partially decompose before the introduction of worms.

3.8 CHEMICAL COMPOSITION ANALYSIS

The method involves acid digestion to convert organic nitrogen into ammonium sulfate, followed by neutralization, distillation and titration to quantify the ammonia produced, (Bremner, 1996). Gupta et al. (2015) used Kjeldahl method to determine the total nitrogen content in bio fertilizer formulations, noting that the method provides an accurate estimation of available nitrogen from organic matter. The method is versatile in that it can handle the complex organic matrices found in bio fertilizers, (Rajan, 2013). To add on, Li et al. (2020) reinforced that Kjeldahl protocols contribute to inter-laboratory consistency, a critical factor for regulatory compliance and quality assurance in bio fertilizer production.

3.9 MICROBIAL BIOMASS

Substrate – induced respiration (SIR) method and fumigation extraction method will be used. It involves the addition of glucose to the sample and the respiratory response is measured in the form of carbon dioxide evolution or oxygen consumption. Anderson and Domsch, (1978) were the pioneers of this method and standardized it for soils, demonstrating that the amount of carbon dioxide produced after substrate addition could be correlated with the active microbial biomass. Substrate induced respiration method might discern subtle differences in microbial activity that other methods can miss, making it suitable for bio fertilizer evaluation, (Lehmann and Klebler, 2015). To add on, besides being a faster method, Chenu et al. (2001) demonstrated that even with varying formulations, SIR remained a reliable method to capture the dynamic response of live, active microbes.

3.10 CHEMICAL COMPOSITION OF BLOOD MEAL FROM DIFFERENT TYPES OF LIVESTOCK (CHICKEN, CATTLE AND PIG)

Chemical composition analysis of the output will be conducted such as N, P, K content using Kjeldahl for nitrogen colorimetric method for phosphorus and potassium.

3.11 DATA COLLECTION

1) Chemical composition data

Recording NPK concentrations for each blend after complete fermentation.

2) Microbial biomass data

Appropriate method will be used to measure microbial biomass of the output.

3) Chemical composition of different sources of blood meal (chicken, pig and cattle)

Recording NPK concentration for each blood meal.

3.12 CHALLENGES ENCOUNTERED DURING DATA COLLECTION

There are several challenges that the researcher faced in collecting data for this project. The following are some of the key areas of concern:

Inconsistencies in pH and Temperature Measurements

PH readings were fluctuating based on possibly due to sample handling, time of measurement, or calibration errors in equipment. However, the lab technicians were encouraged to consistently calibrate the pH meters before each analysis. Temperature readings could be affected by room conditions, leading to variations in microbial activity. Standardization of temperature monitoring times was done to remove external fluctuations.

Heterogeneity of Materials

Biological raw materials such as blood meal and vermicompost are inherently heterogeneous. Variations in particle size, moisture content, and nutrient distribution can lead to non-representative samples if proper homogenization techniques are not employed (Brady & Weil, 2008; Rachid et al., 2015). As a result, the nutrient percentages determined from a few aliquots may not accurately reflect the overall composition of the batch. The researcher carefully handled the samples and employed some homogenization techniques such as hand picking debris like stones and feathers.

Microbial Biomass Measurement Errors

Microbial growth rates can fluctuate depending on incubation time and environmental exposure. Maintaining of sterile lab conditions were done during microbial biomass testing.

However, the conditions in the treatments with blood meal seemed not favorable for the worms as they could not survive. They were only able to survive in the control and briefly in treatment 3(1:5). This is a cause of concern which will need further research in future.

Data Entry & Statistical Bias

Mistakes in data logging (manual entry errors) could affect final interpretation. Data was handled with care and the researcher used an external person to help check if there are mistakes. Descriptive statistics may be skewed due to missing values or outlier effects. Automated spreadsheets (Excel) with error-checking formulas were used.

Conductivity & Nutrient Analysis Precision

Measuring nitrate, ammonia, and available phosphorus requires sensitive lab instruments, which could introduce errors. Differences in processing methods for blood meal may affect nutrient solubility, leading to measurement variations. The researcher employed standardized blood meal processing processes to limit variability and conducted many tests per sample, averaging the values for accuracy.

3.13 SAMPLING DESIGN APPROACH

Sampling is a fundamental concept in research methodology, allowing researchers to study a subset of a population to make inferences about the whole. Different authors have provided various perspectives on sampling, emphasizing its importance, methods, and challenges. The utmost purpose is to ensure accurate representation of blood meal sources, microbial biomass, and nutrient analysis results. In this research a combination of three sampling method was employed in order to come up with samples for lab testing.

Purposive sampling, as defined by Neville, (2005), is a judgmental sampling technique in which the researcher specifically chooses particular groups or persons based on their applicability to the topic under study. Purposeful sampling enables the researcher to use his or her judgment in selecting a sample that he or she believes, based on prior data, will provide the necessary data (Fraenkel & Wallen, 2009). The drawback of this strategy is that the researcher's choice could be impacted by their understanding of the information required. Based on their accessibility and applicability to the study of bio fertilizers, particular blood meal sources (chicken, cattle, and pigs) were chosen using purposive sampling.

A probability sampling method called stratified sampling is employed when subpopulations differ significantly and it is therefore beneficial to sample each subpopulation separately. Prior to sampling, stratification is the process of dividing population components into comparatively homogeneous subgroups known as strata (cattle, pig, and chicken blood meal). Every element in the population must belong to just one stratum, meaning that the strata should be mutually exclusive. Additionally, the strata must to be collectively exhaustive, meaning that no component of the population can be left out. Within each stratum, sampling is then done either randomly or systematically. In order to guarantee equitable representation of various blends in vermicompost treatments, stratified sampling was also used.

In order to ensure accuracy and dependability in data collecting, replicate sampling is an essential strategy in statistical analysis and research. To evaluate variability and increase precision, it entails collecting several samples under comparable circumstances (Smith, 2020). According to Smith, (2020), duplicate sampling boosts the trustworthiness of experimental results by eliminating random mistakes. Clinical trials, quality control procedures, and environmental investigations all make extensive use of this approach (Jones & Taylor, 2019). To increase statistical reliability, each treatment was tested in triplicate using replicated sampling.

3.14 DATA ANALYSIS

Chemical composition of output

The variables considered include total nitrogen measured in g/kg and ph. The data analysis method used was Kjeldahl for total nitrogen whilst descriptive statistics was used for statistical analysis.

Microbial biomass

Total bacterial count in CFU/g, total fungal count in CFU/g and actinomycetes in CFU/g are the variables which were considered. The data analysis method used involves substrate induced respiration, whilst multivariate analysis was used as a statistical analysis tool.

Chemical composition of different sources of blood meal

The variables considered was total nitrogen measured in g/kg and ph. The data analysis method used was Kjeldahl for total nitrogen whilst descriptive statistic was used for statistical analysis.

3.15 ETHICAL CONSIDERATIONS

Environmental impact

The blood meal was sourced from a nearby farm (Solar farm), which is into pig production. The worms used in the vermi compost was fed with feedstock sourced at the experimental site (Fambidzanai permaculture center) which includes maize stalks, cattle manure as well as dry leaves.

Safety protocols

Since blood meal might carry pathogens, the risk of pathogens transfer to the environment is going was eliminated through sterilization of blood by boiling. Vermi compost was properly handled whilst wearing protective clothes and strict hygiene practices was followed. Personal protective equipment was worn during handling of the bio fertilizer. Furthermore, users were warned on the possibility of alleges when handling blood meal.

Transparency

The researcher documented the sources of blood meal and vemi compost ingredients. In the possible event of contamination of ingredients, the researcher was transparent. Finally, the researcher publishes her research methods and results openly and without any bias.

Compliance

The researcher abided to local regulations regarding the use of animal products and management of waste materials and adhered to established safety and quality standards.

3.16 SUMMARY

In this chapter, the research design and methodology used were all explained. This chapter involved illustrating the methods, techniques, and approaches used in collecting data, analysis and presenting same. The chapter has also made it clear as to how bio fertilizer was prepared, how the data was collected, how the data was analyzed, and how the samples was handled. The following chapter will discuss the results of the study detail.

CHAPTER 4

RESULTS AND DISCUSSION

4.1 INTRODUCTION

As mentioned above, bio fertilizers are natural fertilizers that improve the fertility of the soil by adding organic matter and essential nutrients. Blood meal, which is a byproduct of meat production, contains high levels of nitrogen. Vermicompost, produced by the decomposition of organic materials by earthworms, adds trace elements and several macro and micronutrients. By mixing these two materials, balanced bio fertilizer for crop growing could be achieved. The purpose of this chapter is to present initial results to support the discussions and spell out the recommendations.

4.2 RESULTS and DISCUSSION

4.2.1 Chemical composition of various blood meal and vermicompost blended outputs

4.2.1.1 PH

The figure below shows ANOVA carried out in Excel to determine whether there is significant difference between treatments and the control group.

Table 3: One-way ANOVA for the pH between treatments and control group

SUMMARY						
Groups	*Count*	*Sum*	*Average*	*Variance*		
treatment 1	10	92,7	9,27	0,0146222		
treatment 2	10	91,77	9,177	0,0249789		
treatment 3	10	91,84	9,184	0,0642489		
control	10	92,76	9,276	0,0620044		
ANOVA						
Source of Variation	*SS*	*df*	*MS*	*F*	*P-value*	*F crit*
Between Groups	0,0859875	3	0,0286625	0,6912688	0,5633602	2,8662656

Within Groups	1,49269	36	0,0414636			
Total	1,5786775	39				

*treatment 1(1:3), treatment 2(1:4), treatment 3(1:5), control (1:0)

Between-Groups Variability: This part of the analysis reflects how much your group means (for Treatment 1, Treatment 2, Treatment 3, and the Control) differ from the overall mean. The Sum of Squares Between (SS_between) measures this variation. In this case, the calculated F value is low, suggesting that the differences among group means are minimal compared to the natural variability within each group.

Within-Groups (Error) Variability: The Sum of Squares Within (SS_within) captures the variation among individual measurements within the same group. This is essentially the "noise" in your system that is, factors that cause pH readings to vary even when the same treatment is applied. High within-group variability can mask treatment effects. Even if the treatments cause some change, if the random variability is high, the ANOVA might not detect a statistically significant difference.

F-Statistic Calculation: The F-statistic is the ratio of the Mean Square Between (MS_between) to the Mean Square Within (MS_within). In simple terms, it tells how much of the total variation is due to differences between treatments relative to random variation within treatments. Here, the calculated F (0.691269) being lower than F crit (2.866266) leads to failing to reject the null hypothesis.

The p-Value and Statistical Significance: The p-value quantifies the probability that the observed data (or something more extreme) could occur under the assumption that the null hypothesis (no treatment effect) is true. In this case, a p-value of 0.56336 means that there is a 56.3% chance that the variations in pH are due to random fluctuations rather than an effect of the treatments. Since the p-value is well above the conventional threshold of 0.05, we fail to reject H_0 since we do not have sufficient evidence to conclude that any real differences in pH exist among the treatments and control. It's important to note that a non-significant result does not prove that there is no effect; rather, it suggests that under this current experimental conditions and sample size, any differences observed are likely attributable to chance, factors like factors like sample size and experimental power can be considered in order not to let a real effect go unnoticed.

Summary Statement: "The one-way ANOVA revealed no statistically significant differences in pH among the three treatment groups and the control, $F(3, 36) = 0.691269$, $p = 0.56336$, indicating that any observed differences in mean pH are likely due to random

variation rather than treatment effects." This suggests that under the conditions of our experiment, the treatments do not significantly alter the ph. The lack of differences may be due to the small range of pH (8,58-9,53) observed across groups or potential high within-group variability. Additionally, verifying that the underlying assumptions of the ANOVA were met reinforces the reliability of these results.

Future Directions: To further explore these findings, it would be beneficial to conduct additional experiments with a larger sample size or to assess if other environmental factors (like temperature) might interact with treatment effects. Computing and reporting effect sizes would also offer insights into the practical significance of the observed differences.

Table 4: Summary of descriptive statistics generated from EXCEL.

treatment 1		***treatment 2***		***treatment 3***		***control***	
Mean	9,27	Mean	9,177	Mean	9,184	Mean	9,276
Standard Error	0,038239	Standard Error	0,0499789	Standard Error	0,0801554	Standard Error	0,0787429
Median	9,23	Median	9,2	Median	9,26	Median	9,28
Mode	9,16	Mode	9,01	Mode	9,35	Mode	9,51
Standard Deviation	0,1209224	Standard Deviation	0,1580471	Standard Deviation	0,2534736	Standard Deviation	0,2490069
Sample Variance	0,0146222	Sample Variance	0,0249789	Sample Variance	0,0642489	Sample Variance	0,0620044
Kurtosis	-2,2895034	Kurtosis	-2,3564117	Kurtosis	6,1077117	Kurtosis	-2,5578616
Skewness	0,2120856	Skewness	-0,0578882	Skewness	-2,3821398	Skewness	-0,0006736
Range	0,26	Range	0,36	Range	0,83	Range	0,5
Minimum	9,15	Minimum	9	Minimum	8,52	Minimum	9,03
Maximum	9,41	Maximum	9,36	Maximum	9,35	Maximum	9,53
Sum	92,7	Sum	91,77	Sum	91,84	Sum	92,76
Count	10	Count	10	Count	10	Count	10
Confidence Level(95,0%)	0,0865027	Confidence Level (95,0%)	0,1130601	Confidence Level (95,0%)	0,1813241	Confidence Level (95,0%)	0,1781288

The average pH values are nearly identical, treatment 1 is 9.27, treatment 2 is 9.177, treatment 3 is 9.184, and the Control is 9.276. This indicates that, on average, the treatments did not shift the pH in any meaningful way compared to the control. The treatments appear to

maintain a similar pH level, which could suggest that the treatments may not influence pH or that the experimental conditions were too similar. The variances differ among groups: Treatment 1 (0.014622) shows lower variability, while Treatment 3 (0.064249) and the Control (0.062004) exhibit higher variability. Although the mean pH values are similar, the consistency of the measurements varies. Treatment 1, with its lower variance, indicates that all observations in that group are more tightly clustered around the mean. The higher variability in Treatment 3 and the Control suggests a slightly wider spread of pH values, which could be due to natural experimental fluctuations or the inherent variability of that specific treatment or sample.

As previously noted from ANOVA table showing that the Sum of Squares Between Groups (0.085988) is much smaller than the Sum of Squares Within Groups (1.49269). This reveals that most of the total variation in pH comes from differences among individual observations (within groups) rather than from the treatment effect. In practical terms, any variability observed is more likely attributable to natural random error or variation in the samples, rather than the different treatments applied. In summary, the descriptive statistics reveal that the treatments and the control all yield very similar average pH values with modest variability, and the ANOVA confirms that these differences are not statistically significant. This comprehensive analysis suggests that, under the current conditions, the treatments do not alter the pH in any meaningful or replicable way.

4.2.1.2 Temperature

Table 5: One-way ANOVA generated from excel

SUMMARY						
Groups	*Count*	*Sum*	*Average*	*Variance*		
treatment 1	13	313	24,076923	9,5769231		
treatment 2	13	312	24	3,1666667		
treatment 3	13	294	22,615385	3,5897436		
control	13	268	20,615385	3,4230769		
ANOVA						

Source of Variation	*SS*	*df*	*MS*	*F*	*P-value*	*F crit*
Between Groups	102,36538	3	34,121795	6,908501	0,0005845	2,7980606
Within Groups	237,07692	48	4,9391026			
Total	339,44231	51				

*treatment 1(1:3), treatment 2(1:4), treatment 3(1:5), control (1:0)

The treatments appear to elevate the measured value relative to the control. Treatments 1 and 2 have very similar mean values both about 24, whereas Treatment 3 is slightly lower (around 22.62). In contrast, the control group has a notably lower mean (20.62). This descending order suggests that the applied treatments have an effect on increasing the system's temperature.

Notably, Treatment 1 shows a much higher variance compared to the other groups. This indicates that while the average value in Treatment 1 is similar to Treatment 2, there is greater inconsistency or spread among its observations. Such discrepancies in variability could be due to heterogeneous responses within that treatment group, differences in how the treatment was applied, or measurement inconsistencies. In contrast, the other three groups exhibit tighter clustering around their means, which might reflect greater consistency in those conditions.

The significant F value (6.91) and a p-value much less than the common threshold of 0.05 (here, 0.000585) provide strong evidence to reject the null hypothesis. This means that there are statistically significant differences among the groups. In other words, at least one group's mean is different from the others, and the variation between the group means is not simply due to random chance. Given the descriptive statistics, it's clear that the control group differs from each treatment group, and even among treatments, differences are evident particularly given that Treatment 3 has a lower mean compared to Treatments 1 and 2.

The differences between the control and treatment groups are not only statistically significant but also practically noticeable. For instance, the control mean is roughly 20.62, whereas Treatments 1 and 2 exhibit means around 24. This difference of approximately 3.4 units may have meaningful implications depending on your field (e.g., a temperature rise of 3–4°C could significantly affect microbial activity or chemical kinetics). Since the overall ANOVA

shows significant differences, a post-hoc test (e.g., Tukey's HSD) would be useful to determine exactly which group differences are driving the significance.

Table 6: Summary of descriptive statistics

treatment 1		*treatment 2*		*treatment 3*		*control*	
Mean	24,0769231	Mean	24	Mean	22,6153846	Mean	20,6153846
Standard Error	0,85830437	Standard Error	0,49354812	Standard Error	0,52548465	Standard Error	0,51314093
Median	23	Median	24	Median	22	Median	21
Mode	23	Mode	24	Mode	23	Mode	21
Standard Deviation	3,09466041	Standard Deviation	1,77951304	Standard Deviation	1,89466187	Standard Deviation	1,85015592
Sample Variance	9,57692308	Sample Variance	3,16666667	Sample Variance	3,58974359	Sample Variance	3,42307692
Kurtosis	-0,2226738	Kurtosis	1,21510954	Kurtosis	1,13707792	Kurtosis	6,29011948
Skewness	0,89213718	Skewness	0,62917056	Skewness	1,00555927	Skewness	2,16363622
Range	10	Range	7	Range	7	Range	7
Minimum	20	Minimum	21	Minimum	20	Minimum	19
Maximum	30	Maximum	28	Maximum	27	Maximum	26
Sum	313	Sum	312	Sum	294	Sum	268
Count	13	Count	13	Count	13	Count	13
Confidence Level (95,0%)	1,87008457	Confidence Level (95,0%)	1,07534897	Confidence Level (95,0%)	1,14493271	Confidence Level (95,0%)	1,11803803

The treatment groups exhibit comparable and marginally elevated average values (about 24) in contrast to the control group (around 22.6). This difference in means is the main focus of a one-way ANOVA; if it is statistically significant, the researcher may draw the conclusion that the treatments raise the temperature in comparison to the control. Treatment 1 exhibits a significantly higher variation than Treatments 2, 3, and the control, which all exhibit comparable and very low variability. Greater variability within a single group may lessen the sensitivity of an ANOVA to identify differences between groups. Furthermore, Treatment 1's confidence interval (≈1.87) is larger, indicating greater uncertainty surrounding its mean estimate. The treatment groups have a little tail towards higher values, with significant positive skewness (≈0.63 to 1.01). With a higher skewness (≈2.16), the control group has a more noticeable right tail. The moderate positive kurtosis (≈1.14–1.22) of treatments 2 and 3 indicates distributions with somewhat heavier tails than a normal distribution. On the other hand, the control group is significantly more peaked and has

heavier tails due to its extremely high kurtosis value (≈6.29). ANOVA assumptions may be impacted by non-normality, particularly in the control group. The control's high skewness and kurtosis could be a sign of outliers or a non-symmetrical distribution that contradicts the ANOVA's presumptions.

According to the descriptive statistics, treatments are linked to a mean temperature that is greater than the control. However, while interpreting the ANOVA results, subtleties like the increased variability in Treatment 1 and the control group's non-normality (shown by high skewness and kurtosis) should be considered. These factors can change both the power and assumptions behind the test, and further research (through post hoc testing or data visualization) could further uncover how temperature is affected by these treatments.

Post-Hoc Comparisons with Bonferroni Correction (α = 0.008333)

Table 7: Post-Hoc Comparisons with Bonferroni Correction

POST-HOC TEST		
groups	*p-value(t-test)*	*significant?*
t1 vs t2	0,9387163	No
t3 vs t4	0,01186199	No
t1 vs t3	0,15937885	No
t1 vs t4	0,00202603	Yes
t2 vs t4	7,7648E-05	Yes
t2 vs t3	0,06673696	No

ALPHA	
Test	Alpha
ANOVA	0,05
Post-hoc test(Bonferroni corrected)	0,008333

*treatment 1(1:3), treatment 2(1:4), treatment 3(1:5), treatment 4 (1:0)

From the table there is a p-value of 0.00203 ($p < 0.008333$), there is a statistically significant difference in temperature between treatments t1 and t4. With a p-value of 7.76E-05 ($p < 0.008333$), this comparison is also statistically significant, indicating that treatment t4 differs significantly from t2. These comparisons did not yield p-values lower than the Bonferroni-corrected threshold, indicating no significant difference in temperature among these groups.

Blood meal from different sources (poultry, cattle, pig) can vary in nutrient content, particularly nitrogen availability, which directly influences microbial metabolism and, consequently, fermentation temperature. Field, (2009) emphasizes that optimal pH and nutrient conditions drive microbial enzymatic activity, thereby affecting the overall fermentation temperature. The significant differences observed (particularly between t4 and both t1 and t2) suggest that the treatment involving the t4 blood meal or blend influences the fermentation process in a way that either accelerates microbial activity or alters the metabolic pathways resulting in higher or lower temperatures. This aligns with Montgomery, (2013),

who reported that feedstock variations can lead to measurable differences in fermentation dynamics and temperature profiles.

The use of ANOVA followed by Bonferroni-corrected post-hoc tests is well established in process and environmental research (as outlined by Everitt and Skrondal, (2010) and Tabachnick and Fidell, (2013), ensuring that the likelihood of Type I errors is minimized while confirming that the observed differences are robust. The significant differences especially those between t4 and other treatments support the concept that the type of blood meal significantly influences fermentation temperature. This is consistent with previous research indicating that feedstock characteristics, such as nitrogen content and other nutrient profiles, are key determinants in bio-fermentation outcomes. Adjusting formulations based on such parameters can therefore be used as a strategy to optimize product quality in biofertilizer production.

4.2.1.3 Macro nutrients

It is anticipated that the microbial activity will not only decompose organic molecules but also improve the bioavailability of the nitrogen and phosphorus in the final product because the fermentation process changes and "stabilizes" the nutrients. Furthermore, the reliability of the fermentative process has been reinforced by the use of triplicates in each treatment, which has produced solid results with little variance (Sharma et al., 2017).

Nutrient Calculations for Each Treatment

Below is an integrated analysis of the three bio fertilizer treatments along with a control treatment that consists of 100% vermicompost. In this analysis, the vermicompost nutrient values (derived as the mean from the control treatment) are used to calculate weighted averages in the blends. The measured/assumed mean values are as follows:

1. Treatment T1 (1:3):

- **Nitrogen:**

Pig blood meal N: 1 kg × 10.06% = 100.6 g

Vermicompost N: 3 kg × 1.5% = 45 g

Total N: 100.6 + 45 = 145.6 g

Percentage N: (145.6 g / 4000 g) × 100 ≈ **3.64%**

- **Phosphorus:**

Pig blood meal P: 1 kg × 0.23% = 2.3 g

Vermicompost P: 3 kg × 0.2% = 6 g

Total P: 2.3 + 6 = 8.3 g

Weighted P (%): (8.3 g / 4000 g) × 100 ≈ **0.2075%**

2. Treatment T2 (1:4):

- **Nitrogen (N):**

Pig blood meal: 1 kg × 12.05% = **120.5 g**

Vermicompost: 4 kg × 1.50% = **60 g**

Total N: 120.5 g + 60 g = **180.5 g**

Weighted N (%): (180.5 g / 5000 g) × 100 ≈ **3.61%**

- **Phosphorus (P):**

Pig blood meal: 1 kg × 0.315% = **3.15 g**

Vermicompost: 4 kg × 0.20% = **8 g**

Total P: 3.15 g + 8 g = **11.15 g**

Weighted P (%): (11.15 g / 5000 g) × 100 ≈ **0.223%**

- **Potassium (K):**

Pig blood meal K: 1 kg × 0.1% = 1 g

Vermicompost K: 4 kg × 1.0% = 40 g

Total K: 1 + 40 = 41 g

Weighted K (%): (41 g / 5000 g) × 100 ≈ **0.82%**

3. Treatment T3 (1:5):

- **Nitrogen:**

Pig blood meal N: 1 kg × 11.20% = 112 g

Vermicompost N: 5 kg × 1.5% = 75 g

Total N: 112 + 75 = 187 g

Percentage N: (187 g / 6000 g) × 100 ≈ **3.12%**

- **Phosphorus:**

Pig blood meal P: 1 kg × 0.215% = 2.15 g

Vermicompost P: 5 kg × 0.2% = 10 g

Total P: 2.15 + 10 = 12.15 g

Percentage P: (12.15 g / 6000 g) × 100 ≈ **0.2025%**

- **Potassium:**

Pig blood meal K: 1 kg × 0.1% = 1 g

Vermicompost K: 5 kg × 1.0% = 50 g

Total K: 1 + 50 = 51 g

Percentage K: (51 g / 6000 g) × 100 ≈ **0.85%**

4. Control (100% Vermicompost)

Since the control is composed entirely of vermicompost, its nutrient percentages directly represent the measured mean values:

Nitrogen: 1.50%

Phosphorus: 0.20%

Potassium: 1.00%

Table 8: Summary Table of Weighted Nutrient Averages

Treatment	Blend composition	Total Weight(kg)	Nitrogen (%)	Phosphorus (%)	Potassium (%)
Control	**100% vermicompost**	**1(or standardized**	**1.5**	**0.2**	**1.00**
1	**1kg pig blood meal + 3kg vermicompost**	**4**	**3.64**	**0.2075**	**0.775**
2	**1kg pig blood meal + 4kg vermicompost**	**5**	**3.61**	**0.223**	**0.82**
3	**1kg pig blood meal + 5kg vermicompost**	**6**	**3.12**	**0.2025**	**0.85**

Analysis and Implications

Nitrogen (N): The inclusion of blood meal dramatically increases the available nitrogen in treatments T1, T2, and T3 relative to the control (1.50% N). T1 exhibits the highest nitrogen concentration (~3.64%), which can provide a robust vegetative boost for crops with high N demands. In contrast, the control's lower nitrogen level suggests that sole application of vermicompost might be best suited for systems where minimal N input is desired or where gradual nutrient release is preferred. (Brady & Weil, 2008; Fernandes, 2010)

Phosphorus (P): Although phosphorus contributions from blood meal and vermicompost are closer, T2 shows a modest edge (~0.223% P) compared with the control (0.20% P), potentially benefiting crops with intensive root development or high fruiting requirements. T1 and T3 remain very comparable to the control in phosphorus content, indicating that blood meal does not dramatically alter the P content, but slight adjustments may be needed for optimal P availability in specific soils. (FAO, n.d.; Rachid et al., 2015)

Potassium (K): The potassium content in all treatments is predominantly derived from the vermicompost. Although the blood meal contributes only minimally (0.10% by weight), the increased proportion of vermicompost in treatments T2 and T3 slightly boosts the K levels (0.82–0.85%) over T1 (0.775%) yet all remain lower than the control's 1.00% K. For crops where potassium is critical for water regulation and stress tolerance, additional K supplementation or adjustments may be warranted. (Brady & Weil, 2008)

4.2.2 Chemical composition of blood meal from different sources

*Table 9***:** *Mean values for the available macro nutrients.*

Blood Meal	Available-N (%)	Available_P (1st measure, %)	Available_P (2nd measure)
Cattle	12.04	0.31	0.00083
Chicken	10.05	0.23	0.51
Pig	11.20	0.21	0.41

*note that these values are averages of the triplicate samples

Blood meal is generally anticipated to have about 12-15% available nitrogen when applied as an organic fertilizer. This expectation is backed by agronomic research and accepted guidelines that highlight the importance of high nitrogen levels for enhancing plant growth and facilitating rapid nutrient cycling (Field, 2009; Montgomery, 2013). Cattle Blood Meal: 12.04% - This aligns with the lower end of the expected spectrum, indicating that cattle blood meal can serve as an effective nitrogen source. Chicken Blood Meal: 10.05% - This percentage is slightly beneath the typical range, suggesting that chicken blood meal may require mixing with other nitrogen-rich sources to achieve the necessary nutrient balance. Pig Blood Meal: 11.20% - This figure is also somewhat lower than the optimal range but still higher than that of chicken. It can help contribute to nitrogen levels when used in a mixture.

Phosphorus values in blood meal are usually lower than nitrogen. Industry sources generally recommend values around 0.3–0.5% for available phosphorus when measured by certain methods. Using the first measurement, cattle values (≈0.31%) are within the expected range, but chicken (≈0.23%) and pig (≈0.21%) are below. The second measurement shows a different pattern, highlighting the need for standardization. These numbers appear to be in different units or represent a different fraction (possibly water-soluble phosphorus). Here, the values for cattle, chicken and pig are 0.00083, 0.51 and 0.41 respectively. The discrepancy (especially for cattle) suggests that the measurement methodology or units differ. In practice, standardizing these units is essential for meaningful comparisons. Nonetheless, higher numerical values for chicken and pig blood meals on this second measure may indicate greater solubilized phosphorus availability, which is also beneficial for plant uptake.

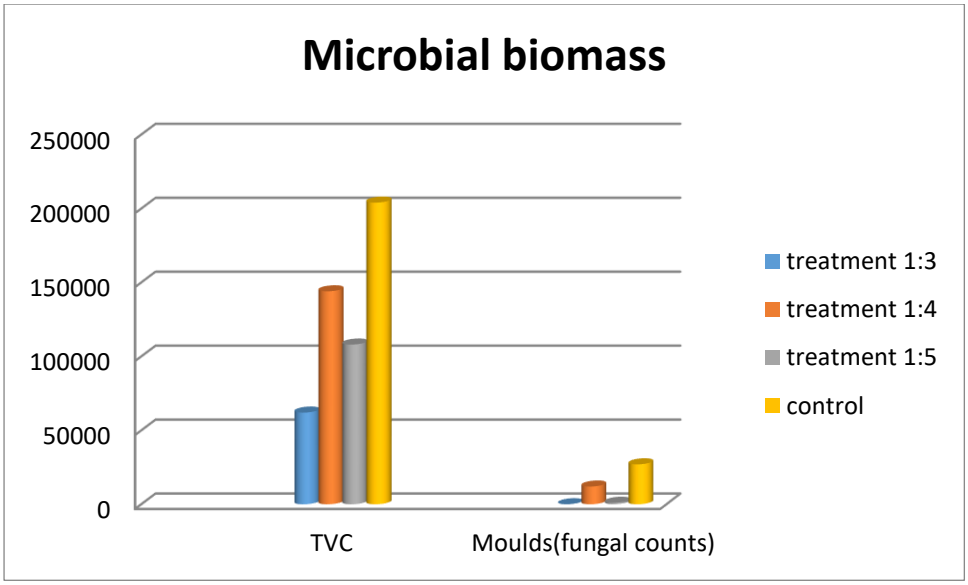
Since cattle blood meal meets the lower threshold of available-N while chicken and pig are somewhat lower, a blend can be formulated that capitalizes on the strength of cattle blood meal. For instance, incorporating a higher percentage of cattle blood meal relative to chicken might help achieve an overall blend with available-N in the optimal 12–15% range. While available P (first measure) for cattle is acceptable, the slightly lower values observed in chicken and pig suggests that, if phosphorus is a critical requirement, the blend's ratio should be adjusted based on consistent and standardized phosphorus measurements. If the second measure reflects water-soluble P, then chicken blood meal may contribute significantly to P availability, supporting plant growth.

4.2.3 Impact of blending on Microbial biomass

Table 10: Microbial biomass for the blends tested after fermentation

Sample ID	TVC (CFU/g)	Moulds (CFU/g)
Treatment 1:3	62,000	0
Treatment 1:4	144,000	12,000
Treatment 1:5	108,000	1,000
Control	204,000	27,000

Figure 2: Microbial biomass of treatments and control



Percentage Reduction in TVC Compared to Control

Treatment 1:3: $\frac{204{,}000-62{,}000}{204{,}000} \times 100 \approx 69.6\%$

Treatment 1:4: $\frac{204{,}000-144{,}000}{204{,}000} \times 100 \approx 29.4\%$

Treatment 1:5: $\frac{204{,}000-108{,}000}{204{,}000} \times 100 \approx 47.1\%$

Treatment 1:3 achieves the greatest reduction in TVC (approximately 70% less than the control), indicating that this treatment may be the most effective in suppressing overall microbial growth.

Percentage Reduction in Mould Counts

Treatment 1:3: Reduction is 100% (complete elimination of detectable molds).

Treatment 1:4: $\frac{27{,}000-12{,}000}{12{,}000} \times 100 \approx 55.6\%$

Treatment 1:5: $\frac{27{,}000-1{,}000}{12{,}000} \times 100 \approx 96.3\%$

Treatment 1:3 exhibited no detectable molds. Treatment 1:5 also achieved a significant reduction (~96%), while Treatment 1:4 have a moderate reduction (~56%). These differences suggest that some formulations inhibit mould growth more effectively than others.

The data indicate that Treatment 1:3 is most effective at reducing both TVC and mold counts relative to the control, while Treatment 1:5 also shows significant reduction especially in mold counts. Treatment 1:4 shows lesser reduction in TVC and molds compared to the other

treatments. Lower counts of total viable microbes and molds may indicate a more stable product with enhanced properties for soil health, which is a key outcome discussed in biofertilizer research (Tabachnick & Fidell, 2013).

Lower microbial biomass (especially potential pathogens such as molds) in Treatments 1:3 and 1:5 may imply a safer and potentially more stable bio fertilizer, assuming that a certain level of beneficial microbes is retained. As highlighted in the literature (Field, 2009; Montgomery, 2013), controlled fermentation conditions can significantly impact microbial dynamics. A treatment that consistently reduces unwanted microbial counts (like Treatment 1:3) aligns with findings that emphasize the importance of process optimization.

These differences might be due to the nature of the feedstock, the interactions between different microbial communities, or specific inhibitory compounds present in one formulation versus another. It was also noted that the worms were able to survive only in the control treatment and fairly in the treatment with highest vermicompost ratio. This may be attributed to higher nitrogen and ammonia content in the blood meals which may pose an environment which is not favorable for them (higher temperature, more carbon). Further research could correlate these findings with other quality parameters (like nutrient content or pH) to optimize the fermentation process.

4.4 CONCLUSION

The researcher has presented, examined, and discussed the research findings in this chapter. The study's findings and suggestions are presented in the upcoming chapter.

CHAPTER 5
SUMMARY, CONCLUSIONS AND RECOMMENDATIONS

5.1 INTRODUCTION

The researcher provides a summary of the study, conclusions, and implications for policy and practice, along with suggestions for additional research. The study's findings, as reported in chapter four, served as the foundation for the conclusions and suggestions.

5.2 SUMMARY

Below is a more detailed summary that captures the breadth and depth of the research

Background and Rationale

The research addresses the growing need for sustainable nutrient sources that can replace or complement synthetic fertilizers, which in Zimbabwe and similar agro-ecosystems have been shown to contribute to soil degradation, loss of soil biodiversity, and increased input costs for smallholder farmers. Previous studies (e.g., Barker and Pilbeam, 2015; Mäder et al., 2020) have documented the benefits of using organic amendments, such as blood meal and vermicompost, yet few have quantified their combined effects on nutrient dynamics and microbial biomass. This study builds on the existing literature by evaluating how blending these two materials can optimize nutrient availability, microbial stability, and, ultimately, soil health.

Research Objectives

1. **Chemical Characterization:** The study set out to precisely measure macronutrients nitrogen (N), phosphorus (P), and potassium (K) in various bio fertilizer blends derived from pig blood meal blended with vermicompost. It also compared nitrogen (N), phosphorus (P), and potassium (K) from chicken, cattle, and pig blood meal to determine which source provided the most favorable nutrient profile for rapid nutrient mineralization and sustained plant growth.
2. **Microbial Analysis:** In parallel, the work assessed how the blending ratios affected microbial biomass (total viable counts and mold counts). The aim was to understand how different formulations promote or suppress microbial communities, as maintaining a beneficial microbiome is critical for nutrient cycling.

3. **Optimization of Blend Ratios:** The overall goal was to identify the optimal ratio of blood meal to vermicompost that yields a product with high nutrient availability, stable fermentation parameters, and a balanced microbial community, while also minimizing potential risks such as ammonia volatilization.

Methodology and Data Precision

A Completely Randomized Design (CRD) was employed with four treatments:

- **Control (1:0):** 100% vermicompost.
- **Treatment 1 (1:3):** 25% blood meal blended with 75% vermicompost.
- **Treatment 2 (1:4):** 20% blood meal with 80% vermicompost.
- **Treatment 3 (1:5):** 16% blood meal with 84% vermicompost.

Each treatment was replicated three times to ensure statistical reliability. The study strictly adhered to standard protocols by calibrating instruments, following the Kjeldahl method for total nitrogen determination (with reported repeatability within ±0.05% N), and employing colorimetric techniques for phosphorus and potassium with similarly high precision. Microbial populations were measured using both the substrate-induced respiration (SIR) method and plate count techniques to capture both the dynamic activity and specific group counts.

Statistical Analysis

One-way ANOVA was used to test the null hypothesis of no differences among groups. For pH, the ANOVA yielded an F-statistic of 0.6913 (with degrees of freedom 3, 36) and a p-value of 0.5634, confirming that pH differences across treatments were statistically non-significant. In contrast, temperature data produced an F-value of 6.91 and a p-value of 0.000585, clearly indicating significant differences among groups. Precision was further ensured by reporting confidence intervals and standard deviations for nutrient measurements, with descriptive statistics showing, for example, mean nitrogen contents of 3.64% for the treatment 1 (1:3) versus 1.5% in the control.

Key Findings

Measured pH values ranged narrowly between 8.58–9.53, and the ANOVA confirmed no statistically significant differences ($p > 0.05$). This stability suggests that blending does not adversely affect the acidity or alkalinity of the bio fertilizer. The treatments significantly impacted fermentation temperature. For example, treatments with blood meal maintained mean temperatures around 24°C compared to approximately 20.62°C in the control. The significantly lower p-value (<0.001) for temperature differences indicates that the nitrogen in blood meal accelerates microbial metabolism.

The inclusion of blood meal increased the available nitrogen considerably. Treatment 1 (1:3) showed 3.64% N, aligning with literature that reports similar boosts when animal by-products are used (Barker and Pilbeam, 2015). Cattle blood meal demonstrated the highest nitrogen content (approximately 12.04%), while chicken blood meal was slightly lower. Treatment 2 (1:4) achieved a weighted average of ~0.223% P, a modest but noticeable enhancement compared to the control's 0.20%. Although largely contributed by vermicompost, treatment 3 (1:5) yielded slight increases in potassium (~0.85% vs. 1.00% in the control) that could benefit crops sensitive to potassium deficiencies.

Treatment 1:3 was particularly effective in reducing total viable microbial counts (approximately a 70% reduction compared to the control) and even eliminated detectable mold levels, suggesting a more stable and pathogen-free product. It was also noted that the worms introduced for fermentation only survived in the control treatment with no blood meal and in the treatment 1:5, they also lasted for a while. These findings echo similar trends reported by previous studies (e.g., Goswami et al., 2021) in which optimal nitrogen content correlated with enhanced microbial dynamics.

5.3 CONCLUSIONS

5.3.1. Chemical composition of bio fertilizers derived from blending blood meal and vermicompost.

A one-way ANOVA was conducted using EXCEL to analyze the means of pH, and it showed that there were no statistically significant differences in pH levels among the three treatment groups and the control, $F(3, 36) = 0.691269$, $p = 0.56336$. This indicates that any differences observed in mean pH are likely the result of random variation rather than the effects of the treatments. This implies that, within the framework of the experiment, the treatments do not

significantly change the pH. However, the absence of differences might be linked to the narrow range of pH values seen across the groups or possibly high variability within the groups. Furthermore, confirming that the basic assumptions of the ANOVA were satisfied bolsters the credibility of these findings.

In conclusion, the descriptive statistics indicated that both the treatments and the control produced very similar average pH values with slight variability, and the ANOVA supports that these differences lack statistical significance. The thorough analysis indicates that, under the prevailing conditions, the treatments do not meaningfully or consistently affect the pH.
The researcher did another one-way ANOVA to see if there are differences in average temperature across the different treatments and the control. The results were pretty clear, the F-value was 6.91, and the p-value was way below 0.05 (0.000585), which strongly suggests we can reject the null hypothesis. Basically, this suggests that there are important differences between at least some of the groups. In simple terms, one or more groups had different average temperatures, and these differences probably aren't just due to random variation. Looking at the descriptive stats, it's pretty obvious that the control group stands out from each treatment group, and even among the treatments, things are different especially since Treatment 3 had a lower average temperature compared to Treatments 1 and 2. To figure out exactly which treatments were making the difference, we did some post-hoc tests with Bonferroni correction. Those showed that the control group was quite different from Treatment 1 and Treatment 2. This suggests that the treatment involving the blood meal or blend in Treatment 4 really impacts the fermentation process maybe by speeding up microbial activity or changing metabolic pathways, which results in higher or lower temperatures.

By using the control treatment as the basis for vermicompost nutrient values, the weighted average nutrient compositions for treatments T1, T2, and T3 were calculated. The analysis shows that, **T1 (1:3)** offers the highest boost in nitrogen relative to the vermicompost control, which may favor crops requiring elevated nitrogen for rapid vegetative growth. **T2 (1:4)** provides a slightly higher phosphorus content compared to other blends, lending it to scenarios where enhanced root development is critical. **T3 (1:5)**, with a higher vermicompost proportion, results in marginally increased potassium, which can be beneficial for water regulation but features the lowest relative nitrogen. This weighted approach underscores how integrating blood meal with vermicompost offers the opportunity to tailor bio fertilizer

nutrient profiles to specific field or crop requirements, thus facilitating sustainable nutrient management practices that optimize yield while minimizing environmental impacts. (Brady & Weil, 2008; Fernandes, 2010; FAO, n.d.; Rachid et al., 2015).

The inclusion of blood meal significantly raised available nitrogen, particularly in T1, which can boost vegetative growth for crops with high nitrogen needs. This aligns with industry expectations (Brady & Weil, 2008; Fernandes, 2010). Improvements of phosphorus are modest, with T2 showing a slight edge that may benefit crops needing enhanced root development and fruiting (FAO, n.d.; Rachid et al., 2015). Since most potassium comes from vermicompost, the treatments display lower levels than the control, suggesting that additional K supplementation might be necessary for crops dependent on high potassium levels.

5.3.2. The impact of blending on microbial biomass and diversity within the bio fertilizer.

According to the data, Treatment 1:3 is the most successful at lowering both TVC and mold counts in comparison to the control, while Treatment 1:5 also demonstrates a notable decrease, particularly in mold counts. In comparison to the other treatments, Treatment 1:4 exhibits a lower reduction in TVC and molds. One important result covered in bio fertilizer research is a more stable product with improved qualities for soil health, which could be indicated by lower counts of total viable microbes and molds (Tabachnick & Fidell, 2013). Assuming a certain amount of beneficial microbes is maintained, lower microbial biomass (particularly potential pathogens like molds) in Treatments 1:3 and 1:5 may indicate a safer and possibly more stable bio fertilizer.

5.3.3. The chemical composition of blood meal from chicken, cattle and pigs.

The averages of nitrogen content from the three triplicates of blood meal samples indicated that cattle blood had the highest concentration which fell into the expected industrial range with pig and poultry in that order suggesting that cattle blood meal can serve as an effective nitrogen source. Pig had the highest potassium content whilst cattle also had the highest phosphorus content. Understanding and utilizing the distinct nutrient profiles of blood meal products can profoundly influence farming practices. By selecting the appropriate source whether high in nitrogen or offering a balanced nutrient spectrum farmer can achieve higher crop yields and more sustainable soil management. In addition, integrating these nutrient

profiles into broader soil fertility plans reinforces the principles of precision agriculture and environmental stewardship.

5.4 RECOMMENDATIONS

Refining experimental design: Since the treatments did not lead to significant shifts in pH, it might be worthwhile to verify if pH was the parameter most likely to be affected by the interventions or to consider whether additional factors (such as temperature, nutrient levels, or microbial activity) might provide more sensitive indicators of an effect. These results might suggest refining the experimental design (for example, increasing sample size, looking at different blend ratios, or measuring additional parameters) if the goal is to detect subtler changes in ph.

Nutrient Monitoring: Regular nutrient analyses should be performed to ensure that the final blended product meets established agronomic standards for both available-N and phosphorus. This practice, as highlighted by Field, (2009) and Montgomery, (2013) is key to maintaining consistent bio fertilizer quality.

Adapting to Crop Needs: The blend ratios can be changed based on the nutrient requirements of the target crop. For crops that need more nitrogen, it may be wise to increase the amount of cattle blood meal. For crops that are more susceptible to phosphorus deficiency, it would be advantageous to ensure a higher contribution from the blood meal with better water-soluble phosphorus (as indicated in the second measure).

Standardization: Because the phosphorus measurements differ between the two methods, it is important to standardize the analysis technique to make reliable comparisons and recommendations. Using a single, consistent method that is in line with industry standards will help to increase the formulation strategy's dependability (Tabachnick & Fidell, 2013; Everitt & Skrondal, 2010).

5.5 IMPLICATIONS FOR PRACTICE AND POLICY

5.5.1 Implications for practice

For Farmers

The blended bio fertilizer can serve as an eco-friendly alternative to synthetic fertilizers, improving soil fertility and crop yields over time. Farmers should consider site-specific

factors such as soil type, crop nutrient requirements, and environmental conditions when applying the bio fertilizer. Furthermore, proper storage (e.g., shaded, ventilated containers) is essential to maintain nutrient quality prevail.

For Agri-Businesses

Commercial production of blood meal-vermicompost blends can be optimized based on ideal mixing ratios and storage protocols. Value-added bio fertilizer products could be marketed for organic farming systems.

Storage Recommendations

Proper storage techniques, such as maintaining optimal temperature and moisture levels, are essential to preserve the bio fertilizer's nutrient content and microbial activity. Developing packaging solutions or stabilizing additives could help extend shelf life and reduce nutrient losses.

Integration with Sustainable Practices

The bio fertilizer can complement other organic inputs and soil amendments, contributing to regenerative agriculture and improved soil carbon sequestration. Its application aligns with sustainable development goals by reducing dependence on synthetic fertilizers and minimizing environmental pollution.

5.5.2 Implications for policy

Promotion of Organic Alternatives

Policymakers can encourage the adoption of bio fertilizers like blood meal and vermicompost blends by offering subsidies, incentives, or educational programs for farmers. Support for research and development in bio fertilizer production can improve their accessibility and affordability hence improving the adoption rate by smallholder farmers.

Standards and Regulations

Establishing quality standards for bio fertilizers, including guidelines on blending ratios, nutrient content, microbial safety guidelines and storage conditions, can ensure consistent performance and safety. Certification programs can help build trust among farmers and consumers regarding the efficacy and sustainability of bio fertilizer products.

Environmental Policies

Policies promoting organic fertilizers can contribute to reducing agricultural emissions, nutrient runoff, and soil degradation, aligning with efforts to combat climate change. Supporting large-scale adoption of bio fertilizers can foster sustainable agricultural practices and enhance food security.

5.6 AREAS FOR FURTHER RESEARCH

Nutrient Availability and Release Patterns - Analyzing how environmental factors (temperature, moisture, soil type) influence nutrient release (N, P, K).

Microbial Diversity and Functionality - Using advanced techniques (e.g., DNA sequencing) to study microbial populations in the bio fertilizer.

Storage Methods and Longevity - Testing how different storage conditions (temperature, humidity, aeration) affect nutrient preservation and evaluating packaging solutions to extend shelf life.

Real-World Agricultural Performance - Conducting field experiments to measure effects on crop productivity and soil quality such comparing effectiveness across a variety of crops (grains, vegetables and pulses).

Comparative Analysis - Compare the performance of this bio fertilizer with other organic fertilizers such as bio char, compost and rock phosphates well as synthetic fertilizers in terms of nutrient release, microbial activity, and crop growth.

Economic Feasibility -Conduct cost-benefit analyses to determine the economic viability of producing and applying the bio fertilizer on a larger scale and explore the adoption rates and challenges faced by farmers in using the product.

Integration with Sustainable Practices - Investigate how this bio fertilizer can be incorporated into sustainable farming practices like intercropping, conservation tillage, or precision agriculture and study its potential applications in organic and regenerative farming systems.

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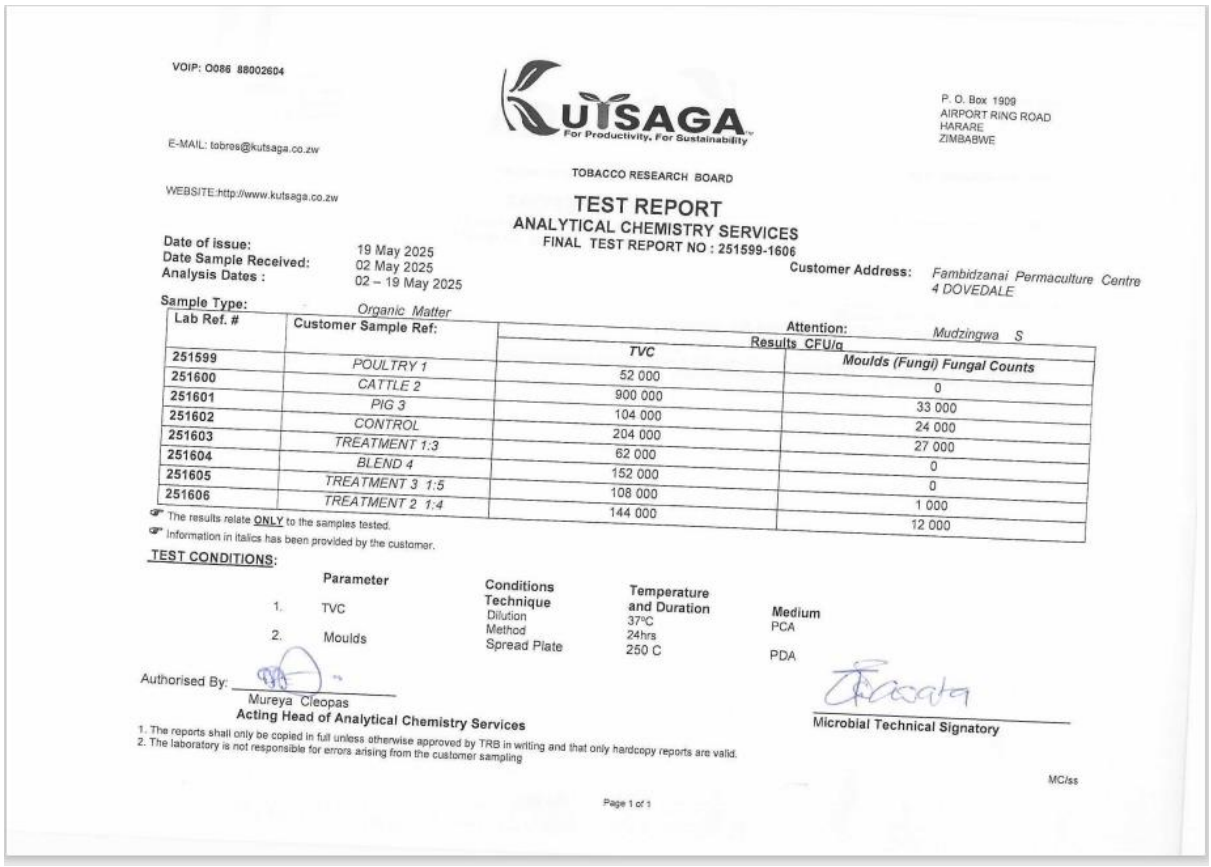
APPENDICES

APPENDIX 1

Map of Fambidzanai Permaculture centre

APPENDIX 2

Original microbial laboratory results



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TEST REPORT
ANALYTICAL CHEMISTRY SERVICES
FINAL TEST REPORT NO : 251599-1606

Date of issue: 19 May 2025
Date Sample Received: 02 May 2025
Analysis Dates : 02 – 19 May 2025

Customer Address: *Fambidzanai Permaculture Centre 4 DOVEDALE*

Sample Type: *Organic Matter*

Attention: *Mudzingwa S*

Lab Ref. #	Customer Sample Ref:	Results CFU/g	
		TVC	Moulds (Fungi) Fungal Counts
251599	*POULTRY 1*	52 000	0
251600	*CATTLE 2*	900 000	33 000
251601	*PIG 3*	104 000	24 000
251602	*CONTROL*	204 000	27 000
251603	*TREATMENT 1:3*	62 000	0
251604	*BLEND 4*	152 000	0
251605	*TREATMENT 3 1:5*	108 000	1 000
251606	*TREATMENT 2 1:4*	144 000	12 000

☞ The results relate ONLY to the samples tested.
☞ Information in italics has been provided by the customer.

TEST CONDITIONS:

	Parameter	Conditions Technique	Temperature and Duration	Medium
1.	TVC	Dilution Method Spread Plate	37°C 24hrs 250 C	PCA
2.	Moulds			PDA

Authorised By: _____
Mureya Cleopas
Acting Head of Analytical Chemistry Services

Microbial Technical Signatory

1. The reports shall only be copied in full unless otherwise approved by TRB in writing and that only hardcopy reports are valid.
2. The laboratory is not responsible for errors arising from the customer sampling

MC/ss

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APPENDIX 3

Original laboratory results of macro-nutrients between raw blood meal samples and final fermented treatments (EXCEL display)

Date of analysis	Source	Type	Sample ID	Ward	pH	Conductivity	Texture	Nitrate-N	Ammonia-N	available-N	Available_P	Available_P
28.04.2025			T1R1		9,41	6,528				3,2	0,061	40.18ppm
28.04.2025			T2R1		9,01	5,126				2,35	0,073	38.16ppm
28.04.2025			T3R1		9,35	6,821				2,09	0,068	41.96ppm
28.04.2025			Control		9,51	12,54				1,5	0,9	39.2ppm
28.04.2025			T1R2		9,39	6,526				3,2	0,06	40.17ppm
28.04.2025			T2R2		9	5,124				2,36	0,071	38.2ppm
28.04.2025			T3R2		9,34	6,819				2,085	0,066	41.99ppm
28.04.2025			Control		9,5	12,55				1,55	0,89	39.22ppm
28.04.2025			T1R3		9,4	6,527				3,3	0,063	40.2ppm
28.04.2025			T2R3		9,03	5,122				2,33	0,074	38.23ppm
28.04.2025			T3R3		9,32	6,822				2,087	0,069	41.97ppm
28.04.2025			Control		9,53	12,52				1,53	0,91	39.3ppm

			BLOOD MEAL SAMPLES							available-N	Available_P	Available_P
28.04.2025			cattle1							12,00%	0,30%	0,00083
28.04.2025			cattle 2							12,10%	0,31%	0,00081
28.04.2025			cattle 3							12,02%	0,33%	0,00084
28.04.2025			chicken 1							10,05%	0,22%	0,5
28.04.2025			chicken 2							10,07%	0,24%	0,52
28.04.2025			chicken 3							10,04%	0,23%	0,51
28.04.2025			pig 1							11,00%	0,20%	0,4
28.04.2025			pig 2							11,40%	0,21%	0,41
28.04.2025			pig 3							11,20%	0,23%	0,42

APPENDIX 4

Experimental setups