Assessing the Nutritive Value, Safety and Consumers' Perceptions of Co-compost made from Dewatered Sewage Sludge and Organic Green Waste

By

SINIKIWE PRECIOUS KHUMALO

Submitted in the partial fulfilment of the academic requirement of,

Master of Science in Agriculture

Discipline of Crop Science

School of Agriculture, Earth, and Environmental Sciences

College of Agriculture, Engineering, and Science

University of KwaZulu-Natal

Pietermaritzburg

South Africa

December 2024

PREFACE

The research in this thesis was carried out and completed in the Discipline of Crop Science, School of Agriculture, Earth, and Environmental Sciences, in the College of Agriculture, Engineering and Science, at the University of KwaZulu-Natal, Pietermaritzburg, South Africa. The content of this work has not been submitted to another university and, except where the work of others is acknowledged in the text and the results reported are due to an investigation by the candidate.

[redacted]

Signed: Prof. Alfred Oduor Odindo (supervisor)

Date: 15 December 2024

DECLARATION 1: PLAGIARISM

I, **Sinikiwe Precious Khumalo**, declare that:

(i) the research reported in this thesis, except where otherwise indicated or acknowledged, is my original work;

(ii) this thesis has not been submitted in full or in part for any degree or examination to any other university;

(iii) this thesis does not contain other persons' data, pictures, graphs, or other information unless specifically acknowledged as being sourced from other persons;

(iv) this thesis does not contain other persons' writing unless specifically acknowledged as being sourced from other researchers. Where other written sources have been quoted, then:

a) their words have been re-written, but the general information attributed to them has been referenced;

b) where their exact words have been used, their writing has been placed inside quotation marks, and referenced;

(v) where I have used material for which publications followed, I have indicated in detail my role in the work;

(vi) this thesis is primarily a collection of material prepared by myself, published as journal articles, or presented as a poster and oral presentations at conferences. In some cases, additional material has been included;

(vii) this thesis does not contain text, graphics, or tables copied and pasted from the Internet unless specifically acknowledged, and the source is detailed in the dissertation

and in the references sections.

[redacted]

Signed: Miss Sinikiwe Precious Khumalo

Date: 15 December 2024

DECLARATION 2: PRESENTATIONS AND PUBLICATIONS

Khumalo S.P*., and Odindo A.O (2023). Parameters for quality co-compost production made from dewatered sewage sludge and organic green waste, 15-18 October 2023, *The 1st International Water Association - Non-Sewered Sanitation (IWA-NSS),* Conference, Emperors Place, Johannesburg, Gauteng, South Africa.

Khumalo S.P*., and Odindo A.O (2023). Effect of feedstock mixing ratios and temperature on quality of co-compost made from human waste and organic green waste, 08-10 November 2023, *The 7th Water Institute for Southern Africa, Young Water Professionals (7th WISA-YWP),* Conference, Protea Hotel, Stellenbosch, Cape Town, Western Cape, South Africa.

Khumalo S.P*., and Odindo A.O (2023). Optimizing mixing ratios using green waste and sewage sludge as feedstocks for co-compost production. 02-03 November 2023, College of Agriculture, Engineering and Science, *Postgraduate Research and Innovation Symposium (PRIS),* Conference, Musgrave Hotel, Durban, KwaZulu-Natal, South Africa.

Khumalo S.P*., and Odindo AO (2024). Assessing fertiliser value of co-compost made from dewatered sewage sludge and organic green waste on maize (*Zea mays*) production. 22-23 May 2024, *The 6th Ukulinga Howard Davis Symposium*, Ukulinga Research Farm, University of KwaZulu-Natal, Pietermaritzburg, KwaZulu-Natal, South Africa.

Khumalo S.P*., Gwara S., Mandizvo T., Rupande L., Sanusi I., Migeri S., Six J., Odindo A.O., (2024), Evaluating Agronomic Value and Consumers' Acceptance of Co-compost made from Human Excreta for Sustainable Agriculture, 29-30 October 2024, College of Agriculture, Engineering and Science, *Postgraduate Research and Innovation Symposium (PRIS)*, Conference, Musgrave Hotel, Durban, KwaZulu-Natal, South Africa.

Khumalo S.P*, **Nkomo N**, **Wilde B.C**, **Mandizvo T**, **Gwara S**, **Migeri S**, **Six J**, **Odindo A., (2024)**. Circular Municipal Waste Management: Assessing Pathogen Deactivation, Nutrient Recovery, and Quality Assurance of Co-Compost Produced from Dewatered Sewage Sludge and Organic Green Waste. (Under review) - **Preliminary study**.

Khumalo S.P*., Mandizvo T., Odindo A.O., (2024), Evaluating the impact of human excreta-based co-compost on soil health and plant growth: assessing nutrient recycling and phytotoxicity on cucumber (*Cucumis sativus*) root morphology. (Under review) – **Chapter 4**

ABSTRACT

The increasing global population which is estimated to reach 9.6 billion in 2050 implies the need to increase food, feed, and fuel production on the limited agricultural lands while carefully managing waste. Chemical fertilisers have primarily been used to enhance plant productivity because they have a high nutrient composition, are water-soluble, readily available to plants, easy to apply, and transport. Although these chemical fertilisers have these benefits, they often leach into the water surface contaminating underground water bodies and causing eutrophication in water courses. Moreover, the Harber-Bosch process which produces these chemical fertilisers is energy-intensive and contributes to greenhouse gas emissions. Furthermore, small-scale farmers in Sub-Saharan Africa often apply less than 10 kg/ha of nitrogen. On the other hand, commercial farmers in developed countries apply excessive amounts of chemical fertilisers, which subsequently degrade the soil and lead to organic matter loss. The nutrients are transported from farms to urban settlements in the form of food and feed leaving the soil nutrient-depleted.

In cities, food, market, and human waste are often landfilled in hazardous landfills; however, they pose health risks and environmental pollution if improperly managed. Co-composting human excreta including dewatered sewage sludge (DSS), and organic green waste (OGW) can prevent the stockpiling of sludge in the wastewater treatment plants (WWTP) and divert OGW from landfills. Moreover, co-composting can recover essential nutrients from residual plant materials and human excreta while subsequently reducing improper waste disposal and management.

The first objective of the study investigated the effect of mixing ratio and turning frequency on the production of high nutritive value and pathogen-free co-compost when using dewatered sewage sludge and organic green waste as feedstocks. The study employed different turning frequencies and mixing ratios to determine the optimum co-composting conditions. The study found that there were significant differences between treatments in pH, electrical conductivity, organic matter, and organic carbon. The highest turning frequency (7 days turning interval) resulted in low pathogen load contamination in two different mixing ratios however it was characterized by low organic matter compared to less frequently turned (28 days turning interval) co-compost piles. The study found all treatments except high turning frequency and low sludge proportion (1DSS:3OGW_28TF) complied with the World Health Organization and some treatments complied with the Fertiliser Act standards of viable helminth eggs in the matured co-compost.

The second objective of the study established a novel, cost-effective method for assessing the phytotoxicity of co-compost through a rhizotron experiment. In context, co-compost treatments outlined in Chapter 3 were compared to sandy loam soil and Gromor commercial compost. Cucumber (*Cucumis sativus*) was used as a test crop because of its sensitivity to phytochemicals that inhibit growth and development of root traits and shoot. The root system architecture measured were convex hull area (CHA), root branch count (RBC), total root length (TRL), root system depth (RSD), root system width (RSW), root mass ratio (RMR), shoot mass ratio (SMR), root-shoot mass ratio (RSMR), shoot-root mass ratio (SRMR), leaf area index (LAI), and leaf number (LN). The study found that co-compost treatments with a higher proportion of sludge resulted into higher above ground biomass and it was positively associated with availability of nutrients in the growing media. The convex hull area was high in the sandy loam soil and roots length was high in the commercial Gromor compost.

The third objective of this study investigated the effect of the combined application of co-compost and synthetic fertiliser on the growth, development, and nutritional composition of maize (*Zea mays*). The maize was grown on two contrasting soils but both acidic with different physical properties a sandy loam and a clay-loam soil. The sandy loam soil was collected from KwaDinabakubo (KD) in Durban while the clay loam soil was collected in Bishopstowe (BS) Pietermaritzburg. The study found that combining co-compost with chemical fertilisers at half the recommended rate improved visual growth parameters of maize including plant height, chlorophyll content, maize cobs, and harvest index. Moreover, the application of co-compost in the sandy loam soil improved organic carbon and organic matter and led to harvestable grain in the poor soil. Essential minerals including nitrogen (N), calcium (Ca), and zinc (Zn) were high in maize grains with the treatment combination of co-compost and synthetic fertiliser in both sandy loam and clay loam soil. These results have provided information and solutions to farmers faced with low productivity because of poor soil organic matter and organic carbon.

The fourth objective of this study assessed the attitudes, perceptions, and willingness of university students to purchase and consume food produced using co-compost. This study aimed at validating assumptions and negative stigmas associated with acceptance of food produced using human excreta-derived fertilisers. The study found that willingness to purchase and consume food produced using co-compost was mainly influenced by students' awareness of the use of human excreta for agricultural purposes. Moreover, the study found that there were differences across gender and male students were more willing to purchase and consume food produced using co-compost compared to female students. The study further unveiled that willingness to consume food produced using co-compost is affected by the type of food or crops grown using this type of soil amendment. In context, students/consumers were less willing to consume food produced using co-compost if it was eaten raw and grew below ground. Interestingly, students were more willing to eat food produced using co-compost of it grows above ground, if it was eaten cooked and if it undergoes processing. These results will assist farmers and policymakers in carefully choosing the types of crops that can be grown or produced using co-compost.

In context, the study revealed that through co-composting challenges of waste management in landfills and wastewater treatment plants can be addressed with an added benefit of ensuing circular nutrient economy. The study further developed a novel approach to determine and assess phytotoxic compost in the matured co-compost using a rhizotron as it shows the growth of both roots and shoots. This approach is useful in identifying phytochemical compounds in the co-compost that can inhibit the growth of crops. The study found that sandy loam and clay loam soil can be improved by integrating synthetic fertilisers and co-compost. The study further recommends simulating awareness campaigns and developing dissemination strategies to enhance public understanding of fertilizers derived from human excreta.

ACKNOWLEDGMENTS

I would like to thank **God** first for the immense love he showed me from the beginning of my career until now. May his name be glorified in the world and the heavens, for he never let me down, and he was right next to me every step of the way.

I thank my supervisor **Professor Alfred Oduor Odindo** for his guidance, support, and ability to see potential in me. He guided me throughout the whole Master's journey and allowed me to develop my ideas and express my research capabilities. He has been the best supervisor ever.

I would like to extend my gratitude to the **Rural-Urban Nexus (RUNRES)** for funding my studies. I also thank the UKZN Postgraduate Scholarship, ML Kisch Scholarship and Postgraduate Research and Innovation Symposium (UKZN) for acknowledging my academic capabilities.

Special thanks to **Dr Takudzwa Mandizvo** and **Dr Simon Gwara** for their support throughout my academic journey. I also thank all of Prof Odindo's postgraduates' students who contributed academically to my work.

I thank **Dr Isaac Sanusi** and **Dr Lorika Beukes** from the microbiology department, **Mr Nhlanhla Magagula** from Soil Science, and **Mr Thokozani Nkosi** from the Crop Science department. I also extend my gratitude to **Mr Mthuthuzeli Tshazi** and **Mr Armstrong** for their assistance and support in the field.

I would like to extend my warmest gratitude to my spiritual parents/leaders, **Mr. J.J. Mtshali** and **Mrs. K. Mtshali**, for teaching me the truth about God's way, for being humble, and for being honest friends whenever I needed them. Thank you so much.

To my biological mother **Mrs T.M Khumalo**, thank you so much for raising me, whenever I saw your eyes full of tears because of the hardship we encountered, I told myself "One day I will be educated and that would be turned to tears of joy".

Lastly, I thank sisters **Nokwanda Mbedu** and **Nobuhle Khumalo**. My brothers **Malibongwe Khumalo**, **Thabiso Khumalo**, **Thabiso Dlamini**, and **Mxolisi Khumalo** for their love and support throughout this journey.

Very warm gratitude to my brother and my business partner **Mr Mbongiseni Khumalo**, "we shall make the Bright Farm successful".

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

ABBREVIATIONS

ABR	Anaerobic baffled reactor
AMBIC	Ammonium bicarbonate
BS	Bishopstowe
C/N	Carbon-to-nitrogen
CAPI	Computer-assisted personal interviewing
CEF	Controlled environment facility
CFU/g	Colony forming unit per gram
CH_4	Methane
CHA	convex hull area
Co	Co-compost
CO_2	Carbon dioxide
DEWATS	Decentralized wastewater treatment system
DSS	Dewatered sewage sludge
EC	Electrical conductivity
F	Chemical fertiliser
FAO	Food and agriculture organization
GHG	Greenhouse gas emissions
H_2O_2	Hydrogen peroxide
H_2S_4	Tetrasulfide
HFCW	Horizontal flow constructed wetlands
K	Potassium
K_2O	Potash
KD	KwaDinabakubo
Kg/ha	Kilograms per hectare
LaDePa	Latrine dehydration and pasteurization
LAI	leaf area index
LAI	Leaf area index
LAN	Lime ammonium nitrate
LN	leaf number
$MgCl_2$	Magnesium chloride
MgO	Magnesium oxide

$MgSO_4$	Magnesium sulfate
ML/day	Megalitre per day
MR	Mixing ratios
mS/cm	Milliesemiens per centimetre
MSW	Municipal solid waste
N	Nitrogen
N_2O	Nitrous oxide
NUC	Nutrified urine concentrate
O3	Ozone
OC	Organic carbon
OGW	Organic green waste
OM	Organic matter
P	Phosphorus
P_2O_5	Phosphorus pentoxide
PCA	principal component analysis
PGFs	Planted gravel filters
PMB	Pietermaritzburg
PSS	Particle System Separation
RBC	Root branch count
RMR	Root mass ratio
RSD	Root system depth
RSMR	Root-shoot mass ratio
RSW	Root system width
SMR	Shoot mass ratio
SRMR	Shoot-root mass ratio
SSP	Single superphosphate
t/ha	Tons per hectare
TF	Turning frequency
TRL	Total root length
UKZN	University of KwaZulu-Natal
UNICEF	United Nations International Children's Emergency Fund
VFCW	Vertical flow constructed wetlands
VIP	Ventilated improved pit

WAE	Week after emergence
WHO	World health organisation
WWTP	Wastewater treatment plants

Chapter 1: Introduction

1.1 Background

Mineral elements such as nitrogen (N), phosphorus (P), potassium (K) and other essential micronutrients contained in the soil are essential for plant growth and crop performance (Yahaya et al., 2023). Current nutrient imbalances exist because of nutrients being transported in food from farms to urban settlements where they are disposed of together with food, market and human waste. This leads to soil fertility being depleted without proper replenishment or return of these nutrients back into the soil. Moreover, the application of chemical fertilizers is often very low less than ten kilograms of nitrogen per hectare (<10kg N/ha) in small-scale farmers, especially in Sub-Saharan Africa (Mwangi, 1996). Most smallholder farmers cannot afford the expensive prices of chemical fertilizers and lack an understanding of fertilizer application in the soil (Gwara et al., 2023).

Moreover, the Haber-Bosch process which converts hydrogen and nitrogen to ammonia produces intensive gasses that contribute to greenhouse gas emissions (GHG) which subsequently lead to climate change. Through the application of these chemical fertilizers on the soil they often leach into the soil surfaces because of low organic matter and poor soil structure thus leading to loss of yield over time, especially among smallholder farmers. For instance, small-scale farmers' yield of maize (*Zea mays*) is less or equal to 2 tons per hectare (ha) compared to 10 tons/ha of maize produced by commercial farmers in developed countries (Mtyobile, 2021).

In conjunction with nutrient depletion, global society faces significant challenges in waste disposal and management in both urban and rural settlements. For instance, research indicates that 66.7% of people without access to proper sanitation live in informal settlements and rural areas, and the vast majority (50%) are from Sub-Saharan Africa (WHO/UNICEF, 2021). Moreover, in South Africa, although 84.1% of the population has access to improved sanitation, about 48% is not connected to water-borne sewerage (STATS_SA, 2022). This proportion of the population uses septic tanks, and pit latrines (ventilated and non-ventilated). In addition to this, there was no plan of emptying these toilets, and therefore municipalities are experiencing serious backlogs with the emptying and disposal of human excreta from these pit latrines.

The problem of disposal of human excreta is a universal challenge globally. The effluent or sewage sludge produced from the wastewater treatment plants (WWTP) is released back to nature (oceans, rivers, and lakes) in large quantities of dilute waste causing water quality issues (Mihelcic et al., 2011; Zhang et al., 2017). Dewatered sewage sludge or biosolids which are the byproduct of sewage sludge treatment are dried on drying beds or landfilled because it is a waste product that needs to be discarded frequently to keep the treatment plant operating optimally.

Thus, there is a need to come up with safe, hygienic, and sustainable ways of addressing the issue of human excreta disposal. According to Rockefeller (1998) the use of human excreta as fertilizer and soil amendments has been used or recorded a long time ago going back to the beginning of agriculture in China and South America. Interestingly, research has revealed that indeed human waste contains essential nutrients including N, P and K. The total nitrogen content of 68.23 g/kg (grams per kilogram) in faeces was reported by (Bai and Wang, 2010), and total Kjeldah nitrogen of 3.35±0.29% was reported by (Manga et al., 2022) in dewatered faecal sludge.

Human faeces in the form of dewatered sewage sludge and faecal sludge can be co-composted to make soil amendments and biofertilizers. Co-composting refers to the simultaneous composting of different types of organic wastes, which can add value or improve compost quality (Zhang and Sun, 2014). Co-composting raw or dewatered faecal sludge alone is impossible because it has a low carbon-to-nitrogen ratio (C/N) and high moisture content (Manga et al., 2021). Therefore organic materials including sawdust (Banegas et al., 2007; Manga et al., 2023), municipal organic waste (Ofei-Quartey et al., 2023; Zhang et al., 2018), coffee husks (Manga et al., 2021), wood chips (Zhao et al., 2015), landscaping waste (Dzulkurnain et al., 2017), agro-industrial waste (Gutiérrez et al., 2017), and wetland plant material (Kwarciak-Kozłowska, 2019) have been reported as alternative feedstocks to improve the C/N ratio when co-composting human excreta. Co-compost can be used as soil amendments to improve organic carbon and organic matter in the soil. Moreover, it has the added benefit of reducing improper deposition of human waste and diverting organic green waste from being landfilled to recycling residual nutrients.

1.2 Problem statement

The disposal management of organic waste including human excreta, food or market waste and garden residues is a major challenge faced by many municipalities globally (de Zeeuw, and Drechsel 2015). These organic wastes contain essential mineral elements (crop plant nutrients) necessary for improved and sustainable crop production. However, these nutrients are mined and disposed of as waste in the form of human excreta and/or food waste contributing to major nutrient imbalances in the soils in farms and environmental pollution in cities. There is a need to correct the nutrient imbalances by recovering the nutrients from human waste and reuse in agriculture. The aerobic process of co-composting by mixing organic green waste with sewage sludge, can provide alternative and sustainable ways of managing waste while at the same time addressing the challenges of soil fertility depletion. Co-compost production requires a carbon-to-nitrogen ratio (C/N) of at least 20-30 (Daiz et al., 2012). The amount of carbon and nitrogen is influenced by the type of organic waste/feedstock and may vary. There is inadequate information on mixing ratios to achieve the correct C/N ratio for good quality co-compost production. The mixing of sewage with organic waste may create poor conditions for aerobic bacteria involved in the biodegradation of complex organic molecules. Research is needed to determine optimal turning frequencies and their effectiveness in eliminating pathogens and improving the quality of the final product. Co-compost often has very low N, P and K levels (1-4%) and cannot meet crop nutrient requirements for optimum growth. Research is needed to assess the effectiveness of using co-compost in combination with chemical fertilizers on agronomic efficiency, and the concentration of beneficial micronutrients taken up by the crops. Many reports in the literature report on above-ground crop responses and yield. There is little information in the literature on the effect of co-compost use on root growth and morphology. It is also important to get an understanding of perceptions that could be associated with the consumption and purchase of food produced using human excreta-derived co-compost.

1.3 Justification

Co-composting dewatered sewage sludge (DSS) with organic green waste (OGW) offers a sustainable solution to the environmental challenges associated with improper DSS disposal, including eutrophication and the proliferation of disease vectors. By integrating aerobic co-composting techniques, this study aims to enhance nutrient recovery (N, P, and K), inactivate pathogens, and produce environmentally safe, phytotoxicity-free compost. The findings will contribute to sustainable waste management practices and help mitigate greenhouse gas emissions from landfill disposal of organic waste.

The resulting waste-based fertilizer (co-compost) can improve soil organic carbon, organic matter content, and water-holding capacity while reducing the negative impacts of improper waste disposal, thereby supporting a circular nutrient economy. Additionally, when combined with chemical fertilizers, co-compost can enhance crop agronomic efficiency by replenishing soil organic matter and carbon.

1.4 Aim

This study aimed to identify optimal processes for producing high-quality co-compost from dewatered sewage sludge and organic green waste under controlled conditions.

1.4.1 Objectives

The specific objectives of the study were to:

i) to investigate the effect of mixing ratios, turning frequency and time on pathogen deactivation, heavy metal contamination, and production of high nutritive co-compost made from dewatered sewage sludge (DSS) and organic green waste (OGW).

ii) to evaluate the root architecture and the phytotoxic effects of co-compost derived from dewatered sewage sludge and organic green waste on cucumber (*Cucumis sativus*).

iii) to determine the effect of combining co-compost with synthetic fertilizers on the agronomic efficiency, and nutritive value of maize (*Zea mays*)

iv) to assess the attitudes, perceptions, and willingness of university students to purchase and/or consumption of food produced using co-compost made from dewatered sewage sludge and organic green waste

1.5 Thesis structure

The thesis consists of 7 chapters:

Chapter 1

This chapter gives a general introduction to the thesis, problem statement, justification, aim and specific objectives of this study.

Chapter 2

This chapter reviews innovative technologies to recover nutrients from organic waste and re-use in plant production. The chapter concludes by providing the most affordable technology used to recover nutrients from waste with a focus on co-compost. In addition, the review identifies key findings, research gaps, and recommendations for future research.

Chapter 3

This chapter was a tunnel experiment that investigated different DSS and OGW mixing ratios and optimized turning frequency to produce a safe and hygienic co-compost. This chapter looks at different compost maturity and stability indicators including temperature, change in organic matter, organic carbon, pH, electrical conductivity, pathogen and helminth egg deactivation.

Chapter 4

This chapter was a rhizotron experiment which assessed the root morphology and architecture of cucumber (*Cucumis sativus*) plants when grown using co-compost. In context, this chapter identified rhizotrons as an inexpensive and alternative way of assessing phytotoxic compounds in the co-compost as it shows root development.

Chapter 5

This chapter is a greenhouse experiment which assessed the application of different soil amendments to reach maize (*Zea mays*) nutrient requirements. In context, it looked at the application of co-compost in plants that were grown and assessed for growth and development parameters as well as the availability of essential micro-nutrients within plants produced.

Chapter 6

This chapter is a socio-economic study where a survey was done assessing attitudes, perceptions, and willingness of university students to purchase and consume food produced from co-compost.

Chapter 7

This chapter is the general conclusion which provides a synthesis of the results and main findings and gives insights into future research.

References

Bai, F., and Wang, X. (2010). Nitrogen-retaining property of compost in an aerobic thermophilic composting reactor for the sanitary disposal of human faeces. *Frontiers of Environmental Science & Engineering in China* **4**, 228-234.

Banegas, V., Moreno, J., Moreno, J. I., Garcia, C., Leon, G., and Hernandez, T. (2007). Composting anaerobic and aerobic sewage sludges using two proportions of sawdust. *Waste Management* **27**, 1317-1327.

de Zeeuw, H. and Drechsel, P., (2015). Productive and safe use of urban organic wastes and wastewater in urban food production systems in low-income countries. In *Cities and Agriculture* (pp. 180-209). Routledge.

Dzulkurnain, Z., Hassan, M. A., Zakaria, M. R., Wahab, P. E. M., Hasan, M. Y., and Shirai, Y. (2017). Co-composting of municipal sewage sludge and landscaping waste: a pilot scale study. *Waste and biomass valorization* **8**, 695-705.

Gutiérrez, M., Serrano, A., Siles, J., Chica, A., and Martín, M. (2017). Centralized management of sewage sludge and agro-industrial waste through co-composting. *Journal of Environmental Management* **196**, 387-393.

Gwara, S., Wale, E., Lundhede, T., Jourdain, D., and Odindo, A. (2023). Ex-ante demand assessment and willingness to pay for human excreta derived co-compost: Empirical evidence from rural South Africa. *Journal of Cleaner Production* **388**, 135570.

Kwarciak-Kozłowska, A. (2019). Co-composting of sewage sludge and wetland plant material from a constructed wetland treating domestic wastewater. *In* "Industrial and municipal sludge", pp. 337-360. Elsevier.

Manga, M., Camargo-Valero, M., Anthonj, C., and Evans, B. (2021). Fate of faecal pathogen indicators during faecal sludge composting with different bulking agents in tropical climate. *International Journal of Hygiene and Environmental Health* **232**, 113670.

Manga, M., Evans, B. E., Ngasala, T. M., and Camargo-Valero, M. A. (2022). Recycling of Faecal Sludge: Nitrogen, Carbon and Organic Matter Transformation during Co-Composting of Faecal Sludge with Different Bulking Agents. *International Journal of Environmental Research and Public Health* **19**, 10592.

Manga, M., Muoghalu, C., Camargo-Valero, M. A., and Evans, B. E. (2023). Effect of turning frequency on the survival of fecal indicator microorganisms during aerobic composting of fecal sludge with sawdust. *International Journal of Environmental Research and Public Health* **20**, 2668.

Mihelcic, J. R., Fry, L. M., and Shaw, R. (2011). Global potential of phosphorus recovery from human urine and feces. *Chemosphere* **84**, 832-839.

Mtyobile, M. (2021). Evaluation of the yield performance of maize cultivars (*Zea mays L.*) in a Semi-Arid Region of the Eastern Cape Province, South Africa. *International Journal of Agricultural Science and Food Technology* **7**, 327-330.

Mwangi, W. M. (1996). Low use of fertilizers and low productivity in sub-Saharan Africa. *Nutrient cycling in Agroecosystems* **47**, 135-147.

Ofei-Quartey, M. N. L., Appiah-Effah, E., Akodwaa-Boadi, K., Ampaw, B., Taylor, T. S., and Millogo, Z. E. N. (2023). Enhancing the economic potential of organic waste by co-composting using ratio modelling toward a circular economy. *Journal of Material Cycles and Waste Management* **25**, 1560-1580.

Rockefeller, A. A. (1998). Civilization and sludge: Notes on the history of the management of human excreta.

STATS_SA (2022). General Household Survey 2021 Media Release. (S. S. Africa, ed.), pp. 2. https://www.statssa.gov.za/publications/P0318/GHS%202021%20MEDIA%20RELEASE.pdf

Szántó, G. L., Letema, S., Tukahirwa, J., Mgana, S., Oosterveer, P., and Van Buuren, J. (2012). Analyzing sanitation characteristics in the urban slums of East Africa. *Water Policy* **14**, 613-624.

WHO/UNICEF (2021). "Progress on household drinking water, sanitation and hygiene 2000-2020: five years into the SDGs," World Health Organisation, United Nations Children's Fund.

Yahaya, S. M., Mahmud, A. A., Abdullahi, M., and Haruna, A. (2023). Recent advances in the chemistry of nitrogen, phosphorus and potassium as fertilizers in soil: a review. *Pedosphere* **33**, 385-406.

Zhang, D., Luo, W., Li, Y., Wang, G., and Li, G. (2018). Performance of co-composting sewage sludge and organic fraction of municipal solid waste at different proportions. *Bioresource technology* **250**, 853-859.

Zhang, J., Sui, Q., Li, K., Chen, M., Tong, J., Qi, L., and Wei, Y. (2017). Influence of natural zeolite and nitrification inhibitor on organics degradation and nitrogen transformation during sludge composting. *Environmental Science and Pollution Research* **24**, 9122-9122.

Zhao, Y., Lu, W., Damgaard, A., Zhang, Y., and Wang, H. (2015). Assessment of co-composting of sludge and woodchips in the perspective of environmental impacts (EASETECH). *Waste Management* **42**, 55-60.

Chapter 2: Innovative Technologies for Nutrient Recovery from Organic Waste, and Reuse in Plant Production: A Review

2.1 Introduction

Due to the increasing population across the world, human waste has been used to extract minerals for its application in agriculture thus making it a valuable resource than ever (Singh et al., 2022). Nevertheless, the application of raw human excreta poses health risks due to the presence of pathogens such as *Helminth* eggs, *E. coli*, and faecal coliforms, (Lourenço and Nunes, 2020).

However, given the pathogen-rich nature of human excreta, researchers have developed various technologies to recover nutrients from it for sustainable agricultural use. Notably, technologies including vermicomposting, explored by (Wang et al., 2014), and vermicompost production from human waste, predominantly sewage sludge, as demonstrated by (Lv et al., 2018), have gained attention. Co-composting is gaining attention among researchers, such as (Hwang et al., 2020; Manga et al., 2021; Manga et al., 2023a; Ofei-Quartey et al., 2023).

Co-composting involves breaking down complex organic matter, comprising two or more feedstocks, into simpler components suitable for soil amendment (Giagnoni et al., 2020). These methods hold promise as a viable and acceptable approach for treating faecal sludge (FS) in both rural and peri-urban settings, offering the added benefit of critical nutrient recovery and reuse (Manga et al., 2022). Moreover, human urine can be used to reclaim nutrients in the process of struvite precipitation, Nutrified Urine Concentrate (NUC), and dried urine (Udert et al., 2015).

In developing countries of Africa and other Latin American countries, very few farmers use synthetic fertilizers, while most produce at the small-scale level using animal excreta, and recycled crop residues (Hidalgo et al., 2021). This inadequate addition of nutrients in the soil results in poor yield which subsequently results in food insecurity and malnourishment. Furthermore, demand for essential macronutrients such as nitrogen, phosphorus and potassium in the form of N, P_2O_5 (Di phosphorus pentoxide), and K_2O (potash) were expected to increase by 1.5, 2.2, and 2.4% respectively, from 2015 to 2020 (FAO, 2017), leading to poor yields for small farmers without their application.

The review aims to gain a current understanding of technologies used to recover nutrients from organic waste for plant production. The review will first examine the challenges of organic waste disposal and explore innovative technologies for recovering essential nutrients for agricultural use. However, special attention will be given to co-composting as a low-cost nutrient recovery technology. The review will further reveal key findings, research gaps and recommendations for future research.

2.2 Challenges with waste disposal and handling

2.2.1 Landfilling of municipal solid waste

Municipal solid waste (MSW) refers to any discarded items that possess the potential for recycling or disposal in landfills (Malinauskaite et al., 2017). This waste category encompasses a range of materials such as paper, plastics, food waste, market waste, wood, glass, metals, and textiles (Nanda and Berruti, 2021). Globally, an annual production of 2.01 billion tonnes of municipal solid waste occurs, and approximately 33% remains uncollected by local authorities (Kaza et al., 2018). The accumulation of organic waste within landfills and the presence of uncollected waste give rise to environmental pollution, leachate formation, and the attraction of disease-carrying flies, potentially leading to diseases like cholera and malaria.

Within landfills, organic waste compaction creates an environment conducive to the generation of landfill gases, which are released into the atmosphere through the soil cover under high pressure (Nanda and Berruti, 2021). In the years, 2005/6, South African landfills contributed to the emission of 16 $MtCO_2e$ (million metric tonnes of carbon dioxide equivalent) of methane, and 380 $MtCO_2e$ of greenhouse gas emissions (including CO_2, CH_4, and N_2O), with methane alone accounting for approximately 4.3% of total methane emissions (Stokke and Thommessen, 2013). The production of greenhouse gas emissions is a prominent challenge intertwined with waste management. Carbon dioxide (CO_2) arises under aerobic conditions, while CH_4 and N_2O emanate from the anaerobic decomposition of organic waste (Pereira et al., 2018).

Landfill leachate exhibits acute and chronic toxicity and is classified as hazardous because it can permeate into groundwater leading to biomagnification (Mishra et al., 2019). As depicted in Figure 2.1, pre-dumping leachate formation stemming from the fresh produce market, along with the presence of flies and offensive odours, occurs before the disposal of such waste in landfills.

Figure 2.1: Poor handling of organic waste which has resulted in the formation of leachates at Mkondeni Fresh Produce Market in Pietermaritzburg (Photo credit: Author)

2.2.2 Challenges with human excreta disposal

Globally, approximately 4.5 billion people don't have access to safely managed sanitation, and approximately 2.5 billion (56%) people do not have access to sanitation at all, this includes 600 million people who share a toilet or latrine with other households, and 892 million people rural areas settlers practising open defecating (Bishoge, 2021). The most common sanitation practises in urban areas are the sewer systems where excreta are diverted to wastewater treatment plants using pipes. In the informal settlements pit latrine or onsite sanitation is mainly used. All these sanitation practices encounter a problem with the disposal of excreta when pits are full and after the tertiary process in the wastewater treatment plant. Conventional pit latrines may pose an environmental risk to the communities if they are poorly maintained and hardly emptied (Szántó et al., 2012). Moreover, in highly populated communities few suction trucks come to empty soakage pits and septic tanks provide a big challenge because the sites are regularly not accessible due to poor or narrow steep roads within the settlements (Bishoge, 2021; Thye et al., 2011). Even if the pits were to be emptied the challenge can be disposal of human excreta. Most informal settlements and rural settlers end up building more pits because their pits are full and not emptied. A scenario from Delfkom, Mkhondo municipality in Mpumalanga province, South Africa, where most families with up to 10 members per household had to build new toilets every 10 years (Figure 2.2). The first toilet (Fig 2.2 **A**) was built by the Mkhondo municipality in 2010 for a family of 10 and was already full in 2018, the household head said that they used ashes and black deep to reduce the smell until in 2023 a new toilet (Figure. 2.2 **B**) which they dug and made for themselves. This shows a clear picture that pit latrines are built without the plan of emptying them and can significantly pose health risks to communities.

Figure 2.2: Scenario at Delfkom, Mkhondo municipality, Mpumalanga, South Africa where the full toilet is closed (**A**) and a new toilet is built by the household member (**B**) to accommodate 10 or more family members (Photo credit: Author).

On the other hand, centralized systems such as Wastewater Treatment Plants (WWTP) produce larger quantities of sewage sludge and its management is becoming a global issue of growing importance (Cieślik et al., 2015). High quantities of sewage sludge occur as a by-product of wastewater treatment processes. Wastewater undergoes primary processing where it is treated to remove viruses, reduce pathogens and subsequently dewater sludge. Quite often this is followed by improper disposal of dewatered sewage sludge (DSS) which causes critical environmental pollution because of its decayable characteristics if discharged into the environment (Zhang et al., 2017). A recent scenario unfolded as depicted in Figure 2.3 in the

KwaZulu-Natal region, specifically in Pietermaritzburg, South Africa. In this situation, DSS is discharged into the environment which is closer to the informal settlement this has created an unsafe environment, particularly affecting children and residents residing in these communities. Pathogenic organisms and other harmful insects that are disease vectors transmit diseases to human beings in the surrounding areas. Therefore, innovative and sustainable ways of counteracting this improper organic waste while recovering nutrients are required.

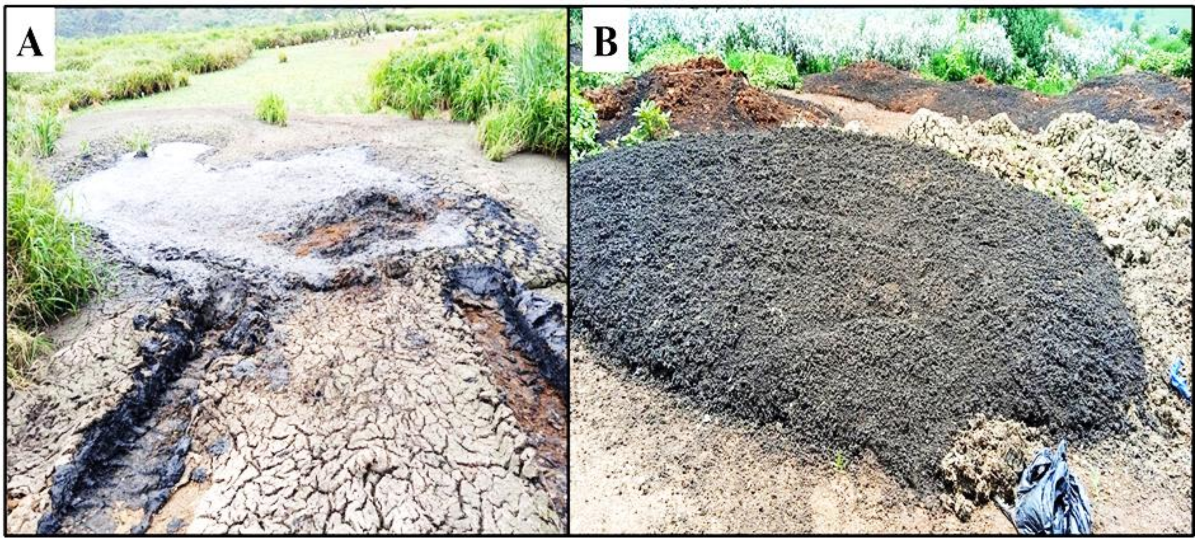


Figure 2.3: Disposal of dewatered sewage sludge (DSS) in the ground or environment in Pietermaritzburg, KwaZulu-Natal, South Africa, **(A)** shows a picture immediately after disposal while (**B)** shows DSS after a few weeks of disposal (Photo creditor: Author).

2.2.3 Nutrient composition of human excreta

Human urine contains approximately 90% water while the remaining 10% are inorganic salts and organic compounds (Rose et al., 2015). Human faeces are composed of 74.6% of water and the remaining 25% is organic matter in terms of solids (Rose et al., 2015). It has N, P, K ranging between 5-7%, 3-5.4%, and 1-2.5% respectively (Rose et al., 2015). These two excrements contain micro-nutrients such as magnesium (Mg) and selenium (Se) which are required by plants and the human body (Harder et al., 2019). Table 2.1 below demonstrates the chemical composition of human faeces and urine; it also clearly demonstrates the quantities of faeces and/or urine produced by a person per day and its nutrient composition.

Table 2.1: Chemical composition of human faeces and urine (Chaggu, 2004).

Approximate Quantity	Faeces	Urine
Quantity (wet) per person per day (g)	70-520	100-1500
Quantity (dry) per person per day (g)	30-70	50-70
Proximate composition (%dry weight/matter)	88-97	65-85
Moisture (%)	66-85	93-99.5
Organic matter (%)	88-97	65-85
Nitrogen (%)	5.0-7.0	15-19
NK_j (gN) (%)	1.4-2.46	5.29
Phosphorus as P2O5 (%)	3.0-5.4	2.5-5.0
PO_4-P (%)	-	0.20
Total Phosphorus (g) (%)	0.69-2.5	1.08-2.2
Total Potassium as K_2O (%)	1.0-2.5	3.0-4.5
Potassium (K) (%)	0.8-2.1	2.5-3.7
Carbon (%)	44-55	11.0-17.0
Calcium as (CaO)	4.5	4.5-6.0
Calcium as Ca	2.9-3.6	3.2-4.3
C:N ratio	5.0-10	0.6-1.1
TS (%)	33	-
pH	-	7.08-9
Proteins (g)	4.0-12.0	0.28
Total lipids (g)	4.0-6.0	-
Polysaccharides (g)	4.0-10.0	0.68

g (grams), **%** (percentage), **C:N ratio** (Carbon to nitrogen ratio),

2.3 Innovative technologies for nutrient recovery

Due to the increasing population across the world, human waste has been used to extract minerals for its application in agriculture thus making it a more important parameter than ever (Singh et al., 2022). Food consumed by humans contains nutrients from crops and during excretion, a nutrient-rich waste (faeces and urine) is produced, which can fulfil 22% of the demand for phosphorus (Ahmed et al., 2019). However, the nutritional value of this waste is very low, and it encompasses bad odour, heavy metals and pathogenic organisms (Mehta et al., 2015). Therefore, innovative technologies for nutrient recovery are required and they are divided into the triangle of nutrient accumulation, extraction, and release (Ahmed et al., 2019). Few of these technologies were developed in the past to highlight the significance of waste in the environment (Mehta et al., 2015). Figure 2.4 underpins the most important organic waste in the concept of nutrient recovery technologies and their reuse in plant production.

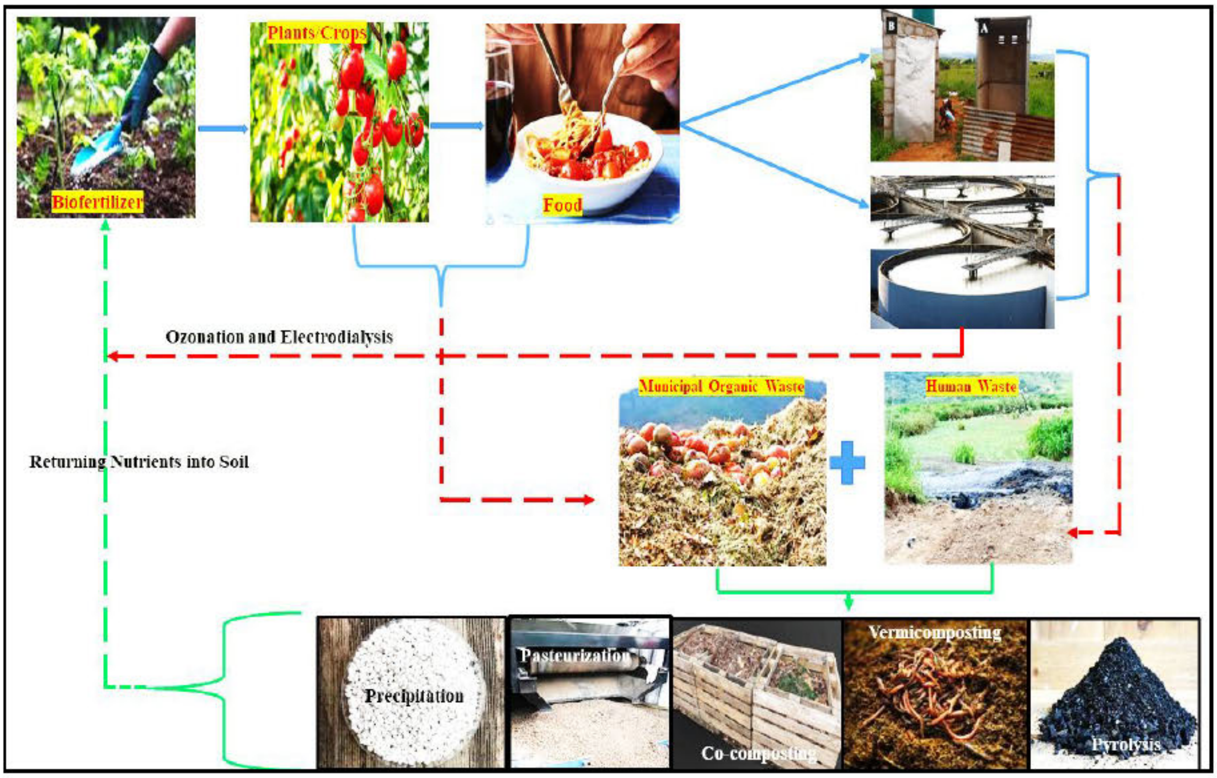


Figure 2.4: The nutrient circular economy model outlining different technologies including pyrolysis, vermicomposting, co-composting, pasteurization, precipitation, ozonation and electrodialysis from different organic waste sources (Sketch: Author; Pictures: Bouhia et al., 2022, Septein et al., 2018).

Research has been done on nutrient recovery technologies such as pyrolysis of biochar (Nkomo et al., 2021), vermicomposting (Singh and Sinha, 2022), composting, Decentralised Wastewater Treatment Systems (DEWATS) (Musazura, 2018), struvite precipitation and nutrified urine concentrate (Li et al., 2019), dried urine, and co-composting (Manga et al., 2022; Ofei-Quartey et al., 2023). There is also ongoing research on osmosis and electro-dialysis also receiving attention from researchers (Xie et al., 2016) and ozonation (Ribeiro et al., 2024). Implementing nutrient recovery technologies is essential for ensuring the hygiene and safe use of human waste-based fertilizers (Harder et al., 2019).

2.3.1 Pyrolysis

Pyrolysis occurs at 400–800°C under oxygen-limited conditions (Ibrahim, 2020), producing biochar as a stable organic carbon material. Biochar can be generated by heating biomass to 300–1000°C under low or zero oxygen conditions (Lehmann and Joseph, 2015) and can also be produced through gasification or hydrothermal carbonisation (Shanmugam et al., 2022). Biochar pyrolysis produced at the temperature of 400 °C has a shapeless structure; however, increasing the temperature to approximately 800 °C displayed a turbostratic carbon structure (Shanmugam et al., 2022).

Different types of feedstocks such as rice stalks, animal waste, wood chips, and maize straws can be used for biochar production (Rasul et al., 2016). Biochar can also be produced from faecal sludge to facilitate the destruction of pathogens and nutrient recovery (Krueger et al., 2020). Nitrogen content in the biochar is often very low, thus it can be mixed with the residual faecal matter which remains after harvesting Black Soldier Flies Larvae to increase N content (Nkomo et al., 2021).

Nutrient use efficiency can be enhanced by biochar by reducing nutrient leaching, improving soil pH and structure, and increasing plant nutrient availability (Das et al., 2020). Biochar is also known to reduce Al and Fe toxicity on plant roots and microorganisms, reduce heavy metals in the soil, and decrease N_2O and CH_4 emissions from agricultural fields (Das et al., 2020). Biochar can be applied at a rate between 5-50 t/ha and has been observed to yield very good results (Huang et al., 2017). It can also be incorporated or added with other amendments such as manure, compost, and co-compost, furthermore, it needs to be applied every year for optimum results (Khan et al., 2017).

2.3.2 Alkaline urine dehydration and precipitation

During precipitation ammonium, magnesium, and phosphates are added to the urine to overcome solubility product under an alkaline environment to recover available phosphorus in the urine (Saidou et al., 2009). Struvite ($MgNH_4PO_4.6H_2O$) is formed by adding magnesium sources such as $MgCl_2$, MgO, and $MgSO_4$ to urine (Udert et al., 2015). Approximately 70% of phosphorus (P) was recovered from human urine while 6% N was left in the solution (Tilley et al., 2014). A report by Grau et al., (2013) demonstrated that 93% of P was recovered while 97% N was left as the supernatant in the solution during struvite precipitation. Moreover, urine can also be nitrified to produce a Nutrified Urine Concentrate (NUC) which subsequently has more nitrate- N than ammonium- N giving (Mchunu et al., 2018). The NUC fertiliser is a nitrogen-concentrated liquid fertiliser which results from nitrification and distillation of urine that was source-separated (Nkomo, 2021). Nitrified urine concentrate (NUC) is a nutrient-rich fertilizer that can be used for crop production. The production of NUC has largely been seen in some European countries and South Africa and reactors have been primarily used to make it (Wilde et al., 2019).

2.3.3 Waterborne systems (Decentralised Wastewater Treatment System)

DEWATS is a community-focused wastewater treatment system designed to handle water-borne waste. It includes a settling tank, an anaerobic baffled reactor (ABR), an anaerobic filter, and planted gravel filters (PGFs) that provide secondary or advanced treatment through horizontal and vertical flow constructed wetlands (HFCW and VFCW) (Phoku, 2020; Sasse, 1998). DEWATS provides a practical alternative for managing sludge in rapidly growing urban areas. Unlike centralized wastewater treatment plants, which can be expensive and take a long time to set up, DEWATS is more cost-effective and quicker to implement (Swan et al., 2023). According to Amoah et al., (2018), this system can remove 95% soil-transmitted helminths and 100% *Taenia spp*., compared to centralized systems which remove 67% and 100%, respectively, thus implying that DEWATS are more efficient for the production of agricultural effluents that meet standards (Musazura and Odindo, 2021). Studies have been conducted on testing the use of DEWATS effluent for agricultural production. A study investigating the effect of irrigating maize (*Zea mays*) grown in different soils using ABR effluent was conducted looking at nutrient holding capacity and leaching of the soil after irrigation (Bame et al., 2013). Moreover, a study to determine the effect of ABR effluent on nutrient uptake and leaching to the soil using Swiss chard (*Beta vulgaris*) (Musazura et al., 2015). DEWATS can offer a promising sanitation solution to communities and residents living in informal settlements while at the same time, the effluent produced can be used for agricultural plant production (Tesfamariam et al., 2018).

2.3.4 Pasteurization

Municipalities have implemented ventilated improved pit (VIP) latrines, but over time, the pits fill up and need to be emptied every five years at no cost to households (Zuma et al., 2015). The human waste produced in this VIP contains nutrients and has the potential to be used in agriculture. However, it also contains a significant number of pathogens such as *E. coli* and helminth eggs, (Lourenço and Nunes, 2020) which can be harmful when it come into contact with crops or food.

The eThekwini municipalities, under eThekwini Water and Sanitation (TWS) in collaboration with Particle System Separation (PSS) developed a Latrine Dehydration Pasteurization (LaDePa) machine to treat VIP sludge (Harrison and Wilson, 2012). Faecal sludge that is collected from the households is extruded using a LaDePa machine which forms pellets that undergo infrared radiation. After undergoing infrared radiation, the pellets are dried and pasteurized which makes them safe to handle, with minimum exposure to pathogen risk, and can be sold as an agricultural product (Harrison and Wilson, 2012; Septien et al., 2018). The LaDePa pellets have phosphorus that is comparable to industrial fertilizers and higher than organic fertilizers, and they are rich in carbon, nitrogen and potassium (Septien et al., 2018). Due to the high operational cost of the LaDePa including electricity and/or diesel, this system may not be feasible to use in developing countries and the product is not stable for use in agriculture (Nikiema and Cofie, 2014).

2.3.5 Ozonation

Ozone (O_3) gas has been extensively used to purify and disinfect drinking water and treat landfill leachates, and industrial and municipal wastewater (Bekbolet et al., 2005; dos Santos Pereira et al., 2018). Regardless of its advantages in treating several wastewaters as mentioned above gas has low solubility in water thus making this technology costly to implement (Ribeiro et al., 2024). To overcome this challenge of gas solubility in water, ozonation can be mixed with hydrogen peroxide (H_2O_2) leading to peroxidation (Oturan and Aaron, 2014). This technology has been implemented and has shown potential for treating potato wastewater. Ozonation combined with catalyst increased efficiency and mineralization thus made suitable for re-use in agriculture (Cao et al., 2022). However, Rossi et al., (2021) found that ozonation in combination with ultrasound effectively treated wastewater because it promoted a circular economy by recovering nutrients and retaining them back in the soil, it also led to reduced synthetic fertilizer use (Rossi et al., 2021). Moreover, ozonation removed 99% of pharmaceuticals in wastewater making it most suitable for agricultural use (Szabová et al., 2020).

2.3.6 Composting

Composting is differentiated into three namely, aerobic, anaerobic, and vermicomposting. In these types of composting one type of feedstock such as food waste, grass clipping and garden waste. The product of these types of composting techniques is affected by different environmental conditions that are required for each composting technique (Shah et al., 2016). The differences may be observed in macro-nutrients (NPK), loss of nitrogen through ammonification, nitrous oxide, and leaching. Composting organic green waste as an alternative way to dumping it in landfills has plenty of benefits including recovering nutrients and converting it to natural fertilisers which can be used as soil amendments to improve soil structure (Zhu-Barker et al., 2017). Organic green waste (OGW) contains recalcitrant organic compounds such as lignin, hemicellulose, and cellulose that are difficult to break down (Reyes-Torres et al., 2018). Composting biodegradable organic green waste is essential and using the compost in agricultural production is the most recognised activity occurring in developed and developing countries (Gómez-López et al., 2022).

2.3.6.1 Aerobic composting

During aerobic composting, aerobic microbes break down complex organic compounds. These microorganisms use oxygen for respiration and in turn release carbon dioxide and water as by-products. Therefore, compost must be well aerated to provide sufficient oxygen for the oxidation of the organic material and to evaporate excess moisture (Chen et al., 2015). Important parameters for composting such as temperature, moisture content, pH, carbon to nitrogen (C/N) ratio, and turning frequency to ensure proper aeration (Diaz et al., 2011), must be maintained at optimum levels. Preserving the moisture content ranging between 50–55%, and the temperature at the thermophilic range (55-65°C) is very essential in aerobic composting for pathogens deactivation (Ruggero et al., 2019).

2.3.6.2 Anaerobic composting

In anaerobic composting degradation of the organic material occurs in the absence of oxygen and the process requires anaerobic bacteria and methanogenic archaea. The microbes present in the anaerobic composting converts organic waste into biogas of approximately 70% methane, 30-40% CO_2, hydrogen (H_2) and tetrasulfide (H_2S_4) to nutrient-rich materials that can be used as soil amendments (Sheets et al., 2015).

2.3.6.3 Vermicomposting

Vermicomposting occurs when earthworms are used to decompose organic waste such as food waste, animal manure and faecal sludge (Arancon et al., 2020). Epigenetic earthworms are identified by bright red and reddy-brown burrow waste during the vermicomposting process (Lee, 1985). These earthworms use the aerobic and anaerobic micro-flora present in their guts to break down the organic waste (Ali et al., 2015). Micro-organisms that are found within the vermicompost are responsible for the biochemical degradation of organic matter while the earthworms alter biological activities and conditions of the substrates (Ali et al., 2015).

Earthworms used for the vermicomposting process can tolerate temperatures between 10-35°C, higher temperatures can cause the death of earthworms (Edwards, 1988). Earthworms excrete faecal black casts which are rich in nutrients after ingesting the organic waste and that is more microbially active than when the organic waste was consumed (Domínguez et al., 2010). Vermicompost tea contains nutrients, microorganisms, and plant growth promoters, and its application has shown an improvement in seed germination, growth, and yield enhancement (Arancon et al., 2020). As compared to compost, vermicompost is produced in a shorter period because of the presence of optimum moisture content that enhances the process of humification, which intensifies the conversion of organic matter (Arancon et al., 2020). Vermicompost increases the stability, organic matter, and nutrient availability of the plants when applied in the soil. The disadvantage of vermicomposting faecal matter is that temperatures between 10-35℃ are not enough to deactivate pathogens such as *E. coli* and helminth eggs.

2.3.7 Co-composting

Co-composting is defined as the method of breaking down complex organic matter comprising two or more feedstocks into simpler parts that can be used to amend soils (Giagnoni et al., 2020). Feedstocks that can be used for co-composting include faecal sludge (Manga et al., 2021; Ofei-Quartey et al., 2023), dewatered sewage sludge (Zhang et al., 2018), chicken manure (Ofei-Quartey et al., 2023), and/or swine manure (Lau et al., 1992). Co-composting is the most desirable method of treating sewage or faecal sludge because of its low energy requirements and ability to recover macro-nutrients such as nitrogen, phosphorous and potassium (Manga et al., 2021). However, faecal and/or sewage sludge cannot be co-composted alone because it has a low Carbon/Nitrogen ratio as well as a high moisture content (Manga et al., 2021). Previous studies show that human waste can be co-composted with different bulking agents such as rice hulks (Mori et al., 1981), wood chips (Golbaz et al., 2021), municipal solid waste (Ofei-Quartey et al., 2023), coffee husks (Manga et al., 2021), and sawdust (Kebibeche et al., 2019). Co-compost can increase soil organic matter, improve the quality of the soil, enhance soil porosity and aeration, and improve water holding capacity, nutrient availability and stability of the soil (Hwang et al., 2020). Co-composting human waste and green organic waste residues can return nutrients to the soil while at the same time ensuring a circular nutrient economy.

2.3.7.1 Factors affecting co-compost production

Co-compost production and quality can be affected by several factors including Carbon-to-Nitrogen (C/N) ratio, temperature, moisture content, aeration methods, aeration frequency, and presence of pathogens (Diaz et al., 2011).

2.3.7.2 Bulking agents and carbon-to-nitrogen ratio

The selection of bulking agents (carbon reach materials) is critical because it shows the relative proportion of carbon to nitrogen ratio that needs to be added to the co-compost heap (Manga et al., 2022). The bulking agent also plays a major role in absorbing the excess moisture content in the sludge and further acts as the desiccant to reduce odour (Zhang et al., 2018). There should be a balance between the carbon and the nitrogen (C/N ratio) in the co-compost to speed up the co-composting process. If there is a high C/N ratio the composting process takes longer to reach maturity because aerobic microbes will have more than the required carbon and thus take longer to break down. A low C/N ratio results in nitrogen destabilization/supplied in access and N being lost as a form of ammonia (Wichuk and McCartney, 2007). The optimum or the standard C/N ratio for co-composting faecal sludge and organic green waste is between 20:1 and 30:1 (Nemet et al., 2021). Sewage sludge, green waste, and food waste were co-composted at low C/N ratios of 8.75, 18.00, and 24,90 conducted for three months where temperature, pH, moisture, carbon, and nitrogen proportion, and germination index as an indicator of co-compost maturity (Grgas et al., 2023). The co-compost produced was within United Nations standards of permissible heavy metal standards and was of good quality as it was characterized by an 89% germination index (Grgas et al., 2023).

2.3.7.3 Co-composting phases and temperature

Temperature conditions during co-composting can be categorised into different phases namely mesophilic, thermophilic, cooling and/or maturation phase. The first composting phase "mesophilic" is characterized by a temperature between 25-45°C (Wichuk and McCartney, 2007). Thermophilic temperature follows mesophilic, in this phase temperature increases from 45°C to 65°C depending on aeration, moisture content and microbial activities taking place (Trautmann and Krasny, 1998). The thermophiles or heat-loving microbes replace mesophile microbes thus breaking down of organic matter and killing of lethal pathogens occurs at a faster rate (Trautmann and Krasny, 1998). Several authors define thermophilic temperature as a co-composting temperature ranging between 45-65°C (Deacon, 2002; Finore et al., 2023). According to Manga et al. (2023b), an increase in microbial diversity occurs if the temperature is 35 to 40°C while 45–55°C is vital for improving the degradation rate, and those greater than 55°C led to compost sanitation. Temperature also plays a crucial role in deactivating or killing pathogens and weed seeds. When the supply of fats, proteins, cellulose and hemicellulose decreases the mesophilic microbes take over and continue to break organics as they approach the cooling and/or maturation stage (Trautmann and Krasny, 1998). In the cooling and maturity phase, slow degradation of lignin and complex matters is observed with the decreased number of bacteria but increased fungi which break down agar and residual lignin (Diaz et al., 2011).

2.3.7.4 Relationship between pile size and temperature

Co-compost heap or pile must be large enough to avoid dissipation of heat but should not be too large that it prevents air circulation. A pile size of 1m^3 is the recommended minimum size for thermophilic co-composting (Trautmann and Krasny, 1998). Research on co-composting faecal sludge and organic waste made with a volume of 3 m^3 attained thermophilic temperature (55-65 °C) which resulted in the deactivation of pathogens (Manga et al., 2021). However, the highest temperature of 49.17 °C for all composting piles with dimensions of 0.58 m height and 0.53 m width and a weight of 9.3 kg was reported by (Ofei-Quartey et al., 2023).

The increase in temperature in the composting piles or windrows also depends on the bulking agent and size of the composting piles. Temperatures above the threshold (<55 °C) are ideal for pathogen (*E coli*, faecal and total coliform) and helminths eggs (*Ascaris*, *Trichuris*, and *Taenia*) deactivation when kept constant or above 55 °C for 10 consecutive days (WHO, 2006). High temperatures can also destroy the germination of weed seeds in the feedstocks (Nemet et al., 2021). However, there are useful micro-organisms such as actinomycetes which break down complex organic matter that can be destroyed at a temperature above 65 °C, therefore maintenance of temperature is always essential (Diaz et al., 2011).

2.3.7.5 Moisture content

Microbiological and chemical processes are important in the co-compost/compost thus moisture content between 50-60% is required to cater for these processes (Nemet et al., 2021). Too low (<35%) moisture content deprives microbes of water needed for their metabolism and inhibits their activities thus resulting in a low decomposition process (Liang et al., 2003). Moreover, dry co-compost piles tend to lose heat because of low microbial activity and a lot of air circulation. High moisture content (>60%) can cause reduced air space thus leading to anaerobic respiration (Chen et al., 2011). This can further lead to offensive or bad odour production in the co-compost. Moisture content can be measured using an oven-dry method where representative samples are dried at 105 °C for 24 hours (Manga et al., 2021; Okalebo et al., 2002). However, farmers practice the "squeeze method" where co-compost samples are squeezed with a hand if it forms a ball then there is no need for moisture correction (Trautmann and Krasny, 1998). Controlling moisture content is essential for controlling the spore-forming bacteria and *Ascaris* eggs' survival as they thrive in excess moisture co-compost (Sanguinetti et al., 2005).

2.3.7.6 Aeration methods and turning interval

Aeration or turning of the co-compost is vital for microbial growth, heat generation and volatilisation of gases (Guo et al., 2012). Aerobic microorganisms breaking down the complex organic matter in the co-compost require oxygen for respiration. Composting with less or excessive aeration influences the decomposition process of the organic material and can reduce temperature and moisture content resulting in nutrient loss and deterioration of compost quality (Kalamdhad and Kazmi, 2009). Researchers have investigated the effect of turning frequency or aeration rate on compost quality. A study conducted by Zhang et al., (2019) to determine the effect of turning frequency (5, 7, 10, 15 days) interval on co-compost quality revealed that a turning frequency of 7 days was optimum and can be implemented during co-composting processes. However, Soto-Paz et al., (2020) investigated the effect of turning frequency (7, 14 and 21 days) on co-compost quality and found that the optimum aeration frequency is 14 days which is different from the 7 days TF reported by (Zhang et al., 2019). A recent study by Manga et al., (2023a) was based on determining the effect of turning frequency (3, 7 and 14-day intervals) on pathogen deactivation in faecal sludge mixed with sawdust. The results showed that there was a significant difference between turning frequencies, the 3-day TF resulted in pathogen inactivation in 8 weeks compared to prolonged turning frequencies (7TF and 14TF) which took 13 weeks for pathogens to be deactivated. Contrasting results on the effect of turning frequency (3- and 10-day intervals) for *Helminth* egg inactivation showed that there was no significant difference between the turning frequency (Koné et al., 2007). Different bulking agents and mixing ratios have a significant effect on the turning frequency of the co-compost being produced. These contrasting observation on turning frequencies based on compost maturity, temperature, and pathogen deactivation indicates that more research is still required to determine the most optimum turning frequency for quality co-compost production with different feedstock. Differences in terms of the study location could also contribute to differences in optimal turning frequency observation.

2.3.7.7 Effect of turning frequency on co-compost volume

High turning frequency increases pathogen deactivation in the co-composting process however it also leads to potentially reduced final compost volume (Manga et al., 2023a). Moreover, the high turning frequency of once every two days led to high organic matter degradation and promoted material degradation of the sheep manure compost (Cai et al., 2023). The study by Getahun et al., (2012) showed that high turning frequency can lead to an increased decomposition rate. However, it can also lead to total C and N reduction and reduction of compost volume. Another study where a 7-day turning interval was implemented revealed that compost volume was reduced due to increased aeration and decomposition efficiency (Zhang et al., 2019). High turning frequency led to a reduction in compost volume, temperature, density and moisture reduction, in addition, first turning decreases the odour emission (Shen et al., 2015).

2.3.7.8 pH and EC of the co-compost

The deactivation of pathogens in the co-compost occurs due to increased pH values (Manga et al., 2023b). The pH increases as a result of CO_2, organic acids and ammonia production which are released as the by-product during the co-composting process (Popat et al., 2010). Therefore, an increase in pH levels has been noted by several authors as a factor that leads to the deactivation of pathogens in the co-composting process (Darimani et al., 2016; Guo et al., 2012). In the later composting periods pH becomes neutral as ammonia is lost into the atmosphere or added to other microbial growth (Trautmann and Krasny, 1998).

The electrical conductivity (EC) of the co-compost/compost represents salinity and can be used as one indicator of compost maturity or stability (Liu et al., 2011). It also indicates the level of decomposition and mineralization of organic matter in the compost (Bazrafshan et al., 2016). Electrical conductivity is often observed by decreasing as composting progresses or at the curing stage reflecting the readiness and stability of the compost. Literature reports indicate that the most optimum EC level should be less or equal to 3 mS/cm (milliesemiens per centimetre) (Manga et al., 2022). However, EC ranging between 5.6-8.1 mS/cm was observed by Bazrafshan et al., (2016), which could contain phytotoxic that may inhibit seed growth when applied to the soil (Said-Pullicino et al., 2007).

2.3.7.9 Implications of co-compost health and safety

The most important parameter for assessing co-compost quality is the deactivation of pathogens. *E. coli* is the most popular within the faecal coliforms category, serving as one of the frequently observed pathogenic microorganism indicators in biological procedures aimed at managing organic waste that includes materials associated with faecal matter (Manga et al., 2023a). According to the literature temperatures above 55℃ that occur during the decomposition of co-compost as the results of microbial activities are essential for pathogen deactivation. Other studies show that turning the compost more often can have a significant impact on pathogen deactivation (Manga et al., 2023a). Pathogen deactivation during FS composting is more complex, and not well understood due to conflicting information in the literature (Manga et al., 2021).

However, the recommended standard for, *E. coli* and faecal coliform concentrations must be <1000 $MPNg^{-1}$ or <3 log units $CFUg^{-1}$ (Nartey et al., 2017). South Africa and other developing countries do not have standards for co-composting thus has adopted United States of America (USA) standards and has championed the concept of pathogen reduction as a number one compost quality standard (USEPA, 1995). Therefore, future research or studies should focus on determining the most accurate way of pathogen deactivation to produce quality co-compost when using human waste as feedstocks.

2.3.8 Effect of waste-based fertilisers on agricultural production

Faecal sludge contains essential macronutrients (potassium, nitrogen, and phosphorus), micronutrients such as copper (Cu), iron (Fe), zinc (Zn), and manganese (Mn), and other organic compounds that improve soil fertility by increasing water retention capacity, aeration, and infiltration, resulting in better soil structure that stimulates plant growth (Guzha et al., 2005). Moreover, these nutrients can be returned into the soil which will not only improve soil structure and fertility but can potentially lead to a circular economy of nutrients. Compost contains nutrients as well as organic matter which builds up the replenished soil (Adugna, 2016). Mineralization of nutrients contained in organic matter and their release in a plant-available form to the soil solution is associated with soil fertility and adding chemical fertilizers alone is not sufficient to retain fertility and water in the soil (Adugna, 2016).

An experiment carried out through the application of 0 (control);5;10 tonnes/ha to measure the nutrients present at harvest indicated that N, P, and K occurred at higher levels of nutrients in all plots with compost mainly in the pot that got 10t/ha of compost (Abdel-Rahman, 2009). Another study was conducted whereby radish (*Raphanus sativus L.*), and pepper (*Capsicum annum L.*) were planted using co-compost made from faecal sludge and municipal solid waste to assess it (Grau et al., 2017). The results indicate that under drought conditions followed by water-saturated soil, co-compost treatments produced comparable yields and significantly improved survival rates compared to the control treated with urea, triple super phosphate, and muriate of potash. An experiment was conducted in Bangladesh to determine the effect of compost supplemented with chemical fertilisers on tomato (*Solanum lycopersicum*) growth, development and yield (Saha et al., 2017). Compost applied at 2 tons/ha supplemented with 50% chemical fertilizer had the highest yield compared to 100% chemical fertilizer. The application of vermicompost at 2 tons per hectare, supplemented with 50% chemical fertilizer, resulted in the lowest observed salinity, which was considered beneficial.

Co-compost made from faecal sludge and oil palm empty fruit bunches (EFB) and/or cocoa pod husk (CPH) was tested for its effects on tomato growth from the seedling stage to maturity (Nartey et al., 2017). Vegetative growth of tomatoes proved to be higher in the treatments with faecal sludge co-compost compared to synthetic fertilisers. Long-term field tests demonstrated that compost has a stabilising influence on seasonal and annual variations in soil moisture, air and heat balance, availability of plant nutrients, and therefore crop yields (Amlinger et al., 2007).

2.3.9 Effect of waste-based fertilisers on greenhouse gas emissions

Composting contributes to and offers the potential to mitigate greenhouse gas (GHG) emissions. The extent of emissions reduction achieved through composting hinges on several factors, encompassing waste composition (particularly the organic fraction) composting technologies employed, the utilization of gas cleaning methods (especially within enclosed systems), and the ultimate purpose of the final compost product (Boldrin et al., 2009). The aeration of co-compost directly or indirectly influences GHG emissions, where heightened aeration corresponds to increased emissions (Han et al., 2018). Additionally, research by Chen et al., (2021) revealed substantial CO_2 and NH_2 emissions during the thermophilic phase of manure and organic waste composting, indicating significant organic matter breakdown and microbial activity within the compost (Wu et al., 2017).

Nonetheless, minimal GHG emission from the composting process was observed, suggesting that composting kitchen waste which constitutes approximately 57% of municipal solid waste could potentially reduce GHG emissions by around 24% (Zhao et al., 2009). Faecal sludge manures also exhibit an advantage in lowering greenhouse gas emissions compared to chemical fertilisers (Rahman et al., 2019). Comparatively, the GHG emissions from landfilling organic waste are higher than those from composting, underscoring the need for a more comprehensive scientific investigation (Khan et al., 2017). Landfills release substantial amounts of GHGs, around 3640 mg CO_2^{-e}/m^2/hour, while vermicomposting only emits 463 mg CO_2^{-e}/m^2/hour (Singh and Sinha, 2022). This major difference implies that an amount of approximately 7.86 times higher emissions in landfills, underscores the imperative of waste management through composting. Optimal moisture content in vermicomposting is crucial; higher moisture content diminished CH_4 and N_2O emissions by 32% and 40%, whereas lower moisture content reduces them by 16% and 23%, respectively (Robin et al., 2011). Thus, maintaining appropriate moisture levels during vermicomposting is essential for mitigating potent GHG emissions.

The introduction of biochar into soil offers a means of climate change mitigation by reducing GHG emissions and enhancing carbon sequestration (Sri Shalini et al., 2021). The inclusion of biochar in soil effectively curbs CO_2 emissions by sequestering C and diminishes GHG emissions (CH_4 and N_2O), resulting in a significant impact (Roberts et al., 2010). A study conducted by Mertenat et al., (2019) in Indonesia indicated that the waste treatment facility utilizing black soldier fly larvae displays lower greenhouse gas emissions (CO_2, CH_4, and N_2O) and energy consumption (electricity and diesel) compared to open windrow composting.

2.4 Key findings

The key findings of this review emphasize that organic waste diversion from landfills is achievable through environmentally friendly technologies that recover nutrients. Research demonstrates the potential for nutrient reclamation from various forms of organic waste, encompassing animal manure, food waste, organics, as well as human waste, including urine, faeces and wastewater. The review additionally uncovers the essential processes necessary to generate pathogen-free soil amendments, like biochar pyrolysis, co-composting, and LaDePa palletization. Furthermore, evidence suggests the recovery of macronutrients from urine via methods like nutrified urine concentrate (NUC) and struvite precipitation. The review also highlights the potential of vermicomposting using earthworms for food waste, animal manure, and human faeces. Yet, the deactivation of pathogens like *E. coli* and coliforms is challenging due to temperature restrictions for earthworms. Aerobic composting, employing a mix of carbon-rich browns (e.g., leaves, wood chips) and nitrogen-rich greens (e.g., food waste), offers an alternative to reduce organic waste in landfills, potentially reaching pathogen-deactivating temperatures. However, an imbalance in the C/N ratio hinders its efficacy. Co-composting can emerge as the cost-effective approach for nutrient recovery from human waste (faecal and sewage sludge) and organic green waste because it sanitizes human excreta without extensive energy use compared to pasteurization. This method can aid wastewater treatment plants grappling with sewage sludge disposal after treatment. Moreover, the co-compost has encompassed high water retention, organic matter, and soil conditioning properties for poor soils. The review also revealed that the adoption of the reviewed technologies for nutrient recovery can contribute to the reduction of greenhouse gas (GHG) emissions. For instance, during the vermicomposting process, only 463 mg CO_2-e/m^2/hour is emitted, whereas waste deposited in landfills releases significant amounts of GHGs, approximately 3640 mg CO_2-e/m^2/hour (Singh and Sinha, 2022). Despite the emission of GHGs associated with these technologies, their quantities are considerably smaller than those generated by organic waste landfilling.

2.5 Research gaps and recommendations for future research

Nutrient recovery technologies such as vermicomposting and co-composting offer solutions to organic waste valorisation. However, there is no consistency in pathogen deactivation when using the time-temperature criteria. Therefore, future research should focus on optimizing parameters such as the mixing ratio of initial feedstocks, turning frequency and moisture regulation to come up with an optimum process for co-compost production. Previous research has primarily focused on the technical feasibility of the technology studies. However, they lack comprehensive cost-benefit analysis and feasibility of the technology. Therefore, future research is needed to determine the economic sustainability and scalability of technologies including but not limited to co-composting, vermicomposting and pyrolysis.

Research on the impact of human excreta-derived product application has been limited to assessing and observing the above-ground performance of selected crop species. Further research is needed to assess the plant response and below-ground performance (root morphological traits) especially using sensitive crop species. Furthermore, these can be used to determine phototoxic compounds in the co-compost.

There is very scant research on societal acceptance of human excreta-derived products, therefore, research is needed to investigate consumer and farmers' attitudes, perceptions and willingness to accept human excreta-derived fertilisers on crop production.

References

Abdel-Rahman, G. (2009). Impact of compost on soil properties and crop productivity in the Sahel North Burkina Faso. *American-Eurasian Journal of Agricultural and Environmental Science* **6**, 220-226.

Adugna, G. (2016). A review on the impact of compost on soil properties, water use and crop productivity. *Academic Research Journal of Agricultural Science and Research* **4**, 93-104.

Ahmed, M., Ahmad, S., Qadir, G., Hayat, R., Shaheen, F. A., and Raza, M. A. (2019). Innovative processes and technologies for nutrient recovery from wastes: a comprehensive review. *Sustainability* **11**, 4938.

Ali, U., Sajid, N., Khalid, A., Riaz, L., Rabbani, M. M., Syed, J. H., and Malik, R. N. (2015). A review on vermicomposting of organic wastes. *Environmental Progress & Sustainable Energy* **34**, 1050-1062.

Amlinger, F., Peyr, S., Geszti, J., Dreher, P., Karlheinz, W., and Nortcliff, S. (2007). Beneficial effects of compost application on fertility and productivity of soils. Literature Study, Federal Ministry for Agriculture and Forestry, Environment and Water Management, Austria. *Online [www. umweltnet. at/filemanger/download/20558]*.

Amoah, I. D., Reddy, P., Seidu, R., and Stenström, T. A. (2018). Removal of helminth eggs by centralized and decentralized wastewater treatment plants in South Africa and Lesotho: health implications for direct and indirect exposure to the effluents. *Environmental Science and Pollution Research* **25**, 12883-12895.

Arancon, N., Cleave, J. V., Hamasaki, R., Nagata, K., and Felts, J. (2020). The influence of vermicompost water extracts on growth of plants propagated by cuttings. *Journal of Plant Nutrition* **43**, 176-185.

Bame, I., Hughes, J., Titshall, L., and Buckley, C. (2013). Leachate characteristics as influenced by application of anaerobic baffled reactor effluent to three soils: a soil column study. *Chemosphere* **93**, 2171-2179.

Bazrafshan, E., Zarei, A., Mostafapour, F. K., Poormollae, N., Mahmoodi, S., and Zazouli, M. A. (2016). Maturity and stability evaluation of composted municipal solid wastes. *Health scope* **5**.

Bekbolet, M., Uyguner, C. S., Selcuk, H., Rizzo, L., Nikolaou, A., Meric, S., and Belgiorno, V. (2005). Application of oxidative removal of NOM to drinking water and formation of disinfection by-products. *Desalination* **176**, 155-166.

Bishoge, O. K. (2021). Challenges facing sustainable water supply, sanitation and hygiene achievement in urban areas in sub-Saharan Africa. *Local Environment* **26**, 893-907.

Boldrin, A., Andersen, J. K., Møller, J., Christensen, T. H., and Favoino, E. (2009). Composting and compost utilization: accounting of greenhouse gases and global warming contributions. *Waste Management & Research* **27**, 800-812.

Cai, R., Cui, X., Cao, X., Jiang, X., Wang, R., and Xu, C. (2023). Crushing combined with high–frequency turning can promote material degradation of sheep manure compost on the Qinghai–Tibet Plateau by improving the microbial metabolic function. *Journal of Environmental Chemical Engineering* **11**, 109535.

Cao, H., Xie, Y., Wang, Y., and Xiao, J. (2022). "Advanced Ozonation Processes for Water and Wastewater Treatment: Active Catalysts and Combined Technologies," *Royal Society of Chemistry*.

Chaggu, E. J. (2004). "Sustainable environmental protection using modified pit-latrines," Wageningen University and Research.

Chen, L., de Haro Marti, M., Moore, A., and Falen, C. (2011). The composting process. *Dairy Manure Compost Production and Use in Idaho* **2**, 513-532.

Chen, Z., Li, Y., Ye, C., He, X., and Zhang, S. (2021). Fate of antibiotics and antibiotic resistance genes during aerobic co-composting of food waste with sewage sludge. *Science of The Total Environment* **784**, 146950.

Chen, Z., Zhang, S., Wen, Q., and Zheng, J. (2015). Effect of aeration rate on composting of penicillin mycelial dreg. *Journal of Environmental Sciences* **37**, 172-178.

Cieślik, B. M., Namieśnik, J., and Konieczka, P. (2015). Review of sewage sludge management: standards, regulations and analytical methods. *Journal of Cleaner Production* **90**, 1-15.

Darimani, H. S., Ito, R., Maiga, Y., Sou, M., Funamizu, N., and Maiga, A. H. (2016). Effect of post-treatment conditions on the inactivation of helminth eggs (*Ascaris suum*) after the composting process. *Environmental Technology* **37**, 920-928.

Das, S. K., Ghosh, G. K., and Avasthe, R. (2020). Application of biochar in agriculture and environment, and its safety issues. *Biomass Conversion and Biorefinery*, 1-11.

Deacon, J. (2002). The microbial world, thermophilic microorganisms. *Biology teaching organization, University of Edinborough, http://helios. bto. ed. ac. uk/bto/microbes/thermo. htm*.

Diaz, L. F., De Bertoldi, M., and Bidlingmaier, W. (2011). "Compost science and technology," Elsevier. https:// www.elsevier.com/books/compost-science-and-technology/diaz/ 978-0-08-043960-0

Domínguez, J., Aira, M., and Gómez-Brandón, M. (2010). Vermicomposting: earthworms enhance the work of microbes. *Microbes at work: from wastes to resources*, 93-114.

dos Santos Pereira, M., Borges, A. C., Heleno, F. F., Faroni, L. R. D. A., and da Silva, J. C. G. E. (2018). Experimental design optimization of dairy wastewater ozonation treatment. *Water, Air, & Soil Pollution* **229**, 1-11.

Edwards, C. A. (1988). Breakdown of animal, vegetable and industrial organic wastes by earthworms. *Earthworms in Waste and Environmental Management/edited by Clive A. Edwards and Edward F. Neuhauser*.

FAO (2017). World Fertilizer Trends and Outlooks for 2020. https://openknowledge.fao.org/handle/20.500.14283/i4324e

Finore, I., Feola, A., Russo, L., Cattaneo, A., Di Donato, P., Nicolaus, B., Poli, A., and Romano, I. (2023). Thermophilic bacteria and their thermozymes in composting processes: a review. *Chemical and Biological Technologies in Agriculture* **10**, 7.

Getahun, T., Nigusie, A., Entele, T., Van Gerven, T., and Van der Bruggen, B. (2012). Effect of turning frequencies on composting biodegradable municipal solid waste quality. *Resources, Conservation and Recycling* **65**, 79-84.

Giagnoni, L., Martellini, T., Scodellini, R., Cincinelli, A., and Renella, G. (2020). Co-composting: An opportunity to produce compost with designated tailor-made properties. *Organic Waste Composting through Nexus Thinking: Practices, Policies, and Trends*, 185-211.

Golbaz, S., Zamanzadeh, M. Z., Pasalari, H., and Farzadkia, M. (2021). Assessment of co-composting of sewage sludge, woodchips, and sawdust: feedstock quality and design and compilation of computational model. *Environmental Science and Pollution Research* **28**, 12414-12427.

Gómez-López, M. D., El Bied, O., Beltrá, J. C., Yanardağ, İ. H., Gómez, C., Faz, Á., and Zornoza, R. (2022). Strategies for the Sustainable Management of the Organic Fraction of Municipal Waste. *Applied Sciences* **12**, 9400.

Grau, F., Drechsel, N., Haering, V., Trautz, D., Weerakkody, W. J. S. K., Drechsel, P., Marschner, B., Dissanayake, D. M. P. S., and Sinnathamby, V. (2017). Impact of fecal sludge and municipal solid waste co-compost on crop growth of *Raphanus Sativus L.* and *Capsicum Anuum L.* under stress conditions. *Resources* **6**, 26.

Grau, M. G., Rhoton, S. L., Brouckaert, C. J., and Buckley, C. A. (2013). Development of a fully automated struvite reactor to recover phosphorus from source-separated urine collected at urine diversion toilets in eThekwini. *Proceeding of WEF/IWA Nutrient Removal and Recovery.*

Grgas, D., Štefanac, T., Barešić, M., Toromanović, M., Ibrahimpašić, J., Vukušić Pavičić, T., Habuda-Stanić, M., Herceg, Z., and Landeka Dragičević, T. (2023). Co-composting of Sewage Sludge, Green Waste, and Food Waste. *Journal of Sustainable Development of Energy, Water and Environment Systems* **11**, 1-14.

Guo, R., Li, G., Jiang, T., Schuchardt, F., Chen, T., Zhao, Y., and Shen, Y. (2012). Effect of aeration rate, C/N ratio and moisture content on the stability and maturity of compost. *Bioresource technology* **112**, 171-178.

Guzha, E., Nhapi, I., and Rockstrom, J. (2005). An assessment of the effect of human faeces and urine on maize production and water productivity. *Physics and Chemistry of the Earth, Parts A/B/C* **30**, 840-845.

Han, Z., Sun, D., Wang, H., Li, R., Bao, Z., and Qi, F. (2018). Effects of ambient temperature and aeration frequency on emissions of ammonia and greenhouse gases from a sewage sludge aerobic composting plant. *Bioresource technology* **270**, 457-466.

Harder, R., Wielemaker, R., Larsen, T. A., Zeeman, G., and Öberg, G. (2019). Recycling nutrients contained in human excreta to agriculture: Pathways, processes, and products. *Critical reviews in environmental science and technology* **49**, 695-743.

Harrison, J., and Wilson, D. (2012). Towards sustainable pit latrine management through LaDePa. *Sustainable Sanitation Practice* **13**, 25-32.

Hidalgo, D., Corona, F., and Martín-Marroquín, J. (2021). Nutrient recycling: from waste to crop. *Biomass Conversion and Biorefinery* **11**, 207-217.

Huang, D., Liu, L., Zeng, G., Xu, P., Huang, C., Deng, L., Wang, R., and Wan, J. (2017). The effects of rice straw biochar on indigenous microbial community and enzymes activity in heavy metal-contaminated sediment. *Chemosphere* **174**, 545-553.

Hwang, H. Y., Kim, S. H., Kim, M. S., Park, S. J., and Lee, C. H. (2020). Co-composting of chicken manure with organic wastes: characterization of gases emissions and compost quality. *Applied biological chemistry* **63**, 1-10.

Ibrahim, H. A.-H. (2020). Introductory chapter: pyrolysis. *Recent Advances in Pyrolysis* **1**.

Kalamdhad, A. S., and Kazmi, A. A. (2009). Effects of turning frequency on compost stability and some chemical characteristics in a rotary drum composter. *Chemosphere* **74**, 1327-1334.

Kaza, S., Yao, L., Bhada-Tata, P., and Van Woerden, F. (2018). "What a waste 2.0: a global snapshot of solid waste management to 2050," World Bank Publications.

Kebibeche, H., Khelil, O., Kacem, M., and Harche, M. K. (2019). Addition of wood sawdust during the co-composting of sewage sludge and wheat straw influences seeds germination. *Ecotoxicology and environmental safety* **168**, 423-430.

Khan, K. Y., Ali, B., Cui, X., Feng, Y., Yang, X., and Stoffella, P. J. (2017). Impact of different feedstocks derived biochar amendment with cadmium low uptake affinity cultivar of pak choi (*Brassica rapa ssb. chinensis L.*) on phytoavoidation of Cd to reduce potential dietary toxicity. *Ecotoxicology and environmental safety* **141**, 129-138.

Koné, D., Cofie, O., Zurbrügg, C., Gallizzi, K., Moser, D., Drescher, S., and Strauss, M. (2007). Helminth eggs inactivation efficiency by faecal sludge dewatering and co-composting in tropical climates. *Water research* **41**, 4397-4402.

Krueger, B. C., Fowler, G. D., Templeton, M. R., and Moya, B. (2020). Resource recovery and biochar characteristics from full-scale faecal sludge treatment and co-treatment with agricultural waste. *Water Research* **169**, 115253.

Lau, A. K., Lo, K., Liao, P., and Yu, J. (1992). Aeration experiments for swine waste composting. *Bioresource Technology* **41**, 145-152.

Lee, K. E. (1985). "Earthworms: their ecology and relationships with soils and land use," https://www.cabidigitallibrary.org/action/doSearch?do=Earthworms%3A+their+ecology+and+relationshipswith+soils+and+land+use.

Lehmann, J., and Joseph, S. (2015). "Biochar for environmental management: science, technology and implementation," Routledge.

Li, B., Boiarkina, I., Yu, W., Huang, H. M., Munir, T., Wang, G. Q., and Young, B. R. (2019). Phosphorous recovery through struvite crystallization: Challenges for future design. *Science of the total Environment* **648**, 1244-1256.

Liang, C., Das, K., and McClendon, R. (2003). The influence of temperature and moisture contents regimes on the aerobic microbial activity of a biosolids composting blend. *Bioresource technology* **86**, 131-137.

Liu, D., Zhang, R., Wu, H., Xu, D., Tang, Z., Yu, G., Xu, Z., and Shen, Q. (2011). Changes in biochemical and microbiological parameters during the period of rapid composting of dairy manure with rice chaff. *Bioresource technology* **102**, 9040-9049.

Lourenço, N., and Nunes, L. (2020). Review of dry and wet decentralized sanitation technologies for rural areas: applicability, challenges and opportunities. *Environmental Management* **65**, 642-664.

Lv, B., Zhang, D., Cui, Y., and Yin, F. (2018). Effects of C/N ratio and earthworms on greenhouse gas emissions during vermicomposting of sewage sludge. *Bioresource technology* **268**, 408-414.

Malinauskaite, J., Jouhara, H., Czajczyńska, D., Stanchev, P., Katsou, E., Rostkowski, P., Thorne, R. J., Colon, J., Ponsá, S., and Al-Mansour, F. (2017). Municipal solid waste management and waste-to-energy in the context of a circular economy and energy recycling in Europe. *Energy* **141**, 2013-2044.

Manga, M., Camargo-Valero, M., Anthonj, C., and Evans, B. (2021). Fate of faecal pathogen indicators during faecal sludge composting with different bulking agents in tropical climate. *International Journal of Hygiene and Environmental Health* **232**, 113670.

Manga, M., Evans, B. E., Ngasala, T. M., and Camargo-Valero, M. A. (2022). Recycling of Faecal Sludge: Nitrogen, Carbon and Organic Matter Transformation during Co-Composting of Faecal Sludge with Different Bulking Agents. *International Journal of Environmental Research and Public Health* **19**, 10592.

Manga, M., Muoghalu, C., Camargo-Valero, M. A., and Evans, B. E. (2023a). Effect of turning frequency on the survival of fecal indicator microorganisms during aerobic composting of fecal sludge with sawdust. *International Journal of Environmental Research and Public Health* **20**, 2668.

Manga, M., Muoghalu, C. C., and Acheng, P. O. (2023b). Inactivation of faecal pathogens during faecal sludge composting: a systematic review. *Environmental Technology Reviews* **12**, 150-174.

Mchunu, N., Odindo, A., and Muchaonyerwa, P. (2018). The effects of urine and urine-separated plant nutrient sources on growth and dry matter production of perennial ryegrass (*Lolium perenne. L*). *Agricultural Water Management* **207**, 37-43.

Mehta, C. M., Khunjar, W. O., Nguyen, V., Tait, S., and Batstone, D. J. (2015). Technologies to recover nutrients from waste streams: a critical review. *Critical Reviews in Environmental Science and Technology* **45**, 385-427.

Mertenat, A., Diener, S., and Zurbrügg, C. (2019). Black Soldier Fly biowaste treatment–Assessment of global warming potential. *Waste management* **84**, 173-181.

Mishra, S., Tiwary, D., Ohri, A., and Agnihotri, A. K. (2019). Impact of Municipal Solid Waste Landfill leachate on groundwater quality in Varanasi, India. *Groundwater for Sustainable Development* **9**, 100230.

Mori, T., Narita, A., Amimoto, T., and Chino, M. (1981). Composting of municipal sewage sludge mixed with rice hulls. *Soil Science and Plant Nutrition* **27**, 477-486.

Musazura, W. (2018). Crop fertigation (nitrogen and phosphorus) with decentralised wastewater treatrment system effluents and effects on soil and groundwater, University of KwaZulu-Natal, South Africa.

Musazura, W., and Odindo, A. O. (2021). Suitability of the Decentralised Wastewater Treatment effluent for agricultural use: Decision Support System approach. *Water* **13**, 2454.

Musazura, W., Odindo, A. O., Bame, I., and Tesfamariam, E. H. (2015). Effect of irrigation with anaerobic baffled reactor effluent on Swiss chard (*Beta vulgaris cicla*.) yield, nutrient uptake and leaching. *Journal of Water Reuse and Desalination* **5**, 592-609.

Nanda, S., and Berruti, F. (2021). Municipal solid waste management and landfilling technologies: a review. *Environmental Chemistry Letters* **19**, 1433-1456.

Nartey, E. G., Amoah, P., Ofosu-Budu, G. K., Muspratt, A., and Pradhan, S. K. (2017). Effects of co-composting of faecal sludge and agricultural wastes on tomato transplant and growth. *International journal of recycling of organic waste in agriculture* **6**, 23-36.

Nemet, F., Perić, K., and Lončarić, Z. (2021). Microbiological activities in the composting process–A review. *COLUMELLA–Journal of Agricultural and Environmental Sciences* **8**, 41-53.

Nikiema, J., and Cofie, O. O. (2014). Technological options for safe resource recovery from fecal sludge. *Resource Recovery and Reuse Series*.

Nkomo, N. (2021). Assessing the fertiliser value of co-composted biochar compost made from black soldier fly larvae faecal residue. University of KwaZulu-Natal, South Africa.

Nkomo, N., Odindo, A. O., Musazura, W., and Missengue, R. (2021). Optimising pyrolysis conditions for high-quality biochar production using black soldier fly larvae faecal-derived residue as feedstock. *Heliyon* **7**, e07025.

Ofei-Quartey, M. N. L., Appiah-Effah, E., Akodwaa-Boadi, K., Ampaw, B., Taylor, T. S., and Millogo, Z. E. N. (2023). Enhancing the economic potential of organic waste by co-composting using ratio modelling toward a circular economy. *Journal of Material Cycles and Waste Management* **25**, 1560-1580.

Okalebo, J. R., Gathua, K. W., and Woomer, P. L. (2002). Laboratory methods of soil and plant analysis: a working manual second edition. *Sacred Africa, Nairobi* **21**, 25-26.

Oturan, M. A., and Aaron, J.-J. (2014). Advanced oxidation processes in water/wastewater treatment: principles and applications. A review. *Critical reviews in environmental science and technology* **44**, 2577-2641.

Pereira, R., Cardoso, E. J. B. N., Oliveira, F., Estrada-Bonilla, G., and Cerri, C. E. P. (2018). A novel way of assessing C dynamics during urban organic waste composting and greenhouse gas emissions in tropical region. *Bioresource Technology Reports* **3**, 35-42.

Phoku, S. T. (2020). Photosynthetic efficiency of hydroponically-grown tomato plants to decentralized wastewater treatment systems (DEWATS) effluents and nitrified urine concentrate (NUC) as nutrient sources. *Acta horticulturae*, pp. 315-320-2020 no.1271.

Popat, S. C., Yates, M. V., and Deshusses, M. A. (2010). Kinetics of inactivation of indicator pathogens during thermophilic anaerobic digestion. *water research* **44**, 5965-5972.

Rahman, N., Bruun, T. B., Giller, K. E., Magid, J., van de Ven, G. W., and de Neergaard, A. (2019). Soil greenhouse gas emissions from inorganic fertilizers and recycled oil palm waste products from Indonesian oil palm plantations. *Gcb Bioenergy* **11**, 1056-1074.

Rasul, F., Gull, U., Rahman, M., Hussain, Q., Chaudhary, H. J., Matloob, A., Shahzad, S., Iqbal, S., Shelia, V., and Masood, S. (2016). Biochar an emerging technology for climate change mitigation. *J Environ Agric Sci* **9**, 37-43.

Reyes-Torres, M., Oviedo-Ocaña, E., Dominguez, I., Komilis, D., and Sánchez, A. (2018). A systematic review on the composting of green waste: Feedstock quality and optimization strategies. *Waste management* **77**, 486-499.

Ribeiro, P. H., Faroni, L. R. D. A., Silva, G. J. d., Heleno, F. F., Cecon, P. R., de Alencar, E. R., and Borges, A. C. (2024). Ozonation with Hydrogen Peroxide for Treating Wastewater from Industrial Potato Processing-a Preliminary Investigation. *Ozone: Science & Engineering* **46**, 53-63.

Roberts, K. G., Gloy, B. A., Joseph, S., Scott, N. R., and Lehmann, J. (2010). Life cycle assessment of biochar systems: estimating the energetic, economic, and climate change potential. *Environmental science & technology* **44**, 827-833.

Rose, C., Parker, A., Jefferson, B., and Cartmell, E. (2015). The characterization of feces and urine: a review of the literature to inform advanced treatment technology. *Critical reviews in environmental science and technology* **45**, 1827-1879.

Rossi, G., Mainardis, M., Aneggi, E., Weavers, L. K., and Goi, D. (2021). Combined ultrasound-ozone treatment for reutilization of primary effluent—a preliminary study. *Environmental Science and Pollution Research* **28**, 700-710.

Ruggero, F., Gori, R., and Lubello, C. (2019). Methodologies to assess biodegradation of bioplastics during aerobic composting and anaerobic digestion: A review. *Waste Management & Research* **37**, 959-975.

Saha, D., Fakir, O., Mondal, S., and Ghosh, R. (2017). Effects of organic and inorganic fertilizers on tomato production in saline soil of Bangladesh. *J. Sylhet Agril. Univ* **4**, 213-220.

Said-Pullicino, D., Erriquens, F. G., and Gigliotti, G. (2007). Changes in the chemical characteristics of water-extractable organic matter during composting and their influence on compost stability and maturity. *Bioresource technology* **98**, 1822-1831.

Saidou, H., Ben Moussa, S., and Ben Amor, M. (2009). Influence of airflow rate and substrate nature on heterogeneous struvite precipitation. *Environmental technology* **30**, 75-83.

Sanguinetti, G., Tortul, C., Garcia, M., Ferrer, V., Montangero, A., and Strauss, M. (2005). Investigating helminth eggs and Salmonella sp. in stabilization ponds treating septage. *Water Science and Technology* **51**, 239-247.

Sasse, L. (1998). "DEWATS: Decentralised wastewater treatment in developing countries," BORDA, Bremen Overseas Research and Development Association.

Septien, S., Singh, A., Mirara, S., Teba, L., Velkushanova, K., and Buckley, C. (2018). 'LaDePa'process for the drying and pasteurization of faecal sludge from VIP latrines using infrared radiation. *South African journal of chemical engineering* **25**, 147-158.

Shah, G., Shah, G., Groot, J., Oenema, O., Raza, A., and Lantinga, E. (2016). Effect of storage conditions on losses and crop utilization of nitrogen from solid cattle manure. *The Journal of Agricultural Science* **154**, 58-71.

Shanmugam, V., Sreenivasan, S., Mensah, R. A., Försth, M., Sas, G., Hedenqvist, M. S., Neisiany, R. E., Tu, Y., and Das, O. (2022). A review on combustion and mechanical behaviour of pyrolysis biochar. *Materials Today Communications*, 103629.

Sheets, J. P., Yang, L., Ge, X., Wang, Z., and Li, Y. (2015). Beyond land application: Emerging technologies for the treatment and reuse of anaerobically digested agricultural and food waste. *Waste Management* **44**, 94-115.

Shen, Y. J., Hou, Y. Q., Zhao, L. X., and Meng, H. B. (2015). Influence of Turning on Physicochemical Properties and Odor Emission during Composting. *Advanced Materials Research* **1092**, 867-873.

Singh, S., Ibrahim, M. A., Pawar, S., and Brdjanovic, D. (2022). Public Perceptions of Reuse of Faecal Sludge Co-Compost in Bhubaneswar, India. *Sustainability* **14**, 4489.

Singh, S., and Sinha, R. K. (2022). Vermicomposting of organic wastes by earthworms: Making wealth from waste by converting 'garbage into gold'for farmers. *In* "Advanced Organic Waste Management", pp. 93-120. Elsevier.

Soto-Paz, J., Oviedo-Ocaña, E. R., Manyoma, P. C., Marmolejo-Rebellón, L. F., Torres-Lozada, P., Barrena, R., Sánchez, A., and Komilis, D. (2020). Influence of mixing ratio and turning frequency on the co-composting of biowaste with sugarcane filter cake: A mixture experimental design. *Waste and Biomass Valorization* **11**, 2475-2489.

Sri Shalini, S., Palanivelu, K., Ramachandran, A., and Raghavan, V. (2021). Biochar from biomass waste as a renewable carbon material for climate change mitigation in reducing greenhouse gas emissions—A review. *Biomass Conversion and Biorefinery* **11**, 2247-2267.

Stokke, O. S., and Thommessen, O. B. (2013). United Nations Framework Convention Climate Change (UNFCCC). *In* "Yearbook of International Cooperation on Environment and Development 2001-02", pp. 87-93. Routledge.

Swan, K. H., Surinkul, N., and Visvanathan, C. (2023). Investigation of Decentralized Wastewater Treatment System in Urban Wastewater Management: Case Study in Yangon, Myanmar. *Sustainability* **15**, 16756.

Szabová, P., Hencelová, K., Sameliaková, Z., Marcová, T., Staňová, A. V., Grabicová, K., and Bodík, I. (2020). Ozonation: effective way for removal of pharmaceuticals from wastewater. *Monatshefte für Chemie-Chemical Monthly* **151**, 685-691.

Szántó, G. L., Letema, S., Tukahirwa, J., Mgana, S., Oosterveer, P., and Van Buuren, J. (2012). Analyzing sanitation characteristics in the urban slums of East Africa. *Water Policy* **14**, 613-624.

Tesfamariam, E., Musazura, W., Odindo, A., and Buckley, C. (2018). Decentralised wastewater treatment effluent fertigation: preliminary technical assessment. *Water SA* **44**, 250-257.

Thye, Y. P., Templeton, M. R., and Ali, M. (2011). A critical review of technologies for pit latrine emptying in developing countries. *Critical Reviews in Environmental Science and Technology* **41**, 1793-1819.

Tilley, E., Ulrich, L., Lüthi, C., Reymond, P., and Zurbrügg, C. (2014). Compendium of sanitation systems and technologies.

Trautmann, N. M., and Krasny, M. E. (1998). "Composting in the classroom: Scientific inquiry for high school students," Kendall/Hunt Publishing Company.

Udert, K. M., Buckley, C. A., Wächter, M., McArdell, C. S., Kohn, T., Strande, L., Zöllig, H., Fumasoli, A., Oberson, A., and Etter, B. (2015). Technologies for the treatment of source-separated urine in the eThekwini Municipality. *Water Sa* **41**, 212-221.

USEPA (1995)." Standards for the Use or Disposal of Sewage Sludge.". *Code of Federal Regulations*.

Wang, J., Hu, Z., Xu, X., Jiang, X., Zheng, B., Liu, X., Pan, X., and Kardol, P. (2014). Emissions of ammonia and greenhouse gases during combined pre-composting and vermicomposting of duck manure. *Waste management* **34**, 1546-1552.

WHO (2006). "World Health Organisation guidelines for the safe use of wasterwater excreta and greywater," World Health Organization.

Wichuk, K. M., and McCartney, D. (2007). A review of the effectiveness of current time–temperature regulations on pathogen inactivation during composting. *Journal of Environmental Engineering and Science* **6**, 573-586.

Wilde, B. C., Lieberherr, E., Okem, A. E., and Six, J. (2019). Nitrified human urine as a sustainable and socially acceptable fertilizer: an analysis of consumer acceptance in Msunduzi, South Africa. *Sustainability* **11**, 2456.

Wu, J., Zhao, Y., Qi, H., Zhao, X., Yang, T., Du, Y., Zhang, H., and Wei, Z. (2017). Identifying the key factors that affect the formation of humic substance during different materials composting. *Bioresource technology* **244**, 1193-1196.

Xie, M., Shon, H. K., Gray, S. 2., and Elimelech, M. (2016). Membrane-based processes for wastewater nutrient recovery: Technology, challenges, and future direction. *Water research* **89**, 210-221.

Zhang, D., Luo, W., Li, Y., Wang, G., and Li, G. (2018). Performance of co-composting sewage sludge and organic fraction of municipal solid waste at different proportions. *Bioresource technology* **250**, 853-859.

Zhang, J., Sui, Q., Li, K., Chen, M., Tong, J., Qi, L., and Wei, Y. (2017). Influence of natural zeolite and nitrification inhibitor on organics degradation and nitrogen transformation during sludge composting. *Environmental Science and Pollution Research* **24**, 9122-9122.

Zhang, J., Ying, Y., and Yao, X. (2019). Effects of turning frequency on the nutrients of Camellia oleifera shell co-compost with goat dung and evaluation of co-compost maturity. *Plos one* **14**, e0222841.

Zhao, W., Van Der Voet, E., Zhang, Y., and Huppes, G. (2009). Life cycle assessment of municipal solid waste management with regard to greenhouse gas emissions: case study of Tianjin, China. *Science of the total environment* **407**, 1517-1526.

Zhu-Barker, X., Bailey, S. K., Burger, M., and Horwath, W. R. (2017). Greenhouse gas emissions from green waste composting windrow. *Waste management* **59**, 70-79.

Zuma, L., Velkushanova, K., and Buckley, C. (2015). Chemical and thermal properties of VIP latrine sludge. *Water Sa* **41**, 534-540.

Chapter 3: The Effect of Turning Frequency and Mixing Ratios on Co-compost Produced Using Dewatered Sewage Sludge and Organic Green Waste

Abstract

Wastewater treatment plants (WWTP) produce large volumes of sewage sludge that have significant disposal challenges. Thus, implying the need to find ways to safely manage and dispose of sewage sludge. The application of sewage sludge to arable land is commonly practised however it poses health risks as pathogens and heavy metals infest the soil. Therefore, this study investigated the optimum mixing ratios (MR) and turning frequencies (TF) to produce a safe and high nutritive value co-compost when using dewatered sewage sludge (DSS) and organic green waste (OGW) as feedstocks. A 2-level of MR (1DSS:2OGW and 1DSS:3OGW) × 3 levels of TF (7-, 14- and 28-day turning interval) factorial experiment was laid out using a completely randomized design (CRD) replicated thrice totalling 18 experimental units (1 m^3 wooden composting boxes). The final co-compost produced had a neutral pH ranging between 7.05 to 7.56. The highest temperature reached was 54.6 °C, attained by 1DSS:3OGW (28TF) compared to other treatments which also failed to reach the recommended 55°C; but was still high enough for helminth egg deactivation. Moreover, *Escherichia coli* (*E. coli*) and *Salmonella enterica* (*Salmonella*) populations were absent in 1DSS:3OGW (7TF) and 1DSS:2OGW (28TF) however they were <30 CFU g^{-1} (colony forming units per gram) in all other treatments. There was a 40% compost volume reduction in the frequently (7TF) turned co-compost in both mixing ratios. All treatments had heavy metal concentrations within European and South African standards and germination index above 80% confirming that the co-compost was safe for use in agricultural fields. In conclusion, this study has demonstrated that DSS can be safely managed via co-composting thus contributing towards an eco-friendly and circular agricultural economy.

Keywords: Co-compost, circular economy, *Salmonella*, *E. coli*, temperature, turning frequencies, waste management

3.1 Introduction

The global population is growing exponentially and is expected to reach 9.6 billion by 2050 (Gerland et al., 2014), thus increasing urbanization by two-thirds (United Nations, 2023). In this context, South African urbanization was approximately 68.34% in 2022 and is forecasted to reach 70% by 2030 (STATS_SA, 2022). This rapid movement to urban settlements has subsequently resulted in the production of large quantities of sewage sludge, organic waste generation, and food insecurity among urban settlers. For instance, sewage sludge is treated or dewatered at wastewater treatment plants (WWTP) to reduce viruses, bacteria, and organic pollutants (Dong et al., 2014).

However, the sewage effluent and dewatered sewage sludge (DSS), which are byproducts of sewage sludge treatment pose significant environmental challenges if not properly managed and disposed of. According to Mainstone and Parr, (2002), sewage sludge causes eutrophication if discharged into rivers, lakes, and dams, thus negatively impacting the riverine ecosystem. This waste significantly causes environmental pollution and attracts disease vectors that pose risks to human health if discharged into the environment or stockpiled in the WWTP. Therefore, these issues call for municipalities and governments to come up with alternative ways of managing waste in landfills and WWTP while carefully ensuring food security among residents.

Co-composting presents an economically feasible way of treating sewage sludge for safe use and preventing its improper disposal (Wong et al., 2011). Co-composting refers to the simultaneous composting of different types of organic wastes, which can add value or improve compost quality (Zhang and Sun, 2014). Furthermore, it enables the recovery of nutrients from faeces, urine, and detergents, allowing their safe reintegration into the soil (Manga et al., 2022). Furthermore, it can convert organic wastes into phytotoxic and pathogen-free compost that can be used to improve soils and plants (Bernal et al., 2009).

However, sewage sludge cannot be composted alone as it has a low Carbon-Nitrogen (C/N) ratio (<20:1), high moisture content, and is highly compacted, limiting oxygen circulation (Banegas et al., 2007). Therefore, organic materials including sawdust (Banegas et al., 2007; Manga et al., 2023a), municipal organic waste (Ofei-Quartey et al., 2023; Zhang et al., 2018), coffee husks (Manga et al., 2021), wood chips (Zhao et al., 2015), landscaping waste (Dzulkurnain et al., 2017), agro-industrial waste (Gutiérrez et al., 2017) and wetland plant material (Kwarciak-Kozłowska, 2019) had previously been used to correct the C/N ratio of sewage/faecal sludge composting or specifically "co-composting". This was usually made efficient with turning frequency and varying the C/N ratio.

Turning frequency (TF) is one of the most important factors or conditions influencing compost quality in aerobic composting (Nakasaki et al., 2009). Adequate aeration or turning can remove excess moisture and CO_2, regulate temperature, and influence O_2 circulation for biological processes (Trautmann and Krasny, 1998). Turning frequency affects the degradation or decomposition rate and the final compost's quality (Ogunwande et al., 2008). Moreover, scientific literature has documented that turning frequency positively correlates with physiochemical properties and indicators of compost maturity (CCQC, 2001). Yuan et al. (2016) found that aeration rate influenced compost stability (pathogen reduction) but did not statistically affect pH and germination index, which are the indicators of quality compost. Moreover, Rasapoor et al. (2009) found that low and medium turning frequencies significantly affected nitrogen, C/N ratio, and thermophilic temperatures while higher TF showed improved or higher electrical conductivity (EC) values.

Interestingly, Manga et al. (2023a) found that higher TF (3-day interval) significantly led to pathogen inactivation over 8 weeks, while moderate TF (7-day interval) pathogens were inactivated after 10 weeks. Moreover, Manga et al. (2023a) suggested that even at low temperatures, pathogens can be inactivated, given that optimum and high TF is employed. Even though high turning frequency improves the maturity, stability, and quality of the co-compost, it can result in high loss of organic matter (OM), organic carbon (OC), and nitrogen losses (Manga et al., 2023a). Moreover, it can result in CO_2 production leading to greenhouse gas emissions. Therefore, there is a need to optimize the mixing ratio and turning frequency which will result in high-quality co-compost. Therefore, the primary objective of this study was to determine the optimal turning frequency for co-composting DSS and OGW to produce high-quality compost while minimizing nutrient and OM losses. This study also evaluated the effect of different DSS and OGW mixing ratios on compost quality, concerning maturity, stability, and nutrient content.

3.2 Materials and methods

3.2.1 Dewatered sewage sludge and organic green waste collection and preparation

The Darville Wastewater Treatment Site in Pietermaritzburg, South Africa, receives approximately 65 megalitres (ML/day) of wastewater per day, including sewage sludge. In the primary stage of wastewater treatment plants, excess sludge generated from sewage treatment undergoes dewatering and subsequent disposal (Hansen, 2015). Therefore, in this study, dewatered sewage sludge (DSS) with a moisture content of 84.22 ± 0.38% was collected from the dumpsite (sludge landfill) in Hollingwood Road, Hayfields, Pietermaritzburg, South Africa (29°60′8″S and 30°44′5″E). The DSS was transported to the point of use in 100 litre (L) buckets by the Sani Squad (Pty/Ltd) company. Organic green waste, including but not limited to garden waste, grass clippings, and pruned tree fellings, is often dumped as waste at Duzi-turf Pietermaritzburg, South Africa (29°36'03.8"S and 30°26'00.9" E) where it can be chipped into 5-10 cm pieces and composted. In this study, chipped OGW was sourced from Duzi-turf using an 8-ton truck from the Isibani Industries (Pty/Ltd) company.

3.2.2 Study area and experimental design

The co-composting experiment was conducted at the University of Kwazulu-Natal, Pietermaritzburg campus at the Controlled Environment Facility (CEF) under controlled conditions (29°61′68″S and 30°39′42″E). The experiment was laid out in a Completely Randomized Design (CRD) in approximately 1 m^3 composting boxes made of wooden planks. The study was laid out as a 2 × 3 factorial design; where two levels of mixing ratio (1DSS:2OGW and 1DSS:3OGW) and three levels of TF, (7-day, 14-day and 28-day) were computed as shown in Table 3.1. The treatments were replicated thrice, resulting in 18 experimental units, which were co-composting bins (≈ 1 m^3) filled with DSS and OGW which were located in tunnel K at CEF. Dewatered sewage sludge was mixed with chipped organic green waste in the ratios explained in Table 3.1 manually using a garden shovel and garden fork on a volume-by-volume (v/v) basis by the Sani Squad company under the author's supervision. The low sludge mixing ratio (1DSS:3OGW) was designed to minimize heavy metal contamination in the final co-compost product. In contrast, the high sludge mixing ratio (1DSS:2OGW) was intended to enhance nutrient content while assessing the presence of heavy metals in the final product.

Table 3.1: Mixing ratio and turning frequency of different co-compost treatments and their descriptions and labels

Mixing ratios	TI (days)	Compost name	Description of the treatment
1DSS:3OGW	7	1:3 (7TF)	Low sludge proportion with high TF
1DSS:3OGW	14	1:3 (14TF)	Low sludge proportion with medium TF
1DSS:3OGW	28	1:3 (28TF)	Low sludge proportion with low TF
1DSS:2OGW	7	1:2 (7TF)	High sludge proportion with high TF
1DSS:2OGW	14	1:2 (14TF)	High sludge proportion with medium TF
1DSS:2OGW	28	1:2 (28TF)	High sludge proportion with low TF

Feedstock	N (%)	C (%)	C:N ratio
DSS	5.02	37.19	7.4:1
OGW	0.89	51.60	57.9:1
1DSS:3OGW	1.92	48.49	24.9:1
IDSS:2OGW	2.26	46.80	20.7:1

TI (turning interval), **DSS** (dewatered sewage sludge), **OGW** (organic green waste), **TF** (turning frequency), **N** (nitrogen), **C** (carbon), **C:N** ratio (carbon to nitrogen ratio)

3.2.3 Temperature monitoring and turning the co-composts

Temperature is the most important parameter used to determine the quality and stability of the co-compost (Karak et al., 2014). In the present study, the temperature was monitored daily using the iButtons Thermochron® DS1921G-F5# (I-Button Link technology, United States of America) which was inserted in the middle of the co-composting bin from the 9th of February 2024 to the 30th of May 2024. The iButtons were shipped to ETH Zurich to extract the temperature data using the iButtons® Log typer ™, HW Rev: Proto2 FW. 0.9, (I-Button Link technology, USA) temperature data logger. In the second month of co-composting from the 6th of March 2024, a 1-meter-long digital compost thermometer sourced from Soil Scopes, Cape Town, South Africa, was used to take daily temperature readings in conjunction with the iButtons. The temperature measurements were taken at 3 points in each co-composting bin. The average temperature of each bin was combined with the average temperatures of the other two bins that received the same treatment. The overall average temperature calculated from these three bins (based on nine data points) represented the temperature profile for each treatment.

Co-composts were turned using the MAC AFRIC 68 CC Earth Auger Drill (Adendorff Machinery Mart, South Africa). Turning interval differed across treatments as explained in Table 3.1. The 7TF were turned every week, 14TF were turned every two weeks and the 28TF were turned once a month. The temperature sensors (iButtons) were removed before turning the co-compost and immediately stored back after turning the co-compost.

3.2.4 *Escherichia coli and total coliforms quantification*

The culturable pathogen indicators were quantified using the dilution plate count technique, with results expressed as Colony Forming Units (CFU) per gram (g) of sample wet weight (CFU/g) (Han et al., 2016). To prepare the samples, 9 mL of distilled water was autoclaved at 121°C for 15 minutes and allowed to cool before adding 1 g of co-compost. Seven serial dilutions were then prepared following the methods of El Hayany et al. (2021) and Ruiz-Llacsahuanga et al. (2021). The compost-water mixture was vortexed to ensure homogeneity before transferring to Petri dishes. Various selective and differential media were prepared to isolate and quantify specific bacterial species. MacConkey agar (51 g/L distilled water) was used for the initial screening of *E. coli* and *Salmonella*. Eosin-methylene blue (EMB) agar (37.5 g/L distilled water) was used for *E. coli* quantification, while Brilliant Green Agar (50 g/L distilled water) was used for *Salmonella spp.* The *Pseudomonas aeruginosa* was isolated using Pseudomonas-Agar P Basis (44 g/L distilled water), and *Staphylococcus aureus* was quantified using Baird-Parker Agar Base (63 g/L distilled water). Total coliforms were quantified using nutrient agar (23 g/L distilled water).

Each medium was autoclaved at 121°C for 15 minutes, allowed to cool to room temperature, and then poured into Petri dishes. Approximately 1 mL of the vortexed co-compost serial dilution was transferred onto the prepared media. The plates were left to solidify before incubation at 37°C for 18 hours following the method by (Manga et al., 2023). After incubation, fully developed colonies were manually counted to determine CFU/g of the wet sample weight.

3.2.5 *Helminths eggs*

To quantify helminth eggs, 25 g of co-compost samples were added to a 100 ml plastic beaker and mixed with 50 ml diluted ammonium bicarbonate [NH_4HCO_3] (AMBIC). The suspension was sieved through a 100 µm mesh sieve placed on top of a 20 µm sieve. The retentate was thoroughly washed over the sieves using distilled H_2O, the retentate on the 100 µm was discarded, and that on the 20 µm mesh sieve was collected. A 3 ml plastic Pasteur pipette was used to collect the retentate into 15 ml plastic conical test tubes. Samples were centrifuged at 1512 g (3000 rpm) for 10 mins, and the supernatant was discarded. Sugar solution (at Specific Gravity of 1.3) was added to the pellet in 3 ml aliquots up to 14 ml while resuspending using a vortex mixer, and the samples were centrifuged at 672 g (2000 rpm) for 5 min. The supernatant was poured onto a 20 µm sieve and washed using clean tap water. The retentate was transferred to the test tubes and centrifuged at 1512 g for 10 min. Wet mount preps of the final pellets were made on glass slides, and all eggs were classified according to species and viability using a light microscope ZEISS Axioscope.A1-2015 (Carl Zeiss AG, Oberkochen, German) with Axiocam 105 colour camera.

3.2.6 Moisture content, organic matter, and organic carbon

Co-compost was sampled at 3 points in each composting bin at 30-70 cm from the surface of the co-compost. Approximately 10 g of the composite co-compost samples were pre-weighed and re-weighed after oven-drying at 105 °C for 24 hrs to determine moisture content (Trautmann and Krasny, 1998). The sample from the oven-dried co-compost sample was ashed in the muffle furnace at 550 °C for 8 hrs to determine the organic matter content (Manga et al., 2022). Organic carbon was derived from organic matter using Equation 3.3, as illustrated in composting guidelines (Trautmann and Krasny, 1998).

Moisture content was calculated using the formulae:

$$MC = \frac{M_{\omega} - M_d}{Md} \times 100 \qquad \text{Equation 3.1}$$

Where: **MC**= moisture content (%) of the co-compost, **Mw**= mass of wet co-compost sample **Md**= mass of dry co-compost sample

Organic matter was calculated using the method by (Trautmann and Krasny, 1998) with the formulae:

$$OM = \frac{M_d - M_a}{Md} \times 100 \qquad \text{Equation 3.2}$$

Where: **OM**= organic matter (%), **Md**= mass of dry at (105 °C), **Ma**= mass of co-compost samples ash after combustion

Organic carbon was calculated using the Equation 3.3:

$$OC = \frac{OM}{1.8} \qquad \text{Equation 3.3}$$

Where: **OC**= organic carbon (%) of the co-compost, **OM**= organic matter (%), **1.8** = standard number developed at the laboratories for organic carbon calculation

3.2.7 pH and electrical conductivity

Co-compost samples were randomly taken at 3 points in each co-composting bin using a 1-meter-long soil test auger. The samples were mixed to make a composite co-compost mixture. Approximately 10 g of each co-compost mixture was weighed using a D&T Electronic balance with an accuracy of 0.01 g LCD and 200 g load. After weighing, samples were transferred to plastic containers with lids, and 100 ml distilled water was added to make a water solute extract of 1:10 weight-by-volume (w/v) (Manga et al., 2022; Manga et al., 2023a). The water-solute was shaken using a reciprocal shaker KS 130 control at 275 rpm per minute for 60 mins. After an hour, the water-solute was filtered using No.1 Whatman filter paper with a diameter of 120 mm. The extract was used to test the co-compost's pH and electrical conductivity (EC). The EC was measured using an EC meter Thermo Scientific ORION STAR A215 calibrated with EC buffer before measurements. The pH was measured using the HANNA edge HI2020-01 pH meter, which was calibrated using buffers with pH 4, 7, and 10 before measurements. After measuring each sample's pH or EC, the probes were rinsed with distilled water and dried with a paper towel to avoid cross-contamination.

3.2.8 Heavy metals and nutrients analysis

Co-compost samples were sieved using a 5 mm sieve and then air-dried at room temperature 25 ± 2 °C. Approximately 0.5 g of the sieved co-compost was transferred into the 50 ml conical flasks. To analyse 18 samples of co-compost, 250 mL of aqua regia was prepared by mixing 62.5 mL of nitric acid (HNO_3) with 187.5 mL of hydrochloric acid (HCl) under fume hood conditions. Approximately 10 ml of each of the prepared aqua regia was used to digest 0.5 g co-compost samples under fume hood conditions. The solution was placed in a hot sand bath maintained at 90 °C and periodically agitated for 2 hrs to ensure complete digestion of the material. Heavy metals were quantified using Inductively Coupled Plasma Optical Emission Spectrometry (ICP-OES) at the Talbot (Pty) Ltd laboratories as per a study by (Mandizvo and Odindo, 2019).

3.2.9 Germination index

The water-soluble extract of 1:10 (w/v) was made by dissolving 10 g of composite co-compost in 100 ml of distilled water. Approximately 5 ml of the extract was sprinkled in the Petri dishes containing 90 mm No.1 Whatman Filter Papers and 10 seeds of radish (*Raphanus sativus*) seed species. The petri dishes were incubated at 27±1°C for 72 hours. Germination percentage was calculated by counting the number of germinated seeds over the total number of seeds incubated and the radicle length was measured using a 30 cm plastic ruler. The germination index was calculated using Equation 3.4 as per study by (Ofei-Quartey et al., 2023; Trautmann and Krasny, 1998).

$$GI = \frac{\%G \times \%L}{100} \qquad \text{Equation 3.4}$$

Where: **GI**= Germination index (%), **%G**= percent germination (mean germination of treatments over mean germination of control), **%L**= percent radical length (mean radical length of treatments over mean germination of control).

3.2.10 Plant growth index

Matured co-compost from different treatments and soil collected from KwaDinabakubo were sieved using a 5 mm sieve and 1 kg pots were used for the plant growth index experiment. Co-

compost was either mixed (50%) with soil or co-compost alone, using the soil alone as a control. The treatments were as follows: Duzi-turf co-compost, 1DSS:3OGW (7TF), 1DSS:2OGW (7TF), 1DSS:3OGW (14TF), 1DSS:2OGW (14TF), 1DSS:3OGW (28TF), 1DSS:2OGW (28TF) and KwaDinabakubo soil alone (control). Two mixing ratios of 50 and 100 % (co-compost to soil) were made, and the experiment was laid out in a completely randomized design that was replicated thrice. Three radish seeds were planted in each pot and each pot was irrigated with 150 ml of distilled water. Radish seedlings were oven-dried at 70 °C for 24 hrs and dry matter was measured and used to calculate the plant growth index as per a study by (Manga et al., 2022).

3.2.11 Data analysis

Analysis of variance for compost maturity and stability indicators was computed using GenStat 23rd Edition (VSN, International, Hemel, Hempstead, United Kingdom). Mean separation was performed using Fisher's protected least significant difference (LSD) test when treatments showed differences at a 5% level of significance. Significant different ($p<0.05$) treatments and their interactions, where applicable, were presented as line and bar graphs using means and standard deviation.

3.3 Results and discussion

3.3.1 pH evolution

The change in pH in the different co-compost treatments in this study is presented in Figure 3.1. Co-compost piles exhibited a higher pH in weeks 8, and 12, with the highest values in week 12. Ofei-Quartey et al. (2023) also found that the pH of the co-compost rapidly increased between weeks 9 and 12. There was a gradual decrease in pH as the co-composting progressed and by the end of the co-composting process in week 16, the pH was neither acidic nor alkaline, with the pH range of 7.05 to 7.56 in all co-composting treatments. The observations agree with those of Zorpas and Loizidou, (2008), who reported a final sewage sludge and sawdust co-compost product with a pH between 7.01 and 7.48. Moreover, Hemidat et al. (2018) emphasized that stable and matured co-composts should have pH values ranging between 6.5 and 9.0. However, Manga et al. (2022) found that the final pH of the co-compost decreased to 7.4, 6.3, and 6.5 for the faecal sludge co-composted with sawdust, coffee husks, and brewery waste, respectively. The added benefit of pH reduction is the reduction of ammonia (NH_3) volatilization during the co-composting process (Raviv et al., 2004). The decrease in pH values might be due to decomposition resulting in organic acid formation, mineralization, or ammonification during the co-composting process (Manga et al., 2022; Zorpas and Loizidou, 2008).

The pH levels often affect metabolic activities, availability of nutrients to micro-organisms, and solubility of heavy metals (El Hayany et al., 2021; Ofei-Quartey et al., 2023). Kazama and Otaki (2011) reported that a slightly alkaline pH of 8.5 or higher is optimal for *E. coli* deactivation. Low final pH in compost can increase the solubility of some anions (e.g., phosphate, sulfate, and nitrate) and enhance their availability. However, acidic conditions can also lead to the leaching of essential cations (e.g., calcium, magnesium, potassium), reducing their availability due to increased competition with hydrogen (H^+) and aluminium (Al^{3+}) ions. High pH tends to increase cation exchange capacity (CEC), leading to greater retention of cations like calcium (Ca^{2+}), magnesium (Mg^{2+}), and potassium (K^+). However, it can precipitate some anions, particularly phosphate, reducing their availability.

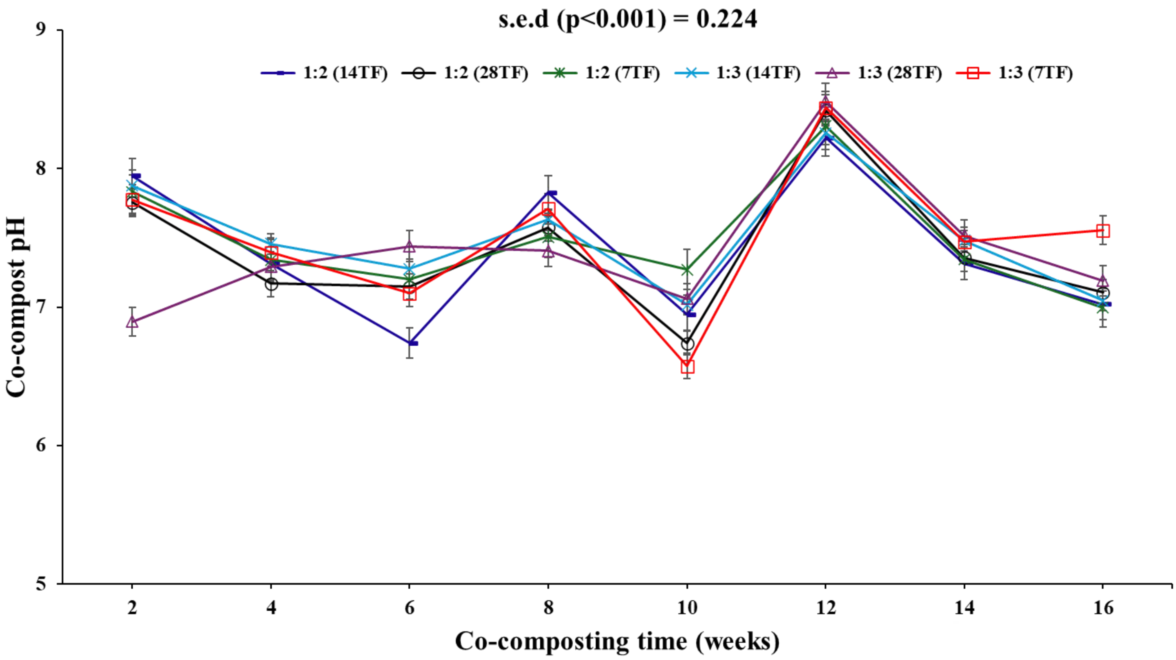


Figure 3.1: The pH evolution of co-compost made from dewatered sewage sludge and organic green waste co-compost over 16 weeks

3.3.2 Electrical conductivity

Electrical conductivity is an important measure of salinity in the co-compost as it also reflects the suitability of the co-compost for plant growth (Zorpas and Loizidou, 2008). According to Liu et al. (2011), EC is used to measure salinity and is an index for evaluating the co-compost quality, maturity, and stability. In this study, the electrical conductivity (EC) fluctuated between the treatments and the lowest EC of 0.54 mS/cm was observed in the 1:3 (14TF) and the highest EC of 1.33 mS/cm was observed in the 1:3 (7TF). The highest EC was obtained in week 12 of co-composting with a value of 3.26 mS/cm being obtained in the 1:2 (7TF). This value was higher than the recommended EC threshold value of 3.0 mS/cm (Richards, 1947). However, at the end of the co-composting process, the EC was below the threshold value for all treatments, with values ranging between 1.02 mS/cm to 1.55 mS/cm. These findings agree with the studies reported in the literature, where there are fluctuations in EC and an increase in EC at the end of the co-composting process (Manga et al., 2022).

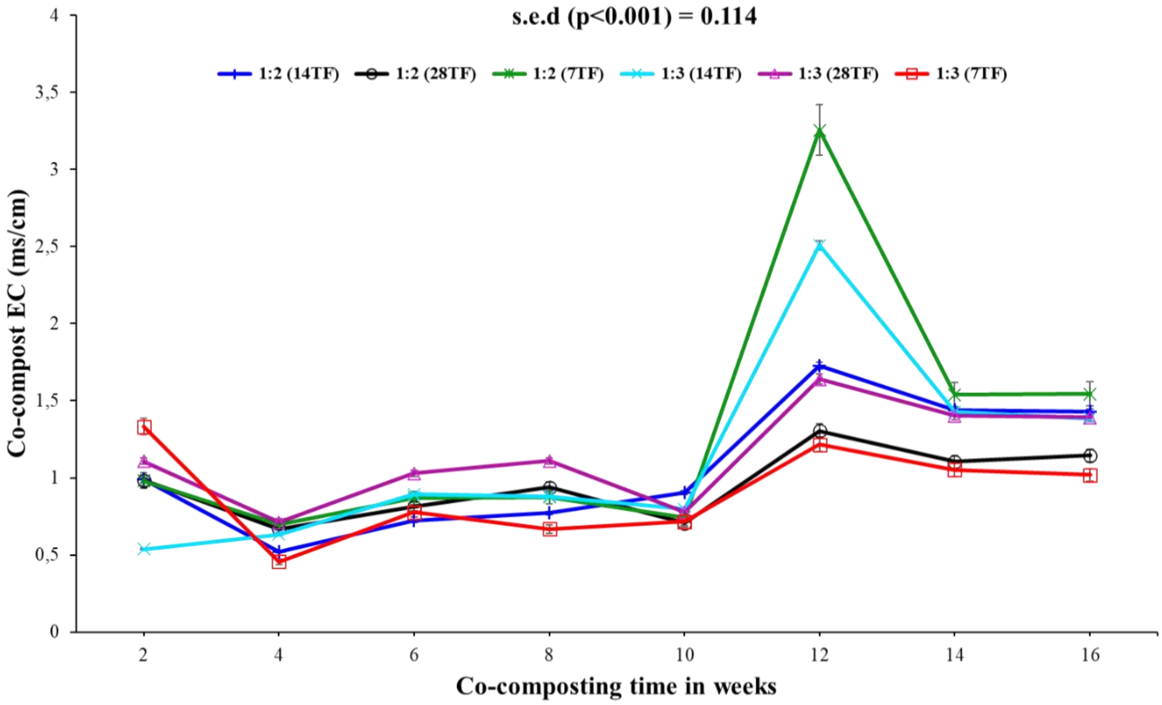


Figure 3.2: Evolution of the electrical conductivity (EC) in the co-compost made from dewatered sewage sludge and organic green waste over time

3.3.3 Organic matter and total organic carbon

There were significant ($p<0.001$) differences in the organic matter (OM) between treatments over time (Figure 3.3). The first measurement of OM occurred in the second week of composting and an optimum value of 52.1% was observed in the 1:2 (7TF) co-compost treatment. The OM gradually decreased in all treatments due to the breaking down of OGW and DSS in the co-compost. At the end of the co-composting process, OM between treatments ranged from 30.02% to 33.35% between all treatments. High OM composition is desirable since organic matter improves crop productivity by enhancing nutrient and water availability to plants in the soil. Also, the higher turning frequency 7TF exhibited a high loss of OM compared to the less frequently turned 28TF compost. The organic matter of the final co-compost was within the recommended 15-45 % suggested by (Trémier, 2012). However, it was slightly lower than the 35 % observed by (Zorpas and Loizidou, 2008). The considerable reduction in total organic carbon (TOC) and OM content observed in this study indicates that considerable mineralisation of OC and OM occurred, thus reducing the volume and weight of the co-compost piles (Manga et al., 2022). According to Varma et al., (2018), microbial degradation led to a decrease in OM and OC in the compost. Microbes convert organic matter to CO_2 and exchangeable carbon. Furthermore, the reduction in OM and OC is thought to occur due to the decomposition of polysaccharides, especially lignocellulose, during the co-composting process (Leifeld et al., 2002).

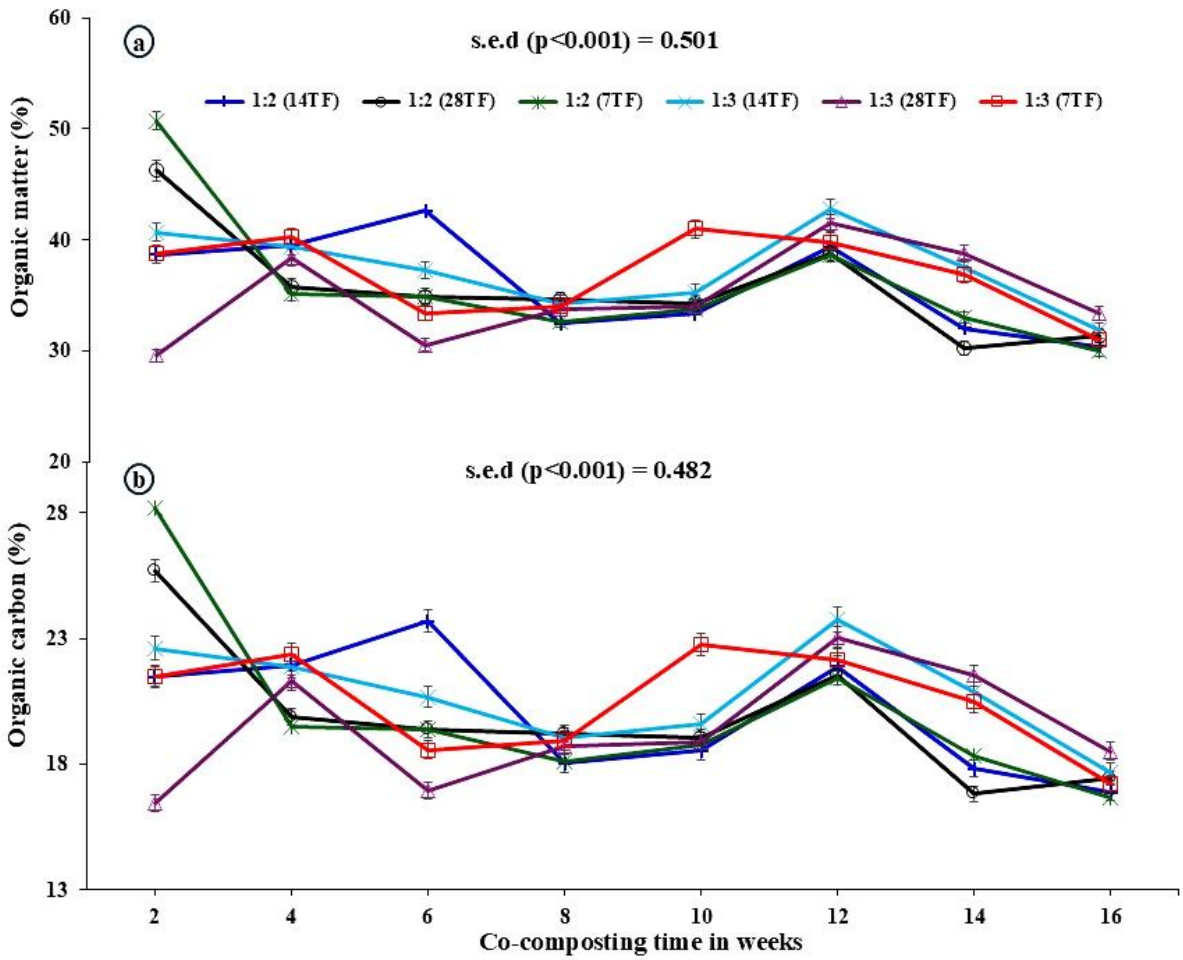


Figure 3.3: Organic matter (**a**) and organic carbon (**b**) of different co-compost treatments made from dewatered sewage sludge and organic green waste over time

3.3.4 Temperature

Temperature monitoring was conducted throughout the co-composting process to assess conditions conducive to pathogens and helminth egg destruction. The iButtons were initially used for temperature recording, but due to malfunctions discovered after shipment to ETH Zurich, temperature data was only reliably captured using a 1-meter-long thermometer from the second to the fourth month of co-composting. In the present study, temperature did not reach the recommended 55 °C for effective pathogen deactivation (Figure 3.4). Moreover, turning frequency combined with mixing ratios did not statistically show a significant difference (p>0.05) in temperature. However, interesting observations were made between temperatures at different mixing ratios. In between the turning frequencies, the highest temperature reached by the co-compost treatment was in the ratio 1:3 (T28), which was 54.6 °C. The treatments with the highest proportion of the sludge (1DSS:2OGW) resulted in higher temperatures in the 7- and 14-days TF with the value of 52.3 °C which was attained by the 1:2 (7TF) on day 21 and 51.5 °C in the treatment 1:2 (14TF). The less-turned treatment (28TF) reached a high temperature, indicating that heat was retained within the pile and not easily lost to the atmosphere.

Sewage sludge is rich in nitrogen and microbial population, which enhances the decomposition rate of feedstocks. Therefore, a high proportion of DSS in the 1DSS:2OGW could have led to increased microbial activities and increased the temperature of the co-compost due to respiration. However, a temperature above 50 °C was not maintained for more than 10 days for effective pathogen deactivation (Preneta et al., 2013). This could probably be attributed to the size of the co-compost pile. The pile size (1 m^3) was not big enough to retain heat inside the co-compost piles, thus resulting in heat loss through convection or conduction processes (Trautmann and Krasny, 1998). These heat loss mechanisms occur between the co-compost and the surroundings, which in this case is from the co-compost to the co-compost bin and/or to the surrounding or atmosphere.

Temperature is essential in deactivating pathogens in the human excreta-derived co-compost. The USEPA (2003) standards indicated that a temperature above 55 °C for approximately 2 weeks is required to deactivate pathogens. Moreover, temperature is an indicator of microbial activities occurring in the co-compost. It is used to determine and distinguish different stages of the co-composting process, namely the mesophilic phase (25-44 °C), thermophilic (45-60 °C) and curing/second mesophilic stage (ambient temperature) (Diaz et al., 2011). Temperature can be affected by various factors including turning frequency, pile size, C/N ratio and moisture content (Getahun et al., 2012; Ofei-Quartey et al., 2023; Wong et al., 2001; Zhao et al., 2015). According to Getahun et al. (2012), the temperature was influenced by turning frequency, wherein the most frequently turned co-compost piles the thermophilic temperatures lasted for 3 days while the less frequently turned co-compost lasted for 15 days. Jiang-Ming (2017) also observed that frequently turned compost piles resulted in high temperatures and excess moisture removal. However, (Manga et al., 2023a) observed that turning frequency did not increase the temperature as all the piles failed to reach the thermophilic temperature (≥55 °C) and the highest temperature was 54 °C for the 3,7 and 14-day turning intervals. However, the present study reported that *E. coli*, *Salmonella*, and viable *Ascaris* eggs were deactivated during the co-composting process and were undetectable in the final co-compost made.

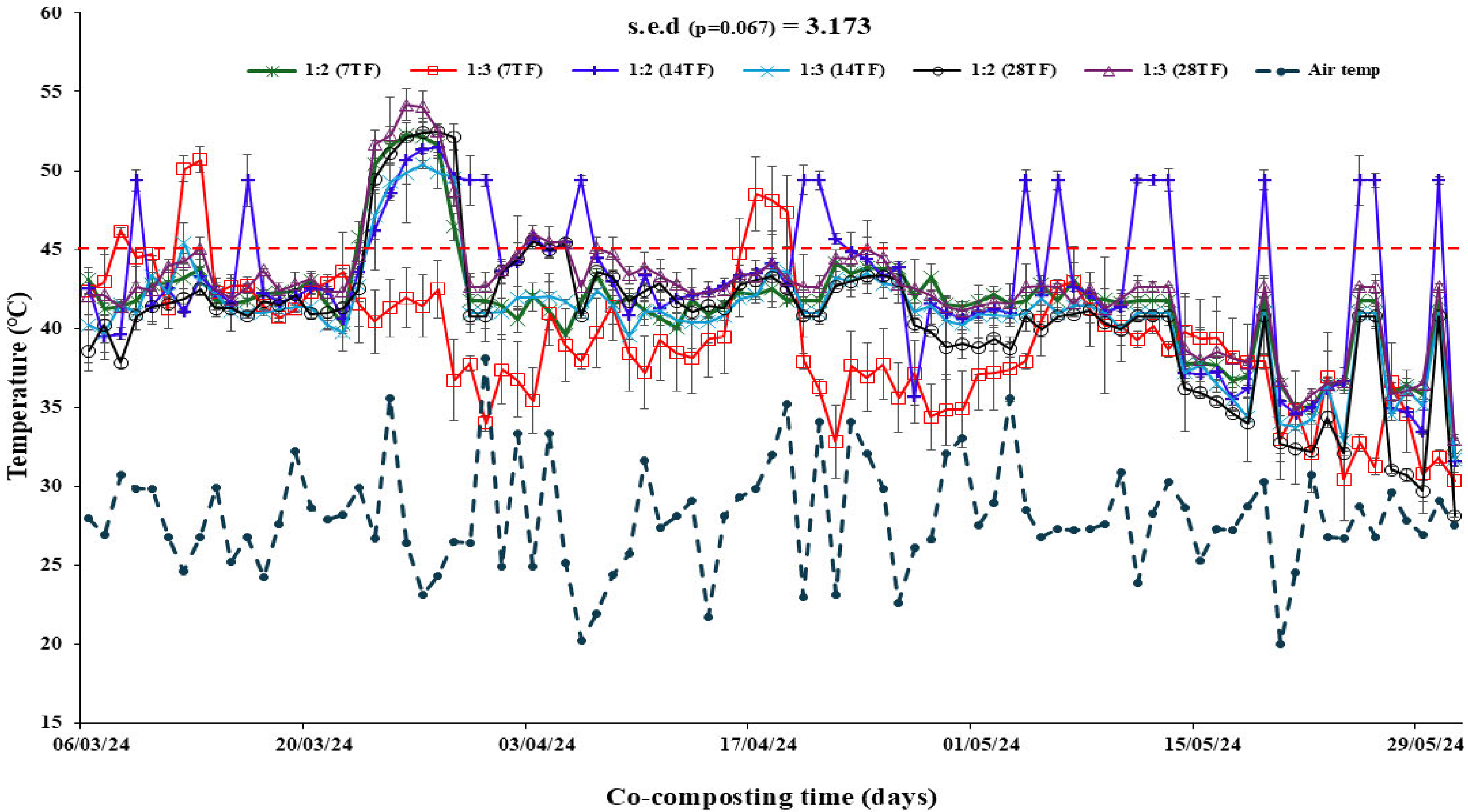


Figure 3.4: Temperature evolution of the co-compost made from dewatered sewage sludge and organic green waste using different mixing ratios and turning frequencies one month later from the initial co-composting process to month four of co-composting, graph showing (n=9 ± standard error)

3.3.5 *Escherichia coli and Salmonella* spp.

Matured co-compost was analysed for *E. coli* and *Salmonella*, indicators of faecal contamination and human pathogenic bacteria (Werner et al., 2023). According to Manga et al. (2023a), *E. coli* is one of the most important faecal matter indicators in the co-compost made from human faeces. Table 3.2 shows the concentration of *E. coli* in the matured co-compost treatments. Thermophilic temperatures of 55 °C for two weeks, 60 °C for six days or 65 °C for three days are required to ensure the destruction of these pathogens in the organic waste compost (BioAbfV, 1998). According to Ceustermans et al., (2007), removal of *Salmonella* spp., in the co-compost requires humidity between 50-60% and temperature greater than 60 °C. However, several authors (Manga et al., 2023a; Preneta et al., 2013) found that a temperature greater or equal to 50 °C can eliminate pathogens in the co-compost. Moreover, Manga et al., (2023a) further implied that turning frequency significantly differed in the deactivation of the pathogens in the co-compost made from faecal sludge and sawdust. The authors further mentioned that higher turning frequency resulted in high pathogen reduction because the heat was evenly distributed among the edges and the middle sections of the co-compost, thus destroying these pathogenic organisms. Controversially, Ofei-Quartey et al. (2023) found that the co-compost had the presence of *E. coli* with the range from 1.2×10^6 CFU/ml to 2.07×10^7 CFU/ml recorded in the different co-compost piles. This was caused by the smaller volume of the piles and the failure of the co-compost piles to reach temperatures high enough to deactivate active pathogenic *E. coli* and *Salmonella*.

Scientific literature has documented that thermal destruction of *E. coli* occurs when the temperature within a co-compost windrow or pile reaches 55 °C and above (Diaz, 2011 et al.; Trautmann, 1998; Guo et al, 2012; Getahun et al., 2012; Manga et al, 2023). Treated dewatered sewage sludge used in this study had an *E. coli* population >300 CFU/g at the first dilution but was less than 30 CFU/g in the second dilution indicating that the *E. coli* was less than 1000 CFU/g. This was an indication that the biosolids or dewatered sewage sludge used in this study was treated before disposal which resulted in less detected concentration of *E. coli*. The *E. coli* was absent in the 1DSS:3OGW (7TF), 1DSS:3OGW (14TF) and 1DSS:2OGW (28TF), indicating that the co-composted material does not pose any risks to human health and is safe for agricultural uses. Moreover, *E. coli* concentrations in 1DSS:3OGW (28TF), 1DSS:2OGW (7TF), and 1DSS:2OGW (14TF) were also too little to count (<30 CFU/g). Temperature and alkaline pH combination could have destroyed *E. coli* (Kazama and Otaki, 2011; Wichuk et al., 2011), thus leading to *Salmonella* deactivation as well. The total coliform, *E. coli* and

Salmonella population was below the standards (1×10^3 cfu/g) recommended by the (WHO, 2006).

Table 3.2: Quantification of *E. coli*, *Salmonella* and total coliform count in the matured co-compost (CFU g^{-1}); a number below 30 represents the detection limit of the method (Werner et al., 2023)

Treatments	**Rep**	***E. coli*** [CFU g^{-1}]	***Salmonella* spp.** [CFU g^{-1}]	**Total coliforms** [CFU g^{-1}]
DSS	-	> 300	> 300	> 300
OGW	-	nd	nd	nd.
1DSS:3OGW (7TF)	1	Absent	Absent	91×10^2
	2	Absent	Absent	85×10^2
	3	Absent	Absent	112×10^2
	Average	**0**	**0**	**96×10^2**
1DSS:2OGW (28TF)	1	Absent	< 30	82×10^2
	2	Absent	Absent	89×10^2
	3	Absent	< 30	82×10^2
	Average	Absent	**< 30**	**83.5×10^2**
1DSS:3OGW (14TF)	1	Absent	Absent	94×10^2
	2	Absent	< 30	114×10^2
	3	< 30	Absent	117×10^2
	Average	**<30**	**< 30**	**108×10^2**
1DSS:2OGW (7TF)	1	< 30	< 30	109×10^2
	2	< 30	Absent	123×10^2
	3	< 30	< 30	126×10^2
	Average	**< 30**	**<30**	**119×10^2**
1DSS:3OGW (28TF)	1	< 30	< 30	131×10^2
	2	< 30	<30	138×10^2
	3	< 30	<30	128×10^2
	Average	**<30**	**<30**	**132.3×10^2**
1DSS:2OGW (14TF)	1	< 30	< 30	126×10^2
	2	< 30	< 30	114×10^2
	3	< 30	< 30	138×10^2
	Average	**< 30**	**< 30**	**126×10^2**

CFU; colony forming units; **n.d** (non-detectable), **TF** (Turing frequency)

3.3.6 Staphylococcus aureus and Pseudomonas aeruginosa

Staphylococcus aureus and *Pseudomonas aeruginosa* are classified as opportunistic bacteria. According to Guo et al. (2023), *S. aureus* can cause skin infections, food poisoning, and other more serious infections. Furthermore, McDougald et al. (2008) observed that *P. aeruginosa* causes immunocompromised infections, subsequently leading to respiratory infections.

Therefore, quantifying these opportunistic bacteria is essential in the compost because they can pose health risks to human beings if the co-compost is not correctly treated. Although *Pseudomonas sp*. could be an opportunistic pathogen, it can also be useful in biodegradable agrochemicals (Sarkar et al., 2009). Microbial biodegradation plays an important role in transforming organic chemicals within the soil. Consequently, bioremediation is regarded as a highly effective and sustainable method for the complete breakdown of external agricultural chemicals introduced into cultivated soils (Kulkarni and Kaliwal, 2014). In this study, there was a significant difference in *Pseudomonas sp*. and *S. aureus* between different co-compost treatments (Figure 3.5).

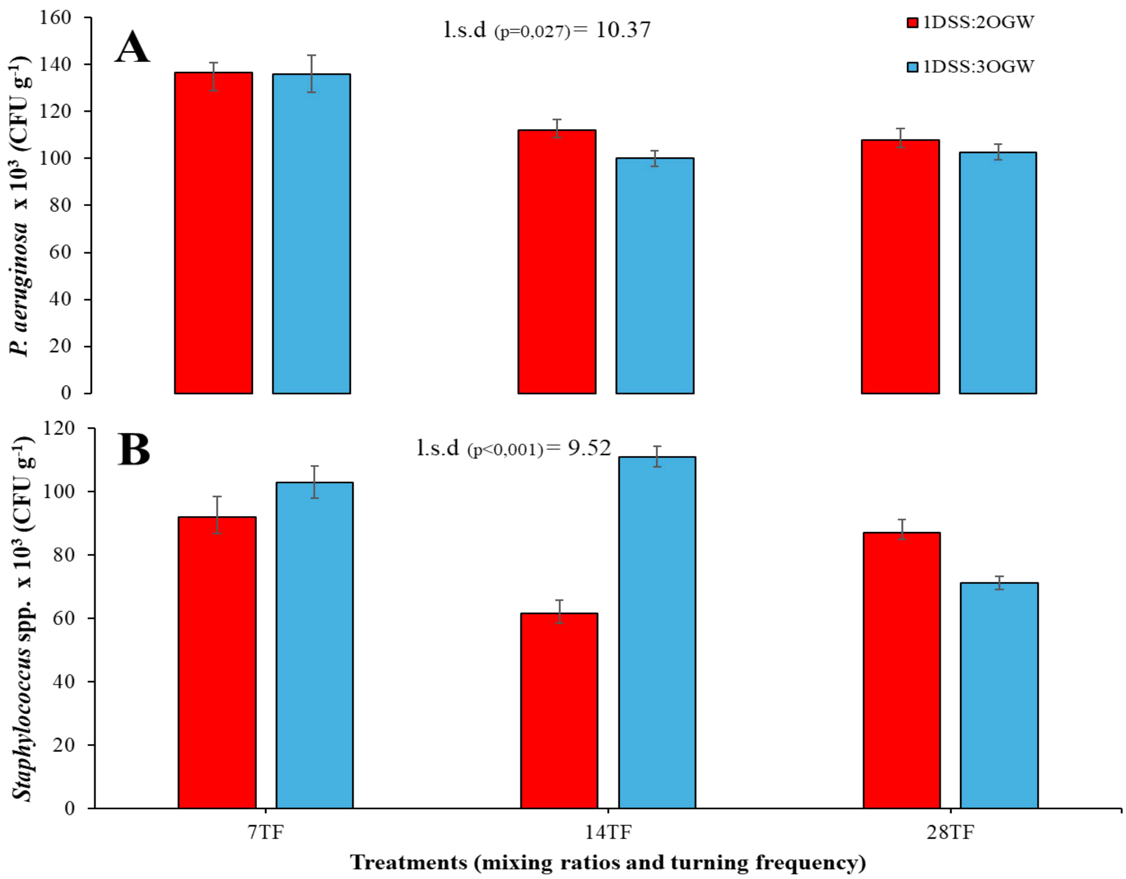


Figure 3.5: Opportunistic bacteria *P. aeruginosa* (**A**) and *Staphylococcus spp.* (**B**) in the matured different co-compost.

3.3.7 Helminths eggs

Helminth eggs are a matter of concern in the co-compost made from human excreta-derived material as they persist in the soil under optimum conditions. According to Manga et al. (2023b), the most persistent helminths in the compost are roundworms (*Ascaris lumbricoides*), tapeworms (*Taenia solium*), whipworms (*Trichuris trichiura*) and hookworms (*Ancylostoma duodenale* or *Necator americanus*). These helminths are the most resistant microbes as they can find alternative hosts and are resistant to heat (temperature), lime and ammonia (Manga et al., 2023b). Moreover, *Ascaris* and *Trichuris* are known to mainly infect young children between the ages of ages 5 and 15 years (Koné et al., 2007). *Ascaris* eggs are usually found in raw sludge at relatively high concentrations with a daily birth rate ranging 200 000 to 240 000 eggs daily (Leung et al., 2020), while *Trichuris* eggs produce 2 000 – 10 000 eggs daily (Feachem, 1978).

According to Manga et al., (2016), *Ascaris* species are the commonly used indicators of matured and safe compost as they persist and can live in soils for up to 7 years Initially, all treatments were highly populated with viable *Ascaris* eggs. However, adverse conditions such as temperatures greater than 45°C during the co-composting process led to the destruction of *Ascaris* eggs (Figure 3.6B). Gaasenbeek and Borgsteede, (1998), indicated that the rough outer shell of *Ascaris* eggs makes it resistance and survive under aerobic and anaerobic conditions and live longer in alternative hosts. According to Harroff et al. (2019), exposing *Ascaris* embryo larvae to high temperatures causes the eggshell to be weak and exposes the larvae to heat enough to inactivate it. Moreover, Maya et al. (2012) further explained that the inactivation of these resistant *Ascaris* eggs occurs during the embryotic phase compared to anembryonic eggs.

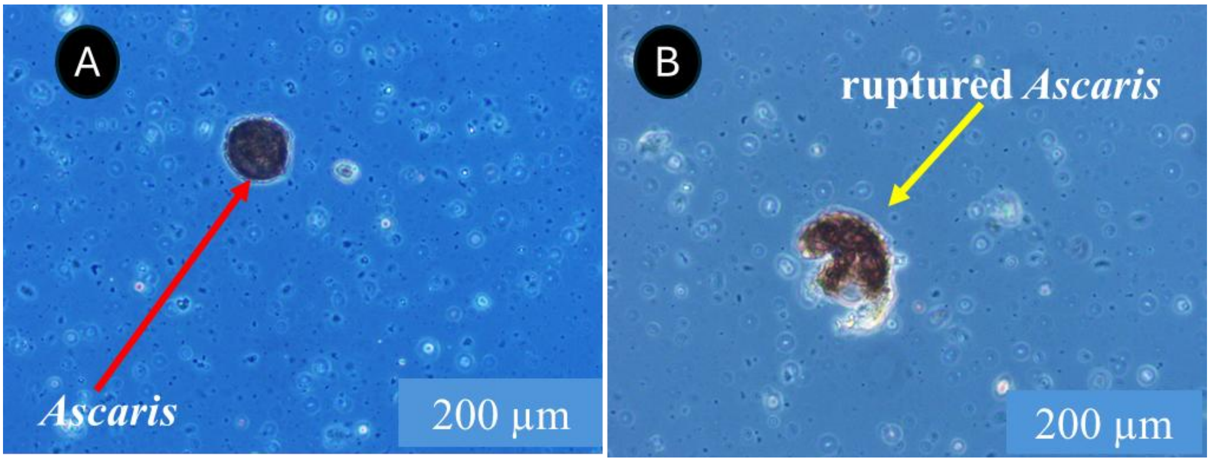


Figure 3.6: Viable *Ascaris* egg (**A**) and ruptured *Ascaris* egg (**B**) in the matured co-compost (×200 magnification)

Helminth eggs are often assessed using viable egg count instead of total count. The total count of eggs includes viable, dead, developing, underdeveloped and non-viable helminth eggs. In the present study, there were abundant *Taenia* eggs, most probably developing or underdeveloped because of their small size and viable eggs. The viable eggs were observed with motile and immotile larvae or developing embryos, as shown in Figure 3.7A. At the maturity stage of the co-compost (matured), most of the *Taenia* eggs ruptured (Figure 3.7B), thus explaining the thermal destruction caused by temperature > 45 °C in the second month of co-composting. According to Koné et al. (2007), temperature > 45 °C for more than 40 days is essential for deactivating helminth eggs in the faecal sludge-based co-compost. Moreover, Harroff et al. (2019) indicated that low or mesophilic temperatures can be used to inactivate pathogens in waste treatment processes but occur over a long period. In conjunction with other studies, alkaline pH ranging between 8 to 12 was discovered as an alternative factor that leads to deactivation of helminth eggs in biosolids or dewatered sewage sludge (Capizzi-Banas et al., 2004). Temperature, pH, and a reduced moisture content in the co-compost are other factors that can subsequently result in reduced or destruction of helminths in eggs in the co-compost (Sanguinetti et al., 2005).

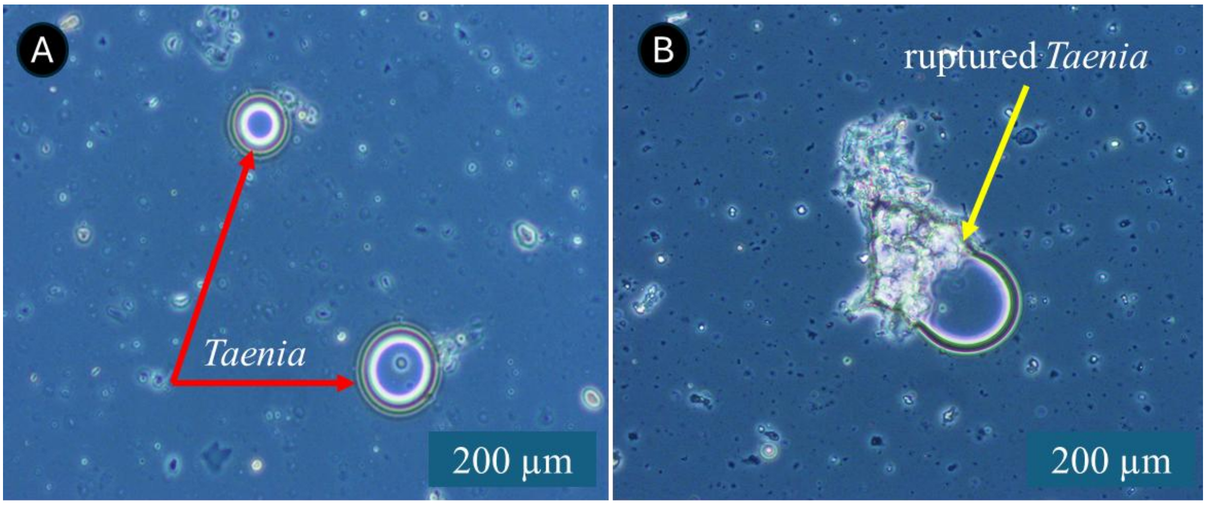


Figure 3.7: The viable *Taenia* eggs (**A**) in the initial stage of co-composting and ruptured *Taenia* eggs (**B**) in the matured co-compost (×200 magnification)

All the co-compost treatments except 1DSS:3OGW (7TF) were slightly populated with viable hookworms (Figure 3.8A) in the initial stage of co-composting. However, in the final co-compost, hookworms were no longer present and ruptured or broken hookworms' traces were found in the co-compost (Figure 3.8B). Typical *Trichuris* were dominant initially in the co-compost treatments. However, at the maturity stage, *Trichuris* eggs were not observed in all co-compost and somehow *Fasciola hepatica* was observed in 1DSS:2OGW (14TF) with some eggs without larvae or ruptured. The *Fasciola hepatica* probably colonised the co-compost through contamination with the snail or cross-contamination from insects and pests from the plants grown in the tunnel.

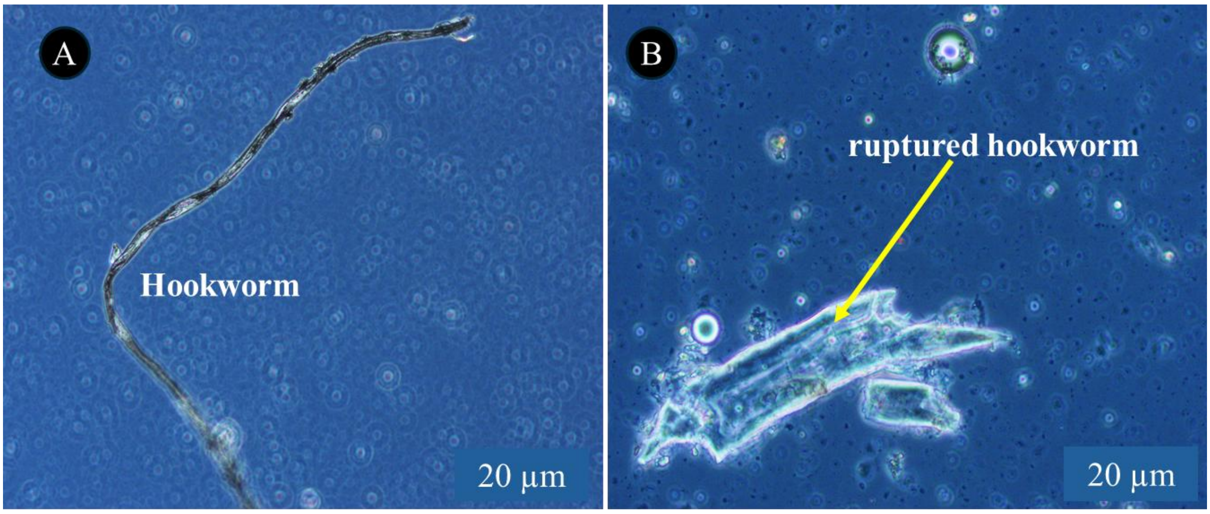


Figure 3.8: Viable hookworm (**A**) and ruptured hookworm (**B**) at (×20 magnification)

In addition to the visual observation of helminth egg deactivation, Table 3.3 shows the co-compost results of helminth eggs and faecal coliforms which were analysed by the Ekurhuleni Water Care Company in Gauteng, South Africa. The results show that most co-compost treatments had absent helminth eggs (0 eggs/4g of dry weight) which comply with the Fertiliser Act standards which indicates that there must be no viable eggs per 4 g of dry matter co-compost (SAG, 2009). The 1DSS:3OGW (7TF) complied with the World Health Organization standards which recommended 0.25 eggs per 4g of co-compost (WHO, 2006). The reduced level of helminth eggs to zero correlates to the observation of helminth eggs in the co-compost under the microscope as shown Figures 3.6-3.8 above. The study surprisingly resulted in higher helminth eggs in the 1DSS:3OGW (28TF) which is the treatment that attained a higher temperature of 54.6 °C. The inability of this treatment to deactivate viable helminth eggs in the co-compost despite reaching the highest temperature could be due to malfunctioning of the i-Button and probably other treatments that reached thermophilic temperature in the first month of co-composting. Reliable temperature data across all phases is critical for drawing accurate conclusions about the relationship between temperature and pathogen destruction. Moreover, additional studies are needed to identify the parameters beyond temperature that contribute to helminth egg destruction, such as microbial diversity, pH levels, moisture content, or biochemical reactions.

Table 3.3: Helminths eggs and faecal coliform quantification in the matured co-compost

Mixing ratio	Turning Frequency	Helminths eggs per 4g	Faecal coliforms	Comment on helminths
1DSS:3OGW	7 days	0	5	Comply with the Fertiliser Act
1DSS:2OGW	28 days	0	<2	Comply with the Fertiliser Act
1DSS:3OGW	14 days	0	<2	Comply with the Fertiliser Act
1DSS:2OGW	7 days	0.25	<2	Comply with WHO standards
1DSS:3OGW	28 days	3	<3	Do not meet the standards
1DSS:2OGW	14 days	1	<3	Comply with WRC standards

DSS (dewatered sewage sludge), **OGW** (organic green waste), **WHO** (World Health Organization), **WRC** (Water Research Commission)

3.3.8 Volume of the co-compost over time

There was a significant difference in volume reduction ($p<0.05$) between the mixing ratios (1DSS:3OGW and 1DSS:2OGW) and different turning frequencies of 7, 14 and 28-day turning intervals (Figure 3.10). The high turning frequency (7TF) resulted in approximately 40% co-compost volume reduction in both mixing ratios. The 14-day turning interval resulted in a relatively moderate (≈30%) loss or reduction of co-compost, with a high loss in the 1DSS:3OGW co-compost. Interestingly, the 28-day turning interval had the minimum organic matter loss and the 1DSS:3OGW had the highest volume of co-composting at the end of the co-composting experiment. This could have been caused by less microbial activities due to less frequent turning. Moreover, organic matter breakdown was prolonged, which eventually caused less degradation. In contrast, there was a high organic matter breakdown in most frequently turned co-compost piles as the microbes received sufficient oxygen for respiration and metabolised the nutrients in the co-compost windrows.

There was an inverse relationship between co-composting time and the volume of the co-compost. As the co-composting period progressed, the volume of the co-compost decreased. Initially, all co-composting experimental boxes had the co-compost filled to the fullest capacity. However, the reduction in the volume of the co-compost occurred gradually between the successive days of day 45 and 90; finally, on day 120, the co-compost volume was at its lowest. The frequently turned co-compost 1:2 (7TF) and 1:3 (7TF) resulted in the smallest co-compost volume at the end of the co-composting process. This could be due to increased turning frequency, which results in high microbial degradation and rapid decomposition rate, which results in less co-composting at the end of the co-composting process. According to El-Saeidy et al. (2016), turning the compost piles 3 times, a week maximizes reduction in the volume of the agricultural residues compost. Moreover, Ugak et al. (2022) observed that turning the food waste compost every 3 days resulted in high volume reduction and organic matter loss, which was efficient for composting.

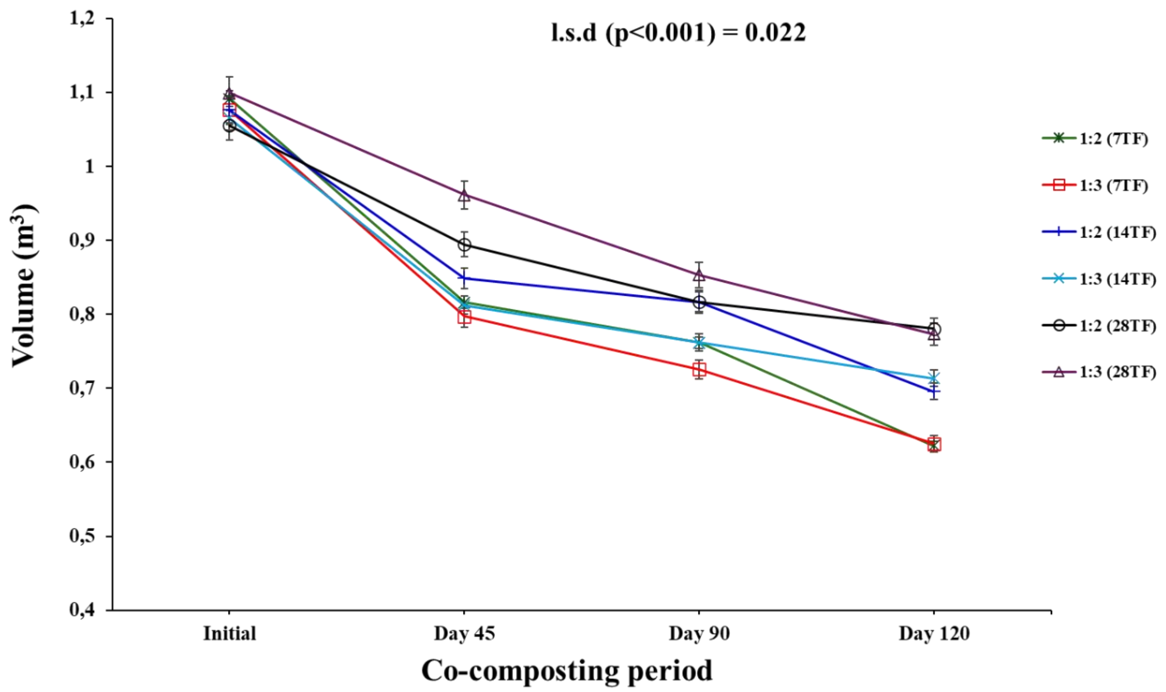


Figure 3.9: Volume reduction of co-compost from the start of the co-composting process to month four of the composting process in different treatments

3.3.9 Germination and plant growth index

Figure 3.11A shows that in the second week after composting, the co-compost exhibited a lower germination index (GI) in all the treatments with values of 9.2%, 25.6 %, 36.4%, 49.3%, and 53.7% in the 1:3 (28TF), 1:2 (7TF), 1:2 (28TF), 1:3 (14TF), 1:2 (14TF), and 1:3 (7TF), respectively. There was a relative increase in the GI in week 4; all co-compost piles exhibited GI greater than 80% implying that it was free from phytotoxic compounds that inhibit growth. However, there was a significant fluctuation and decreased GI for the treatments with a high proportion of sludge (1DSS:2OGW) in all the turning frequencies, attaining a GI value of less than 80% (Figure 3.10). The fluctuation of the GI occurred throughout the co-composting period, during which an increase and decrease in GI were observed (Figure 3.10). However, the highest GI obtained was 374.6% attained by the 1:2 (7TF), indicating that the seedlings were 3.7 times more vigorous than the control. In week 12, all co-compost piles exhibited a reduced GI (less than 80%), which the spike or increase in pH could have caused (Figure 3.1), EC (Figure 3.2), OM (Figure 3.3A) and OC (Figure 3.3B) observed in week 12 could have led to decrease in GI. According to Wang et al., (2022), the pH, EC and OC content influenced seed germination in phytotoxicity tests during sheep manure composting with carbon additives, thus reducing the germination index. Kebibeche et al. (2019) found that a pH between 6.73 and 7.19 is optimal and can enhance GI by reducing phytotoxicity in wood sawdust and sewage sludge co-composts. Therefore, this explains the lower GI obtained in this study because the pH was generally higher than the recommended pH range (Kebibeche et al., 2019). In weeks 14 and 16 of the co-composting process, the GI was greater than 80% in all treatments, and the co-compost was regarded as mature based as time was also considered. A decrease in GI could have occurred due to the degradation and breakdown of organic matter in the co-compost, which released acetic, propionic and valeric acids (Sullivan and Miller, 2001). As observed by Manga et al. (2022), the increase in the GI index over time could have been attributed to the breaking down of phytotoxic compounds such as ammonium ions and volatile fatty acids. Interestingly, the co-compost treatment with the highest sludge proportion and less frequently turned exhibited the highest GI at the end of the co-composting process, indicating the phytotoxic-free compost product. The plant growth index (Figure 3.10B) was even much higher in the co-compost (>80%) in all treatments, 1DSS:2OGW (14TF) exhibited the highest (597.4%) while Duzi-turf co-compost resulted in the lowest (178.3%) plant growth index. Interestingly co-compost in this present study had a plant growth index that was greater than the co-compost that is commercially sold at Duzi-turf, indicating that these co-compost treatments were all free from phytotoxic compounds that inhibit the growth of sensitive crop species.

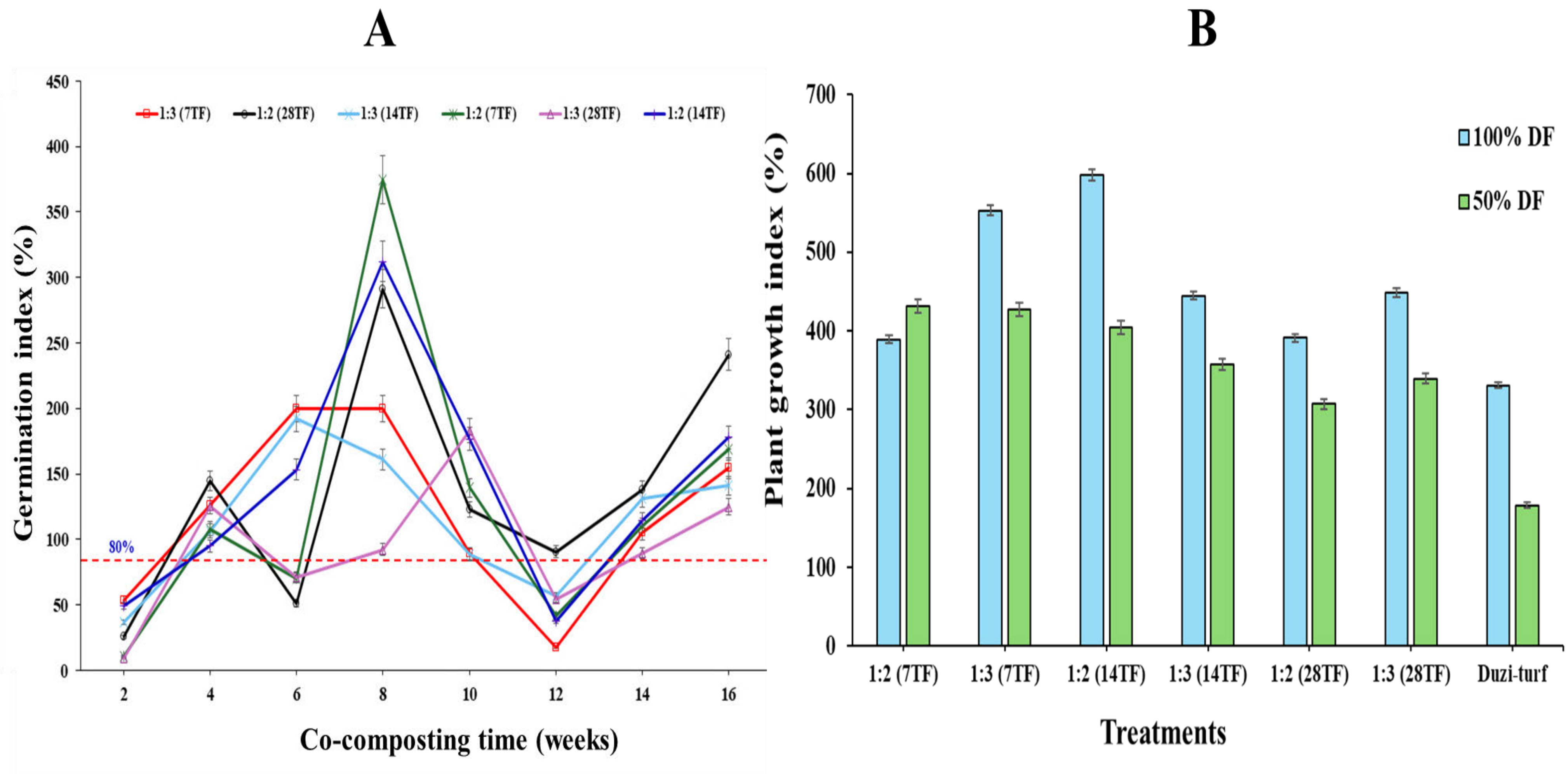


Figure 3.10: Germination index (**A**) over time and plant growth index (**B**) at the end of the co-composting process of different co-composting treatments serving as an indicator of phytotoxicity of the compost over time.

3.3.10 Heavy Metals and nutrients in the co-compost

Heavy metal concentration in the sewage sludge-derived co-compost is one of the major concerns for recycling human excreta-derived co-compost in agriculture since it harms plant growth and development, human health and beneficial soil micro-organisms (Manga et al., 2022). In the documented literature, zinc (Zn), chromium (Cr), cadmium (Cd), copper (Cu), nickel (Ni), mercury (Hg) and lead (Pb) are substances of concern about the application of compost in agricultural soils (Smith, 2009). Therefore, in this study, it was important to investigate further these and other trace elements, that constitute heavy metals and metalloid contaminants in the co-compost that can be toxic to soil microbes, plant growth, and human health. According to Smith, (2009), Zn, Cu, Ni and Cr can result in phytotoxicity or reduced plant growth. Other metals, including aluminium (Al), manganese (Mn), iron (Fe), and sodium (Na), can reduce seed germination, inhibit plant growth, cause stunted growth and chlorosis, and bind with macro-nutrients such as phosphorus, leading to reduced root growth and failure to uptake nutrients (Nadeem et al., 2018). Knowledge of these metals and metalloids can make a meaningful contribution towards simulating standards in compost for maximum permissible values. In the present study, the concentration of Fe and Al were higher, with maximum values of 33 415 mg/kg and 18 643 mg/kg, respectively. However, the permissible thresholds for both these metals in the matured co-compost derived from human excreta are not documented.

Interestingly, all treatments complied with the South African and European standards for safe co-compost (Table 3.4). The macro-nutrients in the matured co-compost were and fell within the recommended standards for nutrient recovery through the composting process (Table 3.4). Less frequent turning frequency (28-day turning interval) and mixing ratio with less sludge (1DSS:3OGW) resulted in high nitrogen (1.41%); and was significantly different from other treatments, which had N ranging between 1.30-1.38%. Turning the co-compost most frequently of the co-compost enhances N loss through ammonium gas, which also escapes to the atmosphere. Interestingly, P, K, Mg and Ca were higher in the less turning (28-day turning interval) in the 1DSS:2OGW, signifying that less turning frequency can be ideal for preserving nutrients in the co-compost.

The neglected metals, including titanium (Ti), vanadium (V), and strontium (Sr) can be beneficial to plants (Eqani et al., 2018). However, they can adversely affect plants if they occur at higher concentrations. According to Lyu et al., (2017), titanium application on leaves and roots can improve enzymatic activities, thus improving chlorophyll content and crop yield; however, excess Ti can compete with Fe, resulting in phytotoxic Ti and plant growth reduction, At low concentrations of V, can improve plant growth and development; however, it can reduce seed germination, inhibit root and shoot growth, depress photosynthesis, and interfere with nutrient uptake if it occurs at higher concentrations (Chen et al., 2021) The concentrations of P and Fe elements in the compost were significantly reduced at higher levels of V, showing competition among these elements for plant uptake and assimilation (Aihemaiti et al., 2020).

The accumulation of Sr in plants occurs through the plasma membrane, which transports calcium and potassium (Burger and Lichtscheidl, 2019). These metals do not have permissible thresholds in compost, yet they have a more pronounced and significant impact on plant growth and human health.

Table 3.4: Macronutrients, micronutrients and metals found in different co-compost treatments at the end of the co-composting process compared to the South African and European standards (Brinton, 2000).

Elements	Units	1:3 (7TF)	1:2 (7TF)	1:3 (14TF)	1:2 (14TF)	1:3 (28TF)	1:2 (28TF)	RSA	EU
Macro-nutrients								%	%
Nitrogen	%	1.36	1.35	1.37	1.30	1.41	1.38	1.5-2.5	1-3.5
Phosphorus	%	0.51	0.57	0.51	0.54	0.47	0.57	0.4-1.2	0.3-1.5
Potassium	%	0.40	0.35	0.37	0.37	0.37	0.43	0.5-1.5	0.5-2.5
Magnesium	%	0.33	0.34	0.33	0.36	0.35	0.39	0.5-1	0.5-1.5
Calcium	%	2.13	2.32	2.18	2.32	2.38	2.58	1.5-4	2-7
Metals								**mg/kg**	**mg/kg**
Manganese	mg/kg	659	637	685	645	673	783	-	-
Sodium	mg/kg	637.9	741.8	595.4	716.2	668.8	765.8	-	-
Zinc	mg/kg	281	380	272	249	293	281	300	200-1500
Copper	mg/kg	91.4	92.9	72.7	74.1	75.2	84.2	150	100-600
Aluminium	mg/kg	17169	18903	15714	15701	16340	18643	-	-
Iron	mg/kg	29549	32130	27499	27994	29027	33415	-	-
Metals and metalloids								**mg/kg**	**mg/kg**
Cadmium	µg/L	8953.2	9432.3	1011.5	9245.8	9.9×10^{-3}	8857.5	3	1.5-3
Chromium	µg/L	1436.79	2504.7	2708	1263	2693.3	1145.7	100	70-100
Boron	µg/L	362	330	371	378.7	393	348.7	5-50	30-300
Nickel	µg/L	176	226	193.7	183.7	213.7	181.3	50	30-90
Arsenic	µg/L	48.3	53.7	62	53.7	53.3	52.3	10	10-40
Barium	µg/L	1395.3	1431.3	1554	1590.3	1613.3	1550.7	-	-
Cobalt	µg/L	261	275.7	287.3	262.7	284.3	271.6	50	20-50
Lithium	µg/L	55	59.7	59	65.3	69.3	57.3	-	-
Lead	µg/L	228	232.7	243	212.7	250	205	150	120-300
Antimony	µg/L	25.7	33.7	35.7	27.3	40.7	26.3	-	-
Silver	µg/L	<10	<10	<10	<10	<10	<10	-	1-5
Molybdenum	µg/L	17.7	18	23	13.6	20.9	14.3	-	2-5
Selenium	µg/L	<13	<13	<13	<13	<13	<13	-	0.7-3
Tin	µg/L	80.3	85.3	77	75.7	82.3	57.7	-	-
Titanium	µg/L	2145.7	1715	1621.7	1953.7	1604.3	1899	-	-
Vanadium	µg/L	396.7	423	443.7	400.7	422.7	387.7	-	-
Strontium	µg/L	383.3	401.7	4224.3	414.7	432	383	-	-
Mercury	µg/L	<13	<13	<13	<13	<13	<13	1.5	0.1-1

3.4 Conclusion

In this study, turning frequency and mixing ratios statistically affected pH, EC, OM and OC ($p<0.05$). The pH at the end of the co-composting process was slightly alkaline, suitable for neutralising acidic soils. The EC was within the recommended range of 1.02 mS/cm to 1.55 mS/cm. The OM and OC were reduced due to microbial degradation of organic waste. The temperature did not significantly differ ($p>0.05$) between turning frequencies. However, it was noted that less turning frequency (28TF) resulted in a high temperature (54.6°C) and was maintained longer than high turning frequency.

There was a significant difference ($p<0.05$) between the co-compost turning frequency and mixing ratio in different co-compost treatments. Higher turning frequency (7TF) resulted in higher volume reduction (40%) compared to less turning interval. Bacteria pathogens *E. coli* and *Salmonella* were deactivated (<30 CFU/g) in the matured co-compost. These results were compiled with threshold values of 1000 CFU/g recommended by the World Health Organisation. In addition to this, heavy metals in the compost treatments were below the maximum permissible values of European and South African standards. Thus, the co-compost is of good quality and is not harmful to plants, animals and human beings. All the treatment combinations exhibited a germination index greater than 80%, indicating that the co-compost was phytotoxic-free since it supported seed germination of a sensitive species. The findings supported the co-utilization of dewatered sewage sludge and organic green waste to produce high-quality co-compost for plant production.

The study further revealed that different turning frequencies impact co-compost volume reduction. In this regard, the lower turning interval resulted in higher quantities of co-compost at the end of the co-composting process compared to the higher turning interval. The lower turning interval may also result in higher temperatures that can last longer thus leading to the deactivation of pathogens. Future research should focus on assessing the cost-benefit of using different mixing ratios and employing different turning frequencies, which could indicate the most cost-effective mixing ratios and turning frequencies for farmers, organisations, and municipalities wishing to start composting facilities.

References

Aihemaiti, A., Gao, Y., Meng, Y., Chen, X., Liu, J., Xiang, H., Xu, Y., and Jiang, J. (2020). Review of plant-vanadium physiological interactions, bioaccumulation, and bioremediation of vanadium-contaminated sites. *Science of the Total Environment* **712**, 135637.

Banegas, V., Moreno, J., Moreno, J. I., Garcia, C., Leon, G., and Hernandez, T. (2007). Composting anaerobic and aerobic sewage sludges using two proportions of sawdust. *Waste Management* **27**, 1317-1327.

Bernal, M. P., Alburquerque, J., and Moral, R. (2009). Composting of animal manures and chemical criteria for compost maturity assessment. A review. *Bioresource technology* **100**, 5444-5453.

BioAbfV (1998). Bioabfallverordnung in der Fassung der Bekanntmachung vom 4. April 2013 (BGBl. I S. 658), die Zuletzt durch Artikel 3 Absatz 2 der Verordnung vom 27. September 2017 (BGBl. I S. 3465) Geändert Worden ist. Federal Ministry for the Environment, Nature Conservation, Nuclear Safety.

Brinton, W. (2000). Compost quality standards & guidelines: An international view, Woods End Research Laboratory. *Inc., USA*.

Bouhia, Y., Hafidi, M., Ouhdouch, Y., Boakhare, M., Mphatso, C., Zeroual, Y., and Lyamlouli, K., .(2022). Conversion of waste into organo-mineral fertilizers: current technological trends and prospects. *Rev Environ Sci Biotechnol* **21**, 425–446 https://doi.org/10.1007/s11157-022-09619-y

Burger, A., and Lichtscheidl, I. (2019). Strontium in the environment: Review about reactions of plants towards stable and radioactive strontium isotopes. *Science of the Total Environment* **653**, 1458-1512.

Capizzi-Banas, S., Deloge, M., Remy, M., and Schwartzbrod, J. (2004). Liming as an advanced treatment for sludge sanitisation: helminth eggs elimination—*Ascaris* eggs as model. *Water research* **38**, 3251-3258.

CCQC, C. M. I. (2001). California Compost Quality Council. *Nevada City, CA*.

Ceustermans, A., De Clercq, D., Aertsen, A., Michiels, C., Coosemans, J., and Ryckeboer, J. (2007). Inactivation of *Salmonella* Senftenberg strain W 775 during composting of biowastes and garden wastes. *Journal of applied Microbiology* **103**, 53-64.

Chen, L., Liu, J.-r., Hu, W.-f., Gao, J., and Yang, J.-y. (2021). Vanadium in soil-plant system: Source, fate, toxicity, and bioremediation. *Journal of hazardous materials* **405**, 124200.

Diaz, L. F., De Bertoldi, M., and Bidlingmaier, W. (2011). "Compost science and technology," Elsevier.

Dong, J., Chi, Y., Tang, Y., Wang, F., and Huang, Q. (2014). Combined life cycle environmental and exergetic assessment of four typical sewage sludge treatment techniques in China. *Energy & fuels* **28**, 2114-2122.

Dzulkurnain, Z., Hassan, M. A., Zakaria, M. R., Wahab, P. E. M., Hasan, M. Y., and Shirai, Y. (2017). Co-composting of municipal sewage sludge and landscaping waste: a pilot scale study. *Waste and biomass valorization* **8**, 695-705.

El-Saeidy, E., Eissa, A., and El-Massry, E. (2016). Effect of Frequently Turning on Some Composting Processes for Agricultural Residuals. *Journal of Soil Sciences and Agricultural Engineering* **7**, 383-388.

El Hayany, B., El Fels, L., Ouhdouch, Y., and Hafidi, M. (2021). Fate of pathogenic microorganisms during lagooning sludge composting and exploration of bacteriophages as indicator of hygienization. *Environmental Technology & Innovation* **21**, 101268.

Eqani, S.A.M.A.S., Tanveer, Z.I., Qiaoqiao, C., Cincinelli, A., Saqib, Z., Mulla, S.I., Ali, N., Katsoyiannis, I.A., Shafqat, M.N. and Shen, H., (2018). Occurrence of selected elements (Ti, Sr, Ba, V, Ga, Sn, Tl, and Sb) in deposited dust and human hair samples: implications for human health in Pakistan. *Environmental Science and Pollution Research*, *25*, pp.12234-12245.

Feachem, R. (1978). "Health aspects of excreta and wastewater management," International Bank for Reconstruction and Development/The World Bank.

Gaasenbeek, C. P., and Borgsteede, F. H. (1998). Studies on the survival of *Ascaris suum* eggs under laboratory and simulated field conditions. *Vet Parasitol* **75**, 227-34.

Gerland, P., Raftery, A. E., Ševčíková, H., Li, N., Gu, D., Spoorenberg, T., Alkema, L., Fosdick, B. K., Chunn, J., and Lalic, N. (2014). World population stabilization unlikely this century. *Science* **346**, 234-237.

Getahun, T., Nigusie, A., Entele, T., Van Gerven, T., and Van der Bruggen, B. (2012). Effect of turning frequencies on composting biodegradable municipal solid waste quality. *Resources, Conservation and Recycling* **65**, 79-84.

Guo, N., Bai, X., Shen, Y., and Zhang, T. (2023). Target-based screening for natural products against Staphylococcus aureus biofilms. *Critical Reviews in Food Science and Nutrition* **63**, 2216-2230.

Gutiérrez, M., Serrano, A., Siles, J., Chica, A., and Martín, M. (2017). Centralized management of sewage sludge and agro-industrial waste through co-composting. *Journal of environmental management* **196**, 387-393.

Han, L., Patil, S., Boehm, D., Milosavljević, V., Cullen, P., and Bourke, P. (2016). Mechanisms of inactivation by high-voltage atmospheric cold plasma differ for *Escherichia coli* and *Staphylococcus aureus*. *Applied and environmental microbiology* **82**, 450-458.

Hansen, K. (2015). Overview of wastewater treatment in South Africa.

Harroff, L. A., Liotta, J. L., Bowman, D. D., and Angenent, L. T. (2019). Current time-temperature relationships for thermal inactivation of *Ascaris* eggs at mesophilic temperatures are too conservative and may hamper development of simple, but effective sanitation. *Water research X* **5**, 100036.

Hemidat, S., Jaar, M., Nassour, A., and Nelles, M. (2018). Monitoring of composting process parameters: A case study in Jordan. *Waste and Biomass Valorization* **9**, 2257-2274.

Jiang-Ming, Z. (2017). Effect of turning frequency on co-composting pig manure and fungus residue. *Journal of the Air & Waste Management Association* **67**, 313-321.

Karak, T., Bhattacharyya, P. and Paul, R.K., (2014). Assessment of Co-Compost Quality by Physico-Chemical and Exploratory Data Analysis. *CLEAN–Soil, Air, Water*, *42*(6), pp.836-848.

Kazama, S., and Otaki, M. (2011). Mechanisms for the inactivation of bacteria and viruses in sawdust used in composting toilet. *Journal of Water and Environment Technology* **9**, 53-66.

Kebibeche, H., Khelil, O., Kacem, M., and Harche, M. K. (2019). Addition of wood sawdust during the co-composting of sewage sludge and wheat straw influences seeds germination. *Ecotoxicology and environmental safety* **168**, 423-430.

Koné, D., Cofie, O., Zurbrügg, C., Gallizzi, K., Moser, D., Drescher, S., and Strauss, M. (2007). Helminth eggs inactivation efficiency by faecal sludge dewatering and co-composting in tropical climates. *Water research* **41**, 4397-4402.

Kulkarni, A. G., and Kaliwal, B. (2014). Bioremediation of methomyl by soil isolate—Pseudomonas aeruginosa. *J Environ Sci, Toxicol Food Technol* **8**, 1-10.

Kwarciak-Kozłowska, A. (2019). Co-composting of sewage sludge and wetland plant material from a constructed wetland treating domestic wastewater. *In* "Industrial and municipal sludge", pp. 337-360. Elsevier.

Leifeld, J., Siebert, S., and Kögel-Knabner, I. (2002). Changes in the chemical composition of soil organic matter after application of compost. *European Journal of Soil Science* **53**, 299-309.

Leung, A. K. C., Leung, A. A. M., Wong, A. H. C., and Hon, K. L. (2020). Human Ascariasis: An Updated Review. *Recent Pat Inflamm Allergy Drug Discov* **14**, 133-145.

Liu, D., Zhang, R., Wu, H., Xu, D., Tang, Z., Yu, G., Xu, Z., and Shen, Q. (2011). Changes in biochemical and microbiological parameters during the period of rapid composting of dairy manure with rice chaff. *Bioresource technology* **102**, 9040-9049.

Lyu, S., Wei, X., Chen, J., Wang, C., Wang, X., and Pan, D. (2017). Titanium as a beneficial element for crop production. *Frontiers in plant science* **8**, 597.

Mainstone, C. P., and Parr, W. (2002). Phosphorus in rivers—ecology and management. *Science of the total environment* **282**, 25-47.

Mandizvo, T., and Odindo, A. (2019). Seed mineral reserves and vigour of Bambara groundnut (*Vigna subterranea L.*) landraces differing in seed coat colour. *Heliyon* **5**.

Manga, M., Camargo-Valero, M., Anthonj, C., and Evans, B. (2021). Fate of faecal pathogen indicators during faecal sludge composting with different bulking agents in tropical climate. *International Journal of Hygiene and Environmental Health* **232**, 113670.

Manga, M., Evans, B., Camargo-Valero, M., and Horan, N. (2016). The Fate of Helminth eggs during the Co-composting of Faecal Sludge with Chicken Feathers and Market waste. *In* "Proceedings of the 13th IWA Specialized Conference on Small Water and Wastewater Systems (SWWS)". Leeds.

Manga, M., Evans, B. E., Ngasala, T. M., and Camargo-Valero, M. A. (2022). Recycling of Faecal Sludge: Nitrogen, Carbon and Organic Matter Transformation during Co-Composting of Faecal Sludge with Different Bulking Agents. *International Journal of Environmental Research and Public Health* **19**, 10592.

Manga, M., Muoghalu, C., Camargo-Valero, M. A., and Evans, B. E. (2023a). Effect of turning frequency on the survival of fecal indicator microorganisms during aerobic composting of fecal sludge with sawdust. *International Journal of Environmental Research and Public Health* **20**, 2668.

Manga, M., Muoghalu, C. C., and Acheng, P. O. (2023b). Inactivation of faecal pathogens during faecal sludge composting: a systematic review. *Environmental Technology Reviews* **12**, 150-174.

Maya, C., Torner-Morales, F., Lucario, E., Hernández, E., and Jiménez, B. (2012). Viability of six species of larval and non-larval helminth eggs for different conditions of temperature, pH and dryness. *Water research* **46**, 4770-4782.

McDougald, D., Klebensberger, J., Tolker-Nielsen, T., Webb, J. S., Conibear, T., Rice, S. A., Kirov, S. M., Matz, C., and Kjelleberg, S. (2008). *Pseudomonas aeruginosa*: a model for biofilm formation. *Pseudomonas: model organism, pathogen, cell factory*, 215-253.

Nadeem, F., Hanif, M. A., Majeed, M. I., and Mushtaq, Z. (2018). Role of macronutrients and micronutrients in the growth and development of plants and prevention of deleterious plant diseases-a comprehensive review. *International Journal of Chemical and Biochemical Sciences* **13**, 31-52.

Nakasaki, K., Idemoto, Y., Abe, M., and Rollon, A. P. (2009). Comparison of organic matter degradation and microbial community during thermophilic composting of two different types of anaerobic sludge. *Bioresource technology* **100**, 676-682.

Ofei-Quartey, M. N. L., Appiah-Effah, E., Akodwaa-Boadi, K., Ampaw, B., Taylor, T. S., and Millogo, Z. E. N. (2023). Enhancing the economic potential of organic waste by co-composting using ratio modelling toward a circular economy. *Journal of Material Cycles and Waste Management* **25**, 1560-1580.

Ogunwande, G., Osunade, J., Adekalu, K., and Ogunjimi, L. (2008). Nitrogen loss in chicken litter compost as affected by carbon to nitrogen ratio and turning frequency. *Bioresource Technology* **99**, 7495-7503.

Preneta, N., Kramer, S., Magloire, B., and Noel, J.-M. (2013). Thermophilic co-composting of human wastes in Haiti. *Journal of Water, Sanitation and Hygiene for Development* **3**, 649-654.

Rasapoor, M., Nasrabadi, T., Kamali, M., and Hoveidi, H. (2009). The effects of aeration rate on generated compost quality, using aerated static pile method. *Waste Management* **29**, 570-573.

Raviv, M., Medina, S., Krasnovsky, A., and Ziadna, H. (2004). Organic matter and nitrogen conservation in manure compost for organic agriculture. *Compost Science & Utilization* **12**, 6-10.

Richards, L. (1947). Diagnosis and improvement of saline and alkaline soils. Vol. 64, pp. 432. LWW.

Ruiz-Llacsahuanga, B., Hamilton, A., Zaches, R., Hanrahan, I., and Critzer, F. (2021). Utility of rapid tests to assess the prevalence of indicator organisms (aerobic plate count, Enterobacteriaceae, coliforms, Escherichia coli, and Listeria spp.) in apple packinghouses. *International Journal of Food Microbiology* **337**, 108949.

SAG (2009). Fertilizers, Farm Feeds, Agricultural Remedies and Stock Remedies Act (Act No. 36 of 1947). (S. A. Government, ed.). Government Printer Pretoria.

Sanguinetti, G., Tortul, C., Garcia, M., Ferrer, V., Montangero, A., and Strauss, M. (2005). Investigating helminth eggs and Salmonella sp. in stabilization ponds treating septage. *Water Science and Technology* **51**, 239-247.

Sarkar, S., Seenivasan, S., and Premkumar, R. (2009). Biodegradation of propiconazole by Pseudomonas putida isolated from tea rhizosphere. *Plant soil environ* **55**, 196-201.

Smith, S. R. (2009). A critical review of the bioavailability and impacts of heavy metals in municipal solid waste composts compared to sewage sludge. *Environment international* **35**, 142-156.

STATS_SA (2022). General Household Survey 2021 Media Release. (S. S. Africa, ed.), pp. 2. https://www.statssa.gov.za/publications/P0318/GHS%202021%20MEDIA%20RELEASE.pdf.

Sullivan, D. M., and Miller, R. O. (2001). Compost quality attributes, measurements and variability.

Trautmann, N. M., and Krasny, M. E. (1998). "Composting in the classroom: Scientific inquiry for high school students," Kendall/Hunt Publishing Company.

Trémier, A. (2012). Home-made composts quality: Methods of assessment and results.

Ugak, M. A. M., Aji, N. A. S., Yaser, A. Z., Lamaming, J., Rajin, M., and Saalah, S. (2022). Food waste-dry leaves composting: mixture formulation, turning frequency and kinetic analysis. *Borneo Science| The Journal of Science and Technology* **43**.

United_Nations (2023). What share of people will live in urban areas in the future? Department of Economic and Social Affairs, Population Division

USEPA (2003). "Environmental Regulations and Technology Control of Pathogens and Vector Attraction in Sewage Sludge."

Varma, V. S., Muthusamy, S., and Rajendran, K. (2018). Organic waste and pollutants reduction through composting. *Waste Bioremediation*, 141-164.

Wang, G., Yang, Y., Kong, Y., Ma, R., Yuan, J., and Li, G. (2022). Key factors affecting seed germination in phytotoxicity tests during sheep manure composting with carbon additives. *Journal of Hazardous Materials* **421**, 126809.

Werner, K. A., Castro-Herrera, D., Yimer, F., Tadesse, M., Kim, D.-G., Prost, K., Brüggemann, N., and Grohmann, E. (2023). Microbial risk assessment of mature compost from human excreta, cattle manure, organic waste, and biochar. *Sustainability* **15**, 4624.

WHO (2006). "WHO guidelines for the safe use of wasterwater excreta and greywater," World Health Organization.

Wichuk, K. M., Tewari, J. P., and McCartney, D. (2011). Plant pathogen eradication during composting: a literature review. *Compost Science & Utilization* **19**, 244-266.

Wong, J., Mak, K., Chan, N., Lam, A., Fang, M., Zhou, L., Wu, Q., and Liao, X. (2001). Co-composting of soybean residues and leaves in Hong Kong. *Bioresource Technology* **76**, 99-106.

Wong, J. W., Selvam, A., Zhao, Z., Yu, S., Law, A. C., and Chung, P. C. (2011). Influence of different mixing ratios on in-vessel co-composting of sewage sludge with horse stable straw bedding waste: maturity and process evaluation. *Waste management & research* **29**, 1164-1170.

Yuan, J., Chadwick, D., Zhang, D., Li, G., Chen, S., Luo, W., Du, L., He, S., and Peng, S. (2016). Effects of aeration rate on maturity and gaseous emissions during sewage sludge composting. *Waste Management* **56**, 403-410.

Zhang, D., Luo, W., Li, Y., Wang, G., and Li, G. (2018). Performance of co-composting sewage sludge and organic fraction of municipal solid waste at different proportions. *Bioresource technology* **250**, 853-859.

Zhang, L., and Sun, X. (2014). Changes in physical, chemical, and microbiological properties during the two-stage co-composting of green waste with spent mushroom compost and biochar. *Bioresource technology* **171**, 274-284.

Zhao, Y., Lu, W., Damgaard, A., Zhang, Y., and Wang, H. (2015). Assessment of co-composting of sludge and woodchips in the perspective of environmental impacts (EASETECH). *Waste Management* **42**, 55-60.

Zorpas, A. A., and Loizidou, M. (2008). Sawdust and natural zeolite as a bulking agent for improving quality of a composting product from anaerobically stabilized sewage sludge. *Bioresource Technology* **99**, 7545-7552.

Chapter 4: Evaluating the Impact of Human Excreta-based Co-compost on Soil Health and Plant Growth: Assessing Nutrient Recycling and Phytotoxicity on Cucumber (*Cucumus sativus*) Root Morphology

Abstract

Co-composting is one of the most cost-effective and safe ways to recover nutrients from faeces and urine. However, phytotoxic compounds and heavy metal contamination are major issue of concern for its application to agricultural lands. Therefore, this study aims to understand the root architecture of phytotoxic sensitive crops like cucumber (*Cucumis sativus*) in response to co-compost. A rhizotron experiment was carried out comparing co-compost treatments to commercial Gromor compost and a control sandy loam KwaDinabakubo soil. The root system architecture measured were convex hull area (CHA), root branch count (RBC), total root length (TRL), root system depth (RSD), root system width (RSW), root mass ratio (RMR), shoot mass ratio (SMR), root-shoot mass ratio (RSMR), shoot-root mass ratio (SRMR), leaf area index (LAI), and leaf number (LN). The data collected was analyzed using variance analysis (ANOVA), principal component analysis (PCA), and correlation. There was a significant difference ($p<0.001$) in the root system architecture results where SRMR was higher with co-compost than soil and commercial compost. Moreover, commercial compost had higher root branch count and length than other treatments, indicating low nutrient availability and water-holding capacity. Significant ($p<0.05$) and positive correlation was observed between RSW and CHA ($r= 0.92$), RBC ($r= 0.76$) and RSD ($r= 0.75$). In conclusion, the study demonstrated that co-compost results in improved above-ground conditions and does not contain toxic compounds that inhibit the growth and development of both roots and shoots.

Keywords: Co-compost, rhizotron, root system architecture, phytotoxins, nutrient

4.1 Introduction

Recycling nutrients from human excreta improves soil fertility and enhances the soil's physical, chemical and biological properties. According to Manga et al. (2022), co-composting human waste presents an alternative and economically viable way of recycling nutrients from faeces. Different microbial communities degrade organic matter into simpler nutrients during composting, thus generating heat (Anwar et al., 2015). When the heat generated reaches temperature ≥ 55 °C (thermophilic temperature), pathogens are deactivated, ensuring the safety of the final product (Amery et al., 2020). Furthermore, several studies have proven that high thermophilic temperature led to the deactivation of faecal matter indicators such as *Escherichia coli*, *Salmonella*, faecal coliforms, faecal streptococci, *Shigella* species, *Enterococcus faecalis* and viable helminths eggs (Burge et al., 1978; El Hayany et al., 2021; Germer et al., 2010; Manga et al., 2021).

Pathogen deactivation in co-compost is one of the essential parameters used to assess the quality and stability of co-compost. However, testing co-compost's ability to promote plant growth and development is essential. Barral and Paradelo (2011) emphasised the importance of evaluating the absence of phytotoxic substances in the compost, which indicates the quality and proper handling of the composting process. Phytotoxic substances can cause a delay in germination and inhibit plant growth (Baumgarten and Spiegel, 2004). Phytotoxic substances (allelochemicals) can inhibit the growth of crops by affecting germination, growth and development (Scognamiglio et al., 2013).

The crop's growth is comprehensively evaluated by examining the plant's above-ground (shoot) and below-ground (root) portions (Putra and Ismoyojati, 2021). Roots are essential components of crop plants, serving critical functions in nutrient absorption and transport from the soil to aerial parts (Mohd-Radzman et al., 2013). Furthermore, root development is a key determinant of canopy growth and overall crop yield, underscoring their fundamental role in plant productivity (Ota et al., 2020). One important factor affecting dry mass accumulation in roots and shoots is nutrient deficiency (Putra and Ismoyojati, 2021). According to Cofie et al., (2005), co-compost made from human excreta is a good soil conditioner and provides recycled nutrient sources such as nitrogen (N), phosphorus (P) and potassium (K). These nutrients are essential for optimum growth and development of the crop. Root morphological traits such as the length of roots indicate a plant’s ability to acquire water and nutrients from the soil (Chen et al., 2018).

Laboratory-grown plants allow for more accessible analysis of root systems using media such as transparent soil, sand, or sieved soil combined with advanced imaging technologies (Giuliani et al., 2024). Rhizotron transparent soil systems designed for direct observation of root development offer an effective platform for examining plant-root interactions in an undisturbed soil environment (Putra and Ismoyojati, 2021). Specific toxic components, such as heavy metals, traditionally require costly and time-consuming analytical methods for detection (Barral and Paradelo, 2011). Rhizotrons, however, may provide a more affordable alternative for assessing phytotoxic compounds in co-compost, as root growth is susceptible to such toxins. Therefore, this study aims to evaluate cucumber (*Cucumis sativus*) root morphology in response to different co-compost amendments and to develop a novel, cost-effective method for assessing co-compost phytotoxicity on sensitive crop species.

4.2 Materials and methods

4.2.1 Composting

The co-composting process was conducted at the Controlled Environment Facility (CEF) at the University of KwaZulu-Natal, Pietermaritzburg campus under controlled conditions. Dewatered sewage sludge (DSS) and organic green waste (OGW) were mixed in two volume-based ratios, 1DSS:2OGW and 1DSS:3OGW, and these mixtures were subjected to turning frequencies (TF) of 7, 14, and 28 days, resulting in six treatments: T1, 1DSS:3OGW with a 7-day TF; T2, 1DSS:2OGW with a 28-day TF; T3, 1DSS:3OGW with a 14-day TF; T4, 1DSS:2OGW with a 7-day TF; T5, 1DSS:3OGW with a 28-day TF; and T6, 1DSS:2OGW with a 14-day TF as explained in. These treatment conditions allowed for investigating the effects of different mixing ratios and turning frequencies on the co-composting process and quality, as described in Chapter 3.

4.2.2 Plant material

Ashley cucumber variety from the Starke Ayres seeds used in this experiment was sourced from the Pick n Pay store, Hayfield Pietermaritzburg, South Africa (29°37'27.8"S and 30°24'33.5" E). Gromor commercial compost was sourced from the university storeroom on the university premises. The Cartref soil, classified as a Typic Hap aquept, was collected from an arable field in the KwaDinabakubo (KD) area, Hillcrest, South Africa, (29°46′48″S and 30°45′46″E).

4.2.3 Growing media

Co-compost treatments (T1, T2, T3, T4, T5 and T6) and sandy soil (KD) were sieved using a 5 mm sieve to remove large debris and larger particles of the soil. The treatments were established by mixing 0.5 kg of KD soil with 0.5 kg of either co-compost (T1, T2, T3, T4, T5 and T6) or Gromor commercial compost (TG) and 1 kg of KD soil which was used as a control (KD). The treatments (T1, T2, T3, T4, T5, T6, TG, and KD) were arranged in a single-factorial design in a Complete Randomized Design (CRD) with three replications.

4.2.4 Rhizotron fabrication

A root rhizotron was fabricated following the method described by (Mandizvo et al., 2022a). Transparent polycarbonate sheets of 2 mm thickness were cut into square sheets (S1 and S3) measuring 20 cm in length and 20 cm in width using a table saw (Ryobi, Hiroshima, Japan). Plastic tubes of 1.25 mm thickness and 1.2 cm diameter were cut into 45 cm pieces (S2). Pieces S1 and S3 were held together using a Grip GV9365 Bench Vice (100 mm) to allow the drilling of aligned pilot holes (Figure 4.1). Circular perforations with a diameter of 3 mm were drilled on S1 and S3 using a Ryobi 16 mm bench drill press on a level surface. Rhizotron pieces were assembled and secured using bolts and wing nuts.

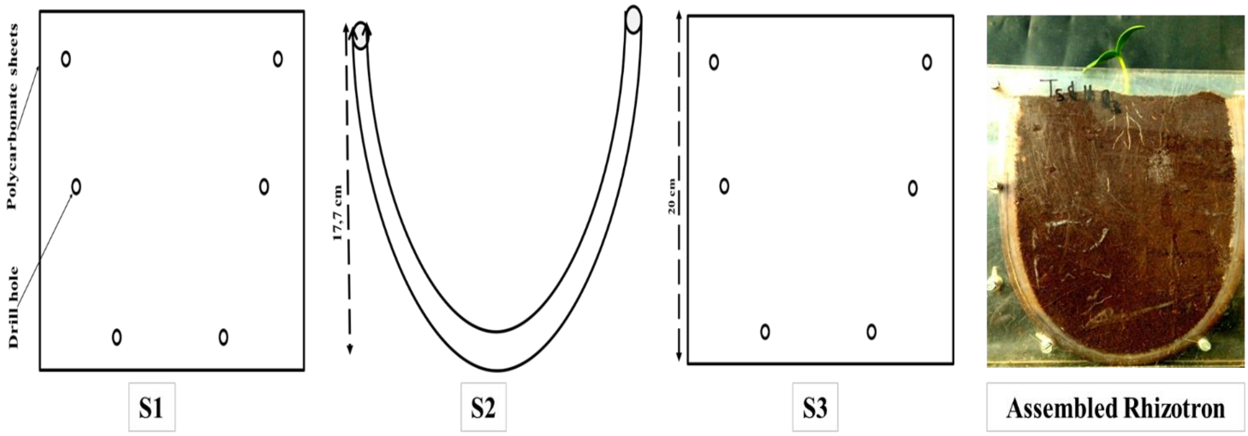


Figure 4.1: Sketch of the rhizotron design, transparent polycarbonate sheet with holes (S1 and S3), curved plastic tube (S2) and assembled rhizotron.

4.2.5 Experimental design and layout

The rhizotron experiment was conducted under controlled conditions in the tunnel at the Controlled Environment Facility (CEF) at the University of KwaZulu-Natal, Pietermaritzburg, South Africa (29°37'36.5" S and 30°24'14.7" E). The study was designed as a single-factorial design with 8 treatments replicated 3 times, giving 24 experimental units laid out in a CRD. Cucumber seeds were sown in each rhizotron filled with 50% co-compost (T1-T6) or compost (TG) and 50% sandy soil (KD).

4.2.6 Capturing root images

The camera positioning method maintained a consistent distance (50 cm) between the camera and the rhizotron, enabling time-series digital recording of root development. Root images were captured on both lateral sides of the rhizotron weekly for 4 weeks. An iPhone 13 camera of 12 megapixels (Apple Inc., One Apple Park Way, Cupertino, CA 95014 USA) was used to capture images. Camera settings included a resolution of 4:3, assistive grid on, exposure 0.0, flash off and timer of 3 s. Images were collected in the raw format with a colour depth of 10-bit (HDR) and an image size of 4032 × 3024 pixels.

4.2.7 Measurement of seedling axis

After terminating the experiment, the rhizotrons were unfastened, and the seedlings that emerged from the compost and sandy soil mixture were uprooted and washed to remove soil particles. Root length and shoot length were measured using a ruler. Measurements were done for root and shoot lengths using a computer-aided digital image analysis. Digital images were acquired at a scanning resolution of 250 dpi using a flatbed scanner (Hewlett Packard Scanjet 4c/t; Palo Alto, Calif.). Root and shoot length were quantified using Image software analysis. The paper towel was used to blot free surface moisture. Shoots, roots, and cotyledons were separated (Figure 4.2) by cutting at the soil line using a scalpel blade and oven-dried at 70 °C to measure the dry matter content of the plants. Based on the dry shoot and mass, the following parameters were calculated using the formula below:

$$\boldsymbol{SMR} = \frac{Total\ shoot\ dry\ mass\ (g)}{Total\ dry\ mass\ of\ the\ plant\ (g)}$$ Equation 4.1

SMR: shoot mass ratio

$$\boldsymbol{RMR} = \frac{Total\ root\ dry\ mass\ (g)}{Total\ dry\ mass\ of\ the\ plant\ (g)}$$ Equation 4.2

RMR: root mass ratio

$$\boldsymbol{RSMR} = \frac{Total\ root\ dry\ mass\ (g)}{Total\ shoot\ dry\ mass\ (g)}$$ Equation 4.3

RSMR: root shoot mass ratio

$$\boldsymbol{SRMR} = \frac{Total\ shoot\ dry\ mass\ (g)}{Total\ dry\ dry\ mass\ (g)}$$ Equation 4.4

SRMR: shoot-root mass ratio

Figure 4.2: Visual presentation of the roots, shoots and cotyledons of cucumber plants grown using different soil growing media, where KD (**a**), T1 (**b**), T2 (**c**), T3 (**d**), T4 (**e**), T5 (**f**), T6 (**g**) and TG (**h**).

4.2.8 Data collection and image analysis

Root Snap software (Version 1.3.2.25, CID Bio-Science Inc., Camas, WA, USA) was used to analyse 2D images of plant roots taken from the sides of the rhizotron, following the method outlined by (Mandizvo et al., 2022b). The software automatically predicted root features, with manual adjustments as needed. Architectural traits of the root system (Table 4.1) were quantified with the help of this user-assisted image analysis software on a Microsoft Surface tablet, where a Surface Pro 4 stylus was used to trace root structures. Data on root system width

(RSW), root system depth (RSD), convex hull area (CHA), total root length (TRL), and root branch count (RBC) were collected weekly.

Table 4.1: Description of the measured traits in the cucumber plants grown with different soil amendments and in the root rhizotron and assessed from week 1 after planting to week 4 after planting

Roots traits	Definition	Units
Root dry mass (RDM)	Total dry mass of roots per plant	g
Root mass ratio (RMR)	Dry mass of roots divided by the total dry mass of the plant	
Root-shoot mass ratio (RSMR)	Total root dry mass divided by total shoot mass	
Shoot dry mass (SDM)	Total dry mass of shoot per plant's dry mass	g
Shoot mass ratio (SMR)	Dry mass of the shoot divided by total dry mass of the plant	
Shoot-root mass ratio (SRMR)	Total shoot dry mass divided by total root mass	
Leaf area index (LAI)	Area of the leaf	cm^2
Leaf number (LN)	Number of leaves per plant	-
Root length (RL)	Length of the tap root	cm
Shoot length (SL)	Length of the shoots	cm
Convex hull area (CHA)	The area that encompasses the root system	cm^2
Root branch count (RBC)	Lateral root number emerging from the primary root	-
Root system width (RSW)	Maximum horizontal distribution of the roots	cm
Root system depth (RSD)	Maximum vertical depth of the root system	cm
Total root length (TRL))	Total root length of seminal and lateral root	cm

4.2.9 Statistical analysis

Analysis of variance for root traits was computed using GenStat 23rd Edition (VSN, International, Hemel, Hempstead, United Kingdom). Mean separation was performed using Fisher's protected least significant difference (LSD) test when treatments showed differences at a 5% level of significance. Principal component analysis (PCA) PC biplots and Pearson correlations were done using Origin 2024b (Origin Lab Corporation).

4.3 Results and discussion

4.3.1 Nutrient composition

Table 4.2 shows the ANOVA results comparing the variance between treatments and residuals (error) for multiple nutrients (N, P, K, Ca, Mg, Fe, Mn, Zn, Cu, Na, Al, and moisture). The analysis indicates a significant difference in nutrient composition among the treatments.

Table 4.2: Analysis of variance showing mean squares and significant tests for macro-nutrients, micro-nutrients and moisture of 8 growing media treatments

S.O.V	**d.f**	**N**	**P**	**K**	**Ca**	**Mg**	**Fe**
Treatment	7	0.798^{**}	0.163^{**}	0.065^{**}	2.798^{**}	0.047^{**}	$1.857\times10^{8**}$
Residual	16	3208×10^{-4}	5.003×10^{-5}	7.551×10^{-5}	1.594×10^{-4}	4.036×10^{-4}	6.570×10^{3}
S.O.V	**d.f**	**Mn**	**Zn**	**Cu**	**Na**	**Al**	**Moisture**
Treatment	7	1.636×10^{5}	4.921×10^{4}	$3.530\times10^{3**}$	$1.079\times10^{5**}$	$3568\times10^{7**}$	326.674^{**}
Residual	16	1.415	4.804×10^{-1}	3.282×10^{-2}	1.025×10^{-1}	1.656×10^{-1}	0.012

S.O.V; source of variation, **d.f**; degrees of freedom, **N**; nitrogen, **P**; phosphorous, **K**; potassium, **Ca**; calcium, **Mg**; magnesium, **Fe**; iron, **Mn**; manganese, **Zn**; zinc, **Cu**; copper, **Na**; sodium, **Al**; aluminium

4.3.2 Nutrient composition of growing media

Table 4.3 below shows the nutrient composition of the growing media (co-compost (T1-T6), KwaDinabakubo soil (KD), and Gromor compost (TG)) used to grow cucumber seedlings. There was a significant difference ($p<0.001$) between growing media in all the nutrients and moisture content. The highest nitrogen content (1.412%) was observed in T5, the co-compost which was made using the 1DSS:3OGW mixing ratio and had a 28-day TF. Moreover, all co-compost treatments exhibited higher N than commercial compost (Gromor), which had 0.434% N and was greater than 0.102% N found in the sandy KD soil. According to Toor et al., (2021), N is an important macronutrient responsible for plants' rapid growth, the quality and sizes of leaves, and enhanced seed and fruit development. Moreover, it has the essential role of forming amino acids that are structural blocks of enzymes and proteins and is also involved in many biochemical processes as a catalysing agent (Leghari et al., 2016).

Phosphorus is a vital plant macronutrient for synthesising energy molecules like adenosine triphosphate (ATP), nucleic acids, and carbohydrates (Toor et al., 2021). The highest P content was observed in the co-compost treatments with values ranging between 0.468-0.571%, while the Gromor (TG) had a P content of 0.055% and the lowest was observed in the KD soil with a value of 0.008%. This implies that essential nutrients were indeed recovered from human excreta (dewatered sewage sludge), as emphasised by (Manga et al., 2022) that nutrients from faecal matter, detergents and urine are recovered during sewage sludge co-composting. Potassium is the second most taken mineral in the soil after nitrogen, thus indicating its importance as a macronutrient in the soil or growing media. High K was observed in the co-

compost treatments; however, there was a statistically insignificant difference between commercial compost (Gromor) and T6, while the sandy KD soil had the lowest K content compared to all treatments.

Micronutrients including zinc (Zn), copper (Cu), molybdenum (Mo), iron (Fe), manganese (Mn) and boron (B) are important plant nutrients which are taken by plants in smaller quantities (Tripathi et al., 2015). According to Toor et al., (2021), micronutrients affect lignin and phenolic content and membrane stability, which signifies their importance in the crops. All co-compost treatments exhibited higher Fe, Mn, Zn, Cu and Al than commercial compost (Gromor) and KD soil. Notably, the KD soil had the lowest Mn (41.200 mg/kg), Cu (6.590 mg/kg), and Zn (8.200 mg/kg), which signifies that most soils have micronutrient deficiencies. Yet the application of micro-nutrients, especially in sub-Saharan Africa, is very scant (Berkhout et al., 2019). Plant growth and proper development depend solely on these nutrients and other essential materials such as water, carbon dioxide, light, and environmental conditions.

Table 4.3: Nutrient composition of co-compost (T1-T6), commercial compost (TG) and KwaDinabakubo sandy soil (KD)

Treatment	**N (%)**	**P (%)**	**K (%)**	**Ca (%)**	**Mg (%)**	**Fe (mg/kg)**	**Mn (mg/kg)**	**Zn (mg/kg)**	**Cu (mg/kg)**	**Na (mg/kg)**	**Al (mg/kg)**	**MC (%)**
T1	1.362^{d}	0.515^{c}	0.400^{b}	2.131^{e}	0.332^{b}	29549^{c}	659.200^{d}	280.700^{c}	91.440^{b}	637.900^{e}	17169^{c}	27.650^{c}
T2	1.383^{b}	0.570^{a}	0.426^{a}	2.583^{a}	0.387^{a}	33415^{a}	782.800^{a}	280.800^{c}	84.240^{c}	765.800^{a}	18643^{b}	29.020^{b}
T3	1.372^{c}	0.510^{c}	0.366^{cd}	2.177^{d}	0.331^{b}	27499^{f}	684.700^{b}	272.200^{d}	72.720^{f}	595.400^{f}	15714^{e}	21.930^{f}
T4	1.348^{e}	0.571^{a}	0.354^{d}	2.321^{c}	0.340^{b}	32130^{b}	637.100^{f}	379.600^{a}	92.950^{a}	741.800^{b}	18903^{a}	29.110^{b}
T5	1.412^{a}	0.468^{d}	0.372^{c}	2.383^{b}	0.347^{b}	29027^{d}	673^{c}	292.600^{b}	75.240^{d}	668.800^{d}	16340^{d}	24.700^{e}
T6	1.304^{f}	0.543^{b}	0.131^{e}	2.317^{c}	0.362^{ab}	27994^{e}	644.600^{e}	248.600^{e}	74.140^{e}	716.200^{c}	15701^{f}	26.980^{d}
TG	0.434^{g}	0.055^{e}	0.131^{e}	0.445^{f}	0.118^{c}	12670^{h}	447.700^{g}	50.700^{f}	12.670^{g}	295.600^{g}	7851^{h}	36.890^{a}
KD	0.102^{h}	0.008^{f}	0.057^{f}	0.082^{g}	0.049^{d}	14365^{g}	41.200^{h}	8.200^{g}	6.590^{h}	295.600^{g}	15316^{g}	1.16^{g}
lsd	0.010	0.012	0.015	0.022	0.035	140.300	2.059	1.200	0.314	0.554	0.705	0.192
cv(%)	0.500	1.700	3.100	0.700	7.100	0.300	0.200	0.300	0.300	0.100	0.100	0.500
p-value	<.001	<.001	<.001	<.001	<.001	<.001	<.001	<.001	<.001	<.001	<.001	<.001

N; (nitrogen), **P**; (phosphorus), **K**; (potassium), **Ca**; (calcium), **Mg**; (magnesium), **Fe**; (iron), **Mn** (manganese), **Zn** (zinc), **Cu** (copper), **Na** (sodium), **Al** (aluminium), **MC** (moisture content), **l.s.d** (least significant difference), **cv** (cumulative variance), **P- value** (probability value)

4.3.3 Root and shoot dry matter of cucumber plants with different treatments

There was a significant difference ($p<0.001$) between the treatments in the root dry mass (RDM), root mass ratio (RMR), and root to shoot mass ratio (RSMR), shoot dry mass (SDM), shoot mass ratio (SMR) and shoot to root mass ratio (SRMR) as shown in Figure 4.2. Plant dry matter accumulation is influenced by multiple factors, with nutrient deficiency being a critical determinant (Putra and Ismoyojati, 2021). Plants with low dry matter accumulation are typically etiolated and produce fewer shoots, primarily due to limited nutrient availability.

Gromor (TG) and KD had the lowest RDM of 0.031 g and 0.028 g, respectively. The highest (0.085 g) RDM was observed in T1, while all treatments with a higher proportion of sludge

(T2, T4 and T6) had a lower RDM. According to Fan Jianbo et al., (2010), increased RL or biomass is greatly influenced by N availability. Soils with less N tend to have increased or invested root biomass production as the roots search for nutrients and water in the soil (Mašková and Herben, 2018). The nutritional results of the growing media shown in Table 4.3 indicate that KD, G and T6 had 0.102%, 0.434% and 1.304% N content, respectively. The low availability of nutrients in KD and TG is coupled and explained by an increased RMR. This increase in RMR indicates that the root biomass was higher in the cucumber seedlings compared to the shoot.

However, the relatively high shoot and root dry mass observed in other co-compost treatments could have been brought by the high N found in those treatments. Moreover, the nutrients supplied by co-compost nutrients to the above ground (shoot) improve its mass. Nitrogen is the main constituent of protein, as well as chlorophyll (Du et al., 2017). The availability of N in the co-compost could have contributed to high root and shoot dry mass in these treatments. Moreover, co-compost is a multi-nutrient source as it supplies both macro-nutrients and essential micro-nutrients.

According to Gruber et al., (2013), root biomass decreased comparatively less than the shoot, and the root-to-shoot mass ratio gradually increased with decreasing N supply. Microbes (bacteria, fungi and nematodes) are known to affect biogeochemical processes and the soil's root morphology (Larsen et al., 2015). The microorganisms decompose the soil organic matter, and consequently, the amounts of nutrients (N and P) increase, affecting the morphological traits in roots (Barrios-Masias et al., 2019). Plants preferentially allocate more biomass to leaves and stems in nutrient-rich environments with intense above-ground competition for light. However, in nutrient-poor soils or environments where below-ground competition for resources is more pronounced, they invest a higher proportion of biomass into root development (Mašková and Herben, 2018).

Significant differences ($p < 0.001$) were observed in RSMR among treatments (Figure 4.2C). Gromor and KD soil had the higher RSMR biomass, indicating that the roots were higher, resulting in very low above-ground biomass. This was due to the low nutrient levels in the Gromor commercial compost and KwaDinabakubo soil, which contained 0.434% and 0.102% nitrogen (N) and 0.055% and 0.008% phosphorus (P), respectively. The results align with those obtained by Lopez et al. (2023), who found that the nutrient-deficient soils had higher RSMR than nutrient-rich soils. Co-compost treatments were comparably different, with the treatments

(T1, T2, T3, and T5) having significantly different RSMRs than T4 and T6. The co-compost treatments T1, T3 and T5 were made using the lower proportion of dewatered sewage sludge (DSS) mixed with organic green waste (OGW) at the ratio of 1-part of DSS and 3-parts of OGW in volume-by-volume bases, meaning the mixture had low nitrogen to carbon content. Moreover, this mixing ratio is not compact. It has a low water-holding capacity compared to 1DSS:2OGW, which has a high water-holding capacity because of the low surface area between compost particles. Similarly, Gruber et al. (2013) found a 48% increase in total RL under N-deficient soils, decreasing shoot length and biomass, thus resulting in a decreased root-to-shoot mass ratio.

The shoot-to-root mass ratio (SRMR) is a valuable indicator of plant performance, reflecting the balance between above-ground biomass and overall growth. Treatment T6 exhibited the highest SRMR (6.82), with a value statistically significantly higher than those of treatments T4 (6.34) and T2 (5.77). According to Lynch et al. (2012), increasing N supply increases shoot and root development, and shoot growth is more prolonged. In contrast to the root-to-shoot mass ratio, the shoot-to-root mass ratio represents the relative mass of above-ground biomass compared to below-ground biomass. A higher SRMR indicates that the mass of the shoot is relatively greater than that of the roots, suggesting an allocation of resources favouring above-ground growth over root development. This metric is useful for understanding how plants allocate biomass under different environmental or treatment conditions, potentially reflecting adaptations to nutrient availability, light, or water resources (Mandizvo and Odindo, 2019; Taiz et al., 2015). Moreover, it shows that nutrients and minerals were readily and abundantly available in the growing media or the soil. This caused the plant roots to easily access and transfer them to the shoot for optimum photosynthesis. The lowest SRMR was obtained in the Gromor commercial compost (TG), while other treatments (T1, T3 and T5) were relatively not statistically significantly different, as shown in Figure 4.2F. All treatments with a higher proportion of sludge (1DSS:2OGW), which were T2, T4 and T6, resulted in a higher SRMR than the 1DSS:3OGW treatments (T1, T3 and T5). This could indicate that nutrients at a higher proportion of sludge are readily available to plants and were quickly acquired by cucumber roots.

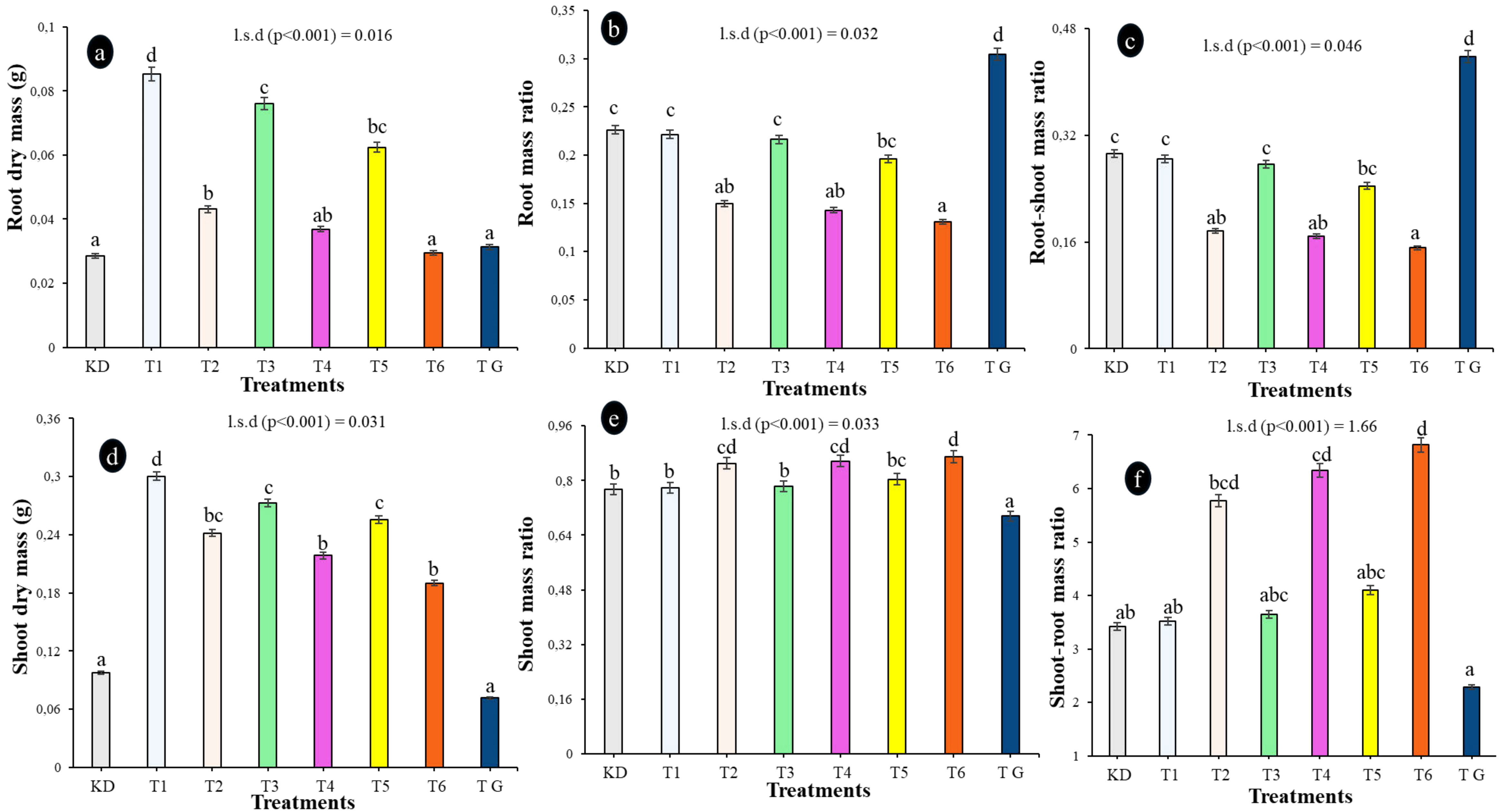


Figure 4.3: Root dry mass (**A**), root mass ratio (**B**), root-shoot mass ratio (**C**), shoot dry mass (**D**), shoot mass ratio (**E**), shoot-root mass ratio (**F**) of cucumber planted using KwaDinabakubo soil (KD), co-compost (T1, T2, T3, T4, T5 and T6), and Gromor commercial compost (TG). The same letters in each graph show statistically insignificant results, while different letters indicate a significant difference between treatments ($p<0.05$) using Tukey's 95% confidence interval.

4.3.4 *Vegetative growth indicators*

Leaf area index (LAI) is the main factor driving carbon and water exchange during plant development (Schickling et al., 2010). LAI is directly linked to the crop yield and can be used to predict yield and optimize planting densities. There was a significant difference ($p<0.01$) in the leaf number (LN) and LAI of the cucumber seedlings in different treatments. The commercial Gromor compost (TG) and sandy soil (KD) had the lowest LAI of 3.24 and 5.53 cm^2, respectively. The co-compost treatments resulted in LAI ranging between 25.27 cm^2 and 42.36 cm^2, the highest of which was obtained in T1. Lower LAI in TG and KD could indicate a lack of nutrients in the media used to grow cucumber plants. This show that co-compost subsequently added beneficial macro and micro-nutrients, which enhanced the above-ground or shoot of the cucumbers, thus contributing to improved LAI in the co-compost treatments. The outstanding performance of T1 (1DSS:3OGW_7TF) could have been constituted by high quality and indicated that the co-compost was matured and phytotoxic-free. There were no significant differences between T3 and T5, which were constituents of 1DSS:3OGW. This reveals that the mixing ratio did not differ despite different turning frequencies of 14 and 28 days, respectively. Moreover, T4 and T6 (1DSS:2OGW) did not statistically differ at the turning interval of 7 and 14 days, as shown in Figure 4.3a.

Seedlings invariably develop roots first (to obtain water); their relative investment into leaves versus roots as carbon- and nutrient-acquiring organs, respectively, can also change over time. Nitrogen deficiency stimulated the growth of a more exploratory root system with longer lateral roots (Gruber et al., 2013). Moreover, a plant's ability to acquire, utilize, and conserve resources is influenced by its leaf and root traits, which are also integral to defining its overall quality strategy (Reich et al., 2003). In this study, co-compost treatment significantly differs from commercial compost and soil ($p<0.01$). This could have been caused by low nutrient levels in both G and KD (Table 4.3) while co-compost resulted in significantly high nutrients. There was a significant difference in the shoot length between treatments ($p<0.01$). Gromor and KD had the shortest shoot length compared to all the co-compost treatments. The highest shoot length was observed in the T1 treatment. Moreover, there was no significant difference between T2, T3 and T4, indicating that nutrients were evenly distributed. Plants distribute higher proportions of biomass into leaves and stems in nutrient-rich environments with intense above-ground competition for light (Mašková and Herben, 2018).

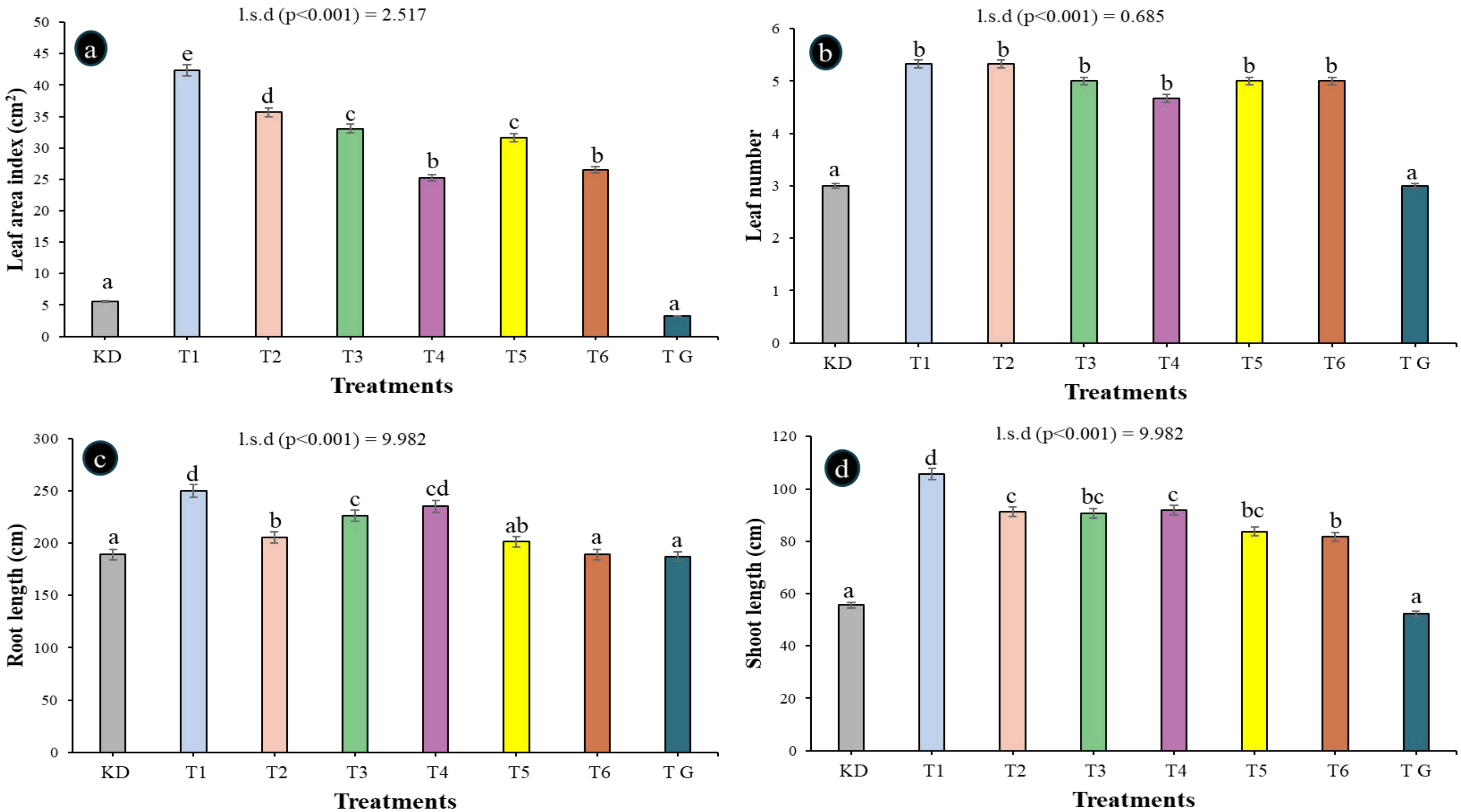


Figure 4.4: Leaf index (**A**), leaf number (**B**), root length (**C**), and shoot length (**D**) of cucumber seedlings in week 4 after planting grown using KwaDinabakubo soil (KD), different co-compost treatments (T1, T2, T3, T4, T5 and T6), commercial Gromor compost (TG), and graphs showing means ± standard deviation and differences using Tukey's 95% confidence interval.

4.3.5 *Root morphological traits*

Different treatments significantly differed ($p<0.001$) for convex hull area (CHA), root branch count (RBC), root system depth (RSD), root system width (RSW) and total root length (TRL) (Figure 4.4). An increase in these variables was also observed from week 1 to week 4 after planting. The highest CHA (172.4 cm^2) in week 4 was observed in KD soil-growing media, while the lowest CHA (110.4 cm^2) was recorded in the co-compost treatments T2 and T3. According to Rangarajan and Lynch (2021), a larger convex hull in roots indicates that roots were expanding in search of nutrients and water in the soil. In the present study, KD resulted in a larger convex hull, which indicates that roots were expanding to access nutrients and water in the soil. The KD soil had fewer macronutrients N (0.102%), P (0.008%) and K (0.057%) and low moisture content (1.16%), which subsequently resulted in low water holding capacity and higher CHA.

Root branch count per week increased at a higher rate in KD, T6 and TG (> 63.3 branch $week^{-1}$) compared to T1, T2, T3, T4, and T5, which were characterised by a small branch rate (≤ 59.0 branch $week^{-1}$). The root system depth was higher in KD, T6, T5 and TG (≥ 15.0 cm) compared to T1, T2, T3, and T4, which were characterised by RSD ranging between 13.1 cm to 14.3 cm. The RSW differed statistically between different treatments ($p<0.001$). In week 4, the highest RSW (≥ 12.7 cm) was observed in the KD treatment, while the smallest RSW was obtained in T3, T4, T5 and TG with a mean ≤ 10.732 cm. Total root length increased rapidly weekly, indicating that the roots expanded across different treatments. In week 4, the highest RL was observed in the KD, T4 and TG treatments, ranging between 70.1 cm to 74.6 cm. The lowest TRL was observed in treatments T3, T5, T6, T1 and T2, respectively. Commercial compost (TG) is composed of larger organic particles, resulting in low water holding capacity, making the plants increase root or have high root development in search for water and nutrients. Furthermore, this observation suggests increased RBC and TRL, reducing shoot growth. This shift likely occurs because the plant reallocates energy and resources toward root development rather than shoot expansion, a strategy often employed under nutrient-limited conditions to enhance nutrient uptake efficiency (Mandizvo et al., 2022b).

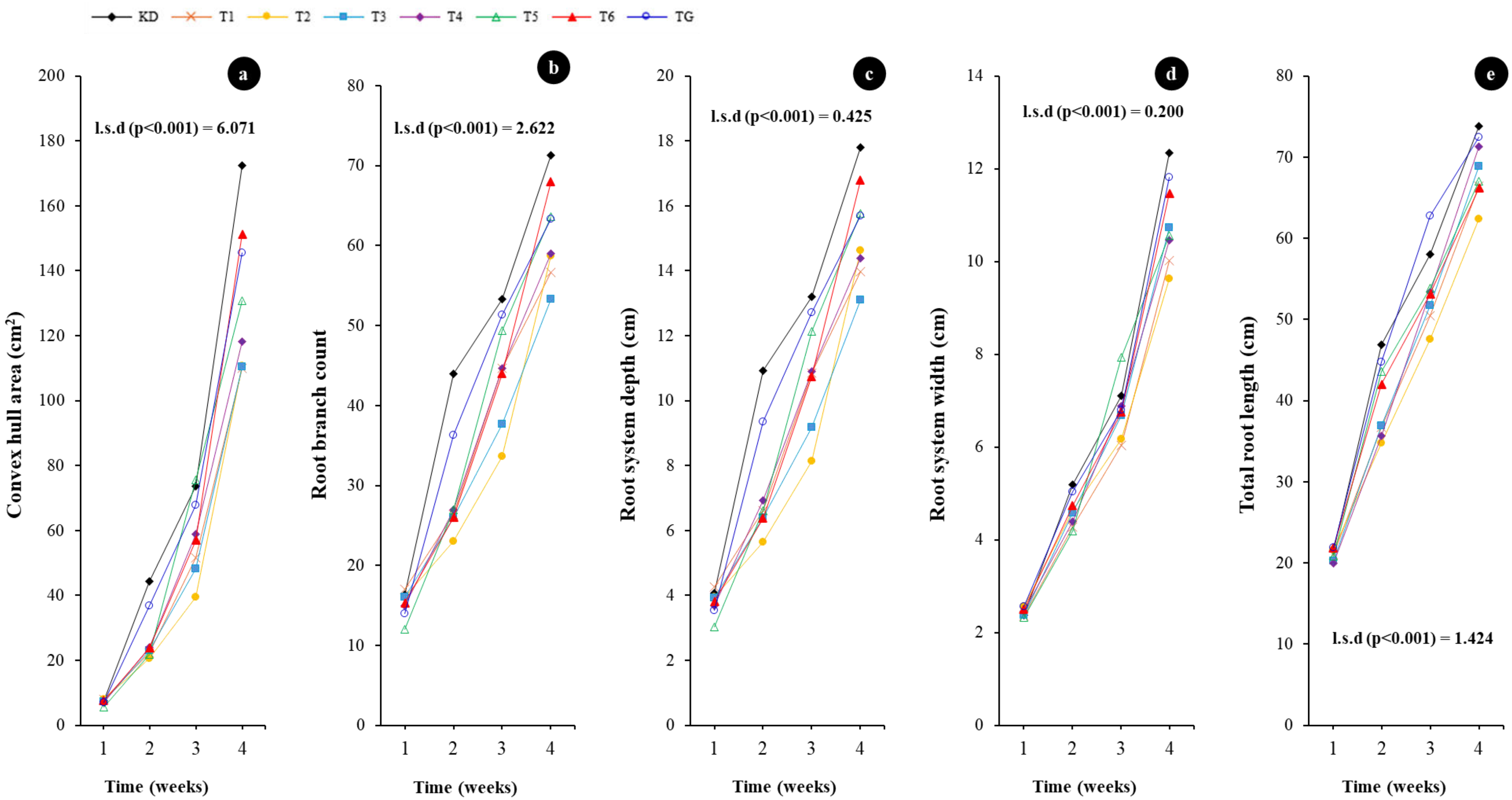


Figure 4.5: Root phenes for different treatments (KD, T1, T2, T3, T4, T5, T6, and TG), (**a**) convex hull area, (**b**) root branch count, (**c**) root system depth, (**d**) root system width, (**e**) total root length.

4.3.6 Rate of growth

The CHA, RBC, RSD, RSW and TRL growth rates differed significantly among treatments ($p<0.05$). The KD treatment obtained the highest CHA, indicating that the media had low nutrient content. According to Bontpart et al. (2020), root growth enlarges the CHA to expand the plant-soil interface depending on nutrient availability. Roots were branching at a higher rate in T5, T6, KD, and TG with values of 17.73, 17.60, 17.43, and 16.29 branch day^{-1}, respectively. These results indicate that these treatments were rapidly branching because a higher proportion of biomass was invested in root formation in search of nutrients and water in the soil or growing media. Root system depth (RSD) and RSW were relatively higher in the KD treatment than in other treatments. This correlates with high RSD and RSW observed in KD, as observed in Figure 4.4. TRL grew faster in KD, TG, and T4, indicating a high investment in underground mining for essential minerals and nutrients. However, T4 had the balance between the below and above-ground biomass as it resulted in an improved shoot-to-root mass ratio (6.34) compared to KD (3.42) and KD (2.28) (Figure 4.2).

Table 4.4: Daily root growth rates of cucumber root traits measured in sandy soil (KD), co-compost treatments (T1, T2, T3, T4, T5, and T6), and commercial Gromor compost (TG) from week 1 to week 4 after planting.

Treatments	**CHA cm week^{-1}**	**RBC branch week^{-1}**	**RSD cm week^{-1}**	**RSW cm week^{-1}**	**TRL cm week^{-1}**
KD	52.36^{e}	17.43^{e}	4.35^{fg}	3.18^{f}	16.79^{e}
T1	33.55^{ab}	13.73^{b}	2.34^{a}	2.47^{b}	15.01^{c}
T2	32.73^{a}	13.87^{b}	3.46^{c}	2.28^{a}	13.43^{a}
T3	32.43^{a}	12.37^{a}	3.03^{b}	2.72^{d}	16.06^{d}
T4	36.83^{b}	14.97^{c}	3.61^{d}	2.65^{c}	17.17^{f}
T5	42.95^{c}	17.73^{e}	4.37^{g}	2.85^{de}	14.91^{c}
T6	46.45^{d}	17.60^{e}	4.32^{f}	2.89^{e}	14.40^{b}
TG	44.63^{cd}	16.29^{d}	3.99^{e}	2.95^{e}	16.98^{e}
l.s.d	2.511	0.441	0.145	0.099	0.191
CV (%)	3.400	1.500	2.100	1.900	0.700
p. value	<0.001	<0.001	<0.001	<0.001	<0.001

CHA (convex hulls area), **RBC** (root branch count), **RSD** (root system depth), **RSW** (root system width), **TRL** (total root length), **l.s.d** (least significant difference), **cv** (cumulative variance)

4.3.7 Pearson correlation analysis showing the relationship between root traits of cucumber seedlings grown in co-compost, commercial compost and sandy soil.

Figure 4.5 shows the Pearson correlation coefficient of root and shoot traits of cucumber plants grown using co-compost, commercial compost and sandy loam soil. Significance differences and positive correlations were observed between LAI and N ($r=0.91$; $p<0.05$), P ($r= 0.90$; $p<0.05$), K ($r=0.85$; $p<0.05$), Ca ($r=0.89$; $p < 0.05$), Fe ($r = 0.90$; $p<0.05$), Mn ($r =0.83$; $p<0.05$), Zn ($r= 0.92$; $p< 0.05$), Na ($r =0.84$; $p<0.05$) and Al ($r=0.71$; $p< 0.05$). According to Lavon et al. (1999), N, K, Mg and Ca are the essential elements required for metabolic and structural function, which enables the plant to improve photosynthetic capacity and produce bigger leaves to capture photosynthetically active radiation (PAR) for optimum plant growth. Significant and negative correlations were observed between LAI and CHA (r=-0.81; p< 0.05), RSW (r= -0.88; p< 0.05), and TRL (r = -0.80; p < 0.005). Plants with a larger convex hull area are often nutrient-deficient, a condition that may arise due to water stress, as observed by (Mandizvo et al. 2022b). These results explain the high CHA observed in KD soil (Figure 4.4c) with low nutrient content (Table 4.2). A significant and negative correlation was observed between RMR and P (r=-0.75; p<0.05), Ca (r=-0.73; p<0.05), Mg (r=-0.71; p< 0.05), and Fe (r=-0.79; p<0.05). Significant differences with negative correlation were also observed in the RSMR, which was similar to the RMR. However, there was a significant and positive correlation between SMR and P (r=0.75; p<0.05), Ca (r=0.73; p< 0.05), Mg (r=0.71; p< 0.05), Fe (r=0.79; p<0.05), Na (r=0.84; p<0.05) and Al (r = 0.82; p< 0.05). Soil nutrient availability promotes greater above-ground biomass in plants, as most biomass is allocated to shoot growth rather than root development, resulting in a higher SRMR and SMR (Mašková and Herben, 2018).

The RMR and SMR had a significant and negative correlation between each other (r=-1.00; p<0.005). Significant (p<0.05) and negative correlation was observed between CHA and nutrients such as N (r =-0.80), P (r = -0.76), K (r = -095), Ca (r =-0.75), Mg (r =-0.75), Fe (r = -0.76), Zn (r=- 0.78), and Cu (r = -0.80). Significant (p< 0.05) and positive correlations were observed between RSW and CHA (r = 0.92), RBC (r =0.76) and RSD (r =0.75). However, significant (p<0.05) and negative correlation were observed between RSW and nutrients including N (r = -0.83), P (r = - 0.82), K (r =-0.96), Ca (r=-0.83), Mg (r =-0.83), Fe (r = -0.87), Zn (r = -0.82), Cu (r = -0.84) and Na (r = -0.80). A significant (p<0.05) and negative correlation was observed in the macronutrient K and RBC (r=-0.83) and RSD (r=-0.82); however, a positive correlation between CHA and RSD (r=0.95) and RBC (r=0.95). This relationship outlines a lack of nutrients in the growing media, which usually invests most of its biomass in the below ground for root development in search of nutrients. The root-to-shoot mass ratio (RSMS) is positively correlated with RMR (r=1.00), while it is negatively correlated with SMR (r=-1.00). These correlations imply that a relationship between nutrients and plants is essential and can determine the optimum growth of the plant.

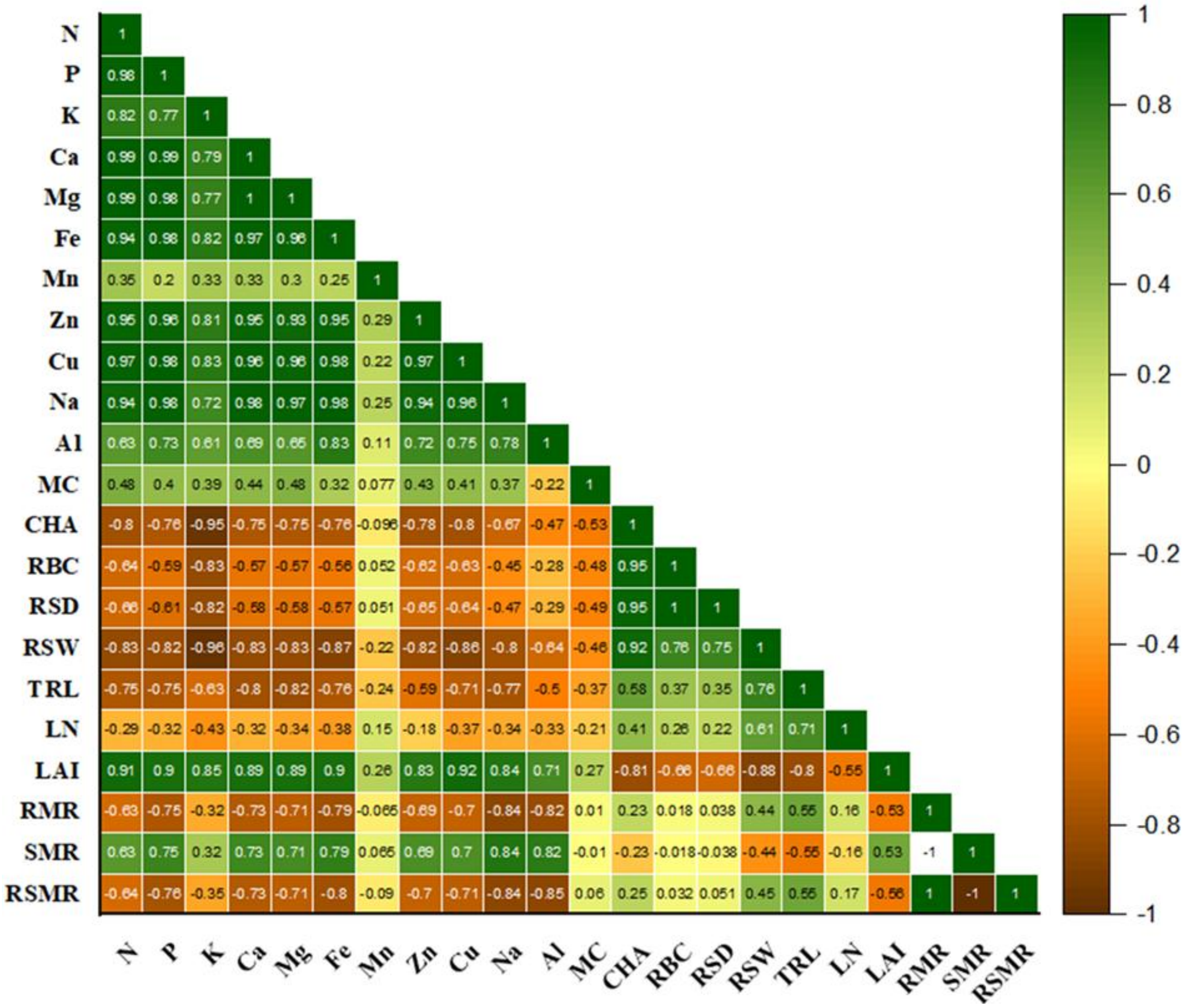


Figure 4.6: Pearson correlation coefficient for root phenes, root system architecture, leaf traits and allometry of cucumber plants grown using different growing media (co-compost, commercial compost and sandy soil).

4.3.8 Principal component analysis (PCA) for the root system architecture of cucumber plants grown using co-compost, commercial compost and sandy soil

Table 4.5 represents factor loadings, eigenvalue, cumulative and variability percentages of evaluated root traits. PC1 accounted for 66.611% variability and was weakly positively correlated with N, P, K, Ca, Mg, Fe, Al, Zn, Cu, MC, LAI and SMR. PC2 was positively correlated with P, Ca, Mg, Fe, Mn, Zn, Cu, Na, Al, CHA, RBC, RSD, RSW, LN and SMR with variability of 15.457% and the cumulative value of 82.068%. PC3 was positively correlated with N, P, K, Mg, Fe, Ca, Mn, Zn, Na, MC, CHA, RBC, RSD, RSW, TRC, LN, RMR and RSMR and there was a 6.131% variability with 88.198% cumulative value. Nitrogen, Ca, Mg, Mn, MC, CHA, RBC, RSD and RSMR were positively correlated with PC4, accounting for 5.378% variability and 93.577% cumulative.

Table 4.5: Factor loading, eigenvalue, variability and cumulative percentages (%) for roots, shoots and leaf traits of cucumber plants grown using eight (8) different growing media and their relationship with nutrients

Variables	PC1	PC2	PC3	PC4
N	0.253	-0.031	0.159	0.059
P	0.256	0.043	0.064	-0.043
K	0.225	-0.208	0.006	-0.013
Ca	0.256	0.033	0.118	0.075
Mg	0.254	0.020	0.094	0.097
Fe	0.259	0.068	0.006	-0.027
Mn	0.063	0.025	0.532	0.527
Zn	0.249	0.009	0.204	-0.117
Cu	0.257	0.006	0.042	-0.046
Na	0.252	0.119	0.053	0.038
Al	0.199	0.233	-0.165	-0.228
MC	0.105	-0.280	0.177	0.265
CHA	-0.216	0.290	0.036	0.135
RBC	-0.169	0.372	0.012	0.291
RSD	-0.171	0.363	-0.022	0.315
RSW	-0.237	0.157	0.143	-0.076
TRL	-0.206	-0.017	0.254	-0.431
LN	-0.110	0.089	0.678	-0.363
LAI	0.243	-0.060	-0.110	0.076
RMR	-0.184	-0.370	0.023	0.097
SMR	0.184	0.370	-0.023	-0.097
RSMR	-0.187	-0.369	0.031	0.105
Eigenvalue	14.654	3.400	1.349	1.183
Variability (%)	66.611	15.457	6.131	5.378
Cumulative (%)	66.611	82.068	88.198	93.577

N (nitrogen), **P** (phosphorus), **K** (potassium), **Ca** (calcium), **Mg** (magnesium), **Fe** (iron), **Mn** (manganese), **Zn** (zinc), **Cu** (copper), **Na** (sodium), **Al** (aluminium), **MC** (moisture content), **CHA** (convex hull area), **RBC** (root branch count), **RSD** (root system depth), **RSW** (root system width), **TRL** (total root length), **LN** (leaf number), **LAI** (leaf area index), **RMR** (root mass ratio), **SMR** (shoot mass ratio), **RSMR** (root shoot mass ratio)

Figure 4.6 shows the PC biplots based on the PCA analysis, which were used to visualize the relationship between cucumber root traits, above-ground traits and nutrients. Traits represented by parallel vectors close to each other were observed to have a strong positive correlation. In our present study, the analysis shows that co-compost treatments T2, T4, and T6 constitute a higher proportion of sludge (1DSS:2OGW) and T5 (1DSS:3OGW) was strongly positively correlated with high SMR. Moreover, these treatments were also positively correlated to Al, Mn, Na, Fe, P, Ca, Mg, Zn and Cu. This scenario's relationship correlates with the high SMR and SRMS observed in T2, T4 and T6 (Figure 4.2). Negatively correlated variables were depicted by vectors posted in opposite directions and oriented at 180 ° from one another. In our present study, SMR was negatively correlated to TG, a commercial compost, resulting in higher roots than shoots due to low nutrient content and water-holding capacity.

Moreover, TG was characterized by high RSMR and RMR, which always have a negative relationship with the SMR and SRMR. Root system width, CHA, RBC, and RSD were positively correlated with KD soil, characterized by low nutrient content and water-holding capacity. Figure 4.3 above illustrates that T1 and T3 resulted in a higher LAI, confirming the positive relationship in Figure 4.6. The plant's roots require mineral nutrients for construction and maintenance (York et al., 2013). Moreover, Lynch and Brown (2008) indicated that roots decrease in diameter and increase in length as the adaptive strategy of acquisition of nutrients in the soil.

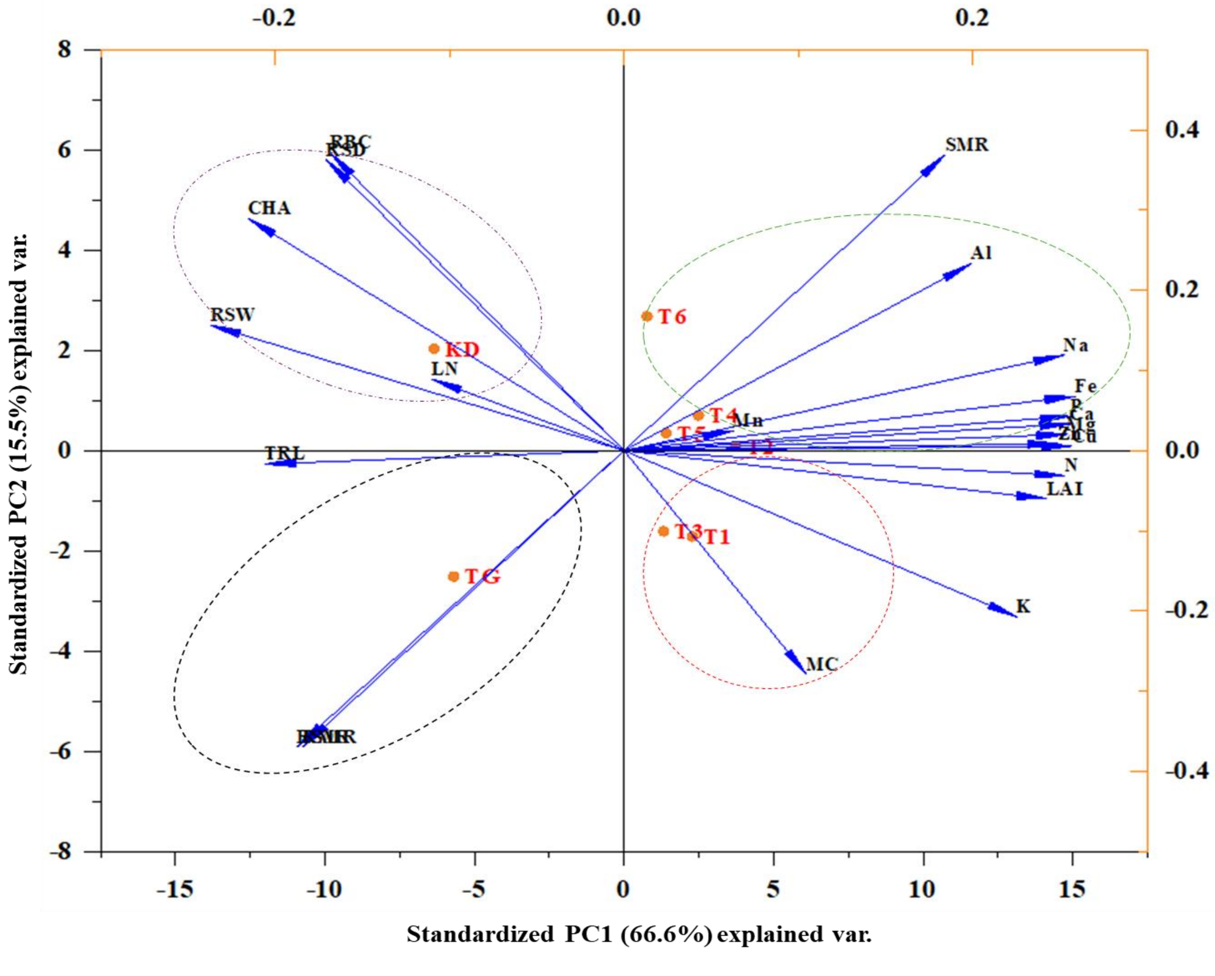


Figure 4.7: Principal component (PC), biplot of PC1 vs PC2 demonstrating the relationship between root and leaf traits of cucumber plants grown using different growing media.

4.4 Conclusion

Co-compost from a mixture of dewatered sewage sludge and organic green waste significantly enhanced above-ground cucumber biomass compared to commercial Gromor compost, yielding relatively lower above-ground growth. Notably, treatments with a higher ratio of dewatered sewage sludge to organic green waste (1DSS:2OGW), specifically treatments T2, T4, and T6, produced the highest shoot-to-root mass ratio (SRMR). In contrast, a lower sludge proportion (1DSS:3OGW) led to a slightly reduced but substantial SRMR. The control soil (KD) and commercial compost (TG) exhibited low SRMR values alongside high root length, convex hull area, root system width, and root mass ratio, indicating limited nutrient availability and water-holding capacity. These conditions compelled plants to allocate more biomass to root development rather than shoot growth.

Additionally, this study revealed a significant positive correlation between the leaf area index and the availability of key nutrients (N, P, K, Ca, Mg, Fe, Zn, Mn, and Na) in the growth media, supporting the role of nutrient-rich media in promoting shoot development. Conversely, there was a significant ($p<0.05$) but negative correlation between the leaf area index and root parameters, including convex hull area ($r=-0.81$), root system width ($r=-0.88$), and total root length ($r=-0.80$). The shoot mass ratio was positively associated with nutrient availability, explaining why the co-compost treatments had a higher shoot mass ratio and SRMR than commercial compost and control soil. These findings confirm that co-compost made from human excreta does not inhibit root growth; instead, it enhances shoot growth, resulting in an improved SRMR relative to commercial compost. Furthermore, this study demonstrates that co-compost with a higher proportion of sludge relative to organic green waste yields better growth outcomes than lower-sludge co-compost, indicating its potential for higher crop yields.

References

Amery, F., Vandaele, E., Körner, I., Loades, K., Viaene, J., Vandecasteele, B., and Willekens, K. (2020). Compost quality indicators. *SoilCom. Aarslev, Denmark; Report*.

Anwar, Z., Irshad, M., Fareed, I., and Saleem, A. (2015). Characterization and recycling of organic waste after co-composting-A review. *Journal of Agricultural Science* **7**, 68.

Barral, M. T., and Paradelo, R. (2011). A review on the use of phytotoxicity as a compost quality indicator. *Dyn. Soil Dyn. Plant* **5**, 36-44.

Barrios-Masias, F. H., Lazcano, C., and Hernandez-Espinoza, L. H. (2019). Advances in understanding vegetable physiology: root systems as the next frontier in improving sustainable vegetable production. *Achieving sustainable cultivation of vegetables*, 3-34.

Baumgarten, A., and Spiegel, H. (2004). Phytotoxicity (plant tolerance). *Agency for Health and Food Safety, Vienna*, 1-36.

Berkhout, E. D., Malan, M., and Kram, T. (2019). Better soils for healthier lives? An econometric assessment of the link between soil nutrients and malnutrition in Sub-Saharan Africa. *PloS one* **14**, e0210642.

Bontpart, T., Concha, C., Giuffrida, M. V., Robertson, I., Admkie, K., Degefu, T., Girma, N., Tesfaye, K., Haileselassie, T., and Fikre, A. (2020). Affordable and robust phenotyping framework to analyse root system architecture of soil-grown plants. *The Plant Journal* **103**, 2330-2343.

Burge, W., Cramer, W. N., and Epstein, E. (1978). Destruction of pathogens in sewage sludge by composting. *Transactions of the ASAE* **21**, 510-0514.

Chen, S., van der Graaff, E., Ytting, N. K., and Thorup-Kristensen, K. (2018). Evaluation of deep root phenotyping techniques in tube rhizotrons. *Acta Agriculturae Scandinavica, Section B — Soil & Plant Science* **69**, 62-74.

Cofie, O. O., Kranjac-Berisavljevic, G., and Drechsel, P. (2005). The use of human waste for peri-urban agriculture in Northern Ghana. *Renewable agriculture and food systems* **20**, 73-80.

Du, Q., Zhao, X., Jiang, C., Wang, X., Han, Y., Wang, J., and Yu, H. (2017). Effect of potassium deficiency on root growth and nutrient uptake in maize (*Zea mays L.*). *Agricultural Sciences* **8**, 1263-1277.

El Hayany, B., El Fels, L., Ouhdouch, Y., and Hafidi, M. (2021). Fate of pathogenic microorganisms during lagooning sludge composting and exploration of bacteriophages as indicator of hygienization. *Environmental Technology & Innovation* **21**, 101268.

Fan Jianbo, Zhang YaLi, Z. Y., Turner, D., Duan YinHua, D. Y., Wang DongSheng, W. D., and Shen QiRong, S. Q. (2010). Root physiological and morphological characteristics of two rice cultivars with different nitrogen-use efficiency. *Pedosphere* **20**.

Germer, J., Boh, M. Y., Schoeffler, M., and Amoah, P. (2010). Temperature and deactivation of microbial faecal indicators during small scale co-composting of faecal matter. *Waste Management* **30**, 185-191.

Giuliani, L. M., Hallett, P. D., and Loades, K. W. (2024). Effects of soil structure complexity to root growth of plants with contrasting root architecture. *Soil and Tillage Research* **238**, 106023.

Gruber, B. D., Giehl, R. F., Friedel, S., and von Wirén, N. (2013). Plasticity of the Arabidopsis root system under nutrient deficiencies. *Plant physiology* **163**, 161-179.

Larsen, J., Jaramillo-Lopez, P., Najera-Rincon, M., and Gonzalez-Esquivel C. E (2015). Biotic interactions in the rhizosphere in relation to plant and soil nutrient dynamics. *Soil Science and Plant Nutrition*.

Lavon, R., Salomon, R., and Goldschmidt, E. E. (1999). Effect of potassium, magnesium, and calcium deficiencies on nitrogen constituents and chloroplast components in Citrus leaves. *Journal of the American Society for Horticultural Science* **124**, 158-162.

Leghari, S. J., Wahocho, N. A., Laghari, G. M., HafeezLaghari, A., MustafaBhabhan, G., HussainTalpur, K., Bhutto, T. A., Wahocho, S. A., and Lashari, A. A. (2016). Role of nitrogen for plant growth and development: A review. *Advances in Environmental Biology* **10**, 209-219.

Lopez, G., Ahmadi, S. H., Amelung, W., Athmann, M., Ewert, F., Gaiser, T., Gocke, M. I., Kautz, T., Postma, J., and Rachmilevitch, S. (2023). Nutrient deficiency effects on root architecture and root-to-shoot ratio in arable crops. *Frontiers in plant science* **13**, 1067498.

Lynch, J., Marschner, P., and Rengel, Z. (2012). Effect of internal and external factors on root growth and development. *In* "Marschner's mineral nutrition of higher plants", pp. 331-346. Elsevier.

Lynch, J. P., and Brown, K. M. (2008). Root strategies for phosphorus acquisition. *In* "The ecophysiology of plant-phosphorus interactions", pp. 83-116. Springer.

Mandizvo, T., and Odindo, A. O. (2019). Seed mineral reserves and vigour of Bambara groundnut (*Vigna subterranea* L.) landraces differing in seed coat colour. *Heliyon* **5**, e01635.

Mandizvo, T., Odindo, A. O., Mashilo, J., Sibiya, J., and Beck-Pay, S. L. (2022a). Phenotypic Variability of Root System Architecture Traits for Drought Tolerance among Accessions of Citron Watermelon (*Citrullus lanatus* var. *citroides* (L.H. Bailey). *Plants (Basel)* **11**.

Mandizvo, T., Odindo, A. O., Mashilo, J., Sibiya, J., and Beck-Pay, S. L. (2022b). Phenotypic Variability of Root System Architecture Traits for Drought Tolerance among Accessions of Citron Watermelon (Citrullus lanatus var. citroides (LH Bailey). *Plants* **11**, 2522.

Manga, M., Camargo-Valero, M., Anthonj, C., and Evans, B. (2021). Fate of faecal pathogen indicators during faecal sludge composting with different bulking agents in tropical climate. *International Journal of Hygiene and Environmental Health* **232**, 113670.

Manga, M., Evans, B. E., Ngasala, T. M., and Camargo-Valero, M. A. (2022). Recycling of Faecal Sludge: Nitrogen, Carbon and Organic Matter Transformation during Co-Composting of Faecal Sludge with Different Bulking Agents. *International Journal of Environmental Research and Public Health* **19**, 10592.

Mašková, T., and Herben, T. (2018). Root: shoot ratio in developing seedlings: How seedlings change their allocation in response to seed mass and ambient nutrient supply. *Ecology and evolution* **8**, 7143-7150.

Mohd-Radzman, N. A., Djordjevic, M. A., and Imin, N. (2013). Nitrogen modulation of legume root architecture signaling pathways involves phytohormones and small regulatory molecules. *Frontiers in Plant Science* **4**, 385.

Ota, R., Ohkubo, Y., Yamashita, Y., Ogawa-Ohnishi, M., and Matsubayashi, Y. (2020). Shoot-to-root mobile CEPD-like 2 integrates shoot nitrogen status to systemically regulate nitrate uptake in Arabidopsis. *Nature communications* **11**, 641.

Putra, F. P., and Ismoyojati, R. (2021). Monitoring of maize root growth on N, P, and K fertilization using rhizotron. *Jurnal Ilmiah Pertanian* **17**, 74-79.

Rangarajan, H., and Lynch, J. P. (2021). A comparative analysis of quantitative metrics of root architecture. *Plant Phenomics*.

Reich, P. B., Wright, I. J., Cavender-Bares, J., Craine, J., Oleksyn, J., Westoby, M., and Walters, M. (2003). The evolution of plant functional variation: traits, spectra, and strategies. *International Journal of Plant Sciences* **164**, S143-S164.

Schickling, A., Graf, A., Pieruschka, R., Plückers, C., Geiß, H., Lai, I.-L., Schween, J., Erentok, K., Schmidt, M., and Wahner, A. (2010). The influence of leaf photosynthetic efficiency and stomatal closure on canopy carbon uptake and evapotranspiration–a model study in wheat and sugar beet. *Biogeosciences Discussions* **7**, 7131-7172.

Scognamiglio, M., D'Abrosca, B., Esposito, A., Pacifico, S., Monaco, P., and Fiorentino, A. (2013). Plant growth inhibitors: allelopathic role or phytotoxic effects? Focus on Mediterranean biomes. *Phytochemistry Reviews* **12**, 803-830.

Taiz, L., Zeiger, E., Møller, I. M., and Murphy, A. (2015). Plant physiology and Development. *In* "Plant physiology and Development" (L. Taiz, E. Zeiger, I. M. Møller and A. Murphy, eds.). Sinauer Associates Incorporated, United States of America.

Toor, M. D., Adnan, M., Rehman, F., Tahir, R., Saeed, M. S., Khan, A. U., and Pareek, V. (2021). Nutrients and their importance in agriculture crop production; A review. *Ind. J. Pure App. Biosci* **9**, 1-6.

Tripathi, D. K., Singh, S., Singh, S., Mishra, S., Chauhan, D., and Dubey, N. (2015). Micronutrients and their diverse role in agricultural crops: advances and future prospective. *Acta Physiologiae Plantarum* **37**, 1-14.

York, L. M., Nord, E. A., and Lynch, J. P. (2013). Integration of root phenes for soil resource acquisition. *Frontiers in Plant Science* **4**, 355.

Chapter 5: Comparative Effects of Co-compost and Chemical Fertilisers on Maize (*Zea mays*) Growth and Soil Nutrient Dynamics

Abstract

The combined application of organic green waste/faecal sludge co-compost and synthetic fertilisers on soils can supply both macro-nutrients and micro-nutrients to plants thus enhancing soil organic matter and overall fertility in the long term. This study investigated the effect of the combined application of co-compost and synthetic fertiliser on the growth, development, and nutritional composition of maize (*Zea mays*). The maize was grown on two contrasting soils both acidic but with different physical properties a sandy loam (KD) and a clay-loam soil (BS). A single factorial design was laid out in the completely randomised design under controlled condition. Plant height significantly differed ($p<0.001$) between treatments, the negative control of the sandy loam soil (KD) was shorter (139.5 cm) while the addition of co-compost improved its growth to 217.5 cm. The negative control of both KD and BS resulted in significantly ($p<0.001$) lower chlorophyll content compared to the addition of either co-compost or synthetic fertilisers in the soil. Combining co-compost with synthetic fertilisers resulted in a higher harvest index (49.3%) in the BS while the negative control of KD resulted in the lowest harvest index (0.0%). Essential minerals including nitrogen (N), calcium (Ca) and zinc (Zn) were high in the treatment combination of co-compost and synthetic fertiliser in both KD and BS and there was a statistically significant difference between treatments ($p<0.05$). In conclusion, combining synthetic fertilisers with co-compost improved growth and the yield of maize compared to the negative control and sole synthetic fertiliser application. Moreover, the study demonstrated improved essential mineral nutrients in maize as the result of combining synthetic fertilisers and co-compost in both sandy loam (KD) and clay loam (BS) soils.

Keywords: Co-compost, chemical fertilisers, pathogens, plant height, harvest index

5.1 Introduction

The increasing global population implies the need to increase food, feed, and fuel production on the limited agricultural lands (Majumdar et al., 2016). Chemical fertilisers have primarily been used to enhance plant productivity because they have a high nutrient composition, are water-soluble, readily available to plants, easy to apply, and transport (Chen et al., 2017). However, applying chemical fertilisers increases the decomposition rate of organic matter in the soil resulting in nutrient loss via fixation, leaching and gaseous emissions (Bhatt et al., 2019). Moreover, research has revealed that they affect soil's biological, physical and chemical properties, leading to soil compaction and acidification and low soil fertility (Lv et al., 2020). Overapplication and use of synthetic fertilisers lead to nutrient imbalance and can result in reduced crop yield over time (Ojeniyi, 2002).

Application of organic fertilisers, such as animal manure, human waste (urine and faeces) or market waste-derived fertilisers, potentially supply an equal nutrient and sustainably release nutrients compared to synthetic fertilisers (Chen, 2006). Moreover, organic fertilisers can improve soil structure, water retention, nutrient availability, cation exchange capacity, and biological activity, enhancing crop yield and quality while reducing the risk of environmental degradation (Qiu et al., 2021). However, the sole application of organic fertilisers may not provide sufficient nutrients for crop growth and development due to their lower immediate nutrient availability and the prolonged mineralization process required to release those nutrients (Miao et al., 2011).

Scientific literature has documented that combining organic and chemical fertilisers increases and sustains soil fertility and crop yields compared to the sole application of either chemical or organic fertilisers (Aguilera et al., 2012). Jatav et al. (2021) proposed integrated nutrient management (INM) as the method of improving soil fertility by supplying plant nutrients to a required level in a sustainable way by combining the benefits of organic and inorganic fertilisers in an integrated manner (Jatav et al., 2021).

This INM method combines organic and inorganic fertilisers to increase nutrient uptake and improve the structure of the soil. According to Ghosh et al. (2016), combining chemical fertiliser and organic fertiliser increased microbial activity, soil organic matter by 16%, and plant production by 67% compared to sole chemical fertiliser application. Moreover, Natsheh and Mousa (2014) found that combining chemical fertiliser and organic fertiliser significantly improved cucumbers' (*Cucumis sativus*) growth, development and yield compared to sole chemical fertiliser and organic fertiliser application. In addition, the study demonstrated a low water requirement for crops fertilised by organic fertiliser (180 m^3/season) compared to those fertilised by chemical fertiliser (217 m^3/season). Moreover, organic fertilisers can return micronutrients, organic matter and organic carbon lost via nutrient mining into the soil thus restoring the quality of the soil and improving crop productivity.

The proposed INM can counteract micronutrient deficiencies which are the main cause of exacerbated abnormal growth and skeletal, cardiovascular, and metabolic disorders (Kiani et al., 2022). This is because the organic fertilisers from human or animal excreta-derived fertilisers will provide micro-nutrients and organic matter while synthetic fertilisers can supply macronutrients for visible growth of plants. Organic fertilisers would subsequently return nutrients lost through food transportation back into the soil and can reduce the use of excessive chemical fertilisers. Therefore, the objective of this study was to determine and assess the impact of combining organic fertilisers (co-compost) with synthetic fertilisers on the growth, development, yield and nutritional composition of maize (*Zea mays*) grown in sandy loam and clay loam under controlled conditions. The study further assessed the regrowth of pathogens from the soils amended with human excreta-derived co-compost.

5.2 Materials and methods

5.2.1 Study site

The study was conducted at the University of Kwazulu-Natal, Pietermaritzburg campus at the Controlled Environment Facility (CEF) (29°61′68″S and 30°39′42″E).

5.2.2 Soil collection and preparation

The Cartref soil, classified as a Typic Hap aquept, was collected from an arable field in KwaDinabakubo (KD) area, Hillcrest, South Africa, (29°46′48″S and 30°45′46″E) and reddish clay loam soil was collected at Bishoptowe (BS), Pietermaritzburg (29°58′71″S and 30°47′93″E). Soil properties of KD and BS soils are presented in Table 5.1. The soil was collected in the top 0-25 cm layer using a pick and shovel, air-dried for 7 days and later sieved with a 5 mm diameter (⌀) rotational sieve. Twenty litres (20 L) of plastic pots, measuring 32.5 cm in height, 37 cm in upper diameter, and 25 cm in lower diameter, were filled with soil collected from both sites. Holes of 2 cm ⌀ were opened beneath to allow drainage after irrigation. The surface area of the pots was calculated using the formula:

$$SA = \pi(R + r^2) + \pi(R + r)\sqrt{(R - r)^2 + h^2} \qquad \text{Equation 5.1}$$

SA (surface area), **R** (upper radius of the bucket), **h** (height), **r** (radius at the bottom)

Table 5.1: Classification of the soil used during the experiment

Property	KwaDinabakubo (Cartref soil)	Bishopstowe soil
Clay (%)	12	51.3
Silt (%)	15	16.3
Sand (%)	73	32.4
Textural class	Sandy loam	Clay loam
Bulk density (kg m^{-3})	1.4	1.1
Field capacity (m^3 m^{-3})	0.24	0.36
Permanent wilting point (m^3 m^{-3})	0.12	0.21
Organic C (%)	1.5	2.5
N (%)	0.07	0.19
P (mg kg^{-1})	3	25
K ($Cmol_c$ kg^{-1})	38	487
Ca ($Cmol_c$ kg^{-1})	535	1712
Mg ($Cmol_c$ kg^{-1})	193	435
Exchangeable acidity ($Cmol_c$ kg^{-1})	0	0
Total cation ($Cmol_c$ kg^{-1})	5	13
Acid saturation (%)	4	1
pH (KCl)	4	5
Zn (mg kg^{-1})	1	18
Mn (mg kg^{-1})	3	17
Cu (mg kg^{-1})	2	7
N requirement (kg ha^{-1})	200	200
P requirement (kg ha^{-1})	60	20
K requirement (kg ha^{-1})	190	0

C= carbon, **N**= nitrogen, **P**= phosphorus, **K**= potassium, **Ca**= calcium, **Mg**= magnesium, **Zn**= zinc, **Mn**= manganese, **Cu**= copper

5.2.3 Environmental conditions

The relative humidity and temperature of the tunnel were recorded daily using Campbell Scientific CR10X Datalogger. Daily temperature and relative humidity are shown in Figure 5.1. The data shows from planting until the maturity of maize. The results show that the tunnel used to grow maize in this experiment had a temperature that ranged between 15 °C -25 °C while the relative humidity was between 50% to 90 %.

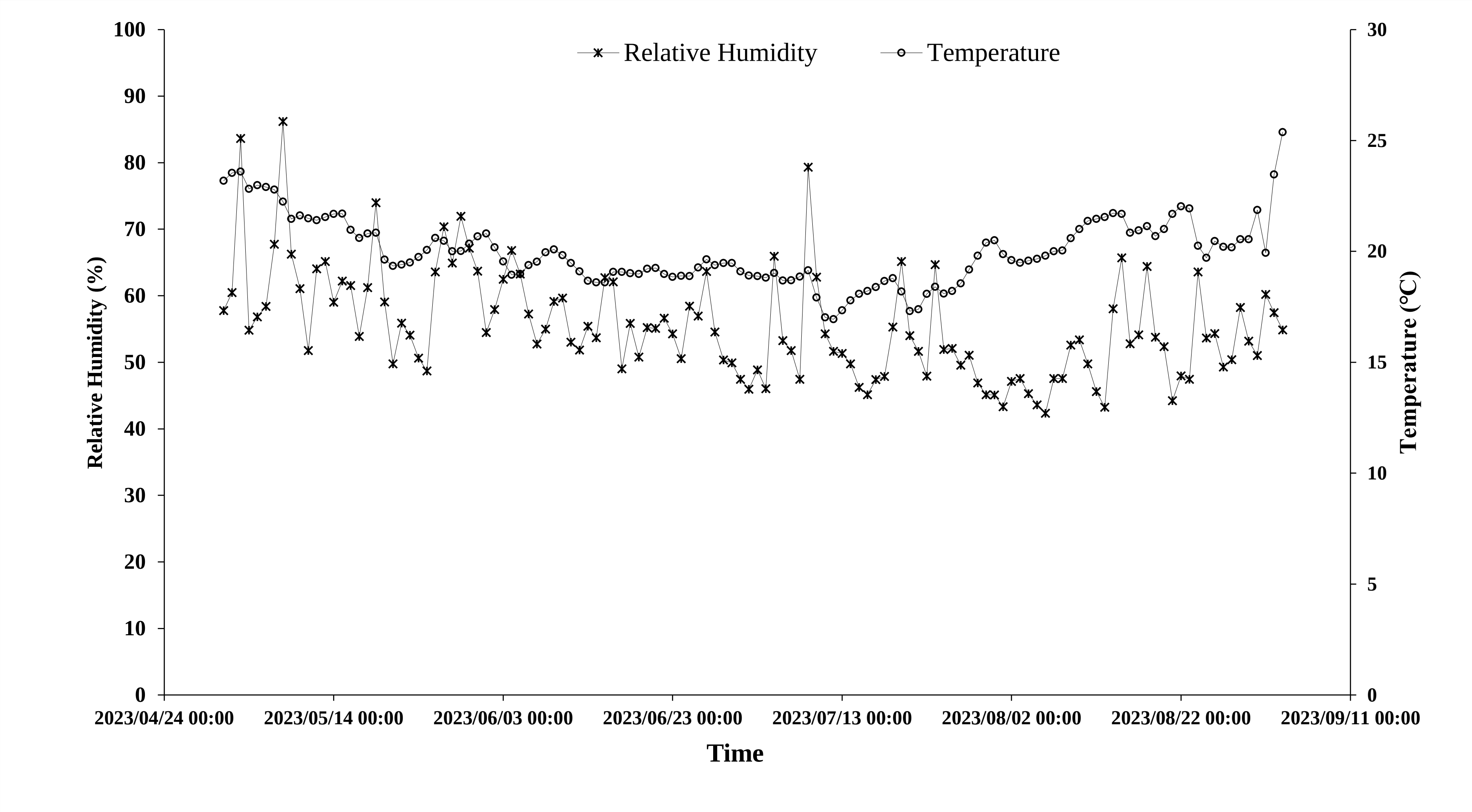


Figure 5.1: Relative humidity and temperature of the tunnel where maize was grown using different soils (KD and BS) and different soil amendments

5.2.4 Experimental design and layout

A pot experiment was conducted under controlled conditions in the tunnel at the Controlled Research Facility (CEF) at the University of KwaZulu-Natal, Pietermaritzburg, South Africa (29°37'36.5" S and 30°24'14.7" E). The study was designed as a single-factor design with 9 treatments replicated 4 times, giving 36 experimental units, which were the 20 L pots filled with soil, and laid out in a completely randomized design (CRD). Table 5.2 shows the treatments, and the description and explanation of each treatment used in this study.

Table 5.2: Descriptions of treatments and different types of soils used in this experiment

Soil type	Treatment	Description
Sandy loam (KD)	KD	Negative control of sandy loam soil (no amendments)
Clay loam (BS)	BS	Negative control of clay loam soil (no amendments)
Sandy loam (KD)	KD+F	Sandy loam amended with NPK
Clay loam (BS)	BS +F	Clay loam amended with NPK
Sandy loam (KD)	KD + Co	Sandy loam at 200 kg/ha N (Co)
Clay loam (BS)	BS +Co	Clay loam at 200 kg/ha N (Co)
Sandy loam (KD)	KD (F+Co)	Sandy loam at 100kg/ha (N), 100kg/ha N (Co), rec (P and K)
Clay loam (BS)	BS (F+Co)	Clay loam at 100kg/ha (N), 100kg/ha N (Co), rec (P and K)
Co-compost (Co)	Co only	Co-compost as the growing media

KD (KwaDinabakubo), **BS** (Bishopstowe), **Co** (Co-compost), **F** (NPK fertilisers) **rec** (recommended), **kg/ha** (kilograms per hectare), **NPK** (nitrogen, phosphorus & potassium).

In this study the NPK used were the straight fertilisers including Lime ammonium nitrate (LAN) containing 46% N, Single super phosphate (SSP) containing 10.5% P and potassium chloride (KCl) containing 50% K. These fertilisers were applied in the treatments with F (Table 5.2) following the method described by (Ebrahimpour et al., 2011). Soils with NPK fertilisers were top-dressed using LAN in week 6 after planting following the recommendations from Cedara analytical laboratory (Appendix 2). Co-compost used in this experiment was made from dewatered sewage sludge and organic green waste using the mixing ratio (1DSS:3OGW) and had a nitrogen content of 1.41%.

5.2.5 Planting and monitoring

Long maturing white grain maize hybrid seeds (SC 701) were sourced from Macdonald Seeds, Mkondeni, Pietermaritzburg (29°65′87″S and 30°41′61″E). Three (3) maize seeds were planted at 5 cm depth to ensure high seedling establishment. The drip irrigation system discharging 20 mm of water per minute was opened daily for 15 minutes (min) during maize seedling, 25 min at a vegetative growth stage, and 30 min at the reproductive stage per day. Small and less vigorous plants were uprooted from the pots two weeks after planting, leaving 2 maize seedlings. During the reproductive stages of maize growth, the water demand was high, and the irrigation system was adjusted according to needs.

5.2.6 Assessing the safety of using Human-Excreta Derived Co-compost (HEDC) for crop production

5.2.6.1 Escherichia coli (E. coli) quantification

Culturable pathogen indicators were quantified using the dilution plate count technique, followed by the expression of resultant counts in the Most Probable Number per gram (MPN g^{-1}) of sample dry weight. A composite mixture of 25 g soil and co-compost was diluted with 100 ml distilled H_2O to form a homogenous solution. The solution was augmented with Colilert reagent agitated until complete dissolution. The prepared solution was poured into an IDEXX Quanti-tray 2000 and incubated at 37 °C for 24 hours. The IDEXX Quanti-tray were examined under ultraviolet (UV) light in a controlled dark room setting, where fluorescent illumination emanating from large and small wells indicated the presence of *E. coli*. Notably, applying appropriate dilution factors facilitated the conversion of *E. coli* values to MPN g^{-1}.

5.2.6.2 Helminths eggs

To quantify helminth eggs, 50 g soil and compost samples were added to a 250 ml plastic beaker and mixed with ammonium bicarbonate (AMBIC). The suspension was sieved through a 100 µm mesh sieve placed on top of a 20 µm sieve. The retentate was thoroughly washed over the sieves using tap water, the retentate on the 100µm was discarded and that on the 20µm mesh sieve was collected. A 3 ml plastic Pasteur pipette was used to collect the retentate into 15 ml plastic conical test tubes. Samples were centrifuged at 1512 g (3000 rpm) for 10 min, and the supernatant was discarded. Zinc sulphate ($ZnSO_4$ at S.G 1.3) was added to the pellet in 3 ml aliquots up to 14 ml while resuspending using a vortex mixer. Samples were centrifuged at 672 g (2000 rpm) for 15 min. The supernatant was poured onto a 20 µm sieve and washed using tap water. The retentate was transferred to the test tube(s) and centrifuged at 1512 g for 10 min. The final pellet(s) were analysed using a light microscope. Wet mount preps were made on glass slides, and all eggs were counted and classified according to species and viability. The *Ascaris* sp. eggs were classified as potentially viable, i.e., undeveloped (a one-celled embryo), developing (two cells or more), developed (with a motile larva) or developed (with an immotile larva or nonviable, i.e., dead (if the wall is damaged, collapsed, or has a deteriorated larva), necrotic (if the larva appears shrivelled) or infertile. Other helminth species were classified as either viable or nonviable. Viable helminth eggs were calculated using Equation 5.2.

$$Viable\ Helminth = \frac{\text{Number of viable eggs}}{\text{Amount of soil processed (g)}}$$ Equation 5.2

5.2.7 Data collection

Plant height was measured weekly from the first week after emergence to week 12, with measurements taken from the soil surface to the tallest part of the plant using a tape measure. Chlorophyll content was measured weekly using SPAD BIOBASE chlorophyll meter model CM-B. A tagged leaf was placed between the Photodiode laminose for approximately 60 s, and the readings were displayed on the screen. Leaf number was determined by counting the number of leaves in each plant per replicate. The average was calculated by summing the total leaves counted and dividing by the number of replications. A data logger was used to measure temperature and humidity daily in the tunnel. Dry matter content was determined by drying the shoot, stover, cob, and roots at 70 °C until constant mass, as per the study (Mandizvo et al., 2022). The laboratory scale was used to measure the mass after drying the samples. The nutrient composition of the maize grains and stover was analysed following the procedure (Mandizvo and Odindo, 2019). The Harvest index was calculated using the formula:

$$Harvest\ Index = \frac{\text{Grain Mass (g)}}{\text{Total Mass (g)}}$$ Equation 5.3

Where total mass = stover mass + grain mass + cob mass

5.3 Results and discussion

5.3.1 Plant growth and deficiency symptoms

Figure 5.2 below shows the trend in maize growth between different soil amendments 10 weeks after emergence (WAE). The pictures were captured before tasselling. Control (KD and BS) were relatively shorter than other treatments; notably, KD showed signs of chlorosis and necrosis by scorching the leaves and yellowing indicating N, P and K deficiencies. Adding co-compost and chemical fertilisers improved the crops; however, KD soil did not show a high improvement in chemical fertilisers alone. Interestingly, combining chemical fertiliser with co-compost improved the maize stalk and height (Figure 5.2 B) in the sandy loam soil (KD). The observation below clearly shows that Co-only treatment was higher and had improved stalk, and the combination of synthetic and organic fertiliser (F+Co) in both the soil resulted in improved plants as well.

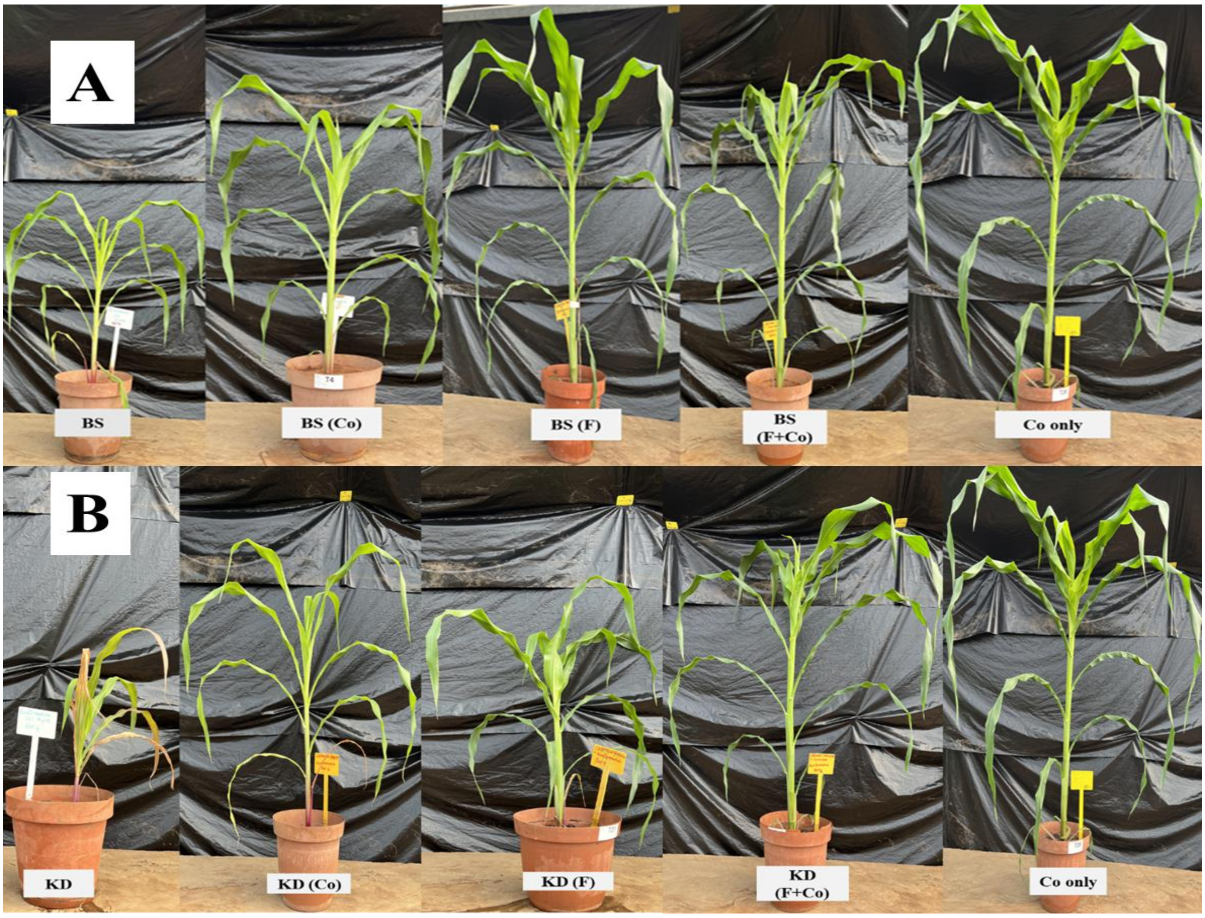


Figure 5.2: Comparison of maize plants grown in **A**-clay loam (BS), and **B**-sandy loam (KD) amended with co-compost and chemical fertilisers at different concentrations with co-compost alone (far right)

5.3.2 *Plant height*

Plant height indicates the crop's vegetative growth to inputs added to the soil (Azhar Ghaffari et al., 2011). In this study, plant height significantly differed ($p<0.001$) between treatments as shown in Figure 5.3. A gradual increase in plant height was observed from the beginning of the experiment in all treatments. In week 12, the lowest plant height (139.5 cm) was observed in the control KD soil while the highest plant (229.5 cm) was observed in the co-compost-only treatments. Adding co-compost into the sandy loam soil (KD) improved plant height compared to the sole chemical fertiliser application. Combining co-compost with chemical fertiliser improved plant height in both sandy loam and clay loam soil implying that co-compost added organic matter and organic carbon into the soil while chemical fertiliser improved the nutrient availability. According to Verma et al. (2006), adding multi-nutrients to the maize crop improved plant height and yields. Amujoyegbe et al. (2007) reported that combining inorganic and organic fertilisers led to the most significant plant height, outperforming the control and solo chemical fertiliser treatments.

As observed in Chapter 3, in Table 3.2, co-compost consists of both micro and macronutrients essential for plant growth and development, while chemical fertilisers contain macronutrients essential for visible growth and development of the plant. These multi-nutrients obtained in the co-compost could have improved plant height for treatments with co-compost. As stated by (Roba, 2018), chemical fertilisers supply nutrients in the early vegetative growth, while co-compost or organic fertilisers undergo mineralisation and supply nutrients in the reproductive stage of maize growth. This agrees with Verma et al., (2006), who observed that application of N, P, K, Zn, and S fertilisers led to improved plant height compared to sole macro-nutrient application in the soil.

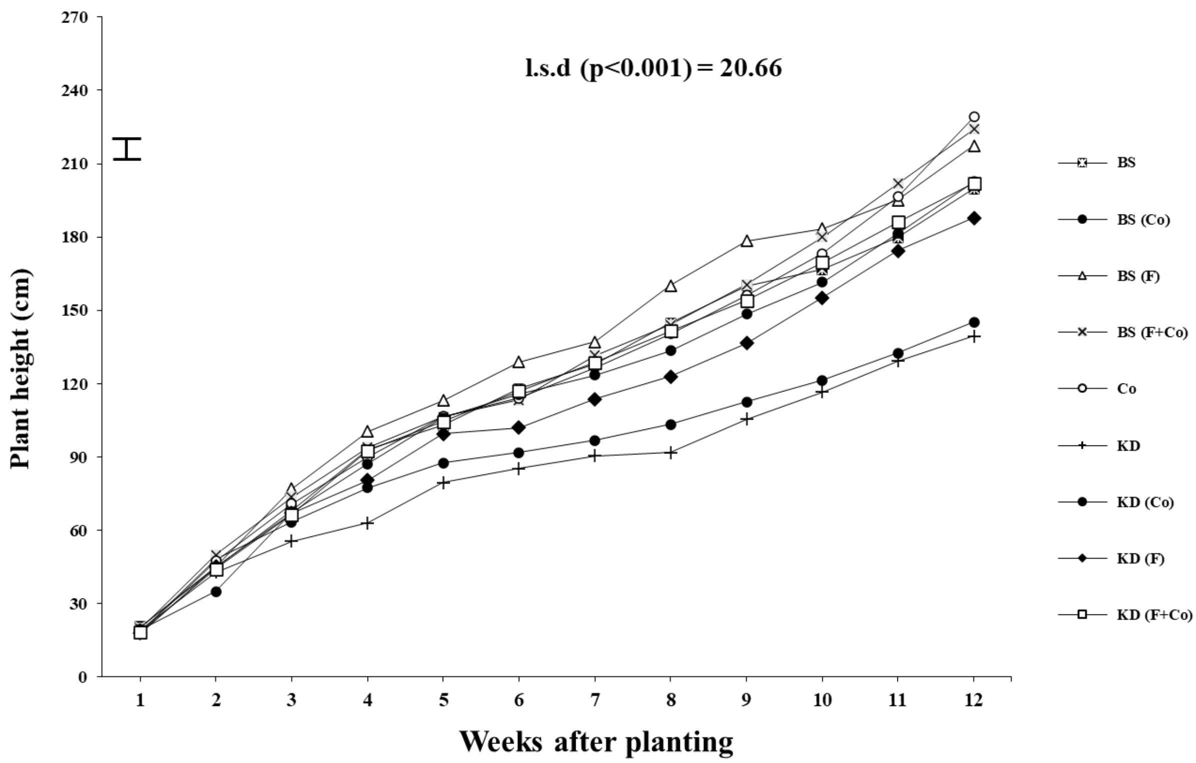


Figure 5.3: Weekly measurements of maize plant height from the first week after emergence to week 12 when maize started tasselling in different treatments

5.3.3 Chlorophyll content

Figure 5.4 shows a significant difference (p<0.001) in the chlorophyll content within different treatments between the first week and the maize tasselling stage. In week 12, the highest chlorophyll content in SPAD units was recorded in Co > KD (F+Co) > BS (F) > BS (F+Co) > KD (F) > KD (Co) > BS (Co) > BS > KD with the values from 60.92 to 34.1 from highest to lowest respectively. The sandy loam (KD) surprisingly resulted in the second highest (59.75) chlorophyll content when it was amended with chemical fertiliser and co-compost. However, the negative control of KD had the lowest chlorophyll content. This was caused by the purpling of leaves in the control which indicated phosphorus deficiencies. A drastic decline in chlorophyll content was observed 5 WAE. The rapid drop in chlorophyll content could have been caused by low temperature (11-17 °C) and it was cloudy and could be the lack of nitrogen in the plants thus affected the measurements of chlorophyll content by the SPAD. According to Reckitt-Benckiser (2015) photosynthetic rate is linked to chlorophyll content and can be affected by light intensity, carbon concentration and temperature. Therefore, altering any of the factors could reduce the photosynthetic rate and subsequently reduce chlorophyll content. The chlorophyll content increased in week 6 as the result of either top dressing as the leaves of the plants were greener and as the result of sunny weather conditions. At the end of the growth stage of maize, the highest chlorophyll content with the SPAD units of 57.75 was observed in the KD (F+Co), indicating that the combination of co-compost with synthetic fertiliser is the optimum practice that can result in high photosynthetic activities in plants, especially in the sandy loam soil.

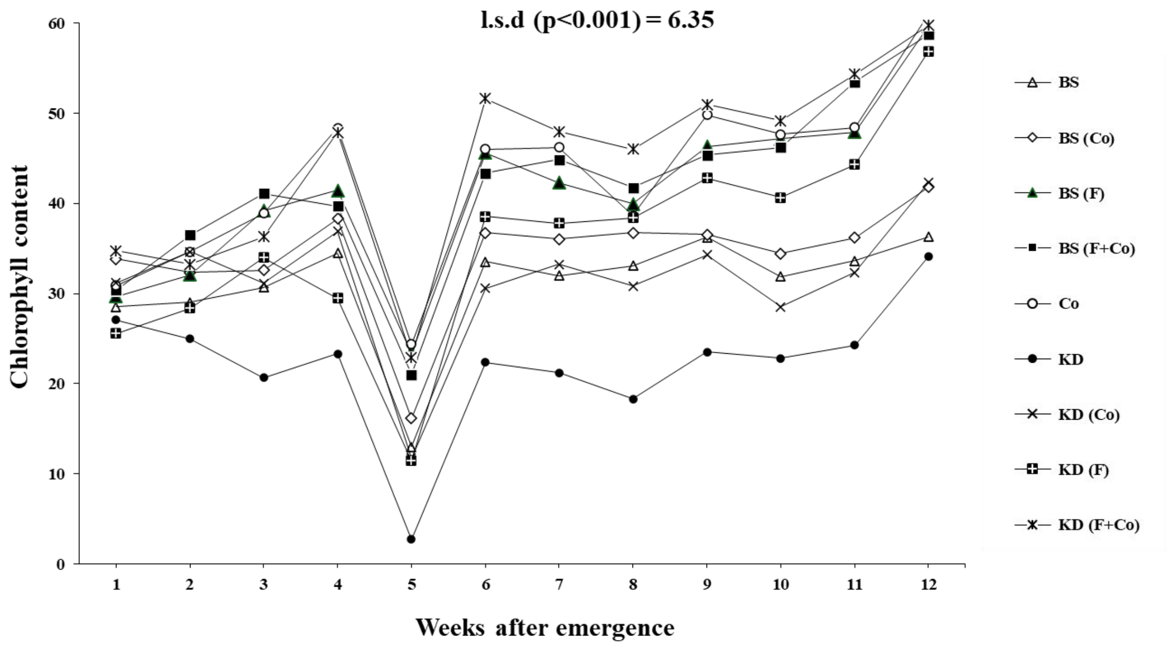


Figure 5.4: Chlorophyll content in SPAD units for maize grown with different soil amendments between the seedling stage (week 1) and the reproductive stage (week 12)

5.3.4 Leaf area index

Figure 5.5 shows a significant difference ($p<0.001$) in leaf area index (LAI) between treatments in week 6. The co-compost alone treatment recorded the highest LAI (316.7 cm^2). Nutrients such as N and P could have improved LAI in treatments with co-compost. However, interesting results were observed when comparing KD and BS. In the BS (control) and BS (F), LAI was higher than the KD soil for both control and fertiliser treatments. However, combining co-compost and chemical fertiliser improved the KD soil and the LAI was higher in KD (202.4 and 200.9 cm^2) compared to BS (190.4 and 174.1 cm^2) in Co and F+Co treatments, respectively. These results align with earlier studies by Slamet, (2017) who observed the feasibility of adding organic amendments to the soil through increased plant height, leaf number, and LAI of crops because of the available N in the organic amendments. Moreover, LAI is among the most important factors in estimating canopy photosynthesis in crop growth simulation models (Amanullah, 2007). Furthermore, it increases with increased nitrogen content (Valentinuz, 2006). It is also an indicator of healthy plants, and a lower LAI can indicate drought stress, disease attack, and nutrient deficiencies (Hongliang-Fang., 2019). Therefore, the highest LAI observed in combining co-compost and chemical fertiliser could have occurred due to improved soil structure and allowed for easy uptake of nutrients in the soil.

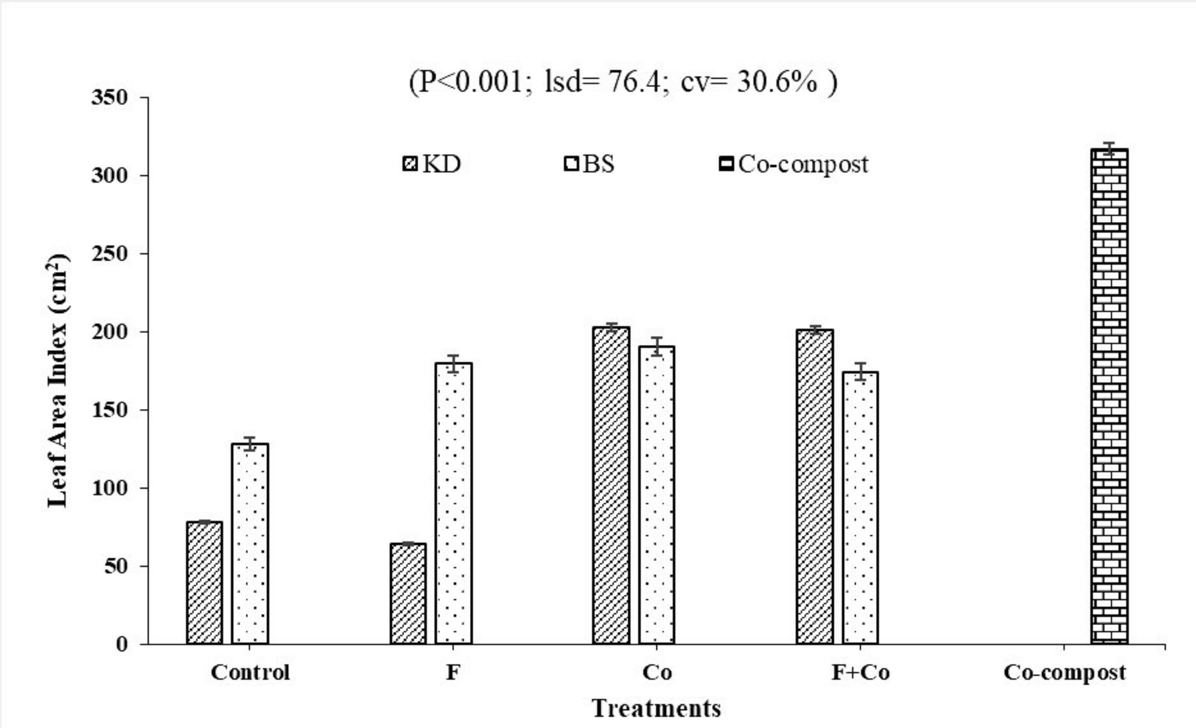


Figure 5.5: Leaf area index of maize grown using contrasting but acidic, sandy loam (KD) and clay loam (BS) soil at different soil amendments

5.3.5 Shoot and root biomass

There was a significant difference ($p<0.001$) in maize plants' shoot and root biomass in week 6. Treatments with co-compost had high shoot biomass of 4.77 g pot^{-1} and 4.28 g pot^{-1} in the KD (F+Co) and KD (Co), compared to the KD and KD (F), which had 1.24 g pot^{-1} and 1.90 g pot^{-1}, respectively. The BS soil resulted in shoot biomass between 4.69 g pot^{-1} and 5.70 g pot^{-1}, higher than KD soils. BS soil-supported shoot biomass (4.69–5.70 g pot^{-1}) was uniformly higher than KD soils across treatments. This implies that BS soil possessed higher intrinsic fertility or a more responsive behaviour with the amendments, than KD soil. The highest shoot biomass (9.85 g pot^{-1}) was observed in the co-compost without soil, indicating that co-compost alone can supply adequate nutrient availability and a growing environment for plants. The increase in shoot biomass in the treatments with co-compost is mainly driven by the availability of nitrogen and organic carbon in the co-compost which are critical for plant growth. Moreover, these results indicate that co-compost played an important role in enhancing the low fertile sandy loam soil (KD) and the use of co-compost as a standalone growing media.

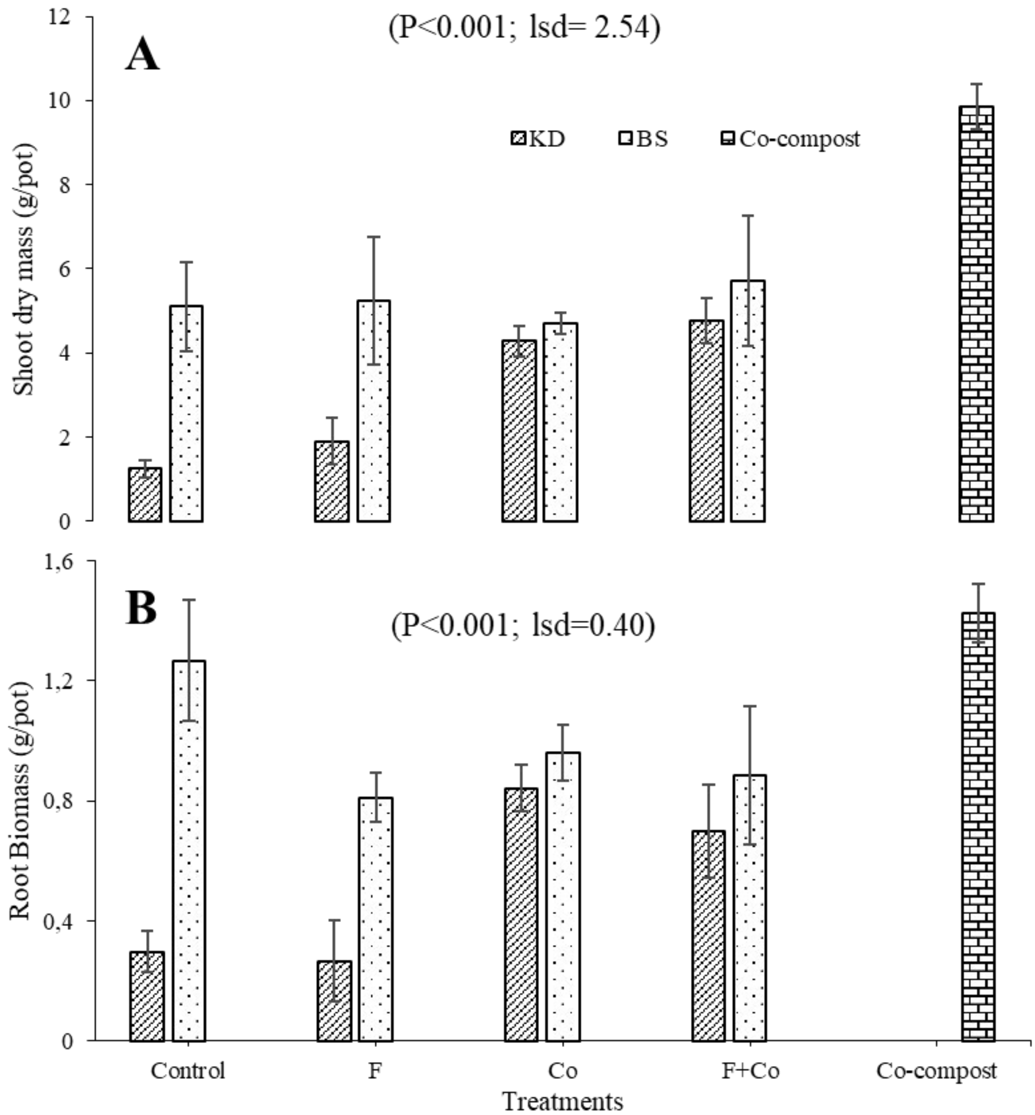


Figure 5.6: The shoot (**A**) and root (**B**) biomass (g pot^{-1}) at 6 weeks after emergence for different treatments during the maize experiment showing means ± standard error where n=4

5.3.6 Plant yield

There was a significant difference (p=0.006) in the grain mass between treatments at harvest (Figure 5.7A). Co-compost alone had the highest grain mass (131.3 g) compared to the treatments with soil. The comparison between treatments with soil shows an increase in grain mass as amendments were applied to the soil. The negative control of clay loam soil (BS) had harvestable maize grains compared to sandy loam soil (KD). Interestingly, co-compost, fertiliser, and combining F+Co addition to the KD soil; improved the average grain mass of maize to 13.5, 26.5, and 32 grams per pot, respectively. It was noted that maize plants with many sheaths could not produce enough harvestable yield. Most energy, minerals, and nutrients contribute to sheath formation compared to the assimilation or formation of grains.

Figure 5.7 B shows a significant difference (p=0.002) in the cob mass between treatments. The greatest difference was observed between treatments, mostly between co-compost alone, which was above 150 g. This could have been caused by mineralization, which led to increased plant height and stalk expansion in diameter. Moreover, F+Co treatments had increased cob mass for both soils compared to control, Co, and F. Figure 5.7 C shows a significant difference (p=0.002) in the stover mass between treatments. Interestingly, combining F+Co resulted in the highest stover mass compared to other treatments, mainly co-compost, as a growing media. Combining co-compost with chemical fertiliser (NPK) increased yield in the sandy loam (KD) and clay loam (BS) soils. These findings correlate with the findings by An et al., (2023), who found that maize grain productivity was higher in the NPK + compost (4.81), NPK fertiliser (4.75), compost (3.77), and least was control with 2.79 Mg ha^{-1} in dry mass basis. However, there were no significant differences in total nitrogen between the treatments as the values ranged between 27.0-33.4 g kg^{-1} at harvest. According to Qasim et al., (2023), the combination of vermicompost and chemical fertiliser improved tomato agronomic and morphological characteristics. This was observed by showing root length increase, improved plant height by 167%, SPAD value of 13.5%, calcium (54%), magnesium (117%), phosphorus (38%) and potassium (128%) compared to the control.

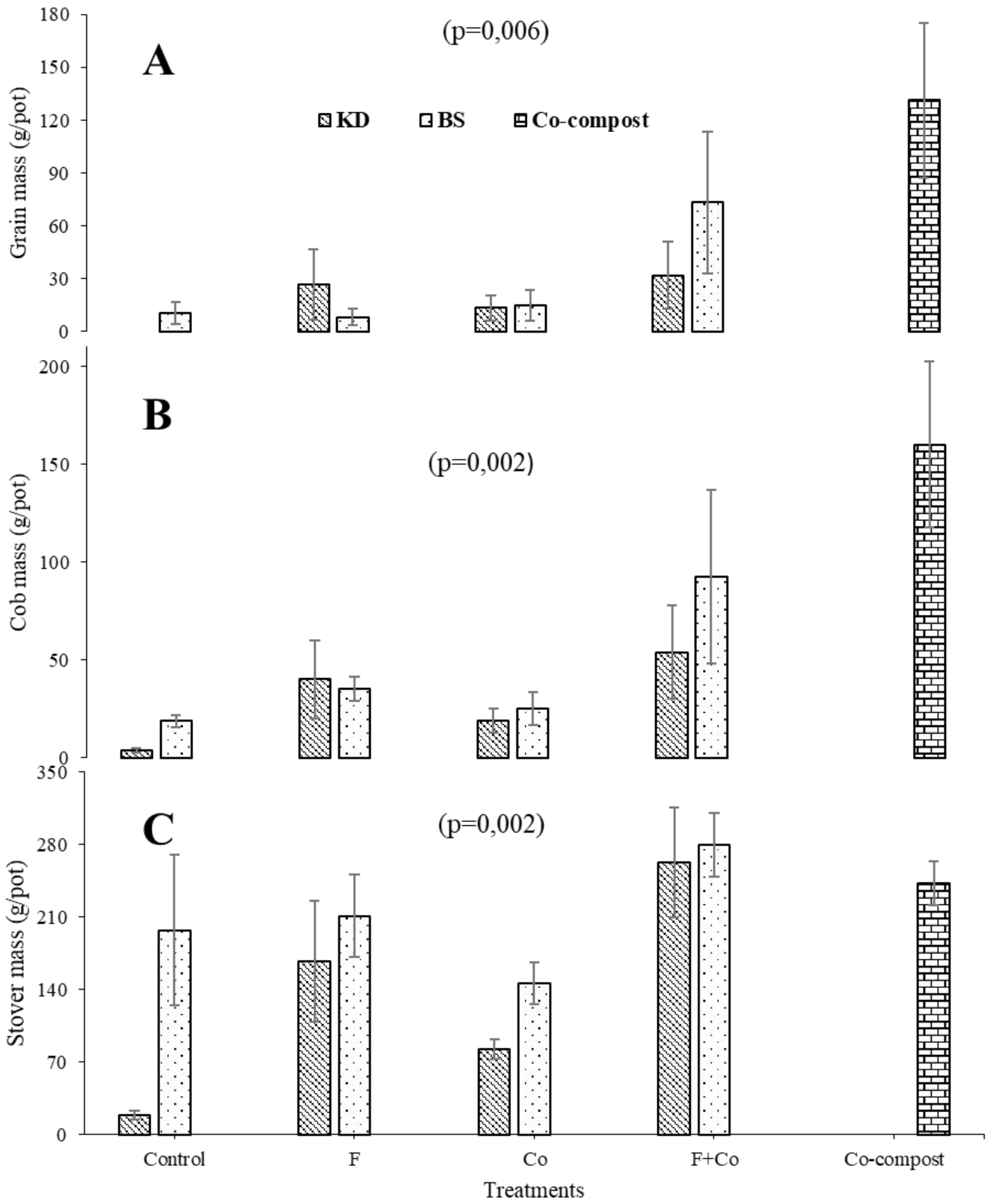


Figure 5.7: Grain (**A**), cob (**B**) and stover (**C**) mass of maize at harvest in grams per pot for clay loam (BS), sandy loam (KD) soil and co-compost with different amendments such as chemical fertilisers and co-compost and integrating both amendments at recommended rate, showing means ± error bars showing standard error where n=4

5.3.7 Maize yield at harvest

There were differences between the yields of different treatments, as shown in Figure 5.8. Co-compost alone had in bigger cobs compared to other treatments; moreover, the sheath was filled with grains. The cob in co-compost alone was also longer than other treatments, which might have occurred because of the macro and micronutrients found in the co-compost. The combination of chemical fertiliser (NPK) and co-compost was also nutrient-rich as the cob length was higher than fertiliser, co-compost, and control. Moreover, a higher number of grains was obtained compared to the control and other treatments. Chemical fertiliser supplied essential macro-nutrients (NPK), and co-compost supplied micro-nutrients. The soils could retain more water and nutrients because of co-compost availability. In treatments where synthetic fertiliser was used as a sole maize grains were scattered in the sheath. An interesting finding is that the BS soil was able to form grains; however, the cob was short and had fewer grains compared to the treatment with a combination of chemical fertiliser and co-compost. The poor sandy loam (KD) soil did not have grains because it took longer to tassel and was nutrient deficient compared to other treatments. This showed that combining organic and inorganic fertilisers can improve sandy loam soil and improve degraded soil structure.

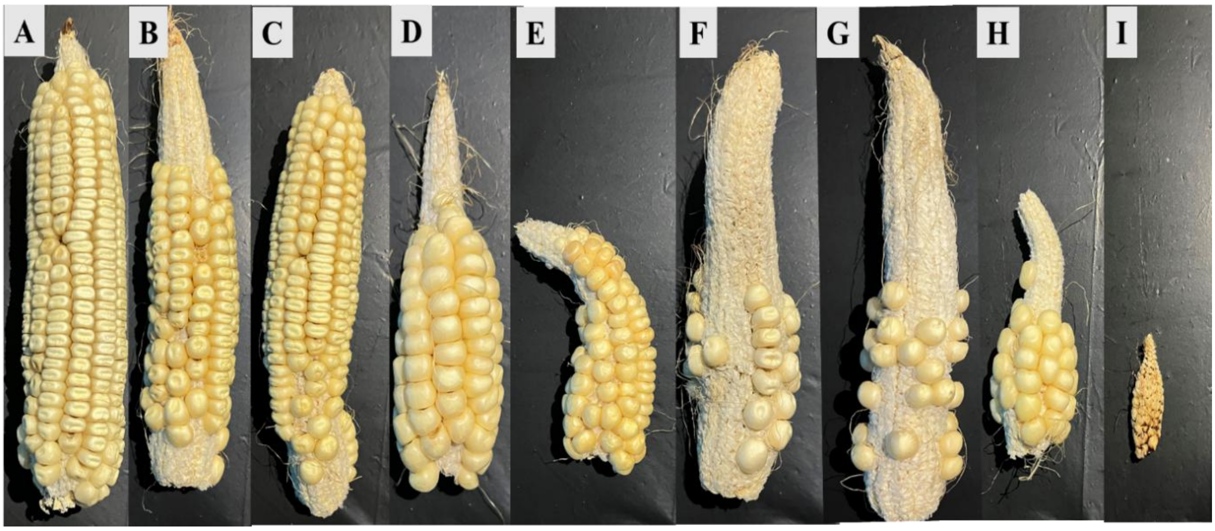


Figure 5.8: Maize cobs from **A** (Co-compost alone), **B** (KD+F+Co), **C** (BS+F+Co), **D** (KD+Co), **E** (BS+Co), **F** (KD+F), **G** (BS+F), **H** (BS) and **I** (KD) after 48 hours of oven dry at 70 °C.

5.3.8 Grain number and cob length

Figure 5.9A shows a significant difference ($p<0.001$) in the cob length of different treatments. The highest cob length (18.47 cm) was recorded in the co-compost treatment, followed by the combination of chemical fertiliser and co-compost at the value of 16.53 and 15.53 cm in KD and BS soils respectively. The shortest cob length (4.04 cm) was recorded in the negative control of the sandy loam soil (KD). Variation in the number of grains per cob was also observed and interestingly there was a significant difference ($p<0.001$) between treatments. Grain numbers per cob ranged from zero (0) grains per plant in the KD soil (control) to a maximum of 275 grains per plant in the co-compost. Moreover, combining chemical fertiliser and co-compost resulted in an improved number of grains in both KD and BS, with BS having lots more grains than KD soil. Adding co-compost (Co) in the soil did not significantly improve yield in BS soil; however, it improved the yield in KD soil from 0 to 29 grains. A study by Girija et al., (2019), revealed that the application of co-compost made from human excrement and municipal waste led to higher sweet potato roots than other treatments.

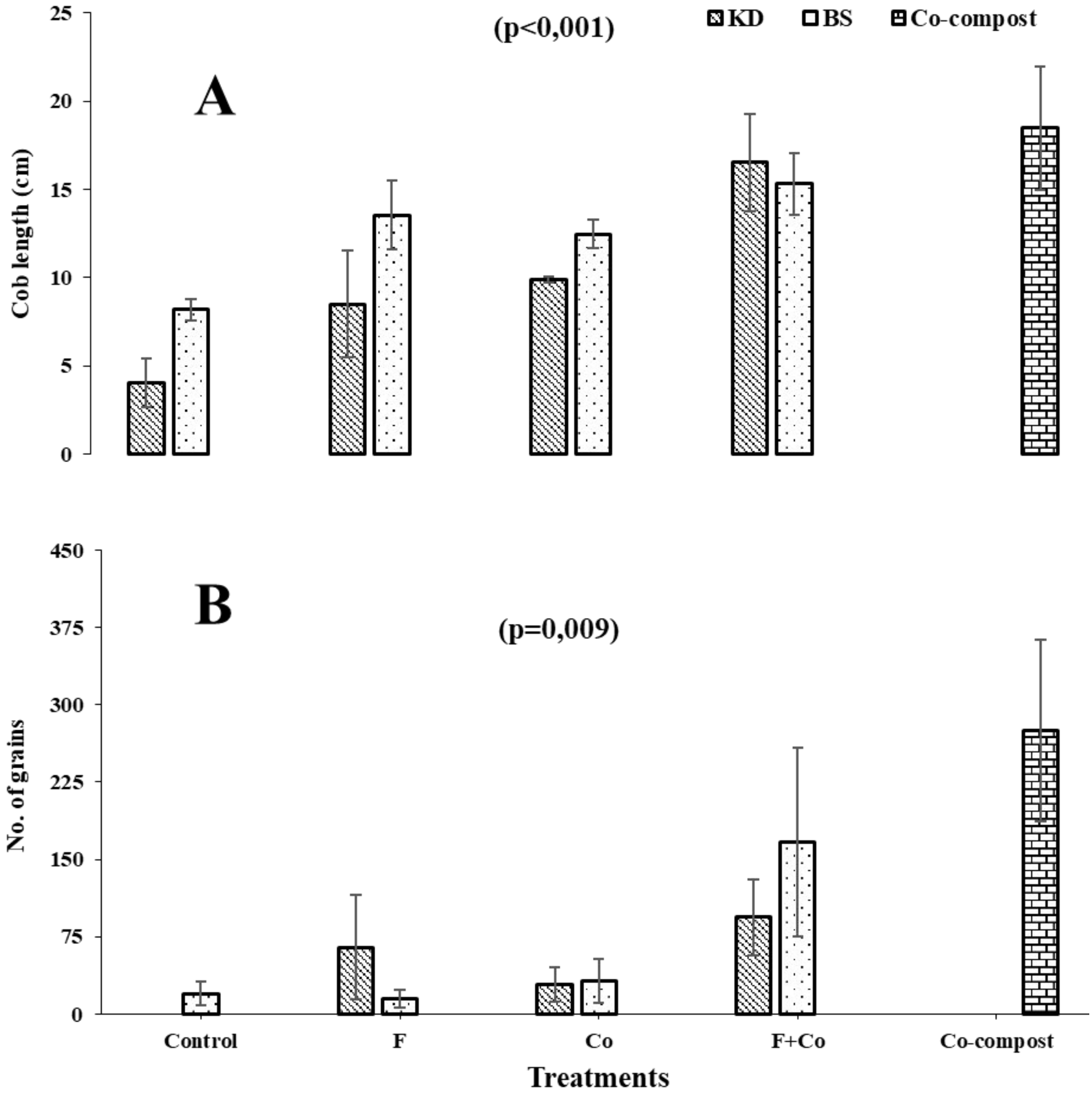


Figure 5.9: Cob length (**A**) and number of grains (**B**) of the maize grown using different soil amendments

5.3.9 Harvest index

The harvest index's (HI) average values ranged from 0% in KD only to 49.4% in the BS (F+Co), interestingly, co-compost alone resulted in the second-highest HI (47.7%) compared to other controls (KD and BS only). This is an indication of the advantage of organic amendments to supply plants with both micro and macro-nutrients. Clay loam soil (BS) without any additives showed the capability to accumulate yield, as shown by the HI of 10.2%, which was higher than when fertiliser was added. Moreover, BS had a higher HI in the F+Co treatment, which shows that co-compost contributed to yield formation. The study by Aly, (2002) extrapolated that applying organic fertiliser at 10 m^3 in a 540 m^3 resulted in a higher cucumber yield and fruits.

Previous studies that assessed different environmental conditions on maize harvest index showed that the harvest index can vary between 30% and 46% (Ion et al., 2015). Moreover, El-Sheikh et al., (1993), indicated that applying organic manure at 45kg/540 m^2 significantly improved yield compared to chemical fertiliser. Moreover, combining organic and synthetic fertilisers helps increase organic matter, increase cation exchange capacity (CEC) and reduce N loss in the soil (Liu et al., 2021). Furthermore, Asaye et al., (2022), found that combining compost and chemical fertiliser increased soil fertility, yield, and agronomic efficiency of maize-potato system plantations in Ethiopia. In addition to this, a comparative maize yield increase of 12-18% in the combined compost and mineral fertiliser compared to the control was observed by (Zerssa et al., 2021). These findings correlated with our study findings where combining co-compost with chemical fertilisers improved harvest index and yield of maize in two contrasting soils.

Combining organic and inorganic fertilisers has the added benefit of reducing greenhouse gas emissions, which are ammonia volatiles by 16% and nitrous oxide by 47% compared to solo chemical fertiliser application (An et al., 2023). Moreover, the combination of synthetic fertiliser and compost resulted in the added benefit of reduced greenhouse gas (N_2O and CO_2) emissions compared to the application of synthetic fertiliser alone (Zerssa et al., 2021).

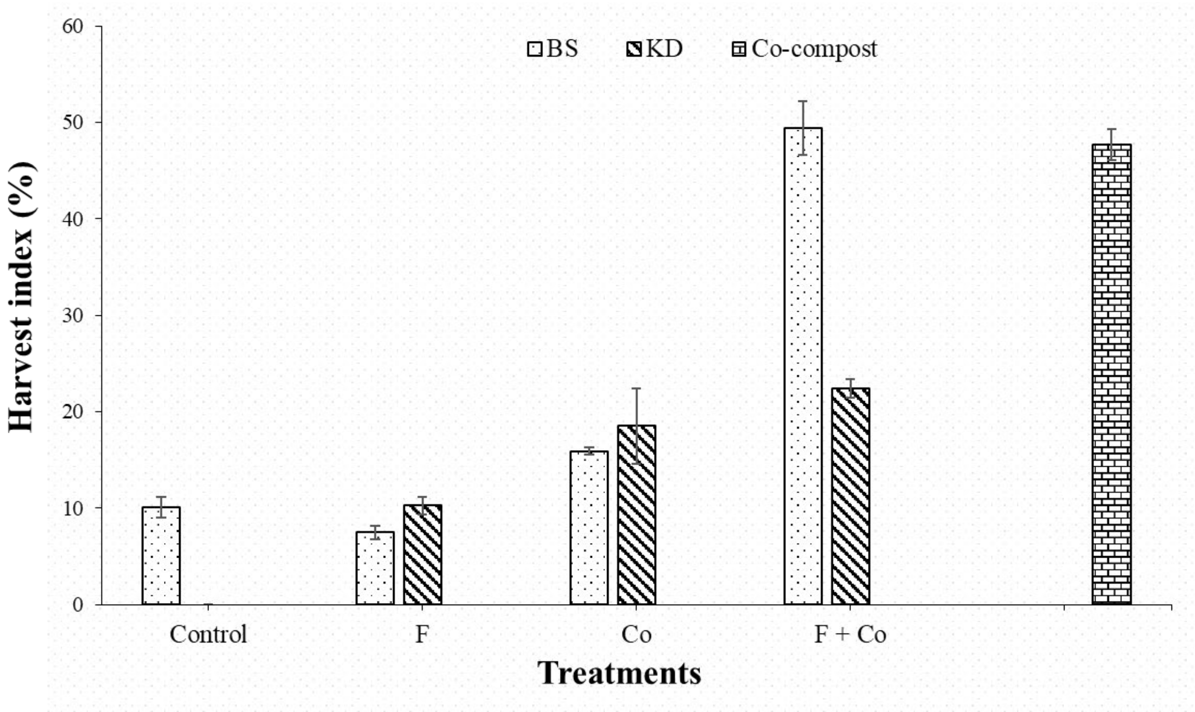


Figure 5.10: Harvest index of maize grown in different soils with different growing amendments graph showing means ± standard error where n=4

5.3.10 Macro and micronutrients in maize grains

Table 5.3 shows that there was a significant difference ($p < 0.05$) in nitrogen (N) content of maize grains across different treatments, highlighting the role of soil amendments in nutrient enrichment. The highest N content (2.2 ± 0.1%) was obtained when sandy loam soil (KD) was mixed with co-compost at the recommended N requirement demonstrating the efficiency of co-compost to adequately supply nutrients to crops. Expectedly, the lowest N content (1.2 ± 0.1%) was recorded in the BS (negative control) treatment, underscoring the inadequacy of clay loam soils to supply sufficient nitrogen.

Potassium (K) concentrations in maize grains also varied significantly between treatments ($p<0.045$). The highest K concentration (388.1 ± 41.2 mg/kg) was achieved with the clay loam soil amended with fertiliser and co-compost treatment (BS+F+Co), while the negative control resulted in the lowest concentration (197.5 ± 41.7 mg/kg). The enhanced K levels in BS+F+Co likely resulted from the combined input of potassium-rich chemical fertilizer (KCl, containing 50% K) and co-compost (containing 0.5% K). Notably, even the nutrient-depleted KD sandy loam soil demonstrated moderate K concentrations (301.2 ± 42.5 to 375.6 ± 20.4 mg/kg), showcasing some improvement with co-compost applications and having high stover nutrients as well (Appendix 3).

Other nutrients, including carbon (C), calcium (Ca), iron (Fe), magnesium (Mg), zinc (Zn), and phosphorus (P), did not differ significantly among treatments ($p>0.05$). However, the BS grains treated with synthetic fertilizer and co-compost (F+Co) showed an improved Zn content of 12 mg/kg which is less than the Zn content in maize grains that accumulates after biofertilization (Xue et al., 2023). The nutrient-depleted, sandy loam KD soil also exhibited relatively high Zn levels, indicating that co-compost application could effectively enhance micro-nutrient availability in soils and subsequent uptake by crops. Even though these findings demonstrate the potential of combined amendments in enriching nutrient-deficient soils and crops, further research is required to understand the mechanisms of nutrient uptake in plants and the assessment of microplastic and other organic pollutants contamination and uptake mechanism to crops.

Table 5.3: Nutrient composition of the maize grains among different treatments, table showing means ± standard error where n=3

Treatments	**N (%)**	**C (%)**	**Ca (mg/kg)**	**Cu (mg/kg)**	**Fe (mg/kg)**	**K (mg/kg)**	**Mg (mg/kg)**	**Mn (mg/kg)**	**Zn (mg/kg)**	**P (mg/kg)**
KD	-	-	-	-	-	-	-	-	-	-
BS	1.2 ±0.1[a]	42.9 ± 1.0[a]	7.5 ± 2.7[b]	0.00 ± 0.0[a]	5.10 ± 2.4[b]	197.5 ± 41.7[a]	74.2 ±15.6[a]	0.01 ± 0.0[a]	4.7 ± 0.3[a]	225.0 ± 41.1[a]
KD (F)	2.0 ± 0.1[cd]	42.3 ± 0.3[a]	7.1 ± 1.3[a]	0.32 ± 0.2[b]	3.77 ± 0.7[ab]	375.6 ± 20.4[c]	113.3 ± 6.6[b]	0.03 ± 0.0[b]	2.8 ± 0.6[a]	331.9 ± 22.2[b]
BS (F)	2.0 ± 0.1[cd]	41.5 ± 1.7[a]	6.1 ± 1.4[a]	0.19 ± 0.1[ab]	1.78 ± 0.6[a]	308.4 ± 11.9[cd]	111.9 ± 8.2[b]	0.02 ± 0.0[a]	3.1 ± 0.4[a]	310.6 ± 15.3[ab]
KD (Co)	2.2 ± 0.1[d]	41.2 ± 0.3[a]	17.7 ± 0.3[bc]	0.09 ± 0.0[a]	2.53 ± 0.2[ab]	302.0 ± 42.5[abc]	105.7 ± 8.1[ab]	0.02 ± 0.0[a]	8.1 ± 4.5[ab]	306.0 ± 29.5[b]
BS (Co)	1.5 ± 0.2[ab]	41.1 ± 0.1[a]	10.3 ± 1.1[abc]	0.00 ± 0.0[a]	1.87 ± 0.3[a]	211.9 ± 10.6[ab]	79.6 ± 2.4[ab]	0.01 ± 0.0[a]	2.9 ± 0.2[a]	224.5 ± 3.5[a]
KD (F+ Co)	2.1 ± 0.0[d]	42.4 ± 0.1[a]	9.2 ± 4.5[b]	0.00 ± 0.0[a]	2.28 ± 0.7[ab]	326.8 ± 70.3[bc]	97.5 ± 19.7[ab]	0.02 ± 0.0[a]	4.1 ± 1.7[a]	285.6 ± 58.4[ab]
BS (F+ Co)	2.0 ± 0.1[cd]	41.5 ± 1.7[a]	20.2 ± 4.0[c]	0.08 ± 0.0[a]	2.44 ± 0.3[ab]	388.1 ± 41.2[c]	114.7 ± 6.3[b]	0.02 ± 0.0[a]	12.0 ± 1.7[b]	330.9 ± 22.9[b]
Co-compost	1.7 ± 0.[1bc]	39.9 ± 1.8[a]	12.9 ± 5.5[abc]	0.00 ± 0.0[a]	2.11 ± 0.4[ab]	284.3 ± 20.1[abc]	90.3 ± 6.7[ab]	0.01 ± 0.0[a]	4.9 ± 1.2[a]	260.0 ± 17.8[ab]
CV (%)	10.6	4.0	48.3	130.7	60.5	21.7	18.7	27.2	63.0	18.7
l.s.d	0.33	2.91	9.33	0.19	2.86	112.5	31.84	0.01	5.68	92.1
p-value	<0.001	0.542	0.050	0.025	0.267	0.022	0.092	0.032	0.072	0.119

N (nitrogen), **C** (carbon), **Ca** (Calcium), **Cu** (copper), **Fe** (Iron), **K** (potassium), **Mg** (magnesium), **Mn** (manganese), **Zu** (zinc), **P** (phosphorus)

5.3.11 Pathogens in the soil

Co-composted sewage sludge typically has undetectable levels of pathogens due to the high temperatures generated during the thermophilic composting. Bacteria growth in the biosolids or co-compost reduces to undetectable limits, however their major concern is that these organisms are susceptible to regrowth (Zaleski et al., 2005). This occurs if bacteria become undetectable due to a loss of viability, this could create conditions conducive to the regrowth of *Salmonella* or *E. coli* when soils are rewetted (Zaleski et al., 2005). This study analysed treatments amended with co-compost for the presence of pathogens and helminth regrowth.

Figure 5.11b shows that sandy loam soil (BS) amended with co-compost only resulted in the highest total coliforms (129.75 MPN/g) concentration. The *E. coli* concentration was higher in the co-compost alone (19.57 MPN/g), all KD soils possessed *E. coli* but was absent in the BS (F+Co). The high amount of co-compost at 200 kg/ha of N was added to the soil, which could have led to an increase in *E. coli* concentration compared to when half the recommended (F + Co) was added to the KD soil as observed in Figure 5.11a. The BS (F + Co) treatment, which combined a reduced amount of co-compost with chemical fertiliser, effectively suppressed *E. coli* proliferation, indicating a potential synergistic effect or reduced microbial input. Moreover, these results indicate that high application rates (200 kg N/ha) of co-compost may increase the risk of microbial contamination, particularly with *E. coli*, which can have implications for crop and human health. These results also signify the need for further research to explore the mechanisms leading to *E. coli* survival in high application rates of co-compost treatments. According to the WHO, (2006), a maximum threshold of 1000 MPN/g of coliforms is permissible for wastewater discharge into the environment thus the results in this study show that the coliforms were below the recommended threshold.

Potentially viable helminth eggs (*Taenia*, *Ascaris* and *Trichuris*) were high (0.02 eggs/g) in the co-compost only, KD (Co), BS (F+Co), however, were below the recommended 0.25 eggs per 4g of compost (Victor et al., 2008; WHO, 2006). The less helminth egg count combined with nutrient nutrient-rich nature of the co-compost implies its potential as a sustainable waste management and nutrient recovery technology. Adding moisture during irrigation may have created favourable conditions for microbial activity, leading to the regrowth of *E. coli* and helminth eggs (Zaleski et al., 2005). According to Skanavis and Yanko (1994), the co-compost application is more hazardous when applied to uncooked vegetables which may be grown in gardens (Skanavis and Yanko, 1994). This is because uncooked food is eaten raw, which can lead to the ingestion of pathogens. However, with maize, the cob is positioned above the soil and is covered with husks, protecting it from any pathogen contamination. Moreover, Musazura and Odindo (2021) observed that using decentralized wastewater for irrigation posed lower microbial contamination risks for maize than cabbage (*Brassica oleracea*) and lettuce (*Lactuca sativa*).

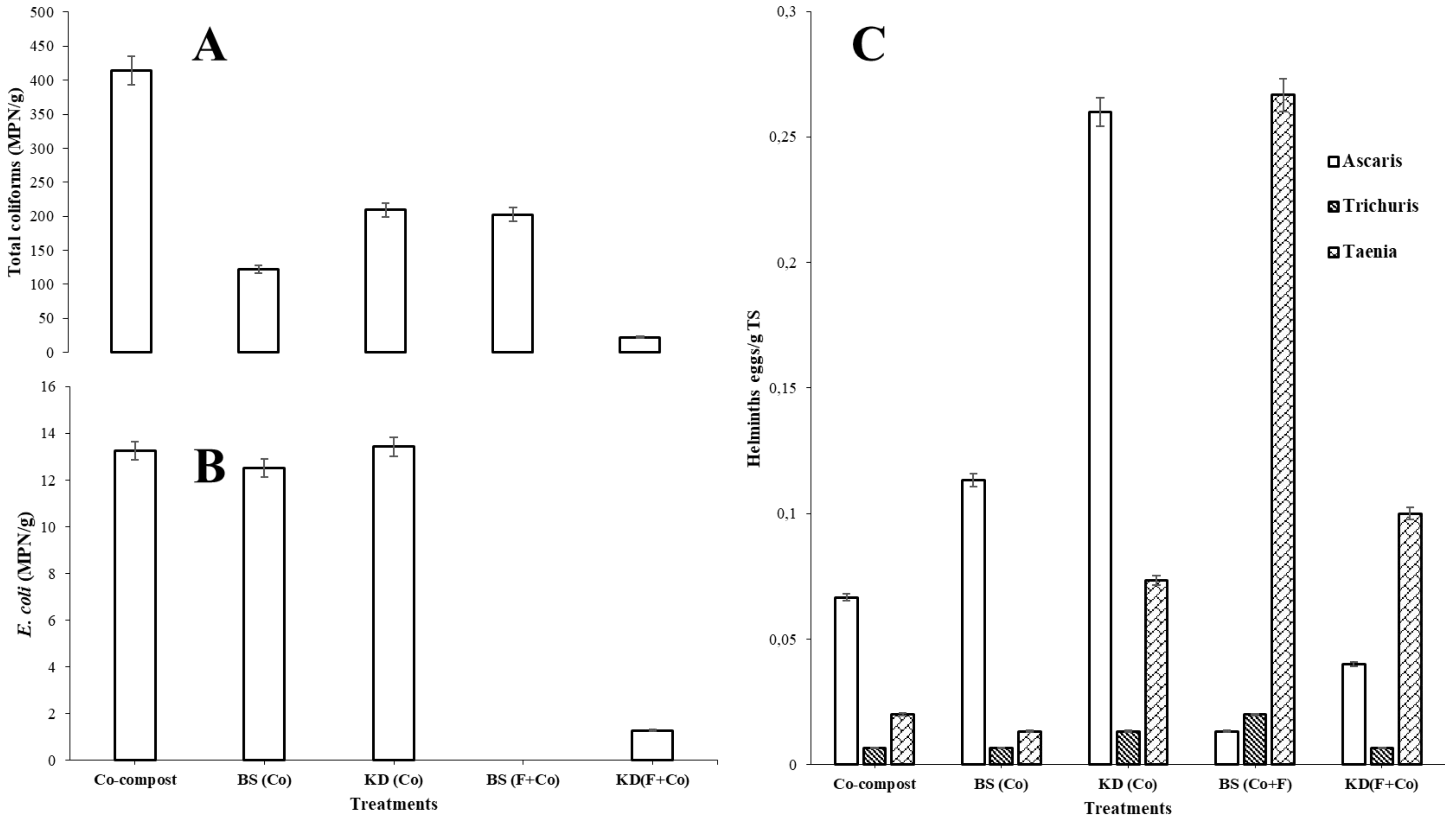


Figure 5.11: Pathogen concentration **A**- *E. coli* (MPN/g), **B**- total coliforms (MPN/g), and **C**- helminth eggs (eggs/g TS) in the treatments with co-compost.

5.4 Conclusion

In conclusion, the study demonstrated that combining co-compost and chemical fertiliser improved plant height of both the sandy loam soil (KD) and the clay loam soil (BS) which is the visual observation of plant growth. Moreover, the study demonstrated that co-compost has the added benefit of improving soil structure because of available N, organic matter and organic carbon and subsequently the benefit of supplying and translocating micro-nutrients into the grains. This was observed where treatments with co-compost improved micro-nutrients in the soil and maize grain, especially Zn, which is one of the essential micro-nutrients. The study further demonstrated that co-compost alone could support maize growth up to maturity however larger quantities would need to be added for the media to reach plant nutrient requirements. Therefore, co-compost alone can be a good growing media for high-value crops that are grown under controlled conditions, such as cucumber (*Cucumis sativus*), tomatoes (*Solanum lycopersicum*), and green pepper (*Capsicum annuum*). Moreover, co-compost can be used as the growing media for seedling production. Applying co-compost to the soil under irrigation did not significantly increase pathogen contamination in the soil; however, total coliforms and *E. coli* were found to be present in the co-compost only but at insignificant levels and still complied with the World Health Organization standards of less than 1000 MPN/g of dry weight. Moreover, the observed low concentrations of helminth eggs and microbial pathogens in the co-compost treatments demonstrate effective pathogen reduction during composting, ensuring compliance with safety guidelines. This supports the use of co-composted materials as safe and sustainable fertilisers in agriculture. Further research should focus on studying the mechanisms by which plants absorb micronutrients from soils amended with co-compost. Additionally, studies should investigate the uptake and removal of micropollutants and microplastics from co-composts to the plants to ensure their safety and sustainability for agricultural use.

References

Aguilera, J., Motavalli, P., Gonzales, M., and Valdivia, C. (2012). Initial and residual effects of organic and inorganic amendments on soil properties in a potato-based cropping system in the Bolivian Andean Highlands. *American Journal of Experimental Agriculture*.

Aly, H. (2002). Studies on keeping quality and storageability of cucumber fruits under organic farming system in greenhouses, M. Sc. Thesis, Fac. Agric., Cairo Univ., Egypt.

Amanullah, M. J. H., Nawab, K. and Ali, A., (2007). Response of specific leaf area (SLA), leaf area index (LAI) and leaf area ratio (LAR) of maize (*Zea mays L.*) to plant density, rate and timing of nitrogen application. . *World Applied Sciences Journal* **2(3)**, 235-243.

Amujoyegbe, B., Opabode, J., and Olayinka, A. (2007). Effect of organic and inorganic fertilizer on yield and chlorophyll content of maize (*Zea mays L.*) and sorghum *Sorghum bicolour* (L.) Moench. *African Journal of Biotechnology* **6**.

An, H., Lee, Y., Lee, J., Yoon, S., Park, S.-J., Hong, S.-C., Kim, J.-H., and Kim, S. Y. (2023). Does Co-Application of Compost and Mineral Fertilizers Reduce Gaseous Nitrogen (NH3 and N2O) Losses and Improve Nitrogen Use Efficiency from Maize-Growing Soil? *Korean Journal of Soil Science and Fertilizer* **56**, 119-125.

Asaye, Z., Kim, D.-G., Yimer, F., Prost, K., Obsa, O., Tadesse, M., Gebrehiwot, M., and Brüggemann, N. (2022). Effects of combined application of compost and mineral fertilizer on soil carbon and nutrient content, yield, and agronomic nitrogen use efficiency in maize-potato cropping systems in southern Ethiopia. *Land* **11**, 784.

Azhar Ghaffari, A. G., Asghar Ali, A. A., Muhammad Tahir, M. T., Muhammad Waseem, M. W., Ayub, M., Asif Iqbal, A. I., and Mohsin, A. (2011). Influence of integrated nutrients on growth, yield and quality of maize (*Zea mays L.*). *American Journal of Plant Sciences*.

Bhatt, M. K., Labanya, R., and Joshi, H. C. (2019). Influence of long-term chemical fertilizers and organic manures on soil fertility-A review. *Universal Journal of Agricultural Research* **7**, 177-188.

Chen, D., Yuan, L., Liu, Y., Ji, J., and Hou, H. (2017). Long-term application of manures plus chemical fertilizers sustained high rice yield and improved soil chemical and bacterial properties. *European Journal of Agronomy* **90**, 34-42.

Chen, J.-H. (2006). The combined use of chemical and organic fertilizers and/or biofertilizers for crop growth and soil fertility. *In* "International workshop on sustained management of the soil-rhizosphere system for efficient crop production and fertilizer use", Vol. 16, pp. 1-11. Citeseer.

Ebrahimpour, F., Eidizadeh, K., and Damghani, A. (2011). Sustainable nutrient management in maize with integrated application of biological and chemical fertilizers. *International Journal of AgriScience*.

El-Sheikh, T., Abd El Rahmen, S., and Dessouky, S. (1993). Effect of polyethylene wrapping on keeping quality and storage of cucumber fruits, Zagazis J. *Agric, Res* **20**, 78.

Ghosh, A., Dey, K., Bhowmick, N., Medda, P., and Ghosh, S. (2016). Impact of different pruning severity and nutrient management on growth and yield of lemon cv. Assam lemon (*Citrus limon Burm.*). *Vegetos* **29**, 25-32.

Girija, R., Shettigar, N. A., Parama, V. R. R., and Gagana, S. (2019). Evaluation of co-composted faecal sludge application in agriculture. *WIT Transactions on Ecology and the Environment* **238**, 701-711.

Hongliang Fang., F. B., Stephen Plummer., Gabriel Schaepman-Strub., (2019). An overview of global leaf area index (LAI): methods, products, validation, and applications. *Rev. Geophys.* **57**, 739–799.

Ion, V., Dicu, G., Dumbravă, M., Temocico, G., Alecu, I. N., Băşa, A. G., and State, D. (2015). Harvest index at maize in different growing conditions. *Romanian Biotechnological Letters* **20**, 10951.

Jatav, H. S., Singh, S. K., Rajput, V. D., and Minkin, T. (2021). "Sustainable Soil Fertility Management," Nova Science Publishers.

Kiani, A. K., Dhuli, K., Donato, K., Aquilanti, B., Velluti, V., Matera, G., Iaconelli, A., Connelly, S. T., Bellinato, F., and Gisondi, P. (2022). Main nutritional deficiencies. *Journal of preventive medicine and hygiene* **63**, E93.

Liu, B., Wang, X., Ma, L., Chadwick, D., and Chen, X. (2021). Combined applications of organic and synthetic nitrogen fertilizers for improving crop yield and reducing reactive nitrogen losses from China's vegetable systems: A meta-analysis. *Environmental Pollution* **269**, 116143.

Lv, F., Song, J., Giltrap, D., Feng, Y., Yang, X., and Zhang, S. (2020). Crop yield and N_2O emission affected by long-term organic manure substitution fertilizer under winter wheat-summer maize cropping system. *Science of the Total Environment* **732**, 139321.

Majumdar, K., Sanyal, S. K., Dutta, S. K., Satyanarayana, T., and Singh, V. (2016). Nutrient mining: addressing the challenges to soil resources and food security. *Biofortification of food crops*, 177-198.

Mandizvo, T., and Odindo, A. (2019). Seed mineral reserves and vigour of Bambara groundnut (*Vigna subterranea* L.) landraces differing in seed coat colour. *Heliyon* **5**.

Mandizvo, T., Odindo, A. O., Mashilo, J., and Magwaza, L. S. (2022). Drought tolerance assessment of citron watermelon (*Citrullus lanatus var. citroides* (LH Bailey) Mansf. ex Greb.) accessions based on morphological and physiological traits. *Plant Physiology and Biochemistry* **180**, 106-123.

Miao, Y., Stewart, B. A., and Zhang, F. (2011). Long-term experiments for sustainable nutrient management in China. A review. *Agronomy for Sustainable Development* **31**, 397-414.

Musazura, W., and Odindo, A. O. (2021). Suitability of the Decentralised Wastewater Treatment effluent for agricultural use: Decision Support System approach. *Water* **13**, 2454.

Natsheh, B., and Mousa, S. (2014). Effect of organic and inorganic fertilizers application on soil and cucumber (*Cucumis sativa* L.) plant productivity. *International Journal of Agriculture and Forestry* **4**, 166-170.

Ojeniyi, S. (2002). Soil management, national resources and environment. *Nigeria Journal of Soil Science* **16**, 131-135.

Qasim, M., Ju, J., Zhao, H., Bhatti, S. M., Saleem, G., Memon, S. P., Ali, S., Younas, M. U., Rajput, N., and Jamali, Z. H. (2023). Morphological and physiological response of tomato to sole and combined application of vermicompost and chemical fertilizers. *Agronomy* **13**, 1508.

Qiu, F., Liu, W., Chen, L., Wang, Y., Ma, Y., Lyu, Q., Yi, S., Xie, R., and Zheng, Y. (2021). Bacillus subtilis biofertilizer application reduces chemical fertilization and improves fruit quality in fertigated Tarocco blood orange groves. *Scientia Horticulturae* **281**, 110004.

Reckitt-Benckiser, R. (2015). Rate of photosynthesis: Limiting factors. Cambridge, UK: Royal Society of Chemistry. https://edu. rsc. org/download.

Roba, T. B. (2018). Review on: The effect of mixing organic and inorganic fertilizer on productivity and soil fertility. *Open Access Library Journal* **5**, 1.

Skanavis, C., and Yanko, W. A. (1994). Evaluation of composted sewage sludge-based soil amendments for potential risks of salmonellosis. *Journal of Environmental Health*, 19-23.

Slamet, W., Purbajanti, E.D., Darmawati, A. and Fuskhah, E., (2017). Leaf area index, chlorophyll, photosynthesis rate of lettuce (*Lactuca sativa L*) under N-organic fertilizer. *Indian Journal of Agricultural Research* **51(4),** , pp.365-369.

Valentinuz, O. R. a. T., M., (2006). Effect of genotype, nitrogen, plant density, and row spacing on the area-per-leaf profile in maize. *Agronomy Journal* **98(1)**, 94-99.

Verma, A., Nepalia, V., and Kanthaliya, P. (2006). Effect of integrated nutrient supply on growth, yield and nutrient uptake by maize (*Zea mays*)–wheat (*Triticum aestivum*) cropping system. *Indian journal of agronomy* **51**, 3-6.

Victor, R., Kotter, R., O'Brien, G., Mitropoulos, M., and Panayi, G. (2008). WHO guidelines for the safe use of wastewater, excreta and greywater, volumes 1–4. *Taylor & Francis*.

WHO (2006). "WHO guidelines for the safe use of wasterwater excreta and greywater," World Health Organization.

Xue, Y.-F., Li, X.-J., Yan, W., Miao, Q., Zhang, C.-Y., Huang, M., Sun, J.-B., Qi, S.-J., Ding, Z.-H., and Cui, Z.-L. (2023). Biofortification of different maize cultivars with zinc, iron and selenium by foliar fertilizer applications. *Frontiers in Plant Science* **14**, 1144514.

Zaleski, K. J., Josephson, K. L., Gerba, C. P., and Pepper, I. (2005). Survival, growth, and regrowth of enteric indicator and pathogenic bacteria in biosolids, compost, soil, and land applied biosolids. *J. Residuals Sci. Technol* **2**, 49-63.

Zerssa, G. W., Kim, D.-G., Koal, P., and Eichler-Löbermann, B. (2021). Combination of compost and mineral fertilizers as an option for enhancing maize (*Zea mays L.*) yields and mitigating greenhouse gas emissions from a Nitisol in Ethiopia. *Agronomy* **11**, 2097.

Chapter 6: Attitudes, Perceptions and Willingness of University Students Towards Purchasing and Consuming Food Produced using Human Excreta-derived Co-compost

Abstract

This study investigated university students' attitudes, perceptions and willingness towards purchasing and consuming food produced using human excreta-derived co-compost. The study interviewed 553 students from the University of KwaZulu-Natal, Pietermaritzburg campus using Computer-Assisted Personal Interviewing (CAPI) technique. The theory of planned behaviour was used to predict behavioural intentions including the factors that affect the attitudes, perceptions and subjective norms towards the use of human excreta-derived co-compost. A descriptive-analytical framework that utilised both descriptive and inferential statistics was used to analyse the data using the Statistical Package for Social Scientists (SPSS) software. The study results show that the majority (76.7%) of the respondents were enrolled for undergraduate bachelor’s degrees. The student attitudes toward food produced using co-compost were mainly influenced by awareness of human excreta reuse in agriculture, gender, settlement type, age, religious practice, and the respondents' study level. Most respondents who were aware of human excreta reuse in agriculture had a positive attitude towards consuming food produced using co-compost. Male students were more positive than female students towards consuming and purchasing food produced using human excreta-derived co-compost. The respondents were more willing to consume food produced using co-compost if it was processed (85.4%), grows above ground (84.2%), and cooked (82.9%) compared to food that is eaten raw (62.3%) and crops that grow below ground (70%). In conclusion, the study findings demonstrate the positive attitude and willingness of students to accept human excreta-derived co-compost for food production. This study recommends designing educational material and dissemination strategies to increase the knowledge and awareness of consumers on the use of human excreta-derived fertilisers in agriculture or food production.

Keywords: Attitudes, agriculture, co-compost, consumers, human-excreta, perceptions

6.1 Introduction

With the increase in the cost of mineral fertilizer and enough quantities of organic waste in many African cities, recovering nutrients from these organic wastes could provide the greatest advantage to smallholder farmers (Cofie et al., 2010). Organic waste material that can be recycled includes municipal solid waste (Soré and Ouoba, 2023), faecal sludge (Koné et al., 2007; Manga et al., 2022; Soré and Ouoba, 2023), sewage sludge (Zhang et al., 2018), urine (Migeri et al., 2023), chicken manure (Ofei-Quartey et al., 2023), and food waste (Waqas et al., 2018). Technologies that were designed to recover nutrients from organic waste including human excreta are biochar pyrolysis (Nkomo et al., 2021), Latrine biosolid Dehydration and Pasteurization (Harrison and Wilson, 2012), black soldier fly larvae (Mertenat et al., 2019), and co-composting (Manga et al., 2023; Ofei-Quartey et al., 2023).

Co-composting is a low-cost technology and low-investment process to add value to organic solid waste through conversion into an organic fertiliser known as compost (Anwar et al., 2015). Moreover, it presents natural processes used to minimize the disposal of solid waste with the added benefit of resource regeneration through stabilizing different types of organic waste (Anwar et al., 2015). The process of composting serves as an alternative, acceptable, and economically viable way of recovering and re-using nutrients from faecal sludge (Manga et al., 2022). This process involves thermophilic temperature which occurs as a result of microbial activities and leads to the deactivation of pathogens (Preneta et al., 2013). The thermophilic temperatures can lead to the deactivation of helminths eggs and *Escherichia coli* (*E. coli*) to the acceptable standard of the World Health Organisation (Koné et al., 2007; Manga et al., 2021; Preneta et al., 2013).

Peer-reviewed studies have demonstrated the benefits of applying co-compost on land in terms of enhanced soil water-holding capacity, nutrient retention, and soil structure (Guzha et al., 2005; Wang et al., 2019). However, the wide-scale acceptance of the co-compost value chain still prevails as a significant challenge and barrier to consumers (Viaene et al., 2016). Examining the factors that shape market demand for co-compost can provide valuable insights into addressing the challenges faced by recovery and reuse innovations, enabling a deeper understanding of the product's market dynamics (Gwara et al., 2022).

The perceived behaviour intention of people about human excreta is influenced by age, gender, religion, education, income, occupation and surroundings (family, friends or colleagues) and situations (Simha et al., 2018). Sociological studies to investigate the social acceptance of human excreta-derived products by farmers have been well-documented in the literature (Cofie et al., 2010; Gwara et al., 2022; Simha et al., 2017). However, Chauha and Ranawat (2024), emphasised that consumers' perceptions and preferences are crucial for producers, marketers, and policymakers to meet market demand. Moreover, Gwara et al. (2022), suggested that the effect of crop type, processing, and cooking could influence attitudes, willingness and perceptions of consumers and farmers towards accepting human excreta-derived fertilizers. However, very scant research has been conducted to verify and assess the willingness of consumers to purchase and/or eat food grown using human excreta-derived products especially co-compost. Moreover, research has not been done to study the factors that influence consumer behaviour based on the fruit or vegetable position in the plant and whether value addition could change consumers' perception of accepting human excreta-derived fertilisers. Therefore, this study aimed to assess the attitudes, perceptions and willingness of university students towards purchasing and consuming food produced using human excreta-derived co-compost.

6.2 Materials and methods

6.2.1 Study area

This study was conducted at the University of KwaZulu-Natal (UKZN), Pietermaritzburg (PMB) campus, (29°62'0" **S** and 30°39'6" **E**). The University of KwaZulu-Natal has five (5) campuses, the Westville campus (main campus), the Nelson R. Mandela School of Medicine, the Howards College campus, the Edgewood campus, and the Pietermaritzburg campus. It further has four colleges including the College of Agriculture, Engineering and Science (CAES), College of Health Science (CHS), College of Humanities (CH), and College of Law and Management Studies (CLMS), which are further differentiated into 19 schools or departments. Moreover, during the time of study, approximately 44991 students were registered as undergraduates and postgraduates. This study was thus carried out in the PMB campus which had 9125 students https://ii.ukzn.ac.za/Report/StudentRegistrations. The number of students registered increases timeously due to students being enrolled for different courses. The PMB campus has different colleges including CAES, CH, and CLMS. Moreover, the students from the Westville campus in the CHS are transferred in their fifth year to the PMB campus thus making the PMB campus a representative of this study.

6.2.2 Survey instruments

A total of 553 consumers were interviewed from the UKZN, PMB campus after obtaining the Humanities and Social Sciences Research Ethics Committee (HSSREC/00005270/2023). The verbal consent was obtained from participants before beginning the survey and participants were free to withdraw at any time of the study. The participants were also provided with the purpose of the study, the of the survey, and the confidentiality of the study provided in the link https://docs.google.com/forms/d/e/1FAIpQLSejp8nDjEoukf060HkUhwdeyP8eQ0PDlkYkVSdIoKqe11uSVQ/viewform?usp=pp_url. Sample power analysis was carried out using G*Power version 3.1.9.7 software based on the power of 0.95, alpha 0.05, and Cohen's effect size of 0.15 which provided a sample size (n) of 146. However, the sample size was increased to 558 to provide more accurate results and to make a representative of the population. The Computer-Assisted Personal Interviewing (CAPI) technique was used to interview 553 students who consented to participate in this survey as per a study by (Bramlett et al., 2024). Responses from the respondents were recorded using a Google form (Google LLC, United States, 2008) and downloaded as an Excel spreadsheet.

Eleven enumerators enrolled for honours and master's degrees at UKZN were trained to improve their face validity, create confidence, and improve the quality of the data. The interviews were facilitated using the IsiZulu language (highly populated in KwaZulu-Natal) for Zulu speakers and English for students whose native language was not IsiZulu. The study surveyed students from UKZN as part of an ongoing research initiative involving small-scale farmers in the Vulindlela community conducted by (Gwara et al., 2022). Individuals or participants were selected randomly from the three campuses of UKZN-PMB which were the agriculture campus, the Main Campus, and the Golf Road campus.

This study assessed the general attitudes of the UKZN students to purchase and/or consume food produced using co-compost which is produced from human excreta (dewatered sewage sludge) and organic green waste (Chapter 3). Close-ended questions required two types of responses where "2" represents agreement (2 = Yes) and "1" represents disagreement (1 = No). Moreover, this led to the mean response of $1 \leq \mu \leq 2$ (Lamichhane et al., 2013; Simha et al., 2018) which allowed us to compute the one-way analysis of variance (ANOVA) test. Significant differences between specific variables were assessed by computing a post hoc test including Tukey's Honestly Significant Different test at alpha less than 10%.

The questionnaire comprised three sections (after seeking participants) consent, demographic information including, gender, age, religious practice, settlement type, source of income, monthly expenditure on food, level of study, and college affiliation. The second section of the questionnaire was based on the perceived risks associated with human excreta handling (4 questions), knowledge of the use of human excreta for agricultural purposes (1 question), willingness to purchase and/or consume food grown using human excreta derived co-compost (5 questions) and factors influencing their attitudes (5 questions). The last part of the questionnaire was the comment section which participant stated their opinion and views about the use of human excreta-derived co-compost on crop production (Appendix 4).

6.2.3 Combined effect

The attitudes towards the behaviour, subjective norms, and perceived behavioural control score were based on a multiple-choice question (Q19, Q20, Q27 and Q28) in the survey instrument. Where attitude towards the behaviour question was investigating whether human excreta can be treated so as not to pose health risks, (Q19), and whether students/consumers think human excreta should be disposed of and never be used (Q20), were coded as either yes = 2 or no = 1. Subjective norms (Q27) indicate that the decision-making of an individual is influenced by friends, family or colleagues, thus participants were asked if they think their friends or colleagues would purchase and consume food fertilised using co-compost, these questions were coded as either yes = 2 or no = 1.

The perceived behavioural control (Q28) assesses individuals' ability to perform a specific behaviour, in this study, participants were asked whether they could consume food produced using co-compost, coded either yes = 2 or 1 = no. The combined effect included all the questions (Q19, Q20, Q27 and Q28) mentioned above. It was used to assess how different variables are influenced by the combined effect of subjective norms, attitudes towards the behaviour and perceived behavioural control. The mean score of the perception was calculated using Equation 6.1 below.

$$\text{Mean score} = \frac{\text{Total score}}{\text{Number of respondents}} \qquad \text{Equation 6.1}$$

For statistical analyses, the perception score <1.5 was treated as a negative attitude, then the attitude score =1.5 was treated as a neutral (neither positive nor negative) attitude, and scores of >1.5 were treated as a positive attitude towards recycling human manure for use in agriculture.

6.2.4 Theory of planned behaviour

According to WHO, (2008), the theory of planned behaviour (TPB) is used to predict human behaviour. These include attitude towards the behaviour, perceived behavioural control, and subjective norms. Perceived behavioural control refers to how a person can undertake new technology and behaviour under investigation in an acceptable manner (Ajzen, 1991). Subjective norms refer to an individual's perception of social pressure from friends, family, and close contacts, which influences their attitudes and behaviours (Gwara et al., 2023). The TPB also looked at the perceived behavioural control which determines the ease and difficulties in performing the behaviour of interest (Ajzen, 2002). The combination of subjective norms, attitude towards the behaviour and perceived behavioural control shape an individual's behavioural intentions (Mariwah and Drangert, 2011). As shown in Figure 6.1 the theory of planned behaviour was used to predict the behaviour of the respondents.

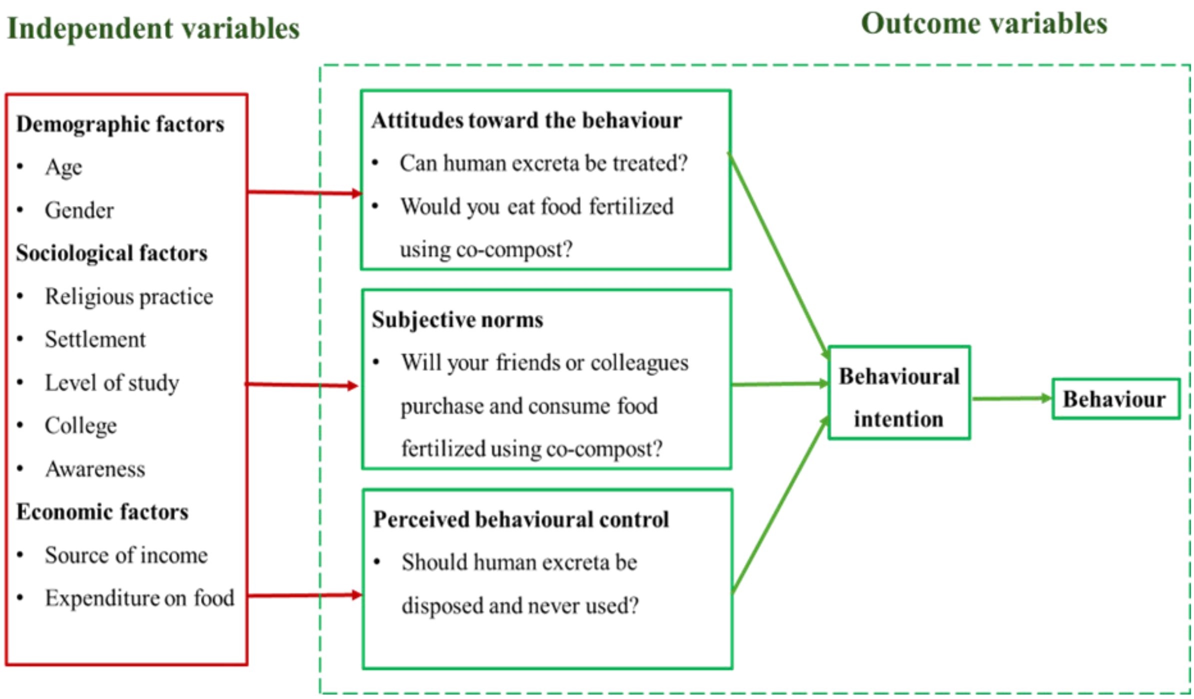


Figure 6.1: Conceptual framework of the factors influencing the willingness of consumers to purchase and/or consume food grown using co-compost derived from human excreta adapted from (Gwara et al., 2022).

6.2.5 Data and statistical analysis

The survey recorded 558 responses in four days, with 553 (99.1%) consenting to participate and only 5 (0.9 %) did not consent to participate. Based on the total population of (N=9125), the sample size of (n=553) with 95% confidence intervals, the marginal error was estimated to be less than 0.05. The data was analysed using the descriptive-analytical approach using IBM SPSS statistics version 29.0.1.0 (IBM Corporation, Armonk, New York, USA). The descriptive statistics included mean, variances, frequencies, and percentages and the inferential statistics which included the Chi-square test and the one-way ANOVA to analyse data.

6.3 Results

6.3.1 Socio characteristics of consumers/students

Reporting the demographic and socioeconomic characteristics of the study participants helps to understand the segregated attitudes of the participants/respondents for customer segmentation (Gwara et al., 2022). Approximately, 51.9% of the 553 students that participated in the survey were female, 46.1% were male and 2.0% (other) did not identify themselves as either male or female (Table 6.1). The majority (44.8%) of the participants were between the age of 20-22 years of age while 24.2% were 17-19 years, 23.0% were between 23-25 years, 6.3% were between 26-30 years, 1.4% were between 31-40 years, and only 0.2% were greater than 41 years. The majority (54.8%) of the participants originally came from rural settlements, 29.8% from urban townships, and 15.4% came from urban suburbs. Most students (37.4%) spend R 501.00 – R 900.00 monthly on the food. The rest of the participants spend either R 901.00 – R 1 200.00 (31.3%), R 1201.00 – R 1 500.00 (12.1%), or R 1 501.00 – R 2 000.00, and 6.1 % of the population spend less or equal to R 500.00 and greater or equal to R 2 001.00 monthly on food.

The majority (66.7%) of the population in this survey were funded by undergraduate bursaries, while 25.2 % were self-funded and the minority (8.0%) received postgraduate scholarships or bursaries. Moreover, out of 140 self-funded students, 73 (52.1%) were postgraduate students while 67 (47.9 %) were undergraduate students. The highest number of participants were from the College of Humanities (CH) and the College of Agriculture, Engineering and Science (CAES) with values of 226 (40.9%) and 225 (40.7%), respectively. The College of Law and Management represented 17.0% of the population while the minority (1.4%) were from the College of Health Science. In the College of Health Science, students are usually enrolled in the Westville campus however their practicals are usually undertaken in Pietermaritzburg when they do 5th year, this could be a reason for the low participation of CHS in the survey. The majority, 55.2% of the participants were unaware of the use of human excreta for agricultural purposes while only 44.8% were aware of the use of human excreta for agricultural purposes.

Table 6.1: Socioeconomic characteristics of the respondents

Variable	Population N	Percentage %
Gender (n=553)		
• Male	255	46.1
• Female	287	51.9
• Other	11	2.0
Age years (n=553)		
• 17-19	135	24.2
• 20-22	248	44.8
• 23-25	127	23.0
• 26-30	35	6.3
• 31-40	7	1.4
• ≥41	1	0.2
Religious practice (n=553)		
• Hinduism	7	1.3
• Islam	8	1.4
• Polytheism	2	0.2
• Christianism	263	47.6
• Shembe/Nazareth	68	12.3
• Atheist (No God)	13	2.4
• Agnostic (Don't know)	9	1.6
• Traditionalism and Christianism	126	22.8
• Traditionalism	50	9.0
• Scientism (I believe in science)	7	1.3
Settlement type (n=553)		
• Rural areas	303	54.8
• Urban (Township)	165	29.8
• Urban (Suburbs)	85	15.4
Source of income (n=553)		
• Self-funded	140	25.3
• Postgraduate bursary	44	8.0
• Undergraduate bursary	369	66.7
Monthly expenditure (n=553)		
• < R 500.00	34	6.1
• R 501 – R 900	207	37.4
• R 901 – R 1 200	173	31.3
• R 1201 – R 1 500	67	12.1
• R 1501 – R 2 000	38	6.9
• ≥ R 2001	34	6.1
Level of study (n=553)		
• Diploma	1	0.2
• Postgraduate diploma	11	2.0
• Bachelor's degree	424	76.7
• Honours degree	63	11.4
• Master's degree	39	7.1
• Doctorate	14	2.5
• Post Doctoral Fellowship	1	0.2
College of study (n=553)		
• CAES	225	40.7
• CH	226	40.9
• CHS	8	1.4
• CLMS	94	17.0
Awareness of human excreta use in agriculture (553)		
• Yes	248	44.8
• No	305	55.2

N (population), **CAES** (College of Agriculture, Engineering and Science), **CH** (College of Humanities), **CHS** (College of Health Science), **CLMS** (College of Law and Management Studies)

6.3.2 Consumers' perceptions of recycling human excreta

According to Gwara et al., (2021), health risk perceptions are among the most reported stigmas regarding accepting and reusing human excreta-derived products in crop production. Moreover, Mugivhisa et al., (2017) found that smell, unhygienic, and fear of diseases and pathogens were the main factors leading to human excreta being unacceptable among farmers. In addition to this Buit and Jansen, (2016), indicated a negative attitude towards farmers accepting fresh urine and faeces however, they were willing to accept products made from them. In our present study, most consumers (89.3%) were aware that human excreta contain pathogens and there was no significant difference ($p>0.1$) across all demographic variables (Table 6.2). Moreover, 83.2% of the population believed that human faeces pose health risks if they come into contact with a person's skin or are contaminated with food. Interestingly, the majority (86.3%) of consumers think that human excreta can be treated to not pose health risks. These results outline a change in consumers' perceptions towards accepting human excreta-derived fertilisers or organic amendments.

The study found a significant association between consumers' age and their attitudes toward the disposal of human excreta ($p=0.008$). Specifically, as participant age increased, individuals were more likely to recognize the value of human excreta and express willingness to recycle it for other uses. Moreover, gender significantly influenced ($p<0.001$) consumer attitudes. Among female students, 54.7% believed that human excreta could be recycled and reused for other purposes, compared to 70.7% of male students, and 72.7% of those identified as 'others. This suggests a comparatively lower acceptance among female students towards recycling human excreta. There was a significant difference in attitudes based on settlement type ($p=0.032$). Among students who live in rural settlements, 66.0% believed that human excreta should not be disposed of compared to 61.8% from urban townships, and 50.5% of urban suburb consumers. People from rural areas are often engaged in farming thus making them both farmers and consumers which could have subsequently led to a positive attitude towards acceptance of human excreta-derived co-compost. Additionally, the type of college affiliation influenced attitudes significantly ($p=0.053$). Among the participants from different colleges, 69.0% from CAES, 59.3% from the CH, 55.3% from the CLMS, and a minority (50.0%) from the CHS expressed willingness to recycle and reuse human excreta for agricultural purposes.

According to Gwara et al., (2022), farmers' and consumers' attitudes toward reusing human excreta-derived products in agriculture are primarily shaped by respondents' awareness of the agricultural applications of human excreta. In our present study, the majority (55.2%) of consumers were not aware of the use of human excreta in agriculture. Moreover, awareness of human excreta reuses in agriculture significantly (p=0.030) differed between different age groups. In addition to this gender also significantly differed (p=0.010), and the majority (61%) of female students and only 49.2% of male students were not aware of the use of human excreta for agricultural purposes. Interestingly, among respondents from the CAES, 52.2% and a minority (25.0%) from the CHS were aware of the use of human excreta for agricultural purposes. The settlement type was significantly different (p=0.012), within each settlement category, 69.4% of the students from urban suburbs, 54.5% from urban townships, and only 51.3% from rural areas were aware of the use of human excreta for agricultural purposes. The study further found that there was a significant difference (p=0.044) between the level of study most (71.4%), where doctoral students were aware of the use of human excreta for agricultural purposes.

The willingness to purchase food grown with co-compost varied significantly by gender (p=0.005). Within each gender category, 81.8% of respondents identified as 'other,' 77.3% of male students, and 62.7% of female students expressed a willingness to buy food produced using co-compost. Female students generally exhibited less acceptance of purchasing food produced using co-compost. Additionally, among students from rural areas, 75.7% indicated they would purchase food produced using co-compost, compared to a smaller proportion of 55.3% from urban townships. Differences in income sources also influenced purchasing decisions (p=0.075); among undergraduate students, 71.4% supported by undergraduate bursaries or self-funded were inclined to buy co-compost-grown food, while a lower percentage (52.3%) of postgraduate bursary recipients expressed the same. Postgraduate students who receive bursaries typically ten times more than undergraduate bursary recipients, giving them more flexibility in their purchasing choices. This increased financial independence may contribute to their more negative attitude toward buying food produced using co-compost.

Price is one important factor that can influence consumers' attitudes and perceptions toward purchasing a certain product as it involves psychological perceptions, economic limitations, and value assessment (Dodds et al., 1991). The influence of price on changing consumers' attitudes towards purchasing food produced using co-compost was studied. There was a significant difference (p=0.076), in gender, within each gender category a lower proportion of female consumers (61.4%) compared to 69.8% male and 80.0% 'other' would be influenced by price towards purchasing food grown using co-compost. Religious practice significantly differed (p=0.031) and amongst the Islam religion only 12.5% would be influenced by the price to purchase food produced using co-compost while in other religious practices, more than 50% of the respondents indicated they can be positively influenced by the price to purchase food produced using co-compost.

The level of study differed (p=0.007) significantly, between each study level category, a higher proportion of doctoral students (79.0%) would not be influenced by price while 68.4% of bachelor's and 100% diplomates and postdoctoral fellowships would be influenced by price in buying food produced using co-compost. The study further found that 78.5% of the consumers would be influenced by the quality of the product including shelf-life on accepting food produced using co-compost. Moreover, the study found that the quality of the product in changing consumers' behaviour was a significant difference between religious practices (p=0.022) and the level of study (p=0.091). The issue of price was coupled with comments from the respondents that food produced using co-compost should cost more than chemically fertilized food because people working with the human excreta to make co-compost will be exposed to pathogen contamination and other health risks. Moreover, one respondent added "*Thina maZulu sikholwa ukuthi amasimba aletha amabhadi, uma uphuphu amasimba noma uwasebenzisa uba namabhadi ngakho abasebenzi abenza lomquba kufanele banxephezelwe*", meaning "*in our Zulu culture we believe that faeces bring bad luck if you dreamt of faeces or working with them you expose yourself to bad luck thus people making co-compost from faeces should be compensated*".

A person's attitude towards accepting a certain practice or technology is mainly influenced by the surrounding people including friends, family members, colleagues, and the surrounding community. When consumers/students were asked if their friends or colleagues would purchase and/or eat food produced using co-compost significant differences were observed in age (p=0.020), gender (p=0.068), and settlement type (p=0.009). Within each group category, 47.5% of students between the age of 17-25 years, compared to the majority (65.7%) above 25 years indicated that their friends would not purchase/consume food produced using co-compost. Moreover, among the settlement category, 61.2% of consumers/students from suburbs had a negative attitude and thought their friends would not purchase/consume food fertilised using co-compost compared to only 43.2% from rural areas and 51.2% from urban townships.

Consuming food produced using co-compost significantly differed across ages (p=0.029). The was an increase in willingness to consume food produced using co-compost as the age of the students increased. Indicating that as a person matures, they see value in human excreta-derived fertilisers thus exhibiting an increase in acceptance and taking decisions about certain technologies. Gender also significantly differed (p=0.010), among female students, only 69.6% were willing to consume food produced using co-compost compared to male students (80.1%) and others (90.9%). The religious practice also significantly differed (p=0.059), within each religious group, 85.7% of the students who believe in science, 71.9% of Christians and a minority (37.5%) of Islam would consume food produced using human excreta-derived co-compost. The scientific worldview might make individuals more open to sustainable practices like waste-to-resource initiatives. The lower acceptance may be influenced by religious teachings or cultural norms associated with cleanliness and purity, which can sometimes shape attitudes toward using human-derived compost in agriculture.

College affiliation significantly differed (p=0.030) and less acceptance was observed in the students from the College of Health Science and the College of Law and Management Studies. Moreover, students from rural areas (78.3%) were more willing to consume food produced using human excreta-derived compost compared to students from urban suburbs (61.2%) thus making it significantly different (p=0.005). One factor that could enhance rural area settlers ' willingness to eat food produced using human excreta-derived compost could be that they are directly exposed to agricultural activities thus making them see value in human excreta as a fertiliser that can improve crop yield.

Change in consumers' perceptions, attitudes, and willingness to eat food produced using co-compost was greatly influenced by processes it undergoes before consumption. In our present study, we observed that 62.3% of consumers would eat food produced using co-compost if it was eaten raw such as lettuce (*Lactuca sativa*) and/or cucumber (*Cucumis sativus*). Eating food raw significantly differed between age (p=0.006), gender (p<0.001), and settlement type (p=0.065). The study found that acceptance increased with age, male students and others were more willing than female students and consumers from rural areas had a positive attitude and were willing to consume food grown using co-compost if eaten raw. Interestingly, there was an increase of 82.9% in consumers' willingness to eat food produced using co-compost if it was cooked or boiled such as Swiss chard (*Beta vulgaris* L. var. cicla) and/or cabbage (*Brassica oleracea*). Leading to significant differences between gender (p=0.002), religion (p=0.092), and settlement type (p=0.022). Approximately, 70.0% of the student population were willing to eat food produced using co-compost if it grows below ground such as beetroots (*Beta vulgaris*) and/or potatoes (*Solanum tuberosum*). Significant differences were observed between age (p=0.009), gender (p=0.017), settlement type (p=0.015), and college affiliation (p=0.039). However, an increase (84.2%) in willingness was observed in the crop that grows above the ground such as maize (*Zea mays*) and/or sorghum (*Sorghum bicolor*). Moreover, significant differences were observed between gender (p=0.019) and settlement type (p<0.001). In addition to this, attitudes towards processed food were even higher (85.4%) indicating that value addition in the crops might temper with consumers' attitudes. Significant differences were observed between gender (p=0.053), settlement type (p=0.009), and religious practice (p=0.023). The findings from this study confirm assumptions by Gwara et al., (2022) who emphasised that farmers' acceptance of food produced using human excreta-derived fertilizers could be changed through food processing.

This study has demonstrated that willingness to consume food produced using co-compost can be affected by the type of food whether it is eaten cooked, raw, boiled, or processed. Moreover, it was observed that the yuck factor is provoked by the fruit/crop in relation to the soil and that most consumers think that pathogens could easily contaminate food if the plant gets into contact with the co-compost. Participants who were not willing to consume food produced using co-compost indicated that they could only eat it if it was produced without their knowledge and if it was not disclosed to them. This was coupled with responses/comments that, "*Just the thought of it makes me lose appetite*", "*I think I will be disgusted if I knew before consumption*", and "*I can't remove the image of faeces being the starting point of what I am consuming*", *"in our society we know you can't touch faeces",* and *"I have less knowledge about co-compost"*.

Most students who were willing to consume food produced using co-compost indicated concern about the environment. This was coupled with responses including, "*It now co-composts so it is safe to eat food produced using it*", and "*I don't see the difference between human excreta and animal excreta*". Some students were more on the nutritional aspect of human excreta, "*I think the food will be more nutritious*", and "*If it's of good quality and can be sold at lower prices*". Others disclosed that their parents are already producing crops near toilets because of the high nutrients next to the toilets. Interestingly, some individuals indicated that "*I want to try it!*", and "*I am curious about the food*". These responses indicate a positive view of consumers towards the consumption of crops grown using human excreta-derived co-compost.

Table 6.2: Results from the Chi-square test indicating whether consumer responses to various questions (Q) varied significantly between different categories of demographic variables

Demographic variable	Q17	Q18	Q19	Q20	Q23	Q24	Q25	Q26	Q27	Q28	Q31	Q31	Q32	Q33	Q34
	Healthy risk?	**Pathogens?**	**Disposed?**	**Treated?**	**Agriculture?**	**Purchase ?**	**Price?**	**Quality?**	**Friends?**	**Consume?**	**Raw?**	**Cooked?**	**Tubber?**	**Above?**	**Processed?**
Age	5.85	0.78	15.59***	3.06	12.35**	15.35	9.01	7.74	13.34**	12.45 **	16.43***	4.93	15.40***	5.28	4.96
Gender	1.02	2.52	15.27***	2.48	9.14**	14.98***	5.14*	2.13	5.37*	9.30***	16.46***	12.06***	8.12**	7.96*	5.86*
Religious	2.23	7.15	7.32	10.84	6.55	30.94	18.35**	19.34*	8.73	16.37**	12.40	14.94**	11.53	8.29	19.29
Settlement	2.34	1.43	6.87**	0.82	8.85**	13.33***	1.42	2.71	9.35**	10.44***	5.45*	7.59**	8.36**	16.27***	9.31***
College	1.51	1.53	7.69*	1.49	11.00***	6.26	0.43	0.46	8.04**	8.93**	1.49	1.47	8.37**	0.87	2.03
Source of income	0.47	3.62	1.56	0.92	3.87	8.49*	7.35*	2.17	0.31	2.26	0.74	1.19	1.73	0.32	3.22
Monthly expenditure	1.33	7.03	4.76	2.24	6.63	8.26	1.43	6.57	6.48	4.76	6.19	2.66	5.28	1.73	1.15
Level of study	1.93	9.67	8.01	13.41	12.95**	9.76	17.62***	10.92*	7.41	4.09	5.07	5.43	3.44	5.06	5.36

*** (p<0.1), ** (o<0.05), *** (p<0.001)

6.3.3 *Attitudinal scores*

A strongly positive attitude toward the treatment of human excreta was observed in all demographic variables with most variables not statistically significant. The level of study and awareness of the use of human excreta in agriculture significantly differed, p=0.036 and p=0.009 respectively (Table 6.3) when consumers were asked about the treatment of human excreta. Moreover, there was a negative attitude (score = 1.00) observed in the level of study where a postdoctoral fellowship thought human excreta could not be treated to reduce its risk. However, this observation could have been influenced by the low participation in a postdoctoral fellowship (n=1). Significant differences were observed in the gender (p<0.001), female students were slightly neutral (score = 1.55) compared to strongly positive attitudes of male students and 'others' with the mean scores of 1.73 and 1.71, respectively.

When asked if human excreta including faeces should be disposed of and never used, there was a positive attitude by the consumers with the majority indicating that it should not be disposed of but be used for other purposes. However, there was a significant difference between gender (p<0.001), age (p=0.008), settlement type (p=0.032), college affiliation with (p=0.053), and awareness of the reuse of human excreta for agricultural purposes (p<0.001). Male students and those identified as 'others' were positive about recycling human excreta for other purposes. Participants from rural areas had a positive attitude (1.66) towards recycling human excreta. All age groups had a positive attitude towards recycling human excreta, interestingly positive attitude increased with age indicating that most grown people are more willing to recycle human excreta for performing other functions. The religious practice did not statistically differ in interestingly, there was a positive attitude in different religious practices towards recycling human excreta for other purposes. However, a negative attitude (1.25) was observed in the Islamic religion. Settlement type showed a significant difference and urban suburbs participants had a neutral attitude towards the treatment of human excreta and re-use for other uses. A negative attitude was observed for the level of study (diplomates) and awareness of using human excreta for agricultural purposes. Indicating that consumers are not aware that human excreta can be treated and used for agricultural purposes.

Negative attitudes were observed in female students and others when the participants whether they thought their friends would purchase and consume food produced using human excreta-derived co-compost. Age group greatly influenced the subjective norms of the participants as most age groups had a negative attitude, moreover, 51.6% of the participants indicated that their friends would consume food grown with co-compost while 48.4% denied that their friends would consume food grown using co-compost. Interestingly knowledge of the use of human excreta in agriculture had a positive attitude with a mean score of 1.63 contrasting with unawareness which had a negative attitude mean score of 1.42. These results indicate that knowledge of the use of human excreta in agriculture is an important factor that influenced subjective norms and made participants think positively about their friends.

Interestingly there was a positive attitude toward the consumption of food produced using co-compost showing great acceptance of human excreta-derived co-compost for agricultural production. The religious practice of Islam showed a negative attitude towards consumption of the food, this is caused by the Islamic belief that human excreta (urine and faeces) are considered impure (Najis). This belief could lead to the Islamic community not being willing to recycle and reuse human excreta products for agricultural purposes. Awareness of the use of human excreta for agricultural purposes was significantly different across all attitudes including the combined attitude (Figure 6.2). Settlement type was the second factor that significantly differed across different attitudinal dimensions. The source of income and monthly expenditure did not statistically differ in all attitudinal scores.

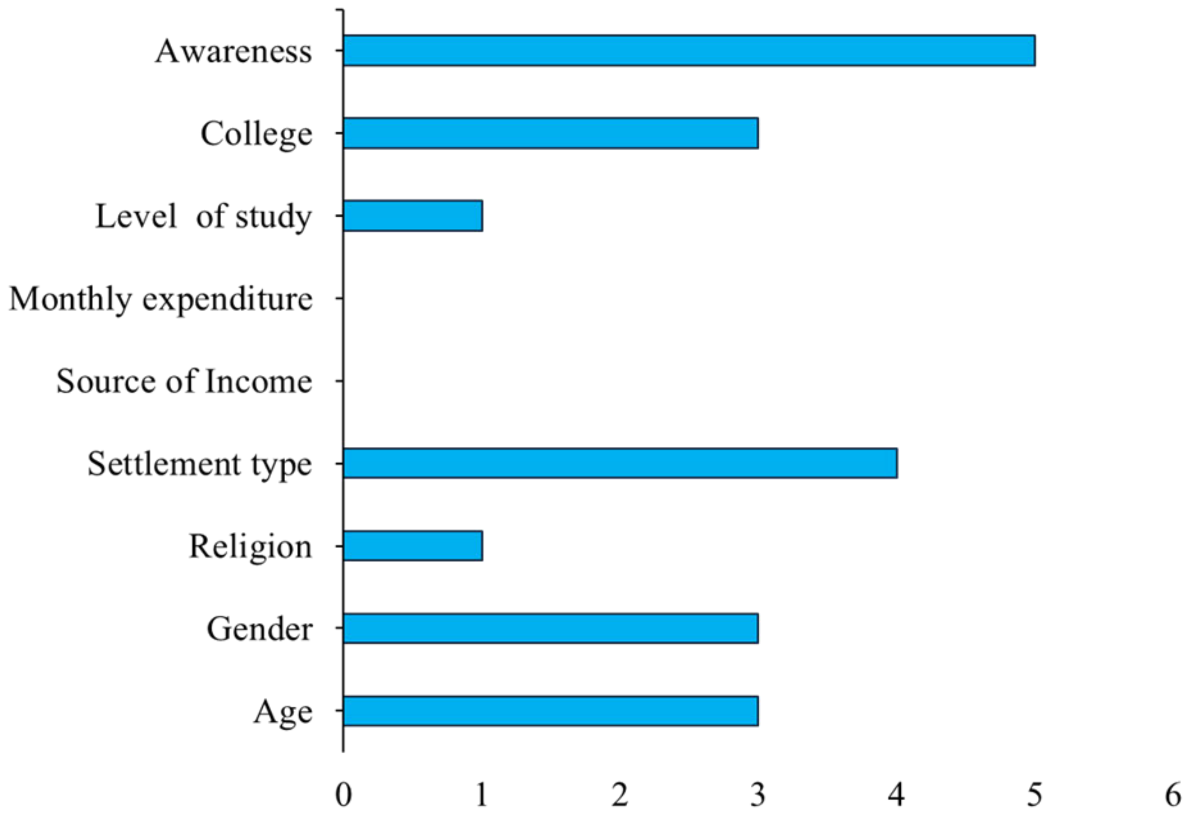


Figure 6.2: Sensitivity of different constructs to consumers/students' attitudes.

Table 6.3: Attitudinal scores of perceived behavioural controls, attitude towards the behaviour and subjective norms and how it affects different socio-demographic variables

Variable		**Q19**			**Q20**			**Q27**			**Q28**			**Combined**	
	µ	**p-value**	**Att.**	**µ**	**p-value**	**Att.**	**µ**	**p-value**	**Att.**	**µ**	**p-value**	**Att.**	**µ**	**p-value**	**Att.**
Gender		0.290			<0.001			0.068			0.009			0.658	
• Female	1.85		+	1.55		+	1.48		-	1.70		+	1.62		+
• Male	1.87		+	1.73		+	1.57		+	1.80		+	1.63		+
• Other	2.00		+	1.71		+	1.36		-	1.91		+	1.64		+
Age (years)		0.694			0.008			0.020			0.029			0.141	
17-19	1.83		+	1.55		+	1.45		-	1.68		+	1.61		+
20-22	1.85		+	1.59		+	1.48		-	1.74		+	1.62		+
23-25	1.87		+	1.70		+	1.64		+	1.80		+	1.66		+
26-30	1.91		+	1.83		+	1.60		+	1.89		+	1.64		+
31-40	2.00		+	1.86		+	1.42		-	1.86		+	1.61		+
≥ 41	2.00		+	2.00		+	1.00		-	1.00		-	1.21		-
Religion		0.288			0.608			0.465			0.058			0.452	
Christianism	1.88		+	1.62		+	1.48		-	1.72		+	1.61		+
Hinduism	1.57		+	1.71		+	1.57		+	1.86		+	1.57		+
Islam	1.87		+	1.25		-	1.38		-	1.38		-	1.59		+
Polytheism	2.00		+	2.00		+	1.00		-	1.50		+/-	1.37		+
Shembe	1.85		+	1.58		+	1.54		+	1.72		+	1.63		+
Atheist	1.69		+	1.69		+	1.46		-	2.00		+	1.62		+
Agnostic	1.89		+	1.67		+	1.56		+	1.89		+	1.67		+
Traditionalism and Christianism	1.85		+	1.64		+	1.59		+	1.80		+	1.65		+
Traditionalism	1.84		+	1.64		+	1.53		+	1.76		+	1.62		+
Scientism	2.00		+	1.71		+	1.71		+	1.86		+	1.71		+
Settlement type		0.667			0.032			0.009			0.005			0.057	
Rural areas	1.87		+	1.66		+	1.57		+	1.78		+	1.64		+
Urban (Township)	1.85		+	1.62		+	1.49		-	1.76		+	1.62		+
Urban (Suburbs)	1.83		+	1.50		+/-	1.39		-	1.61		+	1.58		+
Source of income		0.632			0.460			0.855			0.324			0.368	
Self-funded	1.86		+	1.60		+	1.51		+	1.74		+	1.63		+
Undergraduate bursary	1.86		+	1.72		+	1.52		+	1.76		+	1.63		+
Postgraduate bursary	1.91		+	1.62		+	1.48		-	1.66		+	1.59		+
Monthly expenditure		0.816			0.498			0.264			0.447			0.754	
≤ R 500	1.91		+	1.53		+	1.47		-	1.68		+	1.63		+
R 501 – R 900	1.85		+	1.66		+	1.47		-	1.78		+	1.61		+

R 901 – R 1 200	1.87		+	1.58		+	1.56		+	1.71		+	1.64		+
R 1 201 – R 1 500	1.85		+	1.64		+	1.59		+	1.74		+	1.64		+
R 1 501 – R 2 000	1.85		+	1.69		+	1.47		-	1.79		+	1.64		+
≥ R 2001	1.82		+	1.65		+	1.56		+	1.79		+	1.63		+
Level of study		0.036			0.239			0.286			0.667			0.118	
Diploma	2.00		+	1.00		-	1.00		-	2.00		+	1.75		+
Postgraduate Diploma	2.00		+	1.64		+	1.55		+	1.82		+	1.68		+
Bachelor’s degree	1.85		+	1.60		+	1.49		-	1.74		+	1.62		+
Honours degree	1.92		+	1.66		+	1.63		+	1.80		+	1.61		+
Master’s degree	1.82		+	1.72		+	1.59		+	1.67		+	1.58		+
Doctorate	2.00		+	1.86		+	1.50		+/-	1.86		+	1.63		+
Postdoctoral fellowship	1.00		-	2.00		+	1.00		-	2.00		+	1.25		-
College		0.684			0.053			0.045			0.030			0.118	
CAES	1.85		+	1.69		+	1.50		+/-	1.74		+	1.60		+
CH	2.00		+	1.59		+	1.50		+/-	1.62		+	1.66		+
CHS	1.85		+	1.50		+/-	1.58		+	1.80		+	1.66		+
CLMS	1.87		+	1.55		+	1.40		-	1.65		+	1.59		+
Awareness		0.009			<0.001			<0.001			<0.001			<0.001	
Yes	1.90		+	1.23		-	1.63		+	1.87		+	1.66		+
No	1.83		+	1.49		-	1.42		-	1.65		+	1.60		+

CAES (College of Agriculture, Engineering and Science), **CH** (College of Humanities), **CHS** (College of Health Science), **CLMS** (College of Law and Management Studies), **µ** (mean score), **Att.** (Attitude), **+** (positive attitude), - (negative attitude)

6.3.4 Factors influencing students' attitudes towards food produced using co-compost

The willingness of consumers to recycle human excreta was affected by several factors. Most of the participants were not willing to recycle their faeces because they thought that faeces were disgusting and that they contained pathogens that could cause harm to them (Figure 6.3A). As expected, consumers thought that faeces had a bad odour and thus should be disposed of and never re-used for any other purposes. Interestingly, the subjective norms such as cultural belief, social acceptance, religion, and being mocked by other people did not result in higher attitude prevention.

Availability of nutrients in the human excreta was the major factor that persuaded consumers' behaviour towards accepting human excreta-derived co-compost in agricultural production. The consumers were also concerned about sustainability and circular economy as the factors that would make them recycle human excreta (Figure 6.3B). Moreover, consumers believed recycling human excreta would mitigate improper waste disposal and counteract environmental pollution. Moreover, some consumers believed that recycling human excreta was an indigenous knowledge where their forefathers used urine diluted with water to irrigate their crops with the added advantage of supplying essential nutrients. Moreover, in villages before toilets were introduced people used to defecate in gardens and fields where they grew their crops thus returning nutrients without knowledge. A proportion of consumers believed that recycling human excreta would manage human waste and heal diseases and it's culturally acceptable to recycle it.

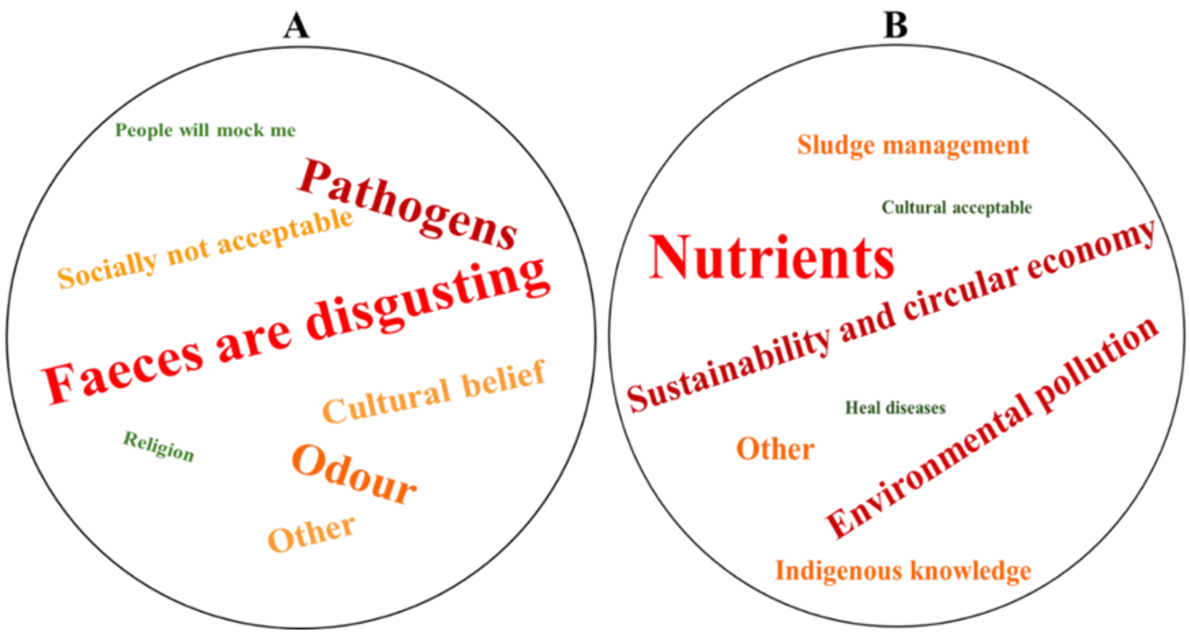


Figure 6.3: Factors influencing consumers' attitude towards recycling human excreta.

6.4 Discussion

6.4.1 Consumers' attitudes towards recycling human excreta

This study explored consumers' recycling behavioural intentions of students at the University of KwaZulu-Natal, Pietermaritzburg campus, based in the KwaZulu-Natal province of South Africa. The theory of planned behaviour was employed as the conceptual framework. The attitude towards human excreta re-use was statistically different between different demographic factors with a moderately positive attitude towards consuming food produced using human excreta-derived co-compost. Awareness of the use of human excreta in agriculture significantly influenced consumers' willingness to recycle it. A strongly positive attitude toward the treatment of human excreta was observed in both individuals who were already aware (1.90) of its agricultural applications and those who were not (1.83). Similarly, Mariwah and Drangert (2011) found that respondents who knew about the use of human excreta in agriculture were more likely to have positive outcomes than those who didn't know about using it on their crops. The majority (86.3%) of students believed that human excreta can be treated so as not to pose health risks which correlates with the results from the previous study by (Simha et al., 2018). This study, however, found that there were differences across gender, settlement type and college students were affiliated with. This positive response towards the treatment of human excreta could significantly play an important role in changing and shaping consumers' attitudes towards the consumption of food produced using human excreta-derived co-compost. According to Gwara et al., (2022), willingness to recycle human excreta is positively correlated with the environmental conscientiousness of farmers. The most preventive barrier towards acceptance of human excreta-based fertilizers in this study was the fear of pathogen contamination and perceived health risks (Figure 6.3A). This finding is similar to the results reported in literature where consumers and farmers had a negative attitude towards urine or faeces-based product use for agricultural productions (Okem et al., 2013; Saliba et al., 2018; Simha et al., 2018). Male students, students between the ages of 25-30, settlers in rural areas, and students that are aware of the use of human excreta in agriculture had a positive attitude towards subjective norms and consuming food produced using co-compost. These consumers thought their friends/colleagues would purchase and consume food produced using co-compost.

6.4.2 Implications of consumers' perceptions towards co-compost

Attitudinal dimensions score in this study were influenced by awareness, settlement type, gender, age, college affiliation, religious practice, and the level of study. Awareness and settlement type positively influenced overall attitudes. Moreover, students'/consumers' awareness of human excreta reuse in agriculture was the most important factor in influencing consumers' behaviour towards food produced using co-compost as also noted by (Gwara et al., 2022). Rural area settlers were more likely to accept food produced using co-compost this attitude could have been attributed to their exposure to the use of chicken and cattle manure which is often used by smallholder farmers to improve fertility in their crops in rural areas. Most studies looking at human excreta re-use in agriculture reported in the scientific literature were testing farmers' attitudes where household heads were mainly male. This study setting was different and most of the participants were female students (Table 6.1). This study found that male students had a positive attitude towards human excreta reuse and consumption of food produced using co-compost compared to female students. In this study, the level of study significantly influenced consumers' perceptions of treating human excreta most levels of study had a positive attitude. In another study, the level of education was found to significantly influence or reduce perceived health risks (Phuc et al., 2006; Samuel, 2016). Behavioural intentions towards consuming food produced using co-compost were mainly influenced by religious practice. The Islamic religion had a negative attitude towards consuming food produced using co-compost while the Polytheism religion was neutral. In the Islamic religion, human excreta are considered ritually impure in Islamic law, and it is a known taboo that transmits diseases (Qaradāwī, 1984). This study recommends implementing awareness campaigns to educate people from diverse religious backgrounds and the public about the benefits of human excreta-derived co-compost. These campaigns should emphasize that co-composting significantly reduces pathogen loads, sanitizing the compost to safe levels that pose no risk of pathogen transmission to crops when used in agricultural production.

The prospect and interesting findings in this study are that the type of food grown using co-compost greatly influenced the attitudes of consumers towards consuming food produced using co-compost. The majority of the consumers were willing to consume food produced using co-compost if it was eaten cooked/boiled, grows above ground, and/or processed. This confirms assumptions by Gwara et al., (2022), that processing food could change people’s behaviour and attitude towards consuming food produced using human excreta-derived co-compost. Some consumers showed unwillingness to consume food produced using co-compost if it was eaten raw or grows below ground, implying that most consumers have a taboo that raw food could still be contaminated with pathogens or has odour which could pose health risks to them. The positive attitude towards cooked food could be induced by boiling meaning the consumers are aware that high temperatures can eliminate pathogens in food.

6.5 Conclusions

Attitudes and perceptions of the university students/consumers were assessed using the theory of planned behaviour. The students had a positive attitude towards recycling human excreta and consuming food produced using human excreta-derived co-compost. The study found that awareness of human excreta reuse in agriculture is the main factor that influences consumers' attitudes towards recycling human excreta and consuming food produced using human excreta-derived co-compost. The study further found that gender differed significantly through recycling human excreta where female students were less willing to recycle and consume food grown using human excreta derived co-compost compared to male students. Attitudes towards consuming food produced using co-compost were also influenced by the background of the participants where consumers from the rural areas were more willing to recycle human excreta for other purposes, and purchase and consume food produced using human excreta-derived co-compost. Students from the College of Agriculture, Engineering and Science were more willing to consume food produced using co-compost indicating their awareness of nutritive value in human excreta. Cooking and processing food that was initially fertilized using co-compost could change consumers' perceptions towards consuming it. As demonstrated in this study most students would eat food produced using co-compost if it was eaten cooked, boiled, didn't get into contact with the soil, and/or processed. This finding suggests that farmers growing crops using human excreta-derived co-compost should focus on growing crops that grow above the ground such as maize, which can be cooked such as Swiss chard, and that can be processed such as chillies (*Capsicum annum L.*). This study recommends designing educational material and dissemination strategies to increase the knowledge and awareness of consumers on the use of human excreta-derived fertilisers in agriculture or food production.

References

Ajzen, I. (1991). The Theory of planned behavior. *Organizational Behavior and Human Decision Processes*.

Ajzen, I. (2002). Perceived behavioural control, self-efficacy, locus of control, and the theory of planned behaviour 1. *Journal of Applied Social Psychology* **32**, 665-683.

Anwar, Z., Irshad, M., Fareed, I., and Saleem, A. (2015). Characterization and recycling of organic waste after co-composting-A review. *Journal of Agricultural Science* **7**, 68.

Bramlett, M. D., Black, L. I., Dahlhamer, J. M., and Zablotsky, B. (2024). Weighting Procedures and Bias Assessment for the 2021-2023 National Health Interview Survey–Teen 30-month File.

Buit, G., and Jansen, K. (2016). Acceptance of human feces-based fertilizers in fecophobic Ghana. *Human Organization* **75**, 97-107.

Chauha , N., and Ranawat , P. (2024). A Study on Consumers' Perception about Sustainable Agriculture Products. *International Journal of Scientific Research in Engineering and Management (IJSREM)* **08**.

Cofie, O., Adeoti, A., Nkansah-Boadu, F., and Awuah, E. (2010). Farmers perception and economic benefits of excreta use in southern Ghana. *Resources, Conservation and Recycling* **55**, 161-166.

Dodds, W. B., Monroe, K. B., and Grewal, D. (1991). Effects of price, brand, and store information on buyers' product evaluations. *Journal of marketing research* **28**, 307-319.

Guzha, E., Nhapi, I., and Rockstrom, J. (2005). An assessment of the effect of human faeces and urine on maize production and water productivity. *Physics and Chemistry of the Earth, Parts A/B/C* **30**, 840-845.

Gwara, S., Wale, E., Lundhede, T., Jourdain, D., and Odindo, A. (2023). Ex-ante demand assessment and willingness to pay for human excreta derived co-compost: Empirical evidence from rural South Africa. *Journal of Cleaner Production* **388**, 135570.

Gwara, S., Wale, E., and Odindo, A. (2022). Behavioral intentions of rural farmers to recycle human excreta in agriculture. *Scientific reports* **12**, 5890.

Gwara, S., Wale, E., Odindo, A., and Buckley, C. (2021). Attitudes and perceptions on the agricultural use of human excreta and human excreta derived materials: A scoping review. *Agriculture* **11**, 153.

Harrison, J., and Wilson, D. (2012). Towards sustainable pit latrine management through LaDePa. *Sustainable Sanitation Practice* **13**, 25-32.

Koné, D., Cofie, O., Zurbrügg, C., Gallizzi, K., Moser, D., Drescher, S., and Strauss, M. (2007). Helminth eggs inactivation efficiency by faecal sludge dewatering and co-composting in tropical climates. *Water research* **41**, 4397-4402.

Manga, M., Camargo-Valero, M., Anthonj, C., and Evans, B. (2021). Fate of faecal pathogen indicators during faecal sludge composting with different bulking agents in tropical climate. *International Journal of Hygiene and Environmental Health* **232**, 113670.

Manga, M., Evans, B. E., Ngasala, T. M., and Camargo-Valero, M. A. (2022). Recycling of Faecal Sludge: Nitrogen, Carbon and Organic Matter Transformation during Co-Composting of Faecal Sludge with Different Bulking Agents. *International Journal of Environmental Research and Public Health* **19**, 10592.

Manga, M., Muoghalu, C., Camargo-Valero, M. A., and Evans, B. E. (2023). Effect of turning frequency on the survival of fecal indicator microorganisms during aerobic composting of fecal sludge with sawdust. *International Journal of Environmental Research and Public Health* **20**, 2668.

Mariwah, S., and Drangert, J.-O. (2011). Community perceptions of human excreta as fertilizer in peri-urban agriculture in Ghana. *Waste management & research* **29**, 815-822.

Mertenat, A., Diener, S., and Zurbrügg, C. (2019). Black Soldier Fly biowaste treatment–Assessment of global warming potential. *Waste management* **84**, 173-181.

Migeri, S., Lawal, M. A., Hughes, J. C., Badza, T., Abafe, O. A., Martincigh, B. S., and Odindo, A. O. (2023). Uptake of selected antiretrovirals by pepper (*Capsicum annum*), radish (*Raphanus sativus*), and ryegrass (*Lolium perenne*) grown on two contrasting soils and fertilized with human urine-derived fertilizers. *Science of the Total Environment* **892**, 164551.

Mugivhisa, L. L., Olowoyo, J. O., and Mzimba, D. (2017). Perceptions on organic farming and selected organic fertilizers by subsistence farmers in Ga-Rankuwa, Pretoria, South Africa. *African Journal of Science, Technology, Innovation and Development* **9**, 85-91.

Nkomo, N., Odindo, A. O., Musazura, W., and Missengue, R. (2021). Optimising pyrolysis conditions for high-quality biochar production using black soldier fly larvae faecal-derived residue as feedstock. *Heliyon* **7**, e07025.

Ofei-Quartey, M. N. L., Appiah-Effah, E., Akodwaa-Boadi, K., Ampaw, B., Taylor, T. S., and Millogo, Z. E. N. (2023). Enhancing the economic potential of organic waste by co-composting using ratio modelling toward a circular economy. *Journal of Material Cycles and Waste Management* **25**, 1560-1580.

Okem, A. E., Xulu, S., Tilley, E., Buckley, C., and Roma, E. (2013). Assessing perceptions and willingness to use urine in agriculture: a case study from rural areas of eThekwini municipality, South Africa. *Journal of water, sanitation and hygiene for development* **3**, 582-591.

Phuc, P.-D., Konradsen, F., Phuong, P., Cam, P., and Dalsgaard, A. (2006). Practice of using human excreta as fertilizer and implications for health in Nghean Province, Vietnam. *Southeast Asian Journal of Tropical Medicine and Public Health* **37**, 222.

Preneta, N., Kramer, S., Magloire, B., and Noel, J.-M. (2013). Thermophilic co-composting of human wastes in Haiti. *Journal of Water, Sanitation and Hygiene for Development* **3**, 649-654.

Qaraḍāwī, Y. a. (1984). The lawful and the prohibited in Islam= Al-halal wal-haram fil Islam.

Saliba, R., Callieris, R., D'agostino, D., Roma, R., and Scardigno, A. (2018). Stakeholders' attitude towards the reuse of treated wastewater for irrigation in Mediterranean agriculture. *Agricultural Water Management* **204**, 60-68.

Samuel, F. (2016). Excreta-related infections and the role of latrines to control the transmission in Ethiopia. *J Community Med Health Educ* **6**, 496.

Simha, P., Lalander, C., Ramanathan, A., Vijayalakshmi, C., McConville, J. R., Vinnerås, B., and Ganesapillai, M. (2018). What do consumers think about recycling human urine as fertiliser? Perceptions and attitudes of a university community in South India. *Water Research* **143**, 527-538.

Simha, P., Lalander, C., Vinnerås, B., and Ganesapillai, M. (2017). Farmer attitudes and perceptions to the re–use of fertiliser products from resource–oriented sanitation systems–The case of Vellore, South India. *Science of the total environment* **581**, 885-896.

Soré, O. A. A., and Ouoba, S. (2023). Monitoring of the Physicochemical Parameters During the Co-Composting of Dehydrated Faecal Sludge and Organic Household Solid Waste: Composts Maturity Assessment. *International Journal of Environmental Research* **17**, 3.

Viaene, J., Van Lancker, J., Vandecasteele, B., Willekens, K., Bijttebier, J., Ruysschaert, G., De Neve, S., and Reubens, B. (2016). Opportunities and barriers to on-farm composting and compost application: A case study from northwestern Europe. *Waste Management* **48**, 181-192.

Wang, Y., Villamil, M. B., Davidson, P. C., and Akdeniz, N. (2019). A quantitative understanding of the role of co-composted biochar in plant growth using meta-analysis. *Science of the total environment* **685**, 741-752.

Waqas, M., Nizami, A., Aburiazaiza, A., Barakat, M., Rashid, M., and Ismail, I. (2018). Optimizing the process of food waste compost and valorizing its applications: A case study of Saudi Arabia. *Journal of Cleaner Production* **176**, 426-438.

WHO (2008). "Advocacy, communication and social mobilization for TB control: a guide to developing knowledge, attitude and practice surveys," Rep. No. 9241596171. World Health Organization.

Zhang, D., Luo, W., Li, Y., Wang, G., and Li, G. (2018). Performance of co-composting sewage sludge and organic fraction of municipal solid waste at different proportions. *Bioresource technology* **250**, 853-859.

Chapter 7: Summary, Conclusions, and Recommendations for Further Research

7.1 Summary and general conclusions

The study addressed the challenges of nutrient imbalances occurring as a result of the transportation of essential mineral elements via food from farms to urban settlements and their disposal together with waste (food, organic green waste, and human waste). Consequently, this leads to linear nutrient flows from soils in farms, soil fertility depletion and environmental pollution. Recovering nutrients from organic waste (including human excreta) and reuse in agriculture can address both challenges of low agricultural productivity due to nutrient-depleted soils and address improper waste disposal thus establishing circular nutrient flows. Human excreta possess a low carbon-to-nitrogen ratio, high moisture content, and is highly compactable limiting the oxygen supply (Banegas et al., 2007). On the other hand, chipped organic green waste has a high carbon content, and low moisture content and is less compactable making it a suitable component to be composted with human excreta such as dewatered sewage sludge (Ofei-Quartey et al., 2023). Co-composting is the process of composting two or more types of organic waste materials to improve the composting processes and quality of the final product. During the co-composting process, microorganisms (from DSS) break down complex organic matter and convert it into simpler forms of organic matter called co-compost. These microorganisms are aerobic bacteria that require oxygen for respiration and moisture to induce enzymatic activities. The process produces heat which effectively deactivates pathogens contained in the faecal matter or sludge; thus, ensuring that the product is safe, hygienic, and of high agronomic value. The co-composting process is a low-cost, less time-intensive procedure that smallholder farmers can mimic and can subsequently improve their soil fertility and overall livelihoods.

The first objective (Chapter 3) investigated the effect of different mixing ratios when using dewatered sewage sludge (DSS) and organic green waste (OGW), (1DSS:3OGW and 1DSS:2OGW) as feedstocks and turning frequency (7-, 14 and 28-day turning interval) to produce high-quality co-compost. Significant differences ($p<0.05$) were observed between co-compost treatments with respect to pH, electrical conductivity, organic matter and organic carbon over time. Higher turning frequencies (7TF) resulted in higher pH and EC values at the end of the co-composting process. Less turning intervals (28TF) for treatments with a lower proportion of sludge (1DSS:3OGW) and higher rates of OM and OC had lower pH and EC values indicating that co-compost degradation occurred at a lower rate than in those treatments that were turned more frequently. Although the temperature was not significantly different

($p>0.05$) between turning frequencies, the study showed that less turning (28TF) resulted in the highest temperature (54.6°C), which was maintained for a longer period compared to when the co-compost pile was turned more frequently. At the end of the co-composting process, the highly turned compost (7TF) had a low volume of compost compared to medium-turned (14TF) and less-turned (28TF) co-compost treatments. Moreover, bacterium pathogens *E. coli* and *Salmonella* were reduced to (<30 CFU/g) in the matured co-compost in all treatments resulting in values below the maximum threshold of 1000 CFU/g recommended by the World Health Organisation (WHO, 2006). The mineral elements (nutrients) recovered during this co-composting process included N, P, K, Ca, Mg, and essential micronutrients including Zn. The heavy metals present in the treatments were all within the maximum permissible South African and European standards as outlined by (Brinton, 2000). These results suggest that co-composting DSS and OGW can produce compost that complies with the WHO standards while recovering nutrients from DSS and OGW. The study concludes that establishing optimal mixing ratios and turning frequencies is critical when making good quality co-compost using treated dewatered sewage sludge mixed with organic green waste.

The second objective (Chapter 4) evaluated cucumber (*Cucumis sativus*) root morphology in response to different co-compost amendments. In addition, the study sought to develop a novel and cost-effective method for assessing co-compost phytotoxicity on sensitive crop species. The co-composts produced using different mixing ratios of OGW and DSS (Chapter 3) were compared with a commercial compost (Gromor) and a sandy loam soil collected from KwaDinabakubo, Hillcrest, Durban, KwaZulu-Natal, South Africa which was used as a control. The study found that treatments with a higher proportion of sewage sludge co-compost (1DSS:2OGW) had the highest shoot mass ratio (SMR) and shoot-to-root mass ratio (SRMR) while the low proportion of sludge co-compost (1DSS:3OGW) resulted into higher leaf area index and shoot length. The commercial compost Gromor (TG) had the highest root biomass with relatively low shoot biomass, high root mass ratio (RMR) and root-to-shoot mass ratio (RSMR) indicating nutrient and water availability stress (Gruber et al., 2013). The KD soil had the highest convex hull area (172.4 cm^2) in week 4 indicating roots were expanding in search of nutrients and water in the soil (Rangarajan and Lynch, 2021). These findings show evidence that the co-compost produced in Chapter 3 was free from phytotoxic compounds which can inhibit root and shoot growth of sensitive plants. In addition, plants produced using co-compost showed improved plant growth compared to the commercial compost.

The third objective (Chapter 5) examined the fertiliser value of combining co-compost made from DSS and OGW and chemical fertilizer applications on the growth, development, and nutritional composition of maize (*Zea mays*) under controlled conditions. Contrasting but acidic soils from two different locations, clay loam soil collected from Bishopstowe (BS), Pietermaritzburg and sandy loam soil collected from KwaDinabakubo (KD), in Durban were used in the study. Combining co-compost with chemical fertiliser improved plant height, chlorophyll content, and harvest index in both BS and KD soils. Moreover, the addition of co-compost increased essential micro-nutrients and macro-nutrients in maize grains resulting in grains that are of good quality. The study further found that the regrowth of pathogens in the soils amended with co-compost was below the maximum threshold set by the World Health Organization. The study concluded that co-compost made from human excreta has the added benefit of adding organic matter and organic carbon in the soil resulting in improved nutrient retention and uptake by a plant.

The fourth objective (Chapter 6) tested and validated consumers' perceptions and social stigmas associated with food produced using co-compost made from DSS and OGW. The study employed the theory of planned behaviour to measure the behavioural intentions using, perceived behavioural control, subjective norms and attitudes towards the behaviour. The study found that consumers' attitudes and perceptions towards purchasing and consuming food produced using co-compost were influenced by gender, age, religious practice, and awareness of the use of human excreta for agricultural purposes. Respondents with prior awareness of the use of human excreta for agricultural purposes highly had a more positive attitude which influenced acceptance of the product. Male students were more willing to consume and purchase food grown using co-compost compared to female students. Age was a factor as shown that the increase in age of the participants had a positive association with higher acceptance of food fertilised using co-compost. Interestingly, religion as a factor showed contrasting responses. Islamic participants had a negative attitude towards food produced using co-compost, while Christians had a moderate but highly positive attitude towards food produced using co-compost. The study further found that consumers were willing to consume food produced using co-compost if it is either cooked, grows above ground or processed than raw and tuber crops as per preliminary assumptions by (Gwara et al., 2022). The study found that consumers' main concern about food fertilised using co-compost made from human excreta was the presence of pathogens and odour and that faeces were disgusting. The research reveals the need for awareness campaigns and demonstration plots to increase awareness of safety and promotions of co-compost products. Such initiatives can convincingly prove that the process of co-composting destroys all biological pathogens making the product completely odourless and pathogen-free. Moreover, these initiatives can dispel misconceptions, generate consumer confidence and help in the introduction of human-excreta-derived co-compost in agriculture as a safe and useful practice.

7.2 Limitations

One potential limitation of this study is that the co-composting process, while effective in pathogen reduction and nutrient recovery, may not be universally applicable across all types of organic waste or geographical locations. The mixing ratios of DSS and OGW, along with turning frequencies, were optimized for the specific materials used in this study. However, variations in the availability and quality of organic waste, as well as environmental conditions such as temperature and moisture levels, may affect the generalizability of these findings. The study's focus on a specific set of organic waste materials and local conditions means that the results may not be directly applicable to other regions or waste streams with different characteristics. Additionally, the scalability of the co-composting process for larger-scale agricultural operations or different climates requires further investigation.

A further limitation of this study is the potential long-term effects of using co-compost made from DSS and OGW on soil health and plant productivity. While the study demonstrated improvements in soil fertility, plant growth, and nutrient content in the short term, the long-term impacts of repeatedly applying co-compost to soil are not fully understood. Prolonged use of co-compost could lead to the accumulation of certain nutrients or compounds, potentially resulting in nutrient imbalances or toxicity over time. Additionally, the breakdown of organic matter in co-compost may vary over extended periods, affecting soil structure and microbial communities in ways that were not assessed in this study. Long-term field trials are necessary to ensure that continuous use of co-compost does not inadvertently degrade soil health, reduce biodiversity, or impact the sustainability of agricultural systems.

Social stigma surrounding the use of human excreta in agriculture, as highlighted in the consumer perception survey. While the study found positive attitudes among certain groups, the widespread acceptance of co-compost made from human excreta is still hindered by concerns about pathogens, odour, and the "disgust" factor associated with human waste. These perceptions could limit the adoption of co-composting practices, even if the technical aspects of pathogen reduction and nutrient recovery are effective. The research suggests that education and awareness campaigns are needed to address these concerns, but changing deeply ingrained cultural attitudes toward human waste may be a long-term process. This challenge could hinder the broader implementation of co-composting as a sustainable agricultural practice in communities where such stigmas are prevalent.

7.3 Recommendations for further research

Co-composting serves as an alternative, sustainable and low-cost technology to recover nutrients from waste products lost through nutrient mining and return them to the soil. Moreover, co-composting has the potential to reduce waste in overfilling landfills and reduce dewatered sewage sludge or biosolids stockpiling in wastewater treatment plants. However, achieving thermophilic temperatures was delayed and the temperatures did not reach levels above 55 °C. A limitation that could have possibly affected the co-compost temperature could be possibly the size of the co-compost piles. The study recommends the use of larger co-composting facilities, increasing pile/windrow size for high-temperature generation, and co-composting under natural conditions. The study further suggests investigation of other factors beyond the temperature which effectively deactivates pathogens in the co-compost. Moreover, the study recommends that future studies should focus on assessing whether co-composting processes increase or reduce greenhouse gas emissions with the comparative assessment of landfill emissions and stockpiling of dewatered sewage sludge (biosolids) emissions in wastewater treatment plants.

Research is still scant on mechanisms and plant responses towards human excreta-derived fertilisers thus this study suggests that future aspects of research should utilise different plant species to test compost quality. This research suggests that future co-composting studies should dwell on assessing plant mechanisms to absorb heavy metals and assess microplastics which are a matter of concern for human excreta-derived fertilisers. Studies on perceptions and attitudes towards acceptance of human excreta-derived products have mainly been conducted with small-scale farmers in rural areas and there is scant information on consumers' acceptance of consuming food produced using co-compost. Moreover, this study's sample size was sufficient for the targeted university, but future research should use larger sample sizes to enhance the generalizability of findings. Further research is also needed to investigate the factors influencing the acceptance of human excreta-derived fertilizers, particularly among women and young people. The study also recommends launching awareness campaigns on social media and within communities could play a crucial role in improving public perceptions of human excreta-derived compost.

References

Banegas, V., Moreno, J., Moreno, J. I., Garcia, C., Leon, G., and Hernandez, T. (2007). Composting anaerobic and aerobic sewage sludges using two proportions of sawdust. *Waste Management* **27**, 1317-1327.

Brinton, W. (2000). Compost quality standards & guidelines: An international view, Woods End Research Laboratory. *Inc., USA*.

Gruber, B. D., Giehl, R. F., Friedel, S., and von Wirén, N. (2013). Plasticity of the Arabidopsis root system under nutrient deficiencies. *Plant physiology* **163**, 161-179.

Gwara, S., Wale, E., and Odindo, A. (2022). Behavioral intentions of rural farmers to recycle human excreta in agriculture. *Scientific reports* **12**, 5890.

Ofei-Quartey, M. N. L., Appiah-Effah, E., Akodwaa-Boadi, K., Ampaw, B., Taylor, T. S., and Millogo, Z. E. N. (2023). Enhancing the economic potential of organic waste by co-composting using ratio modelling toward a circular economy. *Journal of Material Cycles and Waste Management* **25**, 1560-1580.

Rangarajan, H., and Lynch, J. P. (2021). A comparative analysis of quantitative metrics of root architecture. *Plant Phenomics*.

WHO (2006). "WHO guidelines for the safe use of wasterwater excreta and greywater," World Health Organization

Appendices

Appendix 1: Data on moisture content and the amount of water added in each co-compost treatment to correct moisture content

Time (weeks)	Treatments	Mass before (g)	Mass after (g)	Moisture content (%)	Water added (m^3)	Water added (L)	Mass after furnace (g)
2	1:2 (14TF)	22.299	12.154	45.73	0.0321	32.1	7.393
2	1:2 (28TF)	22.394	14.004	37.54	0.0934	93.4	7.476
2	1:2 (7TF)	22.482	14.038	37.53	0.0935	93.5	6.745
2	1:3 (14TF)	20.974	14.148	32.54	0.1309	130.9	8.384
2	1:3 (28TF)	21.061	12.171	42.17	0.0587	58.7	8.668
2	1:3 (7TF)	21.461	14.842	30.84	0.1437	143.7	9.120
4	1:2 (14TF)	10.581	4.974	52.97	-0.0223	-22.3	2.998
4	1:2 (28TF)	10.614	5.479	48.47	0.0115	11.5	3.543
4	1:2 (7TF)	10.779	5.270	51.07	-0.0080	-8.0	3.431
4	1:3 (14TF)	10.392	5.259	49.35	0.0049	4.9	3.186
4	1:3 (28TF)	10.419	5.801	44.46	0.0416	41.6	3.536
4	1:3 (7TF)	11.146	5.249	52.17	-0.0162	-16.2	3.089
6	1:2 (14TF)	10.490	6.303	39.86	0.0761	76.1	3.606
6	1:2 (28TF)	10.336	6.395	38.13	0.0890	89.0	4.160
6	1:2 (7TF)	10.480	6.211	40.73	0.0695	69.5	4.048
6	1:3 (14TF)	10.619	6.480	38.99	0.0826	82.6	4.073
6	1:3 (28TF)	10.489	6.291	40.02	0.0748	74.8	4.359
6	1:3 (7TF)	10.594	6.710	36.66	0.1001	100.1	4.465
8	1:2 (14TF)	10.390	5.592	46.16	0.0288	28.8	3.776
8	1:2 (28TF)	10.253	6.021	41.27	0.0655	65.5	3.939
8	1:2 (7TF)	10.491	5.423	48.28	0.0129	12.9	3.654
8	1:3 (14TF)	10.371	6.084	41.34	0.0649	64.9	3.999
8	1:3 (28TF)	10.495	6.340	39.59	0.0781	78.1	4.205
8	1:3 (7TF)	10.475	5.528	47.20	0.0210	21.0	3.645
10	1:2 (14TF)	10.086	5.760	42.88	0.0534	53.4	3.836
10	1:2 (28TF)	10.323	6.454	37.52	0.0936	93.6	4.229
10	1:2 (7TF)	10.191	5.446	46.56	0.0258	25.8	3.603
10	1:3 (14TF)	10.242	5.517	46.19	0.0286	28.6	3.580
10	1:3 (28TF)	10.183	6.525	35.92	0.1056	105.6	4.309
10	1:3 (7TF)	10.293	5.996	41.73	0.0620	62.0	3.538
12	1:2 (14TF)	10.439	5.275	49.47	0.0040	4.0	3.197
12	1:2 (28TF)	10.363	6.095	41.24	0.0657	65.7	3.744
12	1:2 (7TF)	10.342	5.445	47.36	0.0198	19.8	3.342
12	1:3 (14TF)	10.558	6.185	41.44	0.0642	64.2	3.529
12	1:3 (28TF)	10.753	5.240	51.19	-0.0089	-8.9	3.055
12	1:3 (7TF)	10.513	5.770	45.12	0.0366	36.6	3.465
14	1:2 (14TF)	10.251	6.235	39.21	0.0810	81.0	4.195
14	1:2 (28TF)	10.350	6.425	37.88	0.0909	90.9	4.452
14	1:2 (7TF)	10.509	6.395	39.16	0.0813	81.3	4.272
14	1:3 (14TF)	10.479	6.675	36.29	0.1028	102.8	4.166
14	1:3 (28TF)	10.502	6.875	34.55	0.1159	115.9	4.202
14	1:3 (7TF)	10.361	6.465	37.60	0.0930	93.0	4.082
16	1:2 (14TF)	10.205	6.656	34.79	0.1141	114.1	4.634
16	1:2 (28TF)	10.221	6.219	39.15	0.0814	81.4	4.261
16	1:2 (7TF)	10.284	6.470	37.09	0.0968	96.8	4.528
16	1:3 (14TF)	10.306	6.782	34.21	0.1185	118.5	4.624
16	1:3 (28TF)	10.221	6.572	35.70	0.1072	107.2	4.381
16	1:3 (7TF)	10.077	6.359	36.88	0.0984	98.4	4.391
P value	-	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001
l.s.d	-	0.689	1.097	6.365	0.047	47.730	1.143
C.V	%	3.60	9.70	9.50	46.20	46.20	16.00

Appendix 2: Maize nutrient requirement in the two contrasting soil the sandy loam (KwaDinabakubo) and clay loam (Bishopstowe)

Soil collection	N requirement	P requirement	K requirement
KwaDinabakubo	200 kg/ha	60 kg/ha	190 kg/ha
Bishopstowe	200 kg/ha	20 kg/ha	0 kg/ha

Appendix 3: Nutrients in the maize stover for different treatments

Treatment units	P (%)	N (%)	Ca (%)	Mg (%)	K (%)	Na (mg/kg)	Mn (mg/kg)	Cu (mg/kg)	Fe (mg/kg)	Al (mg/kg)	Zn (mg/kg)
KD only	0.08 ±0.01	0.91 ± 0.13	0.32 ± 0.01	0.44 ± 0.02	0.74 ± 0.12	156.3 ± 7.53	47.67±2.33	3.33 ± 0.35	277.33 ± 66.29	130.67 ± 22.21	31.32 ± 3.35
KD + F	0.26 ± 0.01	1.47 ± 0.24	0.33 ± 0.01	0.29 ± 0.02	1.29 ± 0.06	155.57 ± 14.57	171 ± 23.26	2.17 ± 0.19	113.33 ± 4.48	55.00 ± 5.51	21.6 ± 1.22
KD + Co	0.26 ± 0.06	1.13 ± 0.19	0.35 ± 0.05	0.27 ± 0.02	1.42 ± 0.09	181.47 ± 11.88	67.33 ± 17.13	1.93 ± 0.18	120.67 ± 11.78	70.00 ± 4.73	17.47 ± 1.29
KD + F + Co	0.21 ± 0.02	1.04 ± 0.12	0.37 ± 0.04	0.29 ± 0.03	1.18 ± 0.07	141.4 ± 11.49	116.7 ± 24.88	2.07 ± 0.07	86.3 ± 17.9	47.0 ± 14.8	17.5 ± 0.74
BS only	0.27 ± 0.03	0.89 ± 0.13	0.38 ± 0.05	0.27 ± 0.04	1.62 ± 0.09	163.47 ± 12.8	81 ± 26.95	3.67 ± 0.23	68.3 ± 10.9	34.67 ± 8.70	48.27 ± 4.53
BS + F	0.29 ± 0.03	1.00 ± 0.10	0.50 ± 0.04	0.36 ± 0.0	1.20 ± 0.12	150.83 ± 7.5	98 ± 11.78	3.83 ± 0.48	70.33 ±1.45	31.0 ± 1.15	32.87 ± 2.18
BS + Co	0.32 ± 0.03	0.61 ± 0.08	0.40 ± 0.0	0.33 ± 0.02	1.87 ± 0.07	200 ± 14.7	60.33 ± 6.43	2.5 ± 0.23	88 ± 14.57	50.67 ± 11.46	37.87 ± 3.95
BS + F+ Co	0.24 ± 0.01	1.12 ± 0.10	0.41 ± 0.08	0.28 ± 0.03	1.19 ±0.02	149.7 ± 14.12	66.0 ± 22.03	3.87 ± 0.45	85.3 ± 17.6	36.7 ±10.89	30.61 ± 1.11
Compost	0.28 ± 0.01	1.14 ± 0.06	0.49 ± 0.05	0.30 ± 0.0	1.55 ± 0.12	162 ± 20.02	51.33±16.05	4.0 ± 0.59	105.67 ± 17.42	60.0 ± 11.59	32.46 ± 6.22

Appendix 4: Questioner used to ask students' perceptions and willingness to consume food fertiliser using co-compost

Attitudes, perceptions, and willingness of university students to purchase and/or consume food produced using human excreta-derived fertilizer

Purpose of the study

The purpose of this survey is to understand and document attitudes, perceptions and willingness to purchase and/or consume food grown using co-compost made from human excreta and organic green waste. Co-compost was selected based on the fact that it addresses issues faced in Water and Sanitation and waste management and can be used for agricultural production in most South African communities. Moreover, it is part of Miss Sinikiwe Khumalo's research, which is funded by the *"The rural-urban nexus – Establishing a nutrient loop to improve City Region food system resilience"* also known as the RUNRES project. This study is the continuation of the previous Doctoral study that was done by Dr Simon Gwara in 2021 (Gwara et al., 2022).

Co-compost is amongst many innovations that provide sanitation and return nutrients to the soil by carefully mixing human excreta and organic green waste diverted from landfills. The co-composting process relies on nature for its success where micro-organisms feed on the organic waste and break it down to form fertilizer. When these microorganisms feed on organic waste, they release heat which causes deactivation of disease-causing pathogens that may be found in human excreta. The process requires a correct mixture of carbon-containing material (e.g. organic green waste, sawdust, wood chips etc.) to nitrogen material (dewatered sewage sludge, faecal sludge etc.) for its effectiveness.

How to complete the form?

The survey questions provided in this study were made very simple with most questions allowing you to choose one response or checkbox where you can choose more than one response. You are also given a chance to express your opinion in the form at the end of the survey in the form of a comment or reason. Please kindly note that you are required to provide consent by clicking the I consent to participate in this survey this is done in line with the ethical principles for conducting research.

Time required and compensation

The study takes an estimated time of 15-25 minutes to conduct, and you will be one of approximately 300 participants. You will not be compensated for participating in this study.

Confidentiality and participants' rights

Any or all information collected through this study shall remain anonymous and shall be kept confidential to the extent mandated by South African Law. However, findings from this study may be published and/or shared with other research organizations and interested parties but you will not be identified personally. You are at liberty to stop me or withdraw from the survey at any point. There are no health risks or discomfort from participating in this study.

Contact information

If you have any questions or concerns about this study, please email or contact Miss Sinikiwe Khumalo at [redacted] ([redacted]), or Dr Simon Gwara at [redacted] (+[redacted]) or Prof Alfred Odour Odindo at odindoa@ukzn.ac.za (+27332605425)

Participant Consent

By clicking on the "*Yes I consent to participate in this survey*" button below you acknowledge that you have read and understood the information provided above and that you are informed about the purpose of this survey/study. Moreover, you agree to participate in the survey. Your details will remain anonymous and will not be used for publication and academic research purposes. If you do not wish to participate, please click "No I do not consent to participate" and submit the form.

Please note the following:

*** Indicates a required question**

1. **Do you consent to participate in this survey?** *

Mark only one oval.

- Yes, I consent to participate in this survey *Skip to question 2*
- No, I do not consent (*Submit form)*

Information of the enumerator 2.

Date of the interview

Example: January 7, 2019

3. **Name of the enumerator**

4. **Gender of the enumerator**

Mark only one oval.

- Male
- Female

Demographic Information of the Participant

In this section, we would like to know some information about yourself.

5. **What is your gender?** *

Mark only one oval.

- Male
- Female
- Other

6. **What is your age in years?** *

Mark only one oval.

- 17-19
- 20-22
- 23-25
- 26-30
- 31-40
- 41-50
- >50

7. **What is your religious practice?** *

Mark only one oval.

Hinduism

Islam

Polytheism

Christianism

Shembe/Nazareth

Buddhism

I am an Atheist (There is no God)

I am Agnostic (I do not know about God's existence)

Traditionalism and Christianism

Traditionalism

Scientism (I believe in Science) Other:

8. **How best can you classify where you grew up or come from?** *

Mark only one oval.

Rural areas

Urban (Township)

Urban (Suburbs)

9. **What is your main source of income?** *

Mark only one oval.

- Self-funded
- Postgraduate bursary
- Undergraduate bursary

10. **Approximately how much (in Rands) do you spend on food/grocery per month?** *

Mark only one oval.

- Less or equal to R 500.00
- R 501.00 - R 900.00
- R 901.00 - R 1 200.00
- R 1 201.00 - R 1 500.00
- R 1 501.00 - R 2 000.00
- Greater or equal to R 2 001.00

11. **What is your current level of study?** *

Mark only one oval.

- Diploma
- Postgraduate Diploma
- Bachelor's degree
- Honours degree
- Master’s degree
- Doctorates
- Post Doctoral Fellowship

12. **Which college and school of study are you affiliated with?** *

Mark only one oval.

- College of Agriculture, Engineering and Science *Skip to question 13*
- College of Humanities *Skip to question 14*
- College of Health Science *Skip to question 16*
- College of Law and Management Studies *Skip to question 15*

Skip to question 17

College of Agriculture, Engineering and Science

Choose the school

13. School

Mark only one oval.

- School of Agriculture, Earth and Environmental Sciences
- School of Chemistry and Physics
- School of Engineering
- School of Mathematics, Statistics and Computer Science
- School of Life Science

Skip to question 17

College of Humanities

Choose school

14. School

Mark only one oval.

- School of Art
- School of Applied Human Science
- School of Education
- School of Built Environment and Development Studies
- School of Social Sciences

Skip to question 17

College of Law and Management Studies

Choose school

15. School

Mark only one oval.

- School of Law
- School of Business and Leadership
- School of Accounting, Economics and Finance
- School of Management, Information Technology and Government

Skip to question 17

College of Health Science

Choose school

16. School

Mark only one oval.

- School of Health Science
- School of Clinical Medicine
- School of Nursing and Public Health
- School of Lab Medicine and Medical Science

Skip to question 17

Attitudes and perception towards disposal of human excreta including faeces

In this section we would like to know about your thoughts of human excreta re-use in agriculture.

17. **Do you think fresh or untreated human excreta pose healthy risk to you when it come into contact with your food or skin?**

Mark only one oval.

- Yes
- No

18. **Do you think human faeces contain pathogens (micro-organisms that can cause diseases)?**

Mark only one oval.

- Yes
- No

19. **Do you think human excreta can be treated so as not to pose health risks?**

Mark only one oval.

- Yes
- No

20. **Do you think human faeces should be disposed and never be used for any purposes?**

Mark only one oval.

- Yes *Skip to question 21*
- No *Skip to question 22*

Why do you think human excreta should be disposed and never used for any purposes?

21. **If yes, why?**

Check all that apply.

- It is culturally not acceptable to use human excreta
- It is socially not acceptable to use human excreta
- Human excreta smells bad thus must not be used
- My religion will not allow me to use human excreta
- I am afraid of the micro-organisms that cause diseases in it
- People will make fun of me if I use it for any purpose
- Human excreta looks very bad and disgusting
- Other

Skip to question 23

Why do you think human excreta should not be disposed and should be re-used?

22. **If no, why?**

Check all that apply.

- [ ] My culture allows us to re-use human excreta
- [ ] Human excreta is fertile and thus should be re-used
- [ ] Using human excreta will mitigate environmental pollution
- [ ] It can help emptying our toilets, especially in villages
- [ ] Re-using human excreta can contribute to sustainability and circular economy
- [ ] Our forefathers grew up using human excreta in their gardens
- [ ] It can heal some diseases
- [ ] Other

Skip to question 23

Attitudes and perceptions towards purchasing and consuming food produced using co-compost

In this section, we would like to know if you would purchase and consume food that was fertilized using co-compost.

23. **Are you aware of the use of human excreta in agriculture (i.e. food production)?**

Mark only one oval.

- ( ) Yes
- ( ) No

24. **Would you purchase food that was fertilized using co-compost?**

Mark only one oval.

- ◯ Yes
- ◯ No
- ◯

25. **I would like to know if price can influence your decision to purchase food fertilized using co-compost?**

Mark only one oval.

- ◯ Yes
- ◯ No

26. **I would like to know if quality (e.g., shelf life, taste, nutrition etc.) can influence your decision to purchase food fertilized using co-compost?**

Mark only one oval.

- ◯ Yes
- ◯ No

27. **Do you think your friends or colleagues on the university campus would purchase and consume food that was fertilized using co-compost?**

Mark only one oval.

- ◯ Yes
- ◯ No

28. **Would you consume or eat food that was fertilized using co-compost?**

Mark only one oval.

Yes *Skip to question 29*

No *Skip to question 30*

If YES, why?

29. **I would eat/consume food fertilized using co-compost if:**

Check all that apply.

- [ ] It was produced without my knowledge.
- [ ] It was authorized by the government/relevant authorities If it is more nutritious than the one fertilized by chemicals I don't see the problem in eating it.
- [ ] If it was more sustainable to do so.
- [ ] Only if i was desperate
- [ ] If other people especially my friends and family eat it
- [ ] Other:

Skip to question 31

If NO, why?

30. **I would not eat food fertilized using co-compost because:**

Check all that apply.

- [ ] It is culturally unacceptable.
- [ ] People will make fun of me.
- [ ] I think it tastes bad.
- [ ] It can cause sickness or health issues when ingested.
- [ ] I don't feel like eating it.
- [ ] I think it will smell bad.
- [ ] Other:

____

Skip to question 31

Factors that influence willingness to consume food produced using co-composting.

Considering your response about willingness to eat crops produced using co-compost above, we would like to know if you would......

31. Consume or eat food fertilized using co-compost if it **eaten raw** (for instance lettuce, cucumber, etc.)?

Mark only one oval.

- ( ) Yes
- ( ) No

32. Consume or eat food fertilized using co-compost if it **boiled or cooked** (for instance spinach, kale, etc.)?

Mark only one oval.

Yes

No

33. Consume or eat food fertilized using co-compost if it is a **tuber or root** crop (e.g., potatoes, beetroot, carrots, etc.)?

Mark only one oval.

Yes

No

34. Consume or eat food fertilized using co-compost if it **does not come into contact with the soil** (e.g., maize, sorghum, etc.).

Mark only one oval.

Yes

No

35. Consume or eat food fertilized using co-compost if it was **processed** (e.g., vegetable oil, chili-sauce, potato chips, sugar, etc.)?

Mark only one oval.

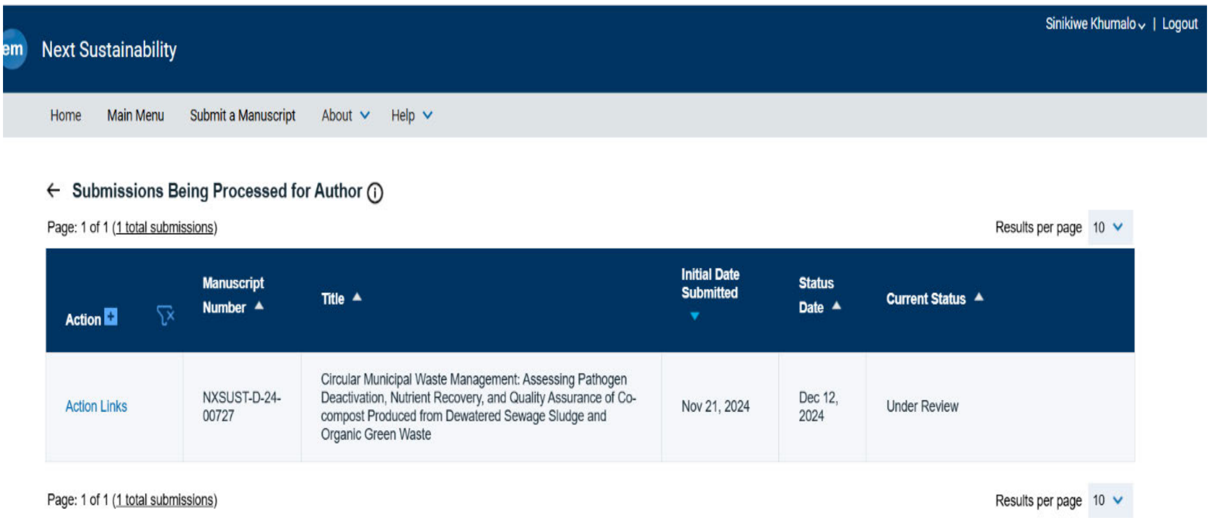
Yes

No

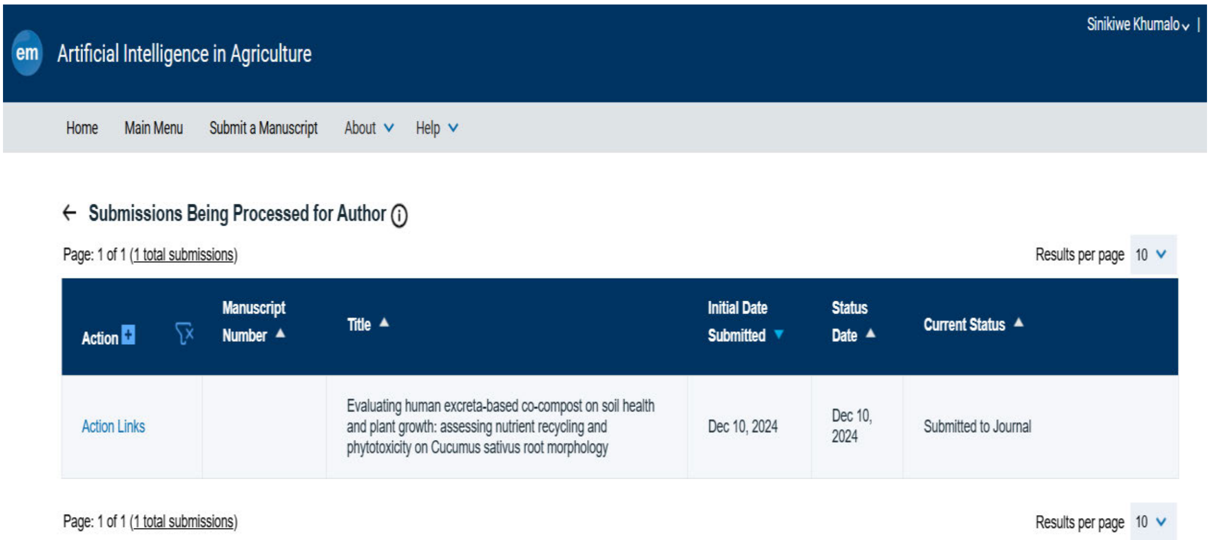
36. **Any comment?**

This content is neither created nor endorsed by Google.

Appendix 5: Publications



Action	Manuscript Number	Title	Initial Date Submitted	Status Date	Current Status
Action Links	NXSUST-D-24-00727	Circular Municipal Waste Management: Assessing Pathogen Deactivation, Nutrient Recovery, and Quality Assurance of Co-compost Produced from Dewatered Sewage Sludge and Organic Green Waste	Nov 21, 2024	Dec 12, 2024	Under Review



Action	Manuscript Number	Title	Initial Date Submitted	Status Date	Current Status
Action Links		Evaluating human excreta-based co-compost on soil health and plant growth: assessing nutrient recycling and phytotoxicity on Cucumus sativus root morphology	Dec 10, 2024	Dec 10, 2024	Submitted to Journal