EVALUATION OF LACTIC ACID PRODUCED BY FERMENTATION OF SELECTED VEGETABLE WASTE AS A TREATMENT IN FAECAL SLUDGE

MWEBIA TYSON MWIMATHIRI

A Thesis Submitted in Partial Fulfillment of Requirements for Conferment of the Degree of Master of Science in Sanitation of Meru University of Science and Technology.

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DECLARATION

This thesis is my original work and has not been presented for a degree in any other institution

EG407/201112/20

Signed _____ Date _____

Mwebia Tyson Mwimathiri

DECLARATION BY SUPERVISORS

This thesis has been submitted with our approval as University supervisors.

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DEDICATION

This thesis is dedicated to my beloved grandmother, Ms. Edith M'Muthuri; her wisdom, strength, and unconditional love have been a constant source of inspiration throughout my life. This work honors her enduring legacy and the values she continually instills in me.

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ABBREVIATIONS, SYMBOLS, AND ACRONYMS

ABR	Anaerobic Baffled Reactor
CFU	Coliform Forming Units
DTF	Decentralized Treatment Facility
FS	Faecal Sludge
FSM	Faecal Sludge Management
GC-MS	Gas Chromatography-Mass Spectrometry
HDPE	High-Density Polyethylene
HTC	Hydrothermal Carbonization
LAB	Lactic Acid Bacteria
LMIC	Low and Middle Income Countries
OD	Open Defecation
OSS	Onsite Sanitation
SDG	Sustainable Development Goals
TON	Threshold Odour Number
TS	Total Solids
TTA	Total Titrable Acidity
VIP	Ventilated Improved Pit
VS	Volatile Solids

ABSTRACT

Effective faecal sludge treatment is essential for pathogen reduction and compliance to World Health Organization standards for disposal or reuse. In developing countries, these treatments are frequently absent, ineffective, or immensely expensive. This study used an experimental research design to investigate the efficacy of lactic acid derived from selected vegetable wastes for the treatment of FS collected from an on-site sanitation system. Equal quantities of fresh cabbage, tomato, and carrot wastes were collected, pre-treated, and subjected to lacto-fermentation at 37 ℃ for six days. Daily monitoring of pH and lactic acid concentrations was performed using an electrode pH meter and a UV-Vis spectrophotometer, respectively. Gas chromatography-mass spectrometry (GC-MS) was used to identify lactic acid in the fermented vegetable wastes through a derivatization reaction. FS treatment was performed in four reactors with varying lactic acid ratios (1:1, 1:0.5, 1:0.35), and a control, with *Escherichia coli* (*E. coli*) as the pathogen indicator for faecal contamination. *E. coli*, total solids, volatile solids, and odour levels were monitored over a 16-day treatment period in the four reactors. The fermentation process was deemed successful as indicated by the decrease in pH levels. Successful GC-MS detection of butyl lactate, the derivatized form of lactic acid was observed in the fermented vegetable wastes. Lactic acid concentrations post fermentation were 1.39 ± 0.09 mg/mL, 1.17 ± 0.13 mg/mL, and 1.61 ± 0.34 mg/mL for cabbage, tomato, and carrot wastes respectively. Statistical analysis revealed no significant differences ($p > 0.05$) in lactic acid concentrations among the vegetable wastes on day six of fermentation. Consequently, cabbage waste-derived lactic acid was selected for subsequent experiments in addition to the local abundance of cabbage waste. Total solids and volatile solids decreased across all the reactors over time. From day four, *E. coli* was undetectable in reactor 1:1; which also showed the highest reduction in odour levels. Therefore, reactor 1:1 treatment produced the optimal *E. coli* elimination and odour reduction conditions. This study demonstrates the potential of cabbage waste derived lactic acid for effective FS treatment.

CHAPTER ONE: INTRODUCTION

1.1 Background of the Study

Faecal sludge (FS) refers to the waste generated from on-site sanitation systems and is not conveyed through a sewer. It typically exists as a partly digested, raw slurry or semi-solid. It is generated through storing, collecting, or treating wastewater, including excreta and black water, either with or without grey water (Tasnim *et al*., 2023).

Ensuring safe management of FS is a crucial objective for global communities. This is as a result of the substantial health hazards linked with exposure to unsanitised FS (Odey *et al*., 2018). The Sustainable Development Goal (SDG) 6 addresses sanitation demands that everyone ought to have a safely-managed sanitation facility in which excreta can be securely disposed off on-site or processed off-site, and that open defecation be eliminated by 2030 (Hyun *et al.*, 2019).

Global coverage of safely managed FS has seen an increase by 8% from 2015 to 2020, however, significant challenges persist, particularly in low-and middle-income countries (LMICs). A report by World Health Organization (2020b) noted that the regions of sub-Saharan Africa and Oceania have fallen 32% behind their SDG 6 target with a sanitation coverage of 30% and 35%, respectively, making them the furthest from achieving their goal, the report also estimated that at current pace no global region will have achieved the SDG 6 target by 2030 and that only 67% of the target will have been achieved by 2030.

More than 3.6 billion people worldwide lack access to fundamental sanitation facilities like private toilets or latrines (World Health Organization & United Nations Children's Fund, 2021). Among them, approximately 494 million individuals still engage in open defecation,

which involves defecating in public places such as street troughs, bushes, or open water bodies (Ubi *et al*., 2021).

Globally, on-site sanitation systems have not been prioritized. It is projected that by the year 2030, on-site sanitation systems will be serving 5 billion people. This also means that a significant amount of FS will be generated, requiring treatment (Zewde *et al*., 2021). In cities that are experiencing rapid growth and poverty, there is often insufficient management of FS, and this leads to the accumulation of sludge in inadequately designed pits, discharge into storm drains or open water, or dumping into waterways, wasteland, and unsanitary landfill sites (Orner & Mihelcic, 2018).

Difficulties in accessing peri-urban and remote areas with exhauster trucks lead to over-reliance on manual emptying methods, which pose enormous health risks (Brands *et al*., 2020). Additionally, these methods often lack a safe way to dispose off FS, thus releasing untreated waste into the environment. Water-borne diseases, for example, intestinal parasitic infections, diarrheal diseases, skin and eye infections, are associated with poor sanitation practices (Chan *et al*., 2021).

Faecal sludge is a significant source of infectious pathogens, containing numerous bacteria, protozoa, viruses, and helminths (Schlosser-Brandenburg *et al*., 2023). Safe handling and treatment of FS are thus key primary barriers in blocking transmission pathways of such pathogens and protecting public health. The uncontrolled spread of faecal matter can cause illness and environmental problems due to its content of pathogens and nutrients (Tortajada & Biswas, 2018).

Among the most promising emerging approaches is the implementation of decentralized treatment systems, which are highly effective in regions with constrained infrastructure

(Rabaey *et al*., 2020). These systems often incorporate technologies such as anaerobic digestion, which breaks down organic matter in the absence of oxygen, producing biogas and a nutrient-rich digestate that can be used in agriculture (Lourenço & Nunes, 2020). The biogas produced is harnessed and used as fuel. These facilities are, however, expensive to build and operate and also require skilled manpower (Hube & Wu, 2021).

Another innovative approach is the use of constructed wetlands for FS treatment. These engineered systems mimic natural wetlands' processes, using plants, soil, and microorganisms to filter and purify wastewater (Rosendo *et al*., 2022). Constructed wetlands are low-cost, require minimal maintenance, and are highly effective at removing pathogens and pollutants from sludge. They also provide the added benefit of creating green spaces that can enhance urban environments and support biodiversity (Parde *et al*., 2021). They, however, have several drawbacks that hinder their application, especially in peri-urban settings, such as large land requirements and seasonal variation since they mimic natural ecosystems (Al Hadidi, 2021)

Composting is gaining popularity as a viable method for treating FS. Through a controlled aerobic process, organic waste is converted into stable, humus-like material that can be safely used as a soil conditioner (Zuberer & Zibilske, 2021). This not only helps in managing waste but also addresses soil degradation issues prevalent in many African regions; however, thermophilic composting needs additional energy in order to inactivate pathogenic bacteria, hence additional costs (Hafliðadóttir *et al*., 2021). It is also not suitable for areas with large volumes of sludge that require rapid treatment due to the long processing time. Additionally, pathogens may not be eliminated if temperatures are not well maintained (Zuberer & Zibilske, 2021)

Pyrolysis, a thermal decomposition process conducted in the absence of oxygen, is also emerging as a cutting-edge technology for FS treatment. This process transforms waste into biochar, a form of charcoal that can improve soil health, sequester carbon, and reduce greenhouse gas emissions (Rahman *et al*., 2020). Pyrolysis units can be designed to be mobile, allowing for flexible deployment in various urban and peri-urban settings (Mutelo, 2023). This method is, however, not suitable for FS treatment on-site sanitation systems due to high energy requirements and safety concerns (Bittencourt & Martins, 2022).

Lactic acid fermentation is a well-established process in various industries, including food, pharmaceuticals, and biodegradable plastics production. Lactic acid, a versatile organic acid, is easily produced through the fermentation of carbohydrate-rich substrates by lactic acid bacteria (Raman *et al*., 2022). Recently, there has been growing interest in utilizing organic waste materials as substrates for fermentation, driven by the dual benefits of waste valorization and sustainable production of valuable chemicals (Odey *et al*., 2018). A few substrates have been investigated for *E. coli* elimination; however, several substrates are ineffective in pathogen elimination, while others have proven expensive (Anderson *et al*., 2015; Odey *et al*., 2018).

Vegetable waste, often abundant and underutilized, presents a viable substrate for lactic acid production. Rich in carbohydrates and other nutrients, vegetable waste can support the growth and metabolic activity of lactic acid bacteria (Wan-Mohtar *et al*., 2023). Utilizing vegetable waste not only addresses waste management issues but also contributes to a circular economy by transforming waste into a commercially valuable product.

This study investigated the feasibility of producing lactic acid through the fermentation of selected vegetable wastes and evaluated its application as a treatment method for FS. In this

study selected vegetable wastes were selected for production of lactic acid through fermentation. Lactic acid from the most abundant local vegetable waste was used to assess pathogen inactivation and odour reduction in FS.

This study utilized a low-cost laboratory set-up that aimed to provide baseline results for treating FS from On-Site Sanitation systems using lactic acid derived from locally abundant vegetable wastes. The use of vegetable waste derived lactic acid for FS treatment has the potential to create synergistic community-led sanitation initiatives that can address both organic waste management and sanitation needs.

1.2 Problem Statement

It is projected that the population in African towns and cities will double by 2050; necessitating a corresponding expansion of sanitation services to meet this growth (Bishoge, 2021). A study by Simiyu *et al.* (2021) indicated that approximately 20% of the population in urban areas use toilets connected to a sewer system, while the remaining depend mainly on-site sanitation facilities and/or open disposal of faecal waste. Faecal sludge from urban households in Kenya utilizing on-site sanitation systems is often discharged into nearby streams, leading to contamination of groundwater and surface water, which presents health risks due to the presence of pathogens (Njue *et al*. 2019). Manetu and Karanja (2021) reported that approximately 9.9 million people in Kenya consume water from polluted surface sources, while around five million engage in open defecation. These practices can result in the spread of diseases.

Several FS treatment methods have proven feasible; however, they are hindered by several drawbacks, hence making them undesirable in LMIC; for instance, some areas are inaccessible to emptying trucks, resulting in seeking services from manual emptiers

(Mallory *et al*., 2021; Riungu *et al*., 2018). The evacuated, highly pathogenic FS is dumped in undesignated areas such as water sources, thus posing a health risk to the residents. Other major disadvantages include the construction and maintenance of treatment facilities and land requirements (Tilley *et al*., 2014).

Vegetable waste, which is often discarded into the environment, holds potential for bioconversion through lactic acid fermentation, a process that can produce lactic acid with antimicrobial properties, potentially useful for pathogen reduction in faecal sludge (Odey *et al*., 2018).

Despite these opportunities, limited research exists on the feasibility and efficacy of using lactic acid produced from fermented vegetable waste as a low-cost, sustainable treatment for faecal sludge (Anderson *et al*., 2015; Zewde *et al*., 2021). This presents a gap in both waste valorization and FS management that could address the environmental and public health related challenges.

This study seeks to evaluate the effectiveness of lactic acid produced by the fermentation of selected vegetable waste as a treatment for FS, exploring its potential for pathogen reduction and practical application in FSM.

1.3 Justification

Simpler and innovative methods, such as vermi and fly larvae treatment, ammonia stabilisation, and aerobic treatment methods, have been proposed for FS treatment; however, they are riddled with several disadvantages; for instance, lime addition to FS has been found to eliminate pathogens to below detectable limits; however, Ali & Shahreen, (2024) noted that the pH declines again after the initial reaction, thereby requiring the addition of high

amounts of lime. This results in higher costs. In addition, bacterial pathogens can re-grow over time.

Faecal sludge is a rich source of nutrients such as nitrogen, phosphorus, and potassium. In human excreta, most organic matter is in feces, while most nitrogen (70-80%) and potassium are in urine (Enayetullah, 2015). Tariq & Mushtaq (2023) estimated that approximately 700 million people in 50 countries eat food from crops irrigated with untreated or inadequately treated wastewater from sewage systems on a total area surface of at least 20 million hectares.

Lactic acid is a naturally occurring C3 α-hydroxycarboxylic acid ($C_3H_6O_3$) that is a product of the anaerobic metabolism of sugars in virtually all living things. In its undissociated form, lactic acid possesses high antimicrobial activity with an inhibitory effect 10-600 times stronger than that of its dissociated forms, a characteristic that has led to its widespread use as a preservative in the food industry (Anderson *et al*., 2015; Antone *et al*., 2022).

Lactic acid can be produced by fermentation of sugars in a simple and economical method using lactic acid bacteria, a fairly large group of bacterial species readily found in foodstuffs and the natural environment (Anderson *et al*., 2015).

1.4 Research Questions

i. What is the quantitative concentration of lactic acid produced through lacto-fermentation of selected vegetable wastes?

ii. How does the number of *E.coli* vary in the FS treated with different ratios of lactic acid derived from selected vegetable waste?

iii. How do the amounts of total solids and volatile solids vary in the FS treated with different ratios of lactic acid derived from selected vegetable waste?

iv. How does the odour level vary in FS treated with different lactic acid levels from selected vegetable waste?

1.5 Research Objectives

1.5.1 Main Objective

To evaluate the effectiveness of lactic acid produced by fermentation of selected vegetable waste in the treatment of FS.

1.5.2 Specific Objectives

i. To quantify the yield of lactic acid from selected vegetable wastes.

ii. To enumerate the levels of *E.coli*, in FS treated with different ratios of lactic acid derived from selected vegetable waste.

iii. To determine the amounts of total solids, and volatile solids in FS treated with different ratios of lactic acid derived from selected vegetable waste.

iv. To determine odour levels in FS treated with different ratios of lactic acid derived from selected vegetable waste.

1.6 Significance of the Study

This research aims to provide a cost- effective and efficient method of treating FS for safe disposal or reuse by pathogen inactivation and odour reduction. The use of vegetable waste contributes to the management and valorization of organic waste contributing to the circular economy approach. As a foundational research study, these investigations will inform future studies.

1.7 Limitations of the Study

Time and financial constraints limited the monitoring of FS treatment for only 16 days, limiting insights into the long-term stability and efficacy of lactic acid treatment for pathogen inactivation and odour reduction.

1.8 Delimitations of the Study

The study deliberately focused on cabbage waste for the FS treatment due to its local abundance in the study area. The research was confined to evaluating lactic acid yield via lacto-fermentation, excluding other fermentation products or treatment methods to streamline the investigation within limited resources.

CHAPTER TWO: LITERATURE REVIEW

2.1 Introduction

This chapter aims to provide an extensive literature review related to FS treatment by use of, among other methods, lactic acid treatment, and it will also provide the knowledge gap that this study aims to fill.

2.2 Faecal Sludge Management

Faecal sludge originates from on-site sanitation technologies and is not collected via a sewerage system. It varies in consistency, quantity, and concentration (Velkushanova *et al*., 2021). It can have significant variations in its characteristics and consistency, and be more or less solid. It could be raw or partially digested, a slurry or semi-solid, and comes from the collection, storage, and treatment of excreta and black water, with or without grey water (Velkushanova *et al*., 2021).

Tasnim *et al.* (2023) emphasize that social and spatial habits influence FS accumulation, highlighting factors such as work schedules, eating patterns, and toilet usage, which directly impact the effectiveness of FSM strategies.

Public health and environmental safety, especially in regions where on-site sanitation systems are prevalent, depend on effective FSM. Research has shown that water-borne infections and environmental pollution are two major risks to public health that can arise from inadequate FSM. Gwenzi *et al.* (2023) state that pathogens can emerge from improperly treated and disposed of FS, which in turn can contaminate water sources and exacerbate public health. Consequently, to lessen the influence of these risks, efficient FSM measures are crucial. In order to achieve this goal, it is essential to implement thorough strategies that cover every step of the sanitation chain, from collection and transportation to

treatment, safe disposal, and reuse. One example is the possibility that composting and anaerobic digestion, two methods for treating sludge, could help accomplish sustainable development goals by transforming trash into biogas and soil conditioners (Cucina, 2023). These processes reduce the environmental footprint of waste and provide economic benefits by generating renewable energy and improving soil fertility.

Faecal sludge management encompasses a comprehensive set of processes that ensure the effective handling of FS from its origin to its final safe disposal or reuse. This management includes the initial storage, where FS is contained in facilities such as latrines, septic tanks, or ventilated improved pit latrines (VIPs). These storage facilities must be designed and maintained to prevent leakage and contamination of the surrounding environment. Following storage, the collection phase involves systematically removing sludge, which can be done manually or mechanized. This step is critical as improper collection can pose significant health risks to workers and the community (Chandana & Rao, 2022).

Transportation is a crucial component of FSM, as it entails the transfer of the accumulated sludge to treatment facilities. This approach requires the use of appropriate vehicles and infrastructure to avoid spills and ensure the safe transportation of sludge in both urban and rural areas (Samal *et al*., 2022). Afterwards, the sludge undergoes treatment processes that aim to remove hazardous substances, decrease the presence of pathogens, and transform it into a more stable form. The anaerobic digestion processes, which may produce biogas as an energy source, and the composting process, which can create soil amendments to enhance agricultural productivity, are often employed treatment methods (Song *et al*., 2021).

The disposal and safe end use of treated FS are equally important, as they prevent environmental pollution and promote resource recovery. Treated sludge can be used as a soil

conditioner to improve soil fertility, contributing to sustainable agricultural practices. Additionally, sludge can be processed into dry or liquid fuels, providing a renewable energy source that can reduce reliance on traditional fossil fuels (Siddiqui *et al*., 2023). Biogas recovery from FS mitigates greenhouse gas emissions and provides a valuable energy source for cooking and electricity generation, particularly in off-grid areas (Raj, 2020).

By integrating these processes, FSM addresses sanitation challenges and offers economic and environmental benefits, highlighting the importance of developing effective and sustainable FSM systems. Various studies on FSM highlight its substantial impact on public health and environmental safety.

A report by the United Nations Children's Fund & World Health Organization (2024) indicated that 21% (1.7 billion people) of the world's population used toilets or latrines where excreta were safely disposed of in situ, making FSM a critical public health issue. Several studies indicate that proper FSM can significantly reduce diarrheal diseases, demonstrating its significant health benefits. Bhatkal *et al.* (2024) reported that improperly managed FS contaminates 60% of surface waters in urban areas. Effective FSM has further been shown to decrease the prevalence of soil-transmitted helminths by up to 60%, further emphasizing its role in disease prevention (World Health Organization, 2020a).

This highlights the critical role of FSM in controlling environmental contamination. Moreover, FSM interventions can lead to a 25% increase in the safe reuse of treated sludge for agriculture, promoting sustainable practices and resource recovery (Ahmad *et al*., 2016). These statistics further highlight the significant positive outcomes that effective FSM can achieve, particularly in regions heavily reliant on on-site sanitation systems, and emphasize

the necessity of improving FSM practices to enhance public health and environmental sustainability.

Most urban and rural people rely on onsite sanitation facilities, especially in low-income countries (Odagiri *et al*., 2021). More than 1.5 billion people still lack access to fundamental sanitation services, such as private toilets or latrines (World Health Organization & United Nations Children's Fund, 2021). Currently, a huge fraction of the people in urban areas of Sub-Saharan Africa (SSA) are served by on-site sanitation technologies; such installations include latrines, VIPs, and septic tanks and constitute the main options for capturing human excreta (Donacho *et al*., 2023). These facilities require regular emptying, mechanically or manually, and the sludge is treated for safe disposal (Wanda *et al*., 2021).

Kenya exemplifies this trend, with a significant portion of its urban and rural populations relying on these systems. Specifically, in Meru County, Kenya, the prevalence of on-site sanitation is a key concern due to the need for regular emptying and proper sludge treatment to ensure safe disposal (Matheka, 2022). Research indicates that inadequate FS management in Kenya has led to environmental contamination and public health issues, stressing the need for improved FSM practices to mitigate these risks and enhance sanitation outcomes (Gitonga *et al*., 2021).

Although common, manual emptying of these facilities poses significant health hazards to workers and can lead to environmental contamination if not appropriately conducted (Conaway *et al*., 2023). Therefore, there is a critical need for mechanized emptying solutions and the establishment of treatment plants that can handle the diverse characteristics of FS.

Studies by Kulabako *et al.* (2010) emphasize the importance of context-specific FSM solutions that consider local socio-economic conditions, infrastructure capacity, and community engagement. Moreover, international collaborations and funding are pivotal in building the necessary infrastructure and providing technical support to enhance FSM practices in low-income regions (Pugel, 2021). By addressing these challenges through targeted interventions and sustainable practices, FSM can significantly improve sanitation outcomes, promote public health, and protect the environment.

2.3 Faecal Sludge Applications

At present, the full potential of FS treatment products is not being utilized; it is common for FS to be disposed of by burying or dumping it into the environment (Semiyaga *et al*., 2015). However, reusing sludge is usually better than sending it to a landfill because selling treated sludge can generate income, and landfill space is often limited (Zewde *et al.*, 2021). There is now a shift in the focus of FS research from disposal to re-use as a fertiliser, soil conditioner, or as a component in value-added products. (Strande & Brdjanovic, 2014; Zewde *et al.*, 2021). Its use is, however, limited due to the dangers associated with it, such as the presence of pathogenic microbes (Odey *et al*., 2018).

2.3.1 Energy Production

Faecal sludge has numerous potential applications beyond agriculture, with energy production being one of the most promising (Tasnim *et al*., 2023). Through the process of anaerobic digestion, treated FS may be turned into biogas. Biogas, which is largely constituted of methane and carbon dioxide, can be utilized as a sustainable energy source for the purposes of cooking, heating, and the generation of electricity (Czekała, 2022). Also, this method helps reduce waste and offers a sustainable energy option, which is great for

areas that don't have enough power. Research has demonstrated that the biogas generated from FS may greatly diminish the dependence on conventional fossil fuels, therefore promoting energy security and environmental sustainability (Ajieh *et al*., 2021).

2.3.2 Construction Materials

Faecal sludge has also been utilized in new ways within the building sector. Processed sludge may be integrated into the manufacturing of construction materials, such as bricks and cement (Agarwal *et al*., 2022). It has been demonstrated that the incorporation of FS ash into cement may enhance the mechanical qualities of concrete, giving it the potential to serve as a viable alternative to traditional materials (Ducoli *et al*., 2021). Furthermore, bricks composed of a blend of FS and clay have demonstrated adequate robustness and longevity to be employed in affordable constructions (Parde *et al*., 2021). In addition to contributing to the reduction of building expenditures and the impact that they have on the environment, these applications provide a sustainable use for FS and give a long-term solution to the problem.

2.3.3 Environmental Remediation

Another important application of treated FS is in environmental remediation. Faecal sludge has strong biosorption capabilities that enable it to efficiently eliminate heavy metals and other contaminants from polluted water bodies (Koetlisi & Muchaonyerwa, 2019). Treated FS can be utilized as an adsorbent to clean industrial effluents, thereby improving water quality and protecting aquatic ecosystems (Mamera *et al*., 2021). Additionally, FS can be utilized to rehabilitate degraded lands. The organic component and nutrient profile of treated sludge help restore soil fertility and promote vegetation growth in barren or polluted areas

(Ali *et al*., 2019). This methodology not only helps in mitigating environmental degradation but also enhances biodiversity and ecosystem resilience.

2.3.4 Material Recovery

Faecal sludge treatment processes also enable the recovery of valuable materials. For instance, the pyrolysis of FS can produce biochar, a stable carbon-rich product that can be used as a soil amendment, water purifier, or even as an additive in composite materials (Cookey *et al*., 2022). Biochar has the potential to sequester carbon, thus contributing to climate change mitigation efforts (Arif *et al*., 2020). Furthermore, the treatment of FS can facilitate the recovery of phosphorus, a critical nutrient in limited supply globally. Phosphorus can be extracted from treated sludge and used in various industrial applications, including the production of fertilizers, detergents, and fire retardants (Kathi *et al*., 2023).

2.3.5 Faecal Sludge Application in Agriculture

Faecal sludge has long been used as a fertiliser by the people in Vietnam and Cambodia in both household gardens and agriculture to increase crop yield (Harper *et al*., 2021). Ungureanu *et al.* (2020) estimated that about 10% of the global population consumes food from crops irrigated with wastewater. Leonel & Tonetti (2021) estimated that across 50 countries, including Kenya, approximately 700 million individuals consume crops irrigated with such wastewater, covering at least 20 million hectares of land. Partly, nutrient recycling is done by on-site sanitation entrepreneurs who empty the faecal sludge on agricultural fields. Although unsafe from a health perspective, these practices have emerged without external support. Thus, the FS presents a value to farmers in the sanitation chain that can be explored by on-site sanitation entrepreneurs (Soeters *et al*., 2021)

The management and utilization of FS in Africa present a critical challenge intertwined with both environmental sustainability and public health concerns. Semiyaga *et al.* (2015) highlighted that the prevailing approach to FS treatment often involves its disposal through burying or dumping, posing significant environmental risks. However, recent studies such as Zewde *et al.* (2021) argue for the economic and environmental benefits of reusing treated sludge, emphasizing its potential as a source of income and a means to alleviate pressure on limited landfill space.

As noted by Strande & Brdjanovic (2014) and Zewde *et al.* (2021), this shift in perspective underscores a growing recognition of FS as a valuable resource rather than a waste product in Africa. The traditional use of human excreta and urine as fertilizers in Vietnam, as documented by (Ho Jr, 2023), resonates with similar practices in parts of Africa, particularly among farmers in rural areas who utilize untreated or poorly treated wastewater for irrigation, covering vast hectares of land (Pratap *et al*., 2021). Moya *et al.* (2019) further highlight the role of on-site sanitation entrepreneurs who engage in FS management, often repurposing it as fertilizer despite health risks.

In Kenya, FSM and utilization pose a critical challenge, traversing environmental sustainability and public health concerns. Studies have highlighted prevalent practices of FS disposal, often involving burial or dumping, which pose significant environmental risks (Awere *et al*., 2020). Equally, recent research by Zewde *et al.* (2021) emphasizes the economic and environmental benefits of reusing treated sludge, underlining its potential as a revenue source and a means to alleviate pressure on limited landfill space.

This shift in perspective is echoed in the global dialogue on FSM, aligning with initiatives in Kenya to reframe FS as a valuable resource rather than ordinary waste. Drawing parallels to

Kenya's context, the traditional use of human excreta and urine as fertilizers, as documented by Scott *et al.* (2004) in Vietnam, finds resonance among farmers in Kenya's rural areas. Like their Vietnamese counterparts, these farmers utilize untreated or poorly treated wastewater for irrigation on a significant scale. Furthermore, Choge (2021) sheds light on the role of on-site sanitation entrepreneurs in Kenya who manage FS, repurposing it as fertilizer despite associated health risks.

This approach highlights the natural importance of FS in Kenya's sanitation system, providing valuable information on possible methods for sustainable farming techniques. Therefore, the ongoing discussion on FS management in Kenya echoes a wider international movement towards utilizing its potential as a valuable asset for agricultural efficiency and environmental preservation, led by actual data and inventive methods.

2.4 Dangers associated with untreated FS in the environment

Discharging untreated FS into the environment has adverse consequences for human well-being and existence. Diarrhoea is recognized as a significant factor in the global burden of disease. This condition is more prevalent in poor nations due to their limited availability of safe drinking water, adequate sanitation, and hygienic behaviors.

Diarrhoea remains a significant cause of morbidity worldwide, accounting for the death of some 1.8 million people annually (Hasan *et al*., 2021). It is one of the significant causes of death among children under five years old. According to Troeger *et al.* (2017), approximately 533,768 deaths in this age group occur annually worldwide due to diarrhoeal diseases. The primary pathogens responsible for most of these deaths are rotavirus, Cryptosporidium spp., and Shigella spp.**,** all of which are transmitted through the faecal-oral route (Adetulubo *et al*., 2023).

In areas where untreated human excreta is used as an alternative fertilizer for vegetables and other crops that are consumed raw, the consumers risk diseases caused mainly by parasites such as *Ascaris spp* and *Trichuris spp*, as well as other food-borne pathogens, while farmers who work in the fields barefoot could face the occupational risk of hookworm infection (Adegoke *et al*., 2018; Chandler *et al*., 2021)

2.5 Faecal Sludge Treatment Methods

Pathogens and odour can be reduced or managed in FS using biological or mechanical treatment methods that utilise different physical, chemical, and biological treatment mechanisms. These include time, temperature, moisture, pH, and solar radiation. Some methods used in FS treatment and odour elimination include anaerobic and aerobic digestion, composting, alkaline treatment, which includes lime and ash addition to FS, and storage (Anderson *et al*., 2015). These methods differ in their efficiency and cost.

2.5.1 Odour Mitigation in Faecal Sludge

Odour treatment in FS management has been a critical area of research due to its implications for public health, environmental quality, and community acceptance of sanitation systems (Zewde *et al*., 2021). Various strategies have been explored to mitigate the offensive odour associated with FS, leveraging chemical, biological, and physical methods.

Chemical treatments have been a prominent focus, particularly the use of oxidizing agents such as hydrogen peroxide and ozone. Hydrogen peroxide has been effective in reducing odour through the oxidation of malodourous compounds, transforming them into less offensive substances (Piccardo *et al*., 2022). This approach significantly reduced volatile sulfur compounds (VSCs) and ammonia, which are primary contributors to the odour

problem (Orlov & Zotkin, 2021). Similarly, ozone treatment has shown promising results in deodorizing FS by breaking down complex organic molecules that contribute to the foul smell (Oliva *et al*., 2023).

Biological treatments have also gained attention due to their sustainability and effectiveness. Biofiltration, using microbial action to degrade odour-causing compounds, has been implemented with varying degrees of success. Studies have demonstrated that bio filters can effectively reduce the concentration of VSCs and other odorants through microbial degradation (González-Cortés *et al*., 2024). Additionally, the use of specific bacterial strains to inoculate sludge has been explored, with some bacteria such as *Bacillus subtilis*, *Bacillus cereus*, and *Schizophyllum commune* showing a high capacity for metabolizing odour-causing compounds (Fan *et al*., 2020; Su *et al*., 2023).

Adsorption methods employing materials such as activated carbon and biochar have been widely researched for their ability to capture and neutralize odorous compounds. With its high surface area and porosity, activated carbon has been particularly effective in adsorbing ammonia and VSCs from FS (Getaneh *et al*., 2021). Biochar, derived from the pyrolysis of organic matter, has also shown potential due to its adsorptive properties and additional benefits, such as soil amendment, when used as a byproduct (Gupta *et al*., 2022).

Combining these methods has been suggested as a way to enhance odour treatment efficiency. For instance, integrating chemical oxidation with biofiltration can immediately reduce odour, followed by sustained microbial degradation of residual compounds (Tripathi & Hussain, 2022). Such integrated approaches may offer more robust solutions to the complex odour control problem in FSM.

While combining methods like chemical oxidation with biofiltration can enhance odour treatment efficiency, there are also notable downsides. One concern is the potential cost implications, as integrating multiple treatment technologies can increase both capital and operational expenses (Ren *et al*., 2019). Chemical oxidation often requires the use of specialized reagents that may not be readily available or affordable in resource-constrained settings, such as many regions in Africa.

Additionally, these chemicals can have negative environmental impacts if not properly managed, leading to potential secondary pollution. Biofiltration, though effective for long-term odour control, may face challenges like clogging, reduced microbial activity under unfavorable conditions, and high maintenance requirements (Su *et al*., 2023). These limitations can compromise the overall sustainability and feasibility of such integrated approaches.

Recent advancements also include the development of sensor technologies for real-time monitoring of odorants. These technologies enable better management and optimization of treatment processes by providing timely data on the concentrations of key odour-causing compounds (Jin *et al*., 2023). Integrating sensor data with treatment systems can enhance the efficiency and effectiveness of odour control measures.

2.5.2 Anaerobic and Aerobic Digestion

Digestion or decomposition is an attractive approach to human waste treatment (Rajagopal *et al*., 2013). This treatment method is employed by large treatment plants, households, and small to large farms.

An important aspect in minimizing pathogens in FS is the ability of the digesting procedures to take place in either mesophilic (30 °C to 38 °C) or thermophilic (50 °C to 60 °C)

environments (Lanko *et al*., 2021). Since thermophilic digestion uses a higher temperature to reduce pathogen levels, it is more effective than mesophilic digestion at killing most bacteria in their vegetative growth stages, as long as the temperature is much higher than their optimal growth temperature and the exposure time is long enough (Jiang *et al*., 2020).

Methane, carbon dioxide, and trace quantities of other gases are produced during anaerobic digestion, along with heat production. Compared to aerobic digestion, the end product is stabilized sludge that has higher nitrogen content. Equally, small amounts of carbon dioxide, ammonia, and other gases, as well as a large quantity of heat, are produced during aerobic digestion, along with a final waste known as sludge (Arthurson, 2008). Anaerobic digestion is preferred since it can stabilize the faecal sludge's organic fraction while generating biogas to offset some energy needs at the treatment plant (Semiyaga *et al*., 2022).

Horan *et al.* (2004) conducted a study that showed the primary sludge digestion stage of mesophilic anaerobic digestion achieved a log removal of 1.66 for *E.coli,* 2.23 for *Listeria monocytogenes,* and 2.23 for *Salmonella senftenberg.* However, the presence of *Campylobacter jejuni* was not affected by primary sludge digestion. The supplementary decrease in population was achieved by the process of secondary sludge digestion, resulting in log removals of 1.70 for *E.coli,* 2.10 for *S. senftenberg,* and 0.36 for *C. jejuni.* Consequently, more secondary die-offs were necessary in order to achieve a substantial reduction in pathogen levels.

Cárdenas-Talero *et al.* (2022) also found that aerobic stabilization did not sufficiently decrease the presence of pathogens and indicator organisms to meet the necessary standards for the unrestricted use of sludge in agriculture.

2.5.3 Co-composting

Co-composting is the deliberate process of breaking down organic materials utilizing several substances in a controlled environment with the presence of oxygen. This process is supported by microorganisms such as bacteria, fungi, and protozoa, and during the maturation stage, it is supported by microfauna such as ants, worms, flies, and nematodes (Cofie *et al*., 2016).

There are two co-composting designs: open and in-vessel (Thomas *et al*., 2020). In open design, the mixed material is piled into heaps called windrows and left to decompose; the material is continuously turned to ensure oxygen flow and to ensure even heat exposure to the material. In-vessel composting requires precise moisture and air supply as well as mechanical mixing. Therefore, decentralized facilities are less favorable (Pandiyan *et al*., 2020; Tilley *et al*., 2014).

Co-composting FS with other organic materials, such as agricultural and kitchen wastes, generates heat when microorganisms break down the organic material, which leads to pathogen reduction in FS while at the same time allowing for the recycling of essential nutrients into agriculture, thereby concluding the nutrient circle (Alamin & Bari, 2022). This process also helps reduce the volume of waste through the biodegradation of organic matter and carbon to carbon dioxide (Zhong *et al*., 2023).

A recent study conducted by Manga *et al.* (2021) in Kampala, Uganda, found that co-composting of FS with chicken waste for eight weeks, with temperatures ranging from 50.7 - 58.7°C, was able to maintain sustained temperatures in the piles for over 31 days. By turning the piles every seven days, this method resulted in complete pathogen inactivation; however, it was noted that pathogen inactivation in composting piles does not entirely

depend on temperature and time, but also other factors such as microbial antagonistic mechanisms or antibiotic action induced by indigenous microbial, moisture content, change in pH, nutrient depletion, and toxic by-products such as ammonia, conditions that are tough to control thus reducing its effectiveness(Millner *et al*., 2014; Vaddella *et al*., 2018).

This method is also plagued with several disadvantages, such as large land area requirements, long storage time, expertise in design and maintenance, and substantial labor requirements, thus making the method expensive (Obaideen *et al*., 2022).

2.5.4 Alkaline Treatment

Alkaline treatment involves adding alkaline material such as wood ash and lime to treat FS and manage odour. This method has proven effective in reducing the pathogenic bacteria to acceptable levels by increasing the pH levels to above 11 (Monney & Awuah, 2015).

Results from an experimental study carried out to determine the efficiency of lime on FS indicated that the addition of 4% and 6%-10% of quicklime to FS maintained the pH level of the resulting mixture above 12 for 20–60 days and three months, respectively (Nobela, 2014). In a similar study by Anderson *et al.*, 2015) using hydrated lime, the *E.coli* levels were reduced to below detectable levels after five hours at pH 10 and one hour at pH conditions above pH 11. The total coliform count was also reduced to undetectable levels after two hours at pH 11.5 and after one hour at pH 12.

According to several studies, lime is a suitable treatment method in FS; its use is, however, unsuitable for FS that will be reused as a soil conditioner (Anderson *et al*., 2015). Pathogenic bacteria have been observed to regrow due to a decrease in pH levels, hence requiring the addition of more lime, resulting in a higher cost of treatment (Ali & Shahreen, 2024).

Wood ash has also, for a long time, been used to treat FS from latrines by increasing the pH to levels toxic to pathogenic bacteria and also to manage odour and flies (Kurola *et al*., 2011; Niwagaba *et al*., 2009; Zindoga, 2016). Its use is, however, unsuitable due to its long duration in eliminating pathogenic bacteria. According to a study by Magri *et al.* (2013), the measured decay in egg inactivation viability of *A. suum* was low. Data on the ideal quantity of ash needed to increase the pH of FS to sanitizing levels also remains unknown (Monney & Awuah, 2015).

2.5.6 Solar Drying

Solar drying leverages the sun's natural energy to reduce faecal sludge's moisture content, thereby decreasing its volume and weight. The dried sludge can be safely disposed of or used as a fuel source (Kodom *et al*., 2024). Solar drying is cost-effective and environmentally friendly, making it an attractive option for low-resource settings. This method is, however, particularly useful in regions with abundant sunlight (An-Nori *et al*., 2021).

2.5.7 Pyrolysis

Pyrolysis is defined as the thermal decomposition process conducted in the absence of oxygen (Soltes & Elder, 2018). This relatively new technology in FSM works by converting FS into biochar, bio-oil, and syngas. Biochar can be used as a soil amendment to improve soil fertility and sequester carbon. Biochar can be utilized as a renewable fuel, and syngas can be used for generating energy (Tan *et al*., 2023). Pyrolysis not only reduces the volume of sludge but also recovers valuable resources, thereby contributing to the circular economy model. Pyrolysis is, however, suitable in the treatment of large volumes of sludge and pre-drying before the process (Ziegler, 2018).

2.5.8 Vermicomposting

Vermicomposting uses earthworms to process FS, accelerating decomposition and enhancing nutrient availability in the compost (Nsiah-Gyambibi *et al*., 2021). The worms consume organic matter, breaking it down into a nutrient-rich product known as vermicompost. One of the significant benefits of vermicomposting is its ability to reduce pathogenic bacteria in the treated waste, making it safer for agricultural use (Wang *et al*., 2024). This reduction in pathogens is achieved through a combination of biological, chemical, and physical processes facilitated by the earthworms (Zeb *et al*., 2020). This environmentally friendly method produces high-quality compost that can improve soil structure and fertility.

2.5.9 Black Soldier Fly Larvae (BSFL) Treatment

BSFL treatment utilizes the larvae of the black soldier fly to rapidly consume FS, reducing its volume significantly (Achieng Oyoo *et al*., 2023). The larvae can be harvested and processed into a variety of products, such as animal feed, biodiesel, and oil (Franco *et al*., 2021). This method is efficient in reducing waste and offers the added benefit of producing valuable by-products. This FS treatment method has also been found to reduce pathogenic bacteria and odour by utilizing their gut micro biota (J. Zhang *et al*., 2020).

BSFL treatment is gaining popularity for its rapid processing time and potential for resource recovery. However, while BSFL can reduce pathogens to a certain extent, they are not as effective as other treatment methods like composting or chemical treatment. Pathogens may persist in the remaining waste, posing health risks if not further treated (Tokwaro *et al*., 2023)

2.5.10 Hydrothermal Carbonization (HTC)

Hydrothermal carbonization (HTC) converts wet FS into hydrochar through a process involving heat and pressure in a water medium (Amenyeku *et al*., 2024). Hydrochar is a stable, carbon-rich material that can be used as a soil amendment or for energy production (Islam *et al*., 2021). HTC is effective in reducing the volume of sludge and recovering resources, as well as utilizing the high temperatures to kill pathogens, thus making it a promising technology for sustainable waste management (X. Zhang *et al*., 2020).

2.5.11 Electrochemical Treatment

Electrochemical treatment uses electrical currents to disinfect and reduce the organic matter in FS. This method can achieve high levels of pathogen reduction and organic matter removal (Hand & Cusick, 2021). Electrochemical treatment is a relatively new method in FS treatment, but it shows potential for small-scale applications, particularly in areas with limited access to traditional treatment methods (Tonanon & Webster, 2023).

2.5.12 Struvite Precipitation

Struvite precipitation recovers phosphorus from FS in the form of struvite ($NH_4MgPO_4 \cdot 6H_2O$), a mineral that can be used as a slow-release fertilizer (Yesigat *et al*., 2022). This process addresses both waste treatment and resource recovery, contributing to sustainable agricultural practices.

Struvite production helps mitigate the issue of phosphorus scarcity and reduces environmental pollution from untreated sludge (Achilleos *et al*., 2022). While this method is promising, it has several drawbacks, such as high operational costs, struvite scaling, blockages, and variability in waste composition (Achilleos *et al*., 2022).

2.5.12 Bio Electrochemical Systems (BES)

Bio-electrochemical systems (BES) use microbial fuel cells or microbial electrolysis cells to treat FS and generate electricity or hydrogen gas (Leicester *et al*., 2020). These systems exploit the metabolic processes of bacteria to convert organic matter into energy. BES offers a dual benefit of waste treatment and energy production, making it a promising technology for integrated waste management solutions (Tota-Maharaj, 2020).

2.6 Lactic Acid

Lactic acid is a weak organic acid that occurs naturally and may be synthesized chemically or biologically, and has found use in many industries, such as cosmetic, pharmaceutical, chemical, and medical industries, and more recently in FS treatment (Anderson *et al.*, 2015; Florou-Paneri *et al.*, 2013).

C.W. Scheele discovered lactic acid in 1780 while examining sour milk. In 1881, Fermi successfully produced lactic acid by fermentation, leading to its subsequent commercial production (Martinez *et al*., 2013). It can be made by the fermentation of sugars using various *Lactobacillus* strains such as *L. manihotivorans*, *L. paracasei,* and *L. plantarum* obtained from renewable resources, which means that it is an eco-friendly product that has attracted a lot of attention in recent years (Komesu *et al*., 2017).

The microbial inhibition mechanism of lactic acid is probably connected to the solubility of the non-dissociated lactic acid within the cytoplasmic membrane and the insolubility of dissociated lactate anion, which results in acidification of the cytoplasm and malfunction of proton motive forces (Florou-Paneri *et al*., 2013). This eventually influences the trans membrane pH gradient and decreases the energy available for cells to grow, thus resulting in pathogenic organism death (Malambo, 2014).

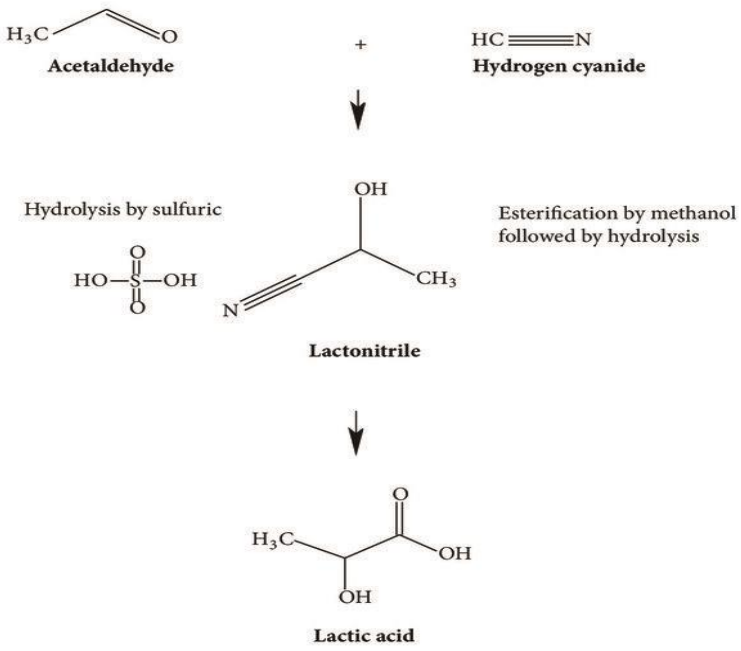
2.6.1 Lactic Acid Production

Lactic acid can be produced via chemical synthesis and fermentation (Malambo, 2014). Via chemical synthesis, acetaldehyde is reacted in the liquid phase under high-pressure conditions in the presence of hydrogen cyanide and a base, forming lactonitrile. After its recovery and refinement by distillation, hydrochloric acid or sulfuric acid is added to hydrolyse lactonitrile to lactic acid, which is then esterified with methanol to produce methyl lactate, and this is recuperated and refined by distillation. The purified methyl lactate is finally hydrolysed in an acidic aqueous solution to lactic acid and methanol, with methanol being recycled in the same process (John *et al.*, 2009; Martinez *et al.*, 2013).

Figure 2.1: The schematic presentation of the production of lactic acid by chemical process.

Figure 2.1

Production of lactic acid by chemical process



Source: *Riaz et al., 2018*

This entire process is optimized to ensure maximum yield and purity of lactic acid, a crucial intermediate for various industrial applications, including the production of biodegradable plastics, pharmaceuticals, and food additives (Swetha *et al*., 2023).

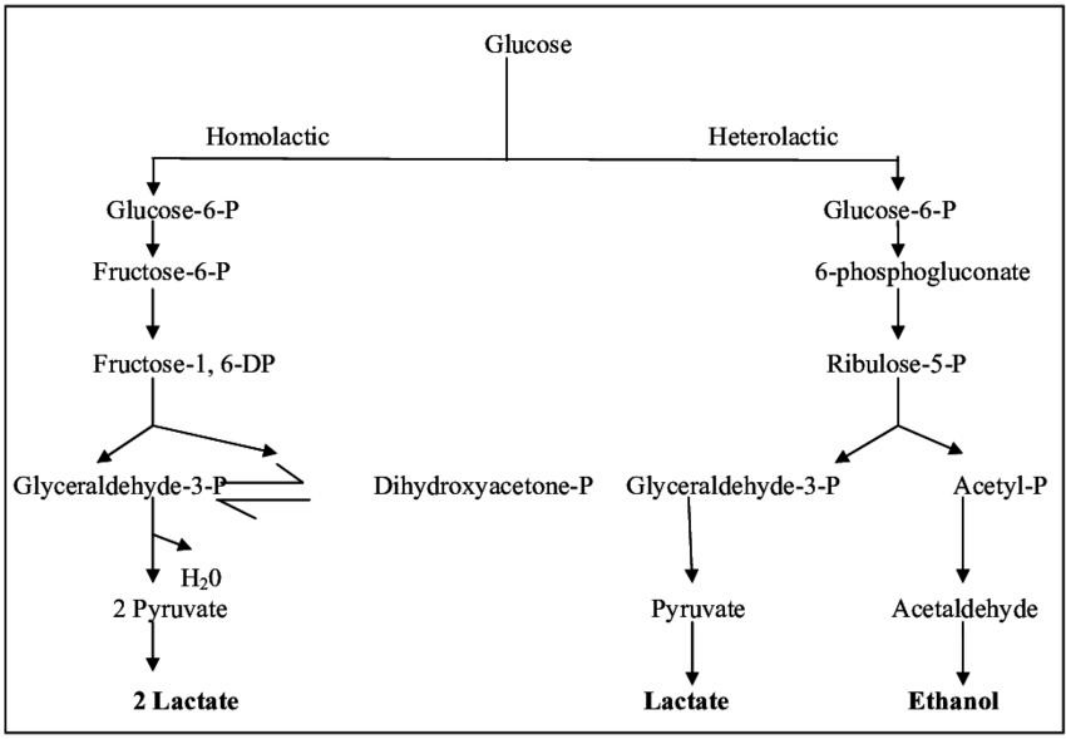
The choice of hydrochloric or sulfuric acid for hydrolysis is carefully considered based on factors such as reaction efficiency, cost, and ease of handling. Throughout the process, careful monitoring of reaction conditions such as temperature, pressure, and pH is essential to prevent by-product formation and to achieve high selectivity. Additionally, the recycling of methanol enhances the economic viability of the process and minimizes environmental impact by reducing waste. The overall process design reflects a balance between chemical efficiency, cost-effectiveness, and sustainability, making it a robust method for lactic acid

production on an industrial scale. However, the overall cost is higher than that of fermentation (Meramo-Hurtado & González-Delgado, 2021; Venus *et al*., 2018).

Lactic acid production via fermentation is rapid and yields high quantities. It can be tailored to selectively produce either one of the two enantiomers of lactic acid or a racemic mixture of both (Auras *et al*., 2022). The process involves the fermentation of sugars or sugar-containing hydrolysates or the single-step conversion of starchy or cellulosic wastes by direct conversion using amylolytic lactic acid-producing microorganisms, LAB (Anderson *et al*., 2015; Nwamba *et al*., 2021). Among lactic acid bacteria, there are two primary sugar fermentation processes. This metabolic process is known as homolactic fermentation, and the end product of glycolysis (the Embden-Meyerhof pathway) is nearly always lactic acid under typical circumstances (Chandel, 2021). The process can also occur in a heterolytic way, leading to the production of lactic acid and ethanol, as shown in Figure 2.2.

Figure 2.2

Fermentation reaction scheme of glucose to lactic acid



Source: (Stephen & Saleh, 2023)

2.6.2 Lactic Acid Pathogen Elimination Mechanisms

The inhibition of gram-negative enteric bacteria such as *E. coli* is challenging due to their resistance to antimicrobials (Breijyeh *et al*., 2020). The reason suggested for this resistance is the inability of the antimicrobials to penetrate the protective outer membrane of the gram-negative bacteria, made up of glycerophospholipids and lipopolysaccharides molecules (Hyldgaard, 2020). Several reports suggest that the synergetic use of chelators (outer membrane disrupting agents) and antimicrobials produced by LAB extends the antimicrobial spectrum to include gram-negative bacteria (PATERSON, 2021; Ribeiro *et al*., 2022).

In a study carried out on *E.coli* O157:H7, *Pseudomonas aeruginosa*, and *Salmonella enterica* serovar Typhimurium, it was observed that lactic acid permeabilises the gram-

negative bacterial outer membrane, thus acting as a potentiator of the effects of other antimicrobial substances in addition to its antimicrobial property due to the lowering of the pH (Alakomi *et al*., 2000). This study focused on lactic acid's effectiveness in treating FS.

2.6.2 Production of Lactic Acid from Vegetable Waste

The production of lactic acid from perishable wastes is a subject of increasing interest due to its potential in sustainable waste management and the generation of valuable by-products. For instance, studies have shown that LAB can be utilized to convert cabbage waste into lactic acid, highlighting the bacteria's role in waste degradation and the production of food additives, bioenergy, and biogas (Novik *et al*., 2017).

Similarly, research on carrot waste has explored the application of LAB in the production of functional foods, such as synbiotic yogurt, where carrot waste extract is used as a substrate for LAB, resulting in the production of lactic acid and other beneficial compounds (Aiello *et al*., 2020). Tomato waste, on the other hand, has been studied for its potential in lactic acid production through eco-friendly processes, with feasibility studies considering the conversion of tomato processing waste into lactic acid, thus contributing to a circular economy approach (Carillo *et al*., 2018).

Numerous studies have investigated lactic acid production from various perishable wastes, aiming to identify efficient and cost-effective substrates for lactic acid fermentation. In-depth analysis reveals that the quantities of lactic acid produced can vary significantly based on the type of waste, the specific strains of LAB used, and the fermentation conditions (Odey *et al*., 2018). For example, the optimization of vegetable wastes for lactic acid production has been a focus, with laboratory-scale approaches demonstrating the potential for significant yields from carrot and tomato wastes (Caldeira *et al*., 2020).

The integration of these processes into FSM and treatment and sustainable food production systems has been emphasized, with lactic acid serving as a key component in extending the shelf-life of food products and enhancing food safety. Common perishable wastes used include fruit peels, vegetable residues, and food processing by-products. For instance, fruit wastes like apple peels, banana peels, and citrus wastes are rich in fermentable sugars, making them effective substrates for lactic acid production (Dwivedi *et al*., 2022; Sánchez *et al*., 2021). Additionally, grape pomace and mango peels have shown high potential due to their high carbohydrate content and availability in large quantities from the juice and fruit processing industries (Iqbal *et al*., 2021).

Using vegetable wastes such as cabbage, carrot, and tomato for lactic acid production presents a sustainable and economically viable approach. Optimizing fermentation conditions and addressing substrate-specific challenges can further enhance the efficiency and yield of lactic acid production. This not only adds value to agricultural by-products but also supports environmental sustainability by reducing waste and promoting a circular economy.

Similarly, vegetable residues such as potato peels, corn husks, and leafy greens have been utilized for lactic acid production, often requiring pretreatment to release fermentable sugars (Rivas *et al*., 2022). Pretreatment methods such as enzymatic hydrolysis, acid hydrolysis, and steam explosion are employed to break down complex carbohydrates into simpler sugars, enhancing the efficiency of the fermentation process (Akhtar *et al*., 2016). Furthermore, broccoli stalks and cauliflower leaves, often discarded during vegetable processing, have been identified as valuable substrates due to their carbohydrate-rich

composition and relatively low lignin content, which facilitates easier breakdown and fermentation (Rivas *et al*., 2022).

Food processing by-products like dairy whey, molasses, and brewery waste have also been extensively studied, providing consistent and reliable sources for fermentation (Komesu *et al*., 2017). Dairy whey, a by-product of cheese production, contains lactose, which is readily fermentable by lactic acid bacteria, making it a highly effective substrate. Molasses, a by-product of sugar production, is rich in sucrose and other sugars, supporting robust lactic acid production (Mladenović *et al*., 2018). Brewery waste, which includes spent grains and yeast, offers a nutrient-rich medium that supports the growth and metabolism of lactic acid bacteria, leading to high lactic acid yields (Liguori & Faraco, 2016). Additionally, the use of cassava bagasse, a by-product of tapioca production, has been investigated due to its high starch content, which can be converted to fermentable sugars through enzymatic processes (Buakeaw *et al*., 2023).

These substrates provide a sustainable approach to waste management and offer a cost-effective alternative for industrial lactic acid production. These perishable wastes' diverse nature and availability make them attractive options for large-scale fermentation processes, contributing to a circular economy and reducing environmental impact.

Focusing on cabbage (*Brassica oleracea*), carrot (*Daucus carota*), and tomato (*Solanum lycopersicum*), these substrates show considerable promise for lactic acid production. Cabbage waste, including outer leaves and trimmings, is rich in fermentable carbohydrates (Das & Mallikarjunarao, n.d.). Studies have demonstrated that submerged fermentation using lactic acid bacteria such as *Lactobacillus plantarum* and *Lactobacillus casei* can yield up to 90 g/L of lactic acid under optimized conditions (Sheeladevi & Ramanathan, 2011).

This high yield is attributed to the readily available fermentable sugars in cabbage waste, making it an excellent substrate for lactic acid production (Abedi & Hashemi, 2020; Nochebuena-Pelcastre *et al*., 2023). The carbohydrate-rich composition of cabbage, particularly glucose, supports these lactic acid bacteria's robust growth and metabolic activity, which is crucial for efficient lactic acid synthesis.

Carrots also show considerable promise as substrates for lactic acid production due to their high sugar content, including sucrose, glucose, and fructose. The high sugar content and beta-carotene in carrot waste enhance fermentation, making it a valuable substrate (Kaur *et al*., 2019). Carrot processing waste, such as peels and pulp, contains these fermentable sugars in significant quantities. Research has shown that lactic acid bacteria like *Lactobacillus delbrueckii, Lactobacillus fermentum,* and *Lactobacillus pentosus* can effectively convert the sugars in carrot waste into lactic acid to produce lactic acid yields ranging between 80 and 100 g/L (Das & Mallikarjunarao, 2008).

Another study reported yields of up to 70 g/L of lactic acid from carrot peels under optimized fermentation conditions (Lorn, 2020). The efficiency of these processes is also influenced by factors such as initial sugar concentration, pH control, and nutrient supplementation. Utilizing carrot waste not only adds value to agricultural by-products but also addresses environmental concerns associated with waste disposal, supporting sustainable agriculture and industrial processes.

Tomato waste, including skins, seeds, and pulp from the food processing industry, is another promising substrate. The high moisture content and sugar profile of tomato waste make it suitable for microbial fermentation. Although the variable composition and high water content of tomato waste can pose challenges, pre-treatment processes such as drying or

concentration can enhance fermentable sugar content and improve lactic acid yields (Hijosa-Valsero *et al*., 2019). The production of lactic acid from perishable wastes, particularly cabbage, carrot, and tomato, presents a sustainable and economically viable approach. These wastes are rich in fermentable sugars, making them suitable substrates for fermentation. Future research should focus on optimizing fermentation conditions and exploring pretreatment methods to enhance lactic acid yields, thereby contributing to waste management and environmental sustainability.

2.6.3 Lactic Acid Use in FS Treatment; Summary and Research Gap

Lactic acid has long been used in the food industry, but few studies have been conducted on its application in FS treatment (Anderson *et al.*, 2015). FS treatment via fermentation and acidification processes is one of the most reliable methods for pathogen inactivation and odour control (Yemaneh *et al*., 2012).

A study by (Odey *et al.*, 2018) showed that fermenting a combination of rice flour and brown sugar could be applied to treat FS. The faecal coliform count reached below the acceptable limit ($<10^3$ CFU/100 mL) on day 15 in the reactor that contained FS and fermented rice water in equal portions. However, rice flour is an expensive commodity, hence making this process unsuitable for use in FS treatment in Low and Middle Income Countries (LMIC).

In a similar study carried out in Blantyre, Malawi, in which lactic acid fermentation was carried out by use of molasses and sugar as substrates, it was noted that lactic acid fermentation by this method had the highest cost for the initial batch at €31.2/m^3 sanitised sludge compared to urea treatment which was estimated to be €20 / m^3 sanitised sludge, and finally by use of lime which was at €12 / m^3 of FS (Anderson *et al*., 2015; Malambo, 2014).

In other studies, several lactic acid bacteria species do not produce effective lactic acids for pathogen inactivation in FS. For example, when lactic acid from fermented cassava flour was used to sanitise FS, the pH and pathogens did not significantly decrease (Odey, 2018).

The identification of cost-effective substrates for lactic acid production in FS sanitization is vital. While sugar and molasses have proven effective, their high costs make them impractical for use in poor settings. Therefore, exploring alternative substrates such as vegetable wastes is critical.

Vegetable wastes are promising candidates due to their high content of fermentable sugars and their abundance as agricultural by-products. These substrates not only offer a potential reduction in costs but also contribute to waste management and environmental sustainability. For instance, cabbage waste, rich in glucose and other fermentable sugars, has shown high yields of lactic acid in submerged fermentation using *Lactobacillus plantarum* and *Lactobacillus casei* (Lima *et al*., 2023).

Similarly, carrot and tomato waste, both high in sugars, could serve as effective substrates for lactic acid production, which in turn could be used for FS sanitization. This approach not only addresses the economic constraints but also leverages locally available resources, making it a sustainable solution for developing regions.

Moreover, the specific characteristics of these vegetable wastes can be optimized to enhance lactic acid production. Factors such as initial sugar concentration, pH control, and nutrient supplementation can significantly influence the efficiency of the fermentation process. For example, maintaining optimal fermentation conditions, such as a pH and a temperature range of 30-37 °C, has been reported to maximize lactic acid yields. Additionally, pre-

treatment processes like drying or concentration can increase the fermentable sugar content of these substrates, further improving lactic acid production.

The presence of bioactive compounds in these wastes, such as lycopene in tomatoes, can also offer added benefits, potentially enhancing the antimicrobial properties of the produced lactic acid (Rodríguez *et al*., 2021). This multifaceted approach not only aims to improve the economic feasibility of lactic acid fermentation for FS sanitization but also enhances the overall sustainability and efficiency of the process. Therefore, continued research and optimization in this area are essential to develop a viable and scalable method for FS treatment, leveraging the untapped potential of vegetable wastes.

This study aided in the identification of types of vegetable wastes that are available locally and can yield high amounts of lactic acid that can lead to pathogen elimination to acceptable levels.

CHAPTER THREE: METHODOLOGY

3.1 Study Site

Sampling took place in Mitunguu town, Meru County. Meru County is served by one centralized sewerage system, which only serves Meru town, while other major towns within the county completely lack a functional centralized sewerage system. As an attempt to tackle this challenge, a decentralized treatment facility was constructed under the Up-scaling Basic Sanitation for the Urban Poor program in Mitunguu town, and it runs under the Ministry of Water, Sanitation, and Irrigation, Water Fund, and IMETHA Water and Sanitation Company. The Government of Germany funded the program via KfW Development Bank. The Decentralized Treatment Facility (DTF) receives sewerage from the surrounding areas via exhauster trucks at the receiving bay; it is then fed into an Anaerobic Baffled Reactor (ABR), where the effluent is treated. The effluent then proceeds to the vertical flow constructed planted drying beds. The effluent is then finally composted for agricultural use.

3.2 Research Design

This study was an experimental research design. In this design, one or more independent variables were controlled by the researcher (as treatments), subjects were randomly assigned to different treatment levels (random assignment), and the results of the treatments on outcomes (dependent variables) were observed (Hafliðadóttir *et al*., 2021).

3.3 Faecal Sludge Sample Collection

Faecal sludge was sourced from the DTF. Sample size and sampling method were acquired according to the method described by (Velkushanova *et al*., 2021). Five kilograms of FS were obtained via a composite grab sampling technique, which involved taking equal sample portions from an exhauster track at set intervals during emptying, as shown in Figure 3.1

below. The portions were then mixed in order to obtain a homogenous mixture and transported in an air-tight, closed HDPE container to Meru University of Science and Technology, Sanitation Research laboratory, where the initial FS characteristics, such as pH, total solids, volatile solids, and *E.coli* were recorded prior to treatment.

Figure 3.1

Collection of FS sample by the researcher

Source: (Researcher, 2024)

3.4 Organic Waste Sample Collection

Fresh cabbage, tomato, and carrot vegetable wastes serving as substrates for lactic acid were sourced from Nkubu market and transported to Meru University of Science and Technology Biological Sciences laboratory, where size reduction was carried out by use of a kitchen knife and a heavy-duty commercial blender (CK-777, China) in order to increase the fermentation process. One thousand grams of each substrate was mixed with 1000 mL of distilled water, and the mixture was homogenized. The wastes were then added to plastic

containers and sealed to make them airtight and incubated at 37 ℃ for seven days, as described by Omar *et al*. (2009).

3.5 Experimental Setup

3.5.1 Medium preparation for lactic acid bacteria growth

De Man–Rogosa–Sharpe agar (MRS) (HIMEDIA) weighed at 5.515 g using an analytical balance (MA2104C, Japan) and dissolved in 100 mL of distilled water. The media was then heated while being stirred using a hot plate with a magnetic stirrer (MS/-h550-S USA) at 350 ℃ for 10 minutes in order to dissolve the media completely. The media was then autoclaved for 15 minutes at 121 ℃, cooled to 50 ℃, and dispensed into sterile petri dishes. After the media had set, 0.1 mL of the homogenized fermented vegetable waste sample was poured on the media and spread using the L glass rod. The plates were then wrapped with parafilm to create an anaerobic environment and incubated at 37 ℃ for 24 hours.

3.5.2 Biochemical Confirmatory Test for LAB.

To confirm the presence of LAB in the fermented samples, a catalase confirmatory examination was conducted as part of the biochemical analysis. Using a sterile inoculating loop, a 24-hour colony was collected and placed onto a microscope slide. Using a dropper, 1 drop of 3% H_2O_2 was placed on the microscope slide, and the slide was observed for copious bubbling.

3.5.3 Qualitative Determination of Vegetable Wastes Derived Lactic Acid

Lactic acid is unsuitable for direct analysis using GC-MS due to its polar and non-volatile nature. Lactic acid was subjected to a derivatization process via acid-catalyzed esterification. The analytes were prepared for GC–MS analysis through derivatization to enhance volatility and improve detection sensitivity. A volume of 10 µL of the sample was accurately

measured into a clean glass vial. To this, 2 mL of n-butanol and two drops of 1 M hydrochloric acid (HCl) were added. The mixture was gently vortexed and subsequently incubated in a water bath at 45 °C for 15 minutes to allow esterification to proceed.

Following incubation, the mixture was cooled to room temperature. Thereafter, 500 µL of HPLC-grade water was added, followed by 1 mL of hexane. The resulting mixture was capped and shaken vigorously for 3 minutes, after which it was allowed to stand undisturbed until phase separation occurred. The upper organic (hexane) layer was carefully separated, and a 100 µL aliquot of this layer was drawn using a micropipette.

The aliquot was filtered through a nylon syringe with a 0.45 µm pore size filter to remove any suspended particulates. The filtrate was transferred into a clean GC–MS auto sampler vial and stored under appropriate conditions until instrumental analysis. The GC-MS was set at parameters shown on Table 3.1.

Table 3.1

GC-MS Parameters

Injection volume	1 µL
Injection mode	Split, with a split ratio of 10:1
Injector temperature	200 °C
Carrier gas	Helium, supplied at a pressure of 11.3 psi
Column flow	1.1 mL/min
Total flow	15.2 mL/min
Linear velocity	38.2 cm/s
Purge flow	3.0 mL/min
Oven temperature program	
Initial temperature	50 °C (hold time 0.0 min)
Ramp 1	15 °C/min to 200 °C (hold 0.0 min)
Ramp 2	10 °C/min to 285 °C (hold 5.5 min)
Total run time	5 Minutes
Column details	BPX5, 30 m × 0.25 mm I.D., 0.25 µm film thickness

Source: (Researcher, 2025)

The mass spectrometer was operated in electron impact ionization (EI) mode at 70 eV, scanning within the range of 40–550 m/z. Data acquisition and chromatogram processing were conducted using the instrument's dedicated software.

3.6 Lactic Acid Recovery

The lactic acid was recovered according to the (Getaneh *et al*., 2021) method with a few modifications: The incubated vegetable wastes were frozen overnight, followed by thawing in a drying oven at 60 ℃ for 2 hours, followed by filtration and centrifugation at 6000 rpm for 10 min using a centrifuge (Hermule Labnet Z216 MK, USA).

3.6.1 Spectrophotometric Quantification of Vegetable Wastes Derived Lactic Acid

Lactic acid was quantified by the use of a cost-effective and highly efficient spectrophotometric technique as described by (Borshchevskaya *et al*., 2016). The technique relies on spectrophotometric analysis to measure the concentration of the colorful compound formed by the interaction between lactate ions and iron (III) chloride at a wavelength of 390 nm.

3.6.2 Construction of a standard lactic acid calibration curve

A lactic acid solution was prepared by diluting 1.2 g of lactic acid (89% purity, density 1.2 mg/mL) with distilled water in a 100 mL volumetric flask. The lactic acid was carefully measured and transferred into the flask, followed by the addition of distilled water to the calibration mark. The solution was then thoroughly mixed to ensure homogeneity. A series of LA solutions was prepared using 2-fold dilutions. Iron (III) chloride (Loba Chemie, Gujarat, India) (0.3 g) was added to a volumetric flask of 100 mL of distilled water for a solution of iron (III) chloride.

A hundred microliters (100 µL) of lactic acid dilution was added to 4 mL of iron (III) chloride and stirred, and the procedure was repeated for all diluted lactic acid solutions. The absorbance of the mixture was measured at 390 nm using a spectrophotometer (Apel PD-UV-3000UV, Japan). Finally, a calibration curve showing the relationship between the

concentration of lactic acid (g/mL) at 2.4, 1.2, 0.6, 0.3, and 0.15 mg/mL and absorbance was plotted. One hundred microliters (100µL) of the sample was mixed with 4 mL of iron III chloride, and the absorbance was noted. The absorbance was then used to determine the concentration of lactic acid in the sample.

3.7 Assessment of Cabbage Waste Derived Lactic Acid in the Reduction of *E. coli* in FS

For assessment of lactic acid in inactivation of pathogens, four reactors containing 1:1, 1:0.5, 1:0.35, FS to lactic acid, and a control were set up as described by (Getaneh, 2021). Cabbage waste was selected as the source of lactic acid among the vegetable wastes due to its local abundance. Samples were collected from all reactor containers after every four days for the pH analysis, volatile solids, total solids, and *E. coli* enumeration for 16 days.

FS treatment efficacy was assessed using *E. coli* as the indicator organism. The ability of lactic acid to inactivate the *E. coli* was determined through a total viable plate count after plating on Violet Red Bile Agar (HIMEDIA), which is a selective media for *E. coli* (Anderson *et al.*, 2015; Finney, Smullen, Foster, Brokx, & Storey, 2003; Digo, 2015). Samples taken from each reactor were serially diluted by adding 1 mL of the sample suspension to 9 mL of distilled water to make a final volume of 10 mL, and the mixture was shaken. This process was repeated up to the last tube (5^{th}), denoted dilution factor 5, and done in triplicate.

The medium was prepared according to the manufacturer's instructions and then poured into the sterile petri dishes. Once solidified, 0.1 mL of the sample solution was pipetted on the petri dish and spread using a sterile glass rod. The petri dishes were then incubated for 18-24 h at 37 ℃. *E. coli* colonies, present as reddish-pink, were then counted using a colony counter.

3.8 Determination of TS and VS Content of FS before and after Treatment

The total solid content of the FS before and after treatment was assessed by adopting the 2540E standard method for wastewater examination. A 10 g sample of thoroughly mixed sample was evaporated to a constant weight in a porcelain crucible in a drying oven at 105 ℃, and the weight noted; the remaining solids were cooled down to room temperature in a desiccator to avoid absorption of air moisture and then re-weighed. The Total solids were calculated based on the following formula:

$$\text{Total solids (g/L)} = \frac{(W3\,(g) - W1\,(g))}{V\text{sample (L)}} \quad (4.1)$$

Where:

W1 = Crucible mass (g)

W3 = Dry sample mass + crucible mass after drying (105 ℃) (g)

V_{sample} = Volume of sample used (L)

The volatile solids were determined by further igniting the sample in a muffle furnace (MC2.5-12, China) at temperatures of 550 ℃ for 1 hour and calculated based on the following formula:

$$\text{Volatile Solids (g/L)} = \frac{(W3\,(g) - W4\,(g))}{V\text{sample (L)}} \quad (4.2)$$

W3 = Weight of residue + crucible after drying at 105 ℃ (g)

W4 = Weight of residue + crucible after ignition in the furnace at 550 ℃ (g).

V_{sample} = Volume of sample used (L)

3.9 Determination of pH

The pH value of the samples was measured using a standard laboratory pH meter (Model Bante 902, Zhejiang, China), standardized using buffer solutions at pH 7, 10, and 4.

3.10 Odour Evaluation of FS treated with Cabbage Waste Derived Lactic Acid

The odour level of the FS was determined according to method 2150B of water and wastewater analysis; Threshold Odour Test (T.O.N). The potency of the perceived odour was investigated by diluting the sample with odour-free water until the odour was just detectable. FS samples were added to labeled 500 mL conical flasks and topped up to 50 mL with odourless water. Odourless water was added to a separate flask for use as a standard. The flasks were covered with petri dishes and placed in a water bath (SY-21 6H, China) pre-heated to 60 ℃ for 10 minutes. The samples were then covered with a foil so that the contents within the flasks were not visible to the panelist. The flasks were randomized using codes and randomly placed on the bench top for the odour test panelists.

$$\text{T.O.N} = \frac{A + B}{A} \qquad (4.3)$$

A = mL of sample

B = mL of odour-free water.

Table 3.1 shows the dilutions and the corresponding threshold numbers.

Table 3.2

Threshold odour Number of various dilutions

Sample volume in mL	Threshold Odour number
50	1
25	2
12.5	4
6.3	8
3.2	16
1.6	32
0.8	64
0.4	128
0.2	256

Source:(Malambo, 2014)

Care was taken that the panelists wore gloves and laboratory coats as personal protective gears. The procedure was repeated until a noticeable odour was detected. The results were negative (-) for no perceptible odour and positive (+) for perceptible odour.

The odour test panelists were selected carefully. Extreme odour sensitivity was not a requirement, but odour insensitive persons were excluded. Peripheral odour sources such as those caused by smoking, eating spicy foods, scented soaps, perfumes, and lotions before the test were avoided. The panelists were free from colds or allergies that may have otherwise affected their odour perception. The test was carried out in an odour-free environment.

3.11 Quality Control

To ensure the reliability and reproducibility of the experimental results, stringent quality control measures were observed throughout the study. All instruments and materials were carefully calibrated, standardized, and handled according to established protocols.

3.11.1 Instrument Calibration and Standardization

The pH meter was calibrated daily using standard buffer solutions (pH 4.0, 7.0, and 10.0) prior to sample analysis to guarantee accuracy. The UV-Vis spectrophotometer was standardized with blank and lactic acid standards of known concentrations to validate absorbance readings. GC-MS was pre-tested using lactic acid standard and a blank before sample analysis.

3.11.2 Sample Handling and Replication

All experiments were conducted in triplicate to minimize random error and ensure statistical reliability. Vegetable wastes were weighed using a digital analytical balance with ±0.001 g precision, while FS samples were collected and homogenized to reduce variability. Sterile glassware and equipment were used during sample preparation to avoid external microbial contamination.

3.11.3 Microbiological Analysis Controls

E. coli was selected as the indicator organism due to its sensitivity and suitability in assessing pathogen reduction in FS treatment. To prevent cross-contamination, aseptic techniques were strictly followed during serial dilution and plating. Positive and negative controls were included in culture runs to verify the reliability of microbial counts.

3.11.4 Data Verification and Statistical Validity

Raw data were recorded immediately during experimentation, and outliers were carefully checked against experimental logs to identify potential errors. Standard deviations (SD) were calculated to measure variability within replicates, while means were reported to represent central tendencies. Where applicable, Analysis of Variance (ANOVA) and Tukey's HSD statistical tests were applied to determine significant differences between treatments at a 95% confidence level.

3.11.5 Safety and Waste Management

Autoclaving was carried out for all microbial cultures and contaminated materials before disposal. Liquid and solid wastes generated from the experiments were sterilized to prevent environmental contamination.

3.12 Data Analysis

3.12.1 Statistical Analysis

The data obtained from each batch was first analysed using Excel by computing the averages of the three trials conducted per batch. The average values of the three batches were then calculated to generate the means, standard deviations, and standard errors. The means obtained were then analyzed using one-way ANOVA in order to determine if there was any significant difference in the means among the treatments.

3.12.2 Data Presentation

Data was presented in the form of tables, graphs, and charts.

CHAPTER FOUR: RESULTS AND DISCUSSION

4.1 Introduction

This chapter presents the information processed from the data collected during the study. Data was collected through experiments and was then analyzed based on the objectives.

The characteristics of the FS collected from the study area can be seen in Table 4.1.

Table 4.1

Initial FS characteristics.

Parameters	**Unit**	**Initial FS characteristics**
		Values
pH		5.63
Total solids	g/L	16.30
Volatile solids	g/L	4.3×10^4
E. coli	CFU/mL	1.02×10^4

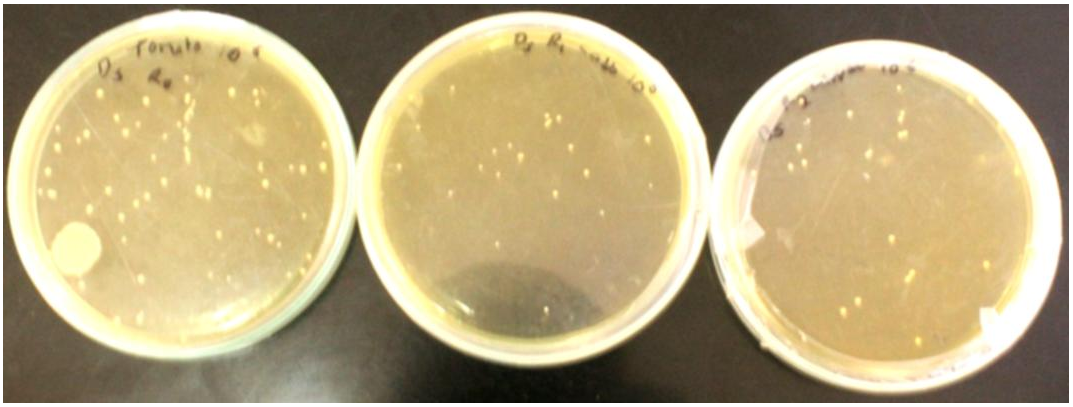
Source: (Researcher, 2024)

4.1.1 Growth of LAB in MRS medium

The presence of LAB in the fermented substrates was confirmed by plating on MRS medium (a selective medium for LAB) and observing for colony growth, followed by a confirmatory biochemical test. There was colony growth on plates containing tomato, cabbage, and carrot wastes, as shown in Figure 4.1, which indicated the presence of lactic acid bacteria.

Figure 4.1

Sampled plates showing the growth of lactic acid bacteria on MRS agar



Source: (Researcher, 2024)

4.1.2 Biochemical test for lactic acid bacteria

The catalase test for the fermented vegetable waste substrates is shown in Table 4.2. The test was negative in all the fermented samples, indicating the absence of aerobic bacteria. There was no bubbling upon the addition of hydrogen peroxide, indicating the absence of catalase activity; LAB usually lack the catalase enzyme (Bryukhanov *et al*., 2022). This finding is crucial as it stresses the anaerobic conditions precisely maintained during the fermentation process, which are vital for the multiplication and function of lactic acid bacteria responsible for lactic acid generation. The negative results from the catalase test further validate the experimental conditions, ensuring that the observed lactic acid production primarily stems from the activity of lactic acid bacteria under anaerobic conditions, thereby supporting the study's findings.

Table 4.2

Catalase biochemical test

Catalase Biochemical Test	
Sample	**Test**
Carrot waste	-
Tomato waste	-
Cabbage waste	-

Note: *(-) sign indicates catalase-negative*

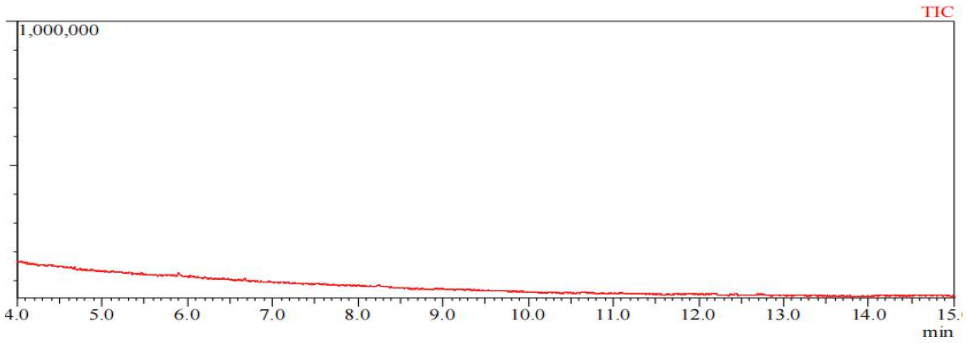
Source: (Researcher, 2024)

4.1.3: Qualitative Determination of Vegetable Waste Derived Lactic Acid using GC-MS

GC-MS was used to qualitatively determine the presence of lactic acid in the fermented vegetable wastes. The results in Figure 4.2 show the chromatogram for the blank. Figure 4.3 show the detection of the standard analytical grade lactic acid in its esterified form butyl lactate derived from esterification of lactic acid and butanol.

Figure 4.2

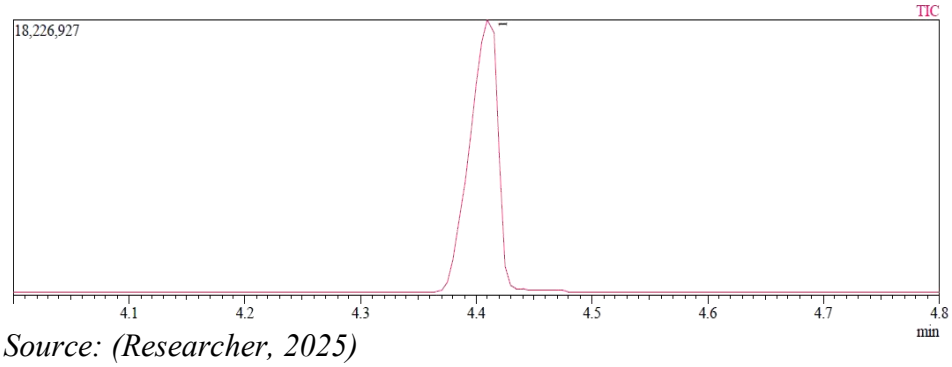
Chromatogram of blank



Source: (Researcher, 2025)

Figure 4.3

Chromatogram of lactic acid standard derivative (butyl lactate)

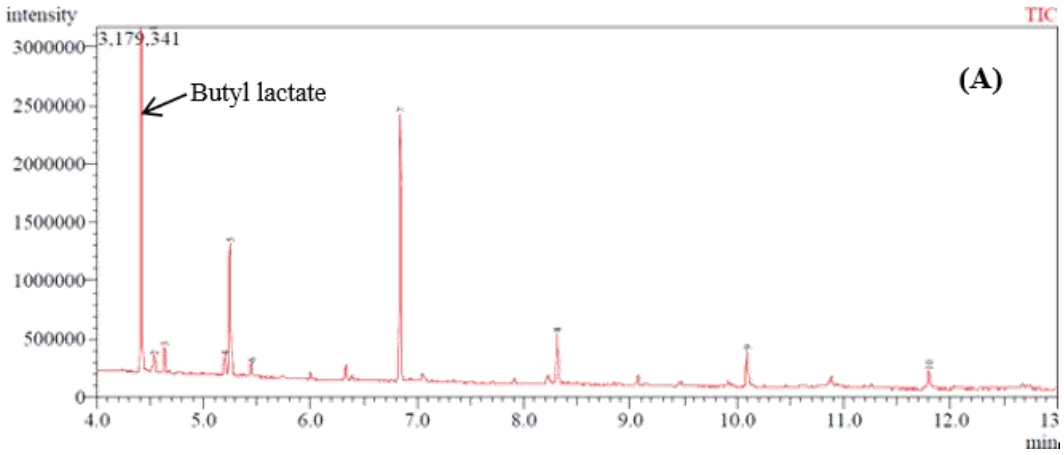


Source: (Researcher, 2025)

GC–MS chromatograms of fermented cabbage, carrot and tomato wastes are shown in Figures 4.4 (A), (B), & (C). These results confirm the production of lactic acid in the fermented vegetable wastes evidenced by the detection of butyl lactate in all the fermented samples. Additionally, low-abundance butane derivatives were detected, which are likely linked to derivatization reactions or from background matrix components.

Figure 4.4 (A)

GC-MS chromatogram of cabbage fermented wastes

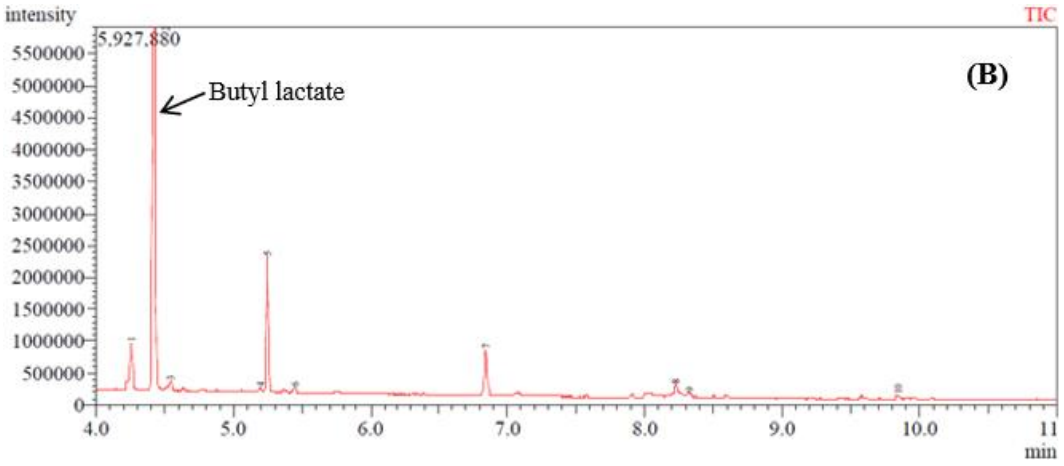


Note: (1) Butyl lactate, (2) 1-Butoxy(methoxy)methoxy)butane, (3) Boronic acid, ethyl-,diethylester, (4) Butane, 1,1'-[ethylidenebis(oxy)]bis-, (5) Dibutoxy(dimethyl)silane, (6) Butane, 1,1'-[(1-methylethylidene)bis(oxy)bis, (7) Butane, 1,1-dibutoxy, (8) Tetradecane, (9) Hexadecane, (10) Heptadecane

Source: (Researcher,2025)

Figure 4.4 (B)

GC-MS chromatogram of carrot fermented wastes.

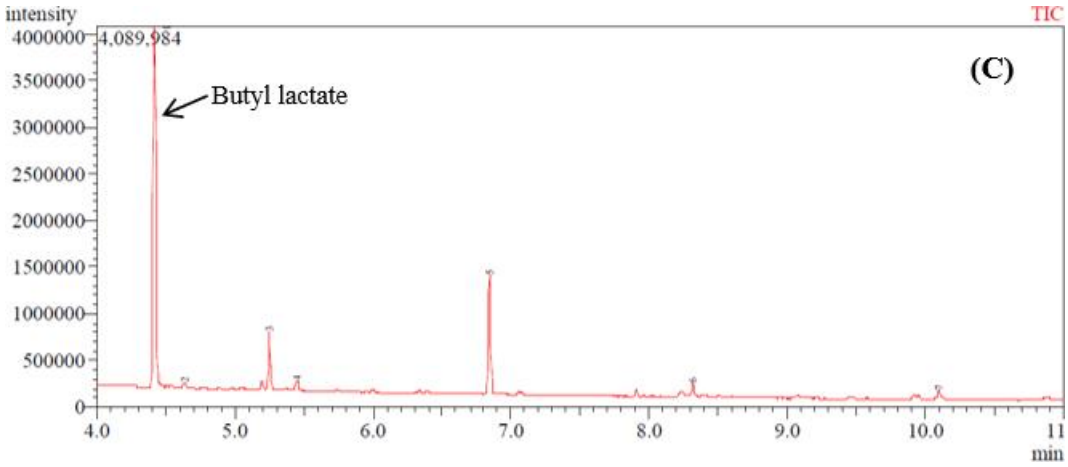


Note: (1) Butanoic acid, butyl ester (2) Butyl lactate (3) 1-Butoxy(methoxy)methoxy)butane, (4) Butane, 1,1'-[ethylidenebis(oxy)]bis-, (5) Dibutoxy(dimethyl)silane, (6) Butane, 1,1'-[(1-methylethylidene)bis(oxy)bis, (7) Butane, 1,1-dibutoxy, (8) Sulphuric acid dibutyl ester, (9) 1,1,3,3,5,5-Hexamethyltrisiloxane, (10) Butanedioic, dibutyl ester

Source: (Researcher,2025)

Figure 4.4 (C)

GC-MS chromatogram of tomato fermented wastes.



Note: (1) Butyl lactate, (2) Boronic acid, (3) Dibutoxy(dimethyl)silane, (4) Butane, 1,1'-[ethylidenebis(oxy)]bis- (5) Butane, 1,1-dibutoxy-, (6) Tetradecane, and (7) Hexadecane

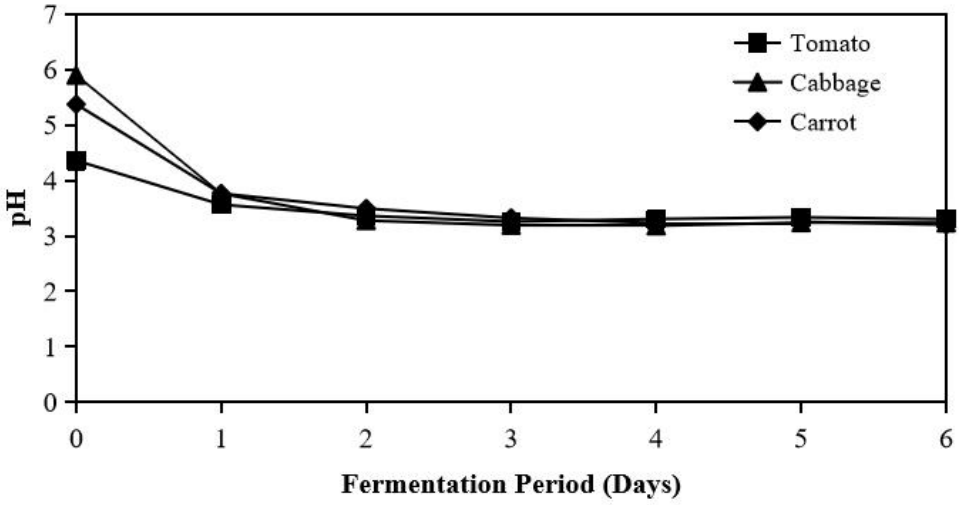
Source: (Researcher, 2025)

4.1.4 pH of Fermeted Vegetable Wastes

The pH variation for carrot, tomato, and cabbage vegetable wastes fermented for 6 days at 37 ℃ is illustrated in Figure 4.5. The pH of the fermented waste decreased in all the vegetable wastes after the 6-day fermentation period. A decrease in the pH levels from 5.37 ± 0.14, 4.36 ± 0.15, and 5.90 ± 0.05 to 3.20 ± 0.16, 3.30 ± 0.07, and 3.23 ± 0.10 for carrot, tomato, and cabbage waste, respectively, showed that fermentation had occurred.

Figure 4.5

pH changes over the fermentation period of the vegetable wastes.



Source: (Researcher, 2024)

pH variation is one of the most crucial parameters that must be observed during fermentation. The initial pH was 5.90±0.05, 5.37±0.14, and 4.36±0.15 and decreased to an average value of 3.23±0.10, 3.20±0.16, and 3.30±0.07 on day 6 for cabbage, carrot and tomato wastes respectively. These conditions favor the rapid growth of LAB during the fermentation process (Supplementary Figure 1). During this process, LAB convert sugars present in the vegetables into lactic acid while causing a decline in the pH (Jabłońska-Ryś *et al*., 2022; Kaur *et al*., 2019). The initial rapid decrease in pH observed in the initial three days for all three vegetables suggests rapid fermentation activity, where LAB quickly utilizes the abundant fermentable sugars to produce energy while producing lactic acid as a metabolite, resulting in acidification (Supplementary Table 1). After which, the pH levels stabilize, indicating that the fermentation process has stopped due to depletion of fermentable sugars (Deshwal *et al*., 2021).

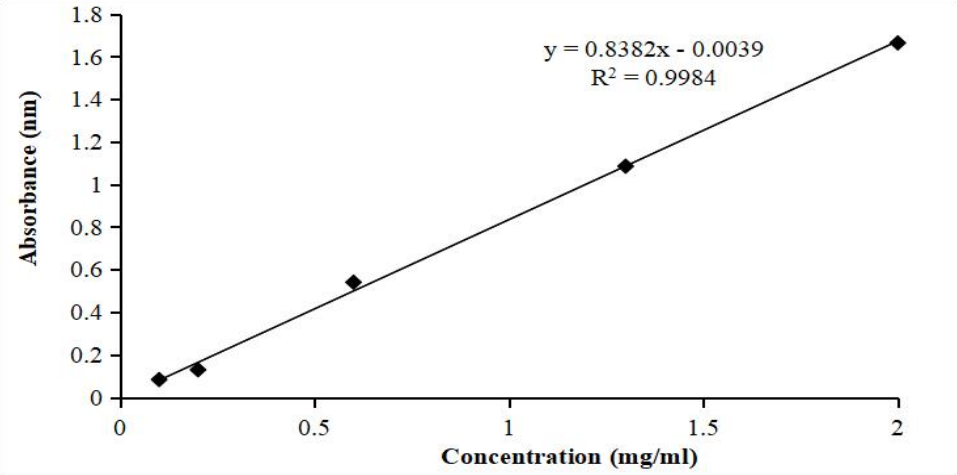
These results suggest that most of the fermentation process occurred earlier in the process, followed by a stabilization phase after three days, which is the period taken to achieve a stable pH. These results agree with findings earlier reported by (Anderson *et al.,* 2015; Getaneh *et al.,* 2021), which reported three days as the period during which the pH appears to stabilize.

4.1.5 Concentration of lactic acid the fermented vegetables

A calibration curve was constructed using the pure standard of lactic acid. The calibration curve was used to calculate the concentration of lactic acid in the fermented vegetable wastes. Figure 4.6 shows the calibration curve for the quantification of lactic acid. The curve plots absorbance (y-axis) against concentration (x-axis) in milligrams per milliliter (mg/mL).

Figure 4.6

Calibration curve of standard lactic acid



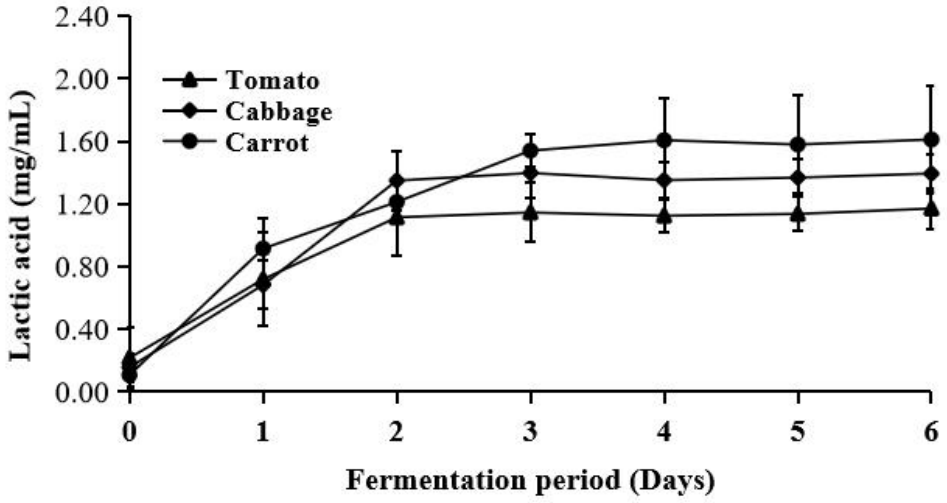
Source: (Researcher, 2024)

Figure 4.7 shows the concentration of lactic acid in the three vegetable waste substrates over a six day period of fermentation at 37 ℃. During the initial 3 days of fermentation, the three vegetable wastes showed a rapid increase in lactic acid concentration, indicating the onset of

fermentation. This is characterized by the activity of lactic acid bacteria that start converting sugars into lactic acid. The increase of lactic acid was similar in all the vegetables reaching a plateau after 3-days.

Figure 4.7

Lactic acid concentration in fermented vegetable wastes during fermentation.



Source: (Researcher, 2024)

The concentrations began to stabilize from the 3rd day of fermentation, indicating that the fermentation process had reached equilibrium. The three vegetable wastes had a final lactic acid concentration of 1.39 ± 0.09 mg/mL, 1.61 ± 0.34 mg/mL, and 1.17 ± 0.13 mg/mL for cabbage, carrot, and tomato respectively.

Statistical analysis was also conducted to determine the significance of lactic acid production across different substrate types. Analysis of variance (ANOVA) was employed to compare the mean lactic acid concentrations among carrot, tomato, and cabbage waste samples after six days. The results indicated no statistical significant difference since $p > 0.05$ ($p = 0.557$) in lactic acid production among the substrates. The data suggest that all three substrates underwent a rapid initial fermentation phase, followed by a slower increase

and eventual stabilization of lactic acid concentrations. The variations in the lactic acid levels among the vegetables could be due to differences in their natural sugar content, microbial communities, and physical structure affecting the fermentation dynamics (Xu *et al*., 2024).

The trends suggest successful lactic acid fermentation for all three vegetables, with slight differences in the extent and timing of lactic acid production. The graph shows that the three substrates attained maximum lacto fermentation within the initial three days, after which the lactic acid production reached equilibrium.

4.2 Faecal Sludge Treatment with Lactic Acid Derived From Selected Vegetable Waste

Cabbage waste was selected as the source of lactic acid among the vegetable wastes due to its local abundance in the study area. FS treatment was performed in four reactors with varying FS to lactic acid ratios (1:1, 1:0.5, 1:0.35) and a control, with *E. coli* as the indicator organism.

4.2.1 pH variation during treatment of FS with different ratios of cabbage waste derived lactic acid

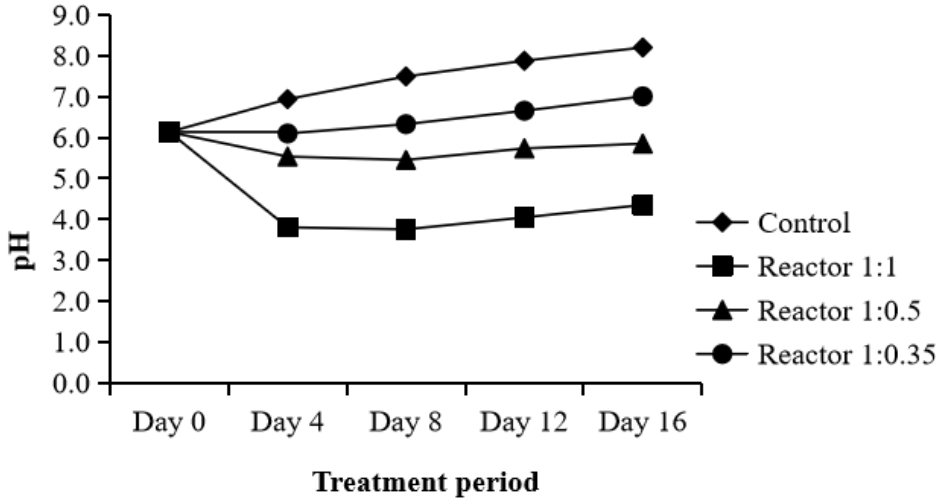
A comparative analysis of pH in the reactors is seen in Figure 4.8. The control showed a steady increase in pH from near neutral 6.13 to moderately alkaline at 8.20. This increase could be attributed to natural biochemical processes in the FS, such as microbial activity leading to urea breakdown and ammonia production (Chang *et al*., 2015).

In reactor 1:1, there was a rapid decrease in pH to acidic levels by Day 4 from the initial pH 6.13 to 4.35, with a slight increase thereafter but the pH remained below pH 5.0**.** The slight increase after Day 4 suggests some buffering capacity or metabolic changes in the sludge

that partially counteracted the acidity. These results are in agreement with (Getaneh *et al*., 2021), which reported a decrease in pH by day five of treatment.

Figure 4.8

pH of FS treated with varying ratios of cabbage waste derived lactic acid



Source: (Researcher, 2024)

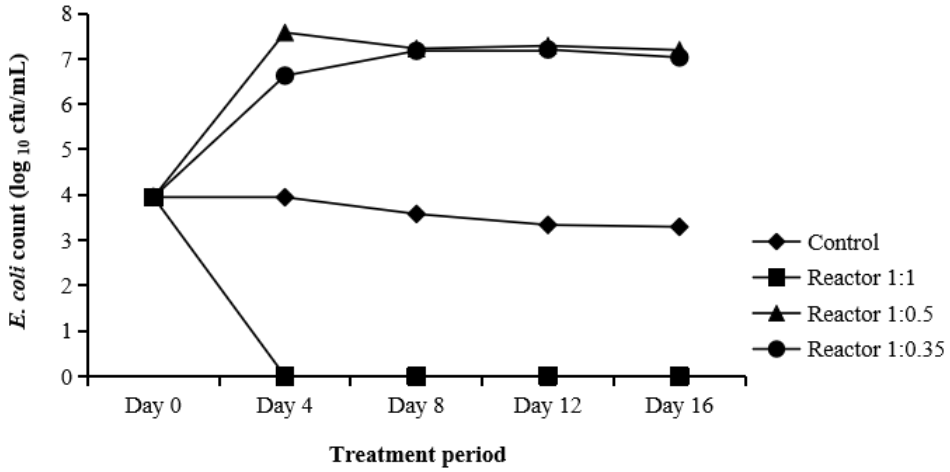
In reactor 1:0.5, the initial pH (Day 0) was 6.13 while the final pH was 5.85. Reactor 1:0.5 exhibited a moderate decrease in pH compared to reactor 1:1. This suggests that the lower lactic acid concentration allowed for some neutralizing processes within the sludge. Reactor 1:0.35 showed the least change in pH, with a minor depression followed by a rise to slightly basic levels. This suggests that the dilution ratio resulted in a low pH. The pH level in the 1:1 reactor is considered to closely match the ideal conditions for FS acidification, as noted by Odey *et al*. (2018), who stated that lactic acid treatment requires a final pH of 4 for FS hygienization. These results suggest that lactic acid concentration significantly influenced the pH of FS.

4.2.2 Levels of *E. coli* in FS treated with different ratios of cabbage waste derived lactic acid

E. coli concentration (cfu/mL) results over a treatment period of 16 days for a FS sample treated with 1:1, 1:0.5, and 1:0.35 FS to lactic acid, and a control is illustrated in Figure 4.9.

Figure 4.9

Levels of E. coli in FS treated with varying ratios of cabbage waste derived lactic acid



Source: (Researcher, 2024)

According to the results there was reduction of *E. coli* in reactor 1:1 whereas, the levels of *E. coli* remained high in reactors 1:0.5, 1:0.35 and the control. From day four, *E. coli* was not detected in reactor 1:1. On the other hand, the levels of *E.coli* slightly decreased in the control, indicating that in the control setup, conditions were not conducive to *E. coli* survival or growth over time due to factors such as high pH or depletion of nutrients, thus resulting in a natural die-off of *E. coli* (Malambo, 2014; Van den Bergh *et al*., 2016). Reactor 1:1 was highly effective in eliminating *E. coli*. This aligns with findings indicating that low pH inhibits *E. coli* growth, as acidic pH levels fall outside the optimal range for *E. coli*'s growth, likely leading to its inactivation. Other research suggests that a pH range of 3.51 to 4.2 can effectively eliminate various pathogens (Odey et. al, 2018, Rodrigues *et al*., 2020). The

antimicrobial property of lactic acid could have contributed to the limited growth of *E. coli* (Anderson *et al*., 2015; Getaneh *et al*., 2021).

In reactor 1:0.5, the *E. coli* concentration increased significantly by day 4. It remained relatively high, fluctuating slightly from day 8 to day 16. This suggests that in reactor 1:0.5, the growth conditions of *E. coli* were promoted. In reactor 1:0.35, similar to reactor 1:0.5, there was a significant increase in *E. coli* levels by Day 4. The concentration remained high from day 8 to day 16 of the treatment. The conditions in Reactor 1:0.35 also appeared to support *E. coli* growth.

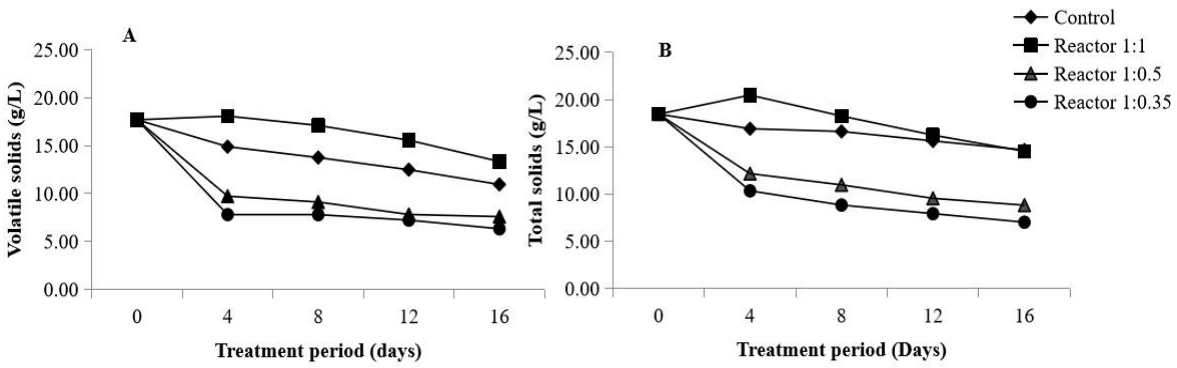
These findings are in agreement with (Malambo, 2014), who reported an initial increase in *E. coli* count for the treatment reactors within the initial four days of treatment. The growth might have been supported by the utilization of different carbon sources such as glucose, lactose, and galactose that might have been available in the lactic acid extract (Cutrim *et al*., 2016). These findings demonstrate that certain treatments had a greater impact on lowering *E. coli* concentrations than others. These findings further suggest that reactors 1:0.5 and 1:0.35 conditions did not contain the threshold lactic acid amount or acidification levels required to eliminate or significantly lower the *E. coli* levels to acceptable levels. Reactor 1:1 had the best conditions for *E. coli* elimination.

4.2.3 Trend of VS of FS treated with varying ratios of cabbage waste derived lactic acid

Volatile solids are the organic matter content in the sludge, which can be decomposed through microbial activity (Chynoweth & Pullammanappallil, 2020). Monitoring the reduction in VS provides insight into the efficiency of biodegradation processes. Figure 4.10 (**A**) shows the trend of volatile solids during the 16-day treatment period of FS with different ratios of cabbage derived lactic acid.

Figure 4.10

VS and TS of FS treated with varying ratios of cabbage waste derived lactic acid



*Note: (**A**) Volatile Solids (VS) and (**B**) Total Solids*

Source: (Researcher, 2024)

The results revealed a decrease in VS in all the reactors. The control showed a gradual reduction in VS concentration over the 16 days, from 17.70 g/L to 10.95 g/L. This reduction is expected due to natural microbial activity and degradation processes occurring in the sludge without any additional treatments. At the same time, in reactor 1:1, the VS concentration initially increases slightly from 17.70 g/L to 18.08 g/L by Day 4. This was attributed to low initial microbial activity, leading to low breakdown of complex organic materials. After Day 4, there is a gradual reduction in VS, reaching 13.35 g/L by Day 16. This pattern indicated a slower but steady decomposition process compared to other reactors. Reactor 1:0.5 showed a significant and rapid reduction in VS concentration from 17.70 g/L to 9.73 g/L within the first four days, followed by a more gradual decline to 7.59 g/L by Day 16. This rapid initial decrease suggested a highly efficient microbial degradation process due to an optimal environment for microbial activity and organic matter breakdown.

Reactor 1:0.35, similar to reactor 1:0.5, demonstrated a sharp decline in VS from 17.70 g/L to 7.81 g/L by Day 4, indicating a highly effective reduction in organic matter. The concentration stabilized around 7.80 g/L from Day 4 to Day 8 and decreased slightly to 6.32 g/L by Day 16. The sharp initial drop followed by slower degradation suggested that the conditions in reactor 1:0.35 maximized the microbial efficiency. The high microbial load in reactor 1:0.5 and 1:035, as shown in Figure 4.9, led to the rapid decrease in VS due to the increased microbial activity.

The efficiency of VS reduction in FS treatment is critical for reducing the sludge's volume and potential environmental impact (Zewde *et al*., 2021). Effective treatment results in lower organic load, making the remaining sludge more stable and less odorous (Demirbas *et al*., 2017). High microbial activity and optimal environmental conditions, such as appropriate moisture content, temperature, and pH, facilitate the breakdown of organic matter in FS (Cofie *et al*., 2016). The results highlight the effectiveness of different reactor setups in reducing volatile solids in FS. Reactors with a lower concentration of lactic acid, i.e., 1:0.5 and 1:0.35, displayed more rapid initial VS reduction. This can be attributed partially by the high pH in the reactors, indicating more efficient microbial degradation.

4.2.4 Trend of TS of FS treated with varying ratios of cabbage waste derived lactic acid

Total solids refer to the sum of dissolved and suspended solids in a given volume of liquid, which includes both volatile and non-volatile components. Analysis of total solids in FS treatment provides insights into the overall reduction of solids and the efficiency of the treatment process. Figure 4.10 (B) shows the TS for the control group and three different reactors (1:1, 1:0.5, and 1:0.35) over a period of 16 days. The TS were measured at intervals of 0, 4, 8, 12, and 16 days.

The control TS concentration gradually decreased from 18.47 g/L to 14.67 g/L over the 16-day treatment period. The steady decline indicated natural microbial degradation of solids without additional treatment. The reduction is consistent with typical sludge stabilization processes observed in untreated or minimally treated FS (Strande & Brdjanovic, 2014).

In reactor 1:1, TS concentration initially increased from 18.47 g/L to 20.48 g/L by Day 4. Following this peak, the TS gradually decreased to 14.53 g/L by Day 16, indicating ongoing microbial degradation. The spike at Day 4 suggests temporary microbial activity or aggregation of solids due to lactic acid's inhibitory effects on initial microbial metabolism (Paul & Liu, 2012).

The data indicate that the treatment reactors (1:0.5 and 1:0.35) were significantly more effective in reducing total solids compared to the control and reactor 1:1. The reactor 1:0.35 consistently showed the greatest reduction in TS, demonstrating its potential for optimizing FS treatment.

The steady decline in TS suggests efficient biodegradation, likely due to a balanced pH and microbial synergy. This is supported by previous findings indicating that low lactic acid concentrations help minimize microbial inhibition, thereby promoting robust microbial activity (Cofie *et al*., 2016). These results agree with those of Odey *et al*. (2018), who reported a final TS of 33% in the 1:1 (FS: lactic acid) reactor, which was higher than the control (13%) and the 1:2 reactor (19.2%).

4.3 Odour Levels of FS treated with varying ratios of cabbage waste derived lactic acid

High Threshold Odour Number (T.O.N) levels indicate a significant presence of odour, suggesting the need for effective odour management strategies, while a low TON indicates effective odour treatment. Table 4.3 shows the mean T.O.N levels measured during the FS

treatment process with different ratios of lactic acid derived from cabbage waste. The data includes the initial T.O.N level and the levels observed in the control and the three different reactors (1:1, 1:0.5, and 1:0.35) after the 16-day treatment period.

Table 4.3

TON levels of FS treated with varying ratios of cabbage waste derived lactic acid

	Mean level T.O.N
Initial FS	37.33
Control	25
Reactor 1:1	7.33
Reactor 1:0.5	42.67
Reactor 1:0.35	26.67

Source: (Researcher, 2024)

The initial FS odour level was relatively high (37.33 TON). This acted as the reference point measurement against which the other treatments were compared. The control condition demonstrates a decrease in odour levels to 25.00 TON. This reduction from the initial condition indicates some level of odour reduction. Reactor 1:1 showed a significant decrease in odour levels, reaching 7.33 TON, the lowest among all reactors. This suggests that the 1:1 ratio was highly effective in controlling and reducing odour. The odour reduction was likely due to the low pH and the high concentration of lactic acid in the reactor, which inhibited the growth of bacteria that degrade proteins, such as sulfur-reducing bacteria. These bacteria typically produce odorous compounds, including dimethyl trisulfide, hydrogen sulfide (H_2S), dimethyl disulfide, methyl mercaptan, and dimethyl sulfide (Choudhury *et al*., 2024;

Newman, 2023). Low pH in the range 3-4.5 has been reported to reduce urea hydrolysis to ammonia by inactivating urease-producing bacteria (Ray *et al*., 2018).

The ratio 1:0.5 reactor exhibited an increased odour level to 42.67 TON, which was even higher than the initial condition. This indicated that this ratio was ineffective for odour control and might even contribute to the growth of odour-causing bacteria. Possible reasons could have been due to the low lactic acid concentration and the relatively high pH that allowed *E. coli* and other microorganisms to persist at high levels throughout the treatment period (Anderson *et al*., 2015; Andreev *et al*., 2017). Continued microbial activity led to the breakdown of organic material, thereby producing large amounts of odour-causing compounds (Zewde *et al*., 2021).

The ratio 1:0.35 reactor indicated an odour level of 26.67 TON, which was lower than the initial condition but higher than the control. This indicated a moderate level of effectiveness in odour reduction. While it was better than the initial condition, it did not perform as well as the control or the ratio 1:1 reactor. Similar results were reported by Anderson *et al.* (2015); Getaneh *et al.* (2021); Odey *et al.* (2018), who reported decreased odour levels in higher lactic acid concentrations and increased odour levels in lower lactic acid concentrations.

CHAPTER FIVE: CONCLUSIONS, RECOMMENDATIONS AND PUBLICATION

5.1 Introduction

In this chapter, the conclusion and recommendations in relation to the study objectives are given.

5.2 Conclusion

This research aimed to determine whether carrot, tomato, and cabbage vegetable wastes could serve as an effective substrate for lactic acid production, which could be used for effective treatment of FS treatment. It was noted that all three vegetable wastes underwent significant fermentation as evidenced by the increase in lactic acid concentration and the corresponding decrease in pH by the third day. The presence of LAB also confirmed the lacto-fermentation process. This shows that carrot, cabbage, and tomato wastes can be utilized in the production of lactic acid though in different concentrations attributed to the composition of sugars in the vegetable wastes.

The 1:1 ratio of FS to lactic acid achieved complete elimination of *E. coli* within four days, demonstrating greater pathogen inactivation compared to lower ratios (1:0.5, 1:0.35) and the control. It was noted that in higher lactic acid concentrations (1:1), volatile solids were not reduced due to microbial inhibition, while lower ratios (1:0.5, 1:0.35) showed rapid VS and TS degradation, indicating enhanced microbial activity at lower acidity.

The analysis of TON levels across different reactors revealed significant variations in odour management effectiveness. The ratio 1:1 reactor demonstrated the most effective odour control, achieving the lowest TON level of 7.33, indicating that this ratio optimizes conditions for reducing odour. On the other hand, the ratio 1:0.5 reactor is the least effective, with an increased odour level of 42.67, suggesting that this ratio was unsuitable for odour

management. The control and ratio 1:0.35 reactors show moderate effectiveness, with TON levels of 25.00 and 26.67, respectively.

These findings suggest that careful optimization of treatment ratios is crucial for effective odour management in FS treatment. The ratio 1:1 appeared to be the best option among those tested, while the ratio 1:0.5 should be avoided due to its poor performance in controlling odour. Further research could explore the mechanisms behind the varying effectiveness of these ratios and investigate additional ratios or treatment conditions to enhance odour control in FS treatment systems.

5.3 Recommendations

Based on the findings of this research, several recommendations can be made to enhance the effectiveness of lactic acid production from vegetable wastes and its application in FS treatment, particularly focusing on odour management and pathogen reduction:

Further studies can be conducted to optimize the precise concentration of lactic acid that maximizes pathogen reduction as well as prevents the growth of *E. coli* . Investigating other vegetable wastes or agricultural by-products that might offer similar or better efficiency in lactic acid production. This could diversify the options for substrate selection and enhance the sustainability of the process. Optimization of the fermentation conditions such as temperature, pH, use of inoculants, and nutrient availability in order to improve the overall yield and purity of lactic acid. Combining lactic acid treatment with other microbial or chemical treatments can synergistically enhance pathogen reduction. Finally, to upscale the research to field trials to investigate the efficacy of lactic acid derived from vegetables in treating FS.

5.4 Publication

Mwebia, T. M., Mbogoh, E., Muthuri, G. G., & Mugo Mwenda, C. N. (2025). Evaluation of vegetable waste-derived lactic acid from fermentation of selected vegetable wastes for faecal sludge treatment. *African Journal of Science, Technology and Social Sciences*, *4*(2), PAS 93–103. https://doi.org/10.58506/ajstss.v4i2.336

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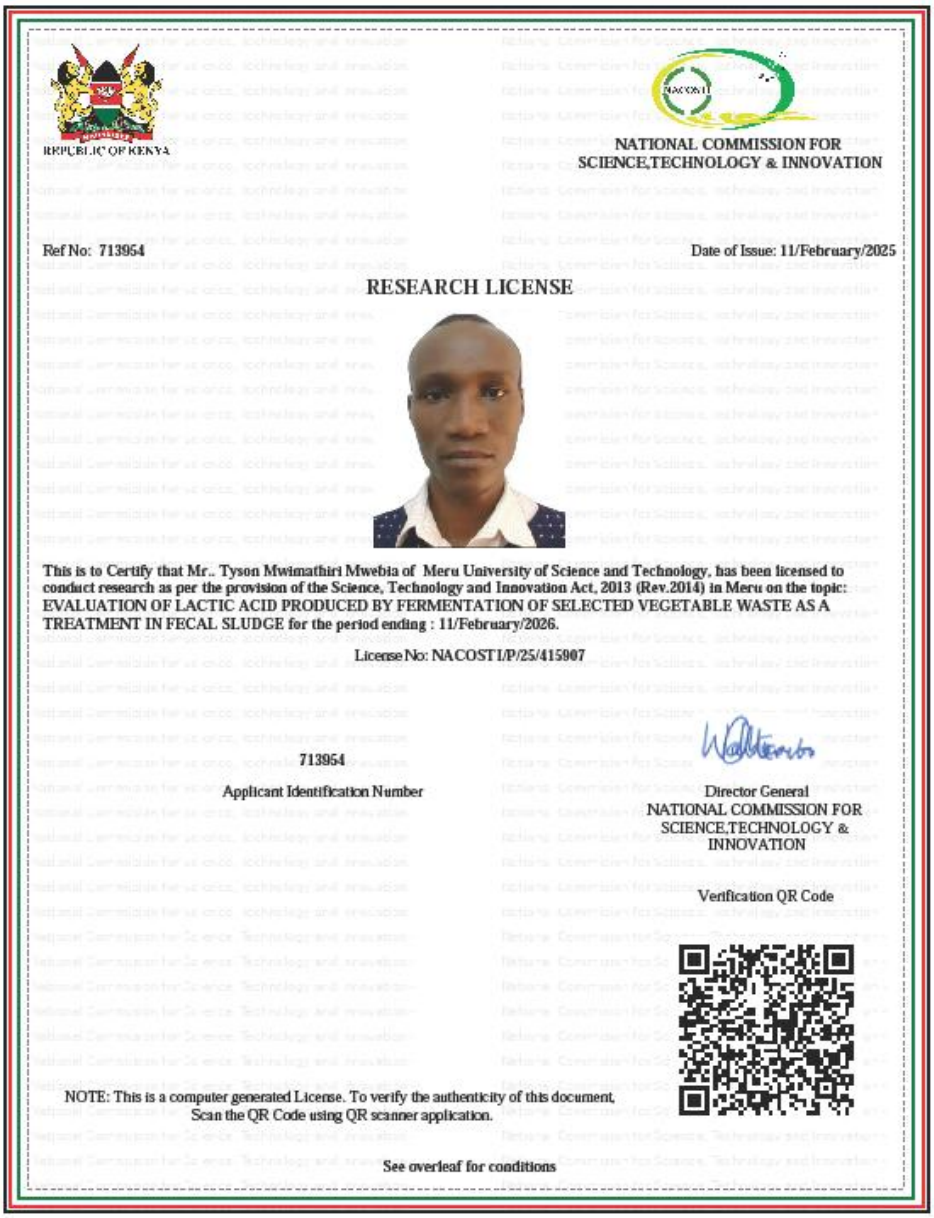
APPENDICES

Appendix A: NACOSTI Research License



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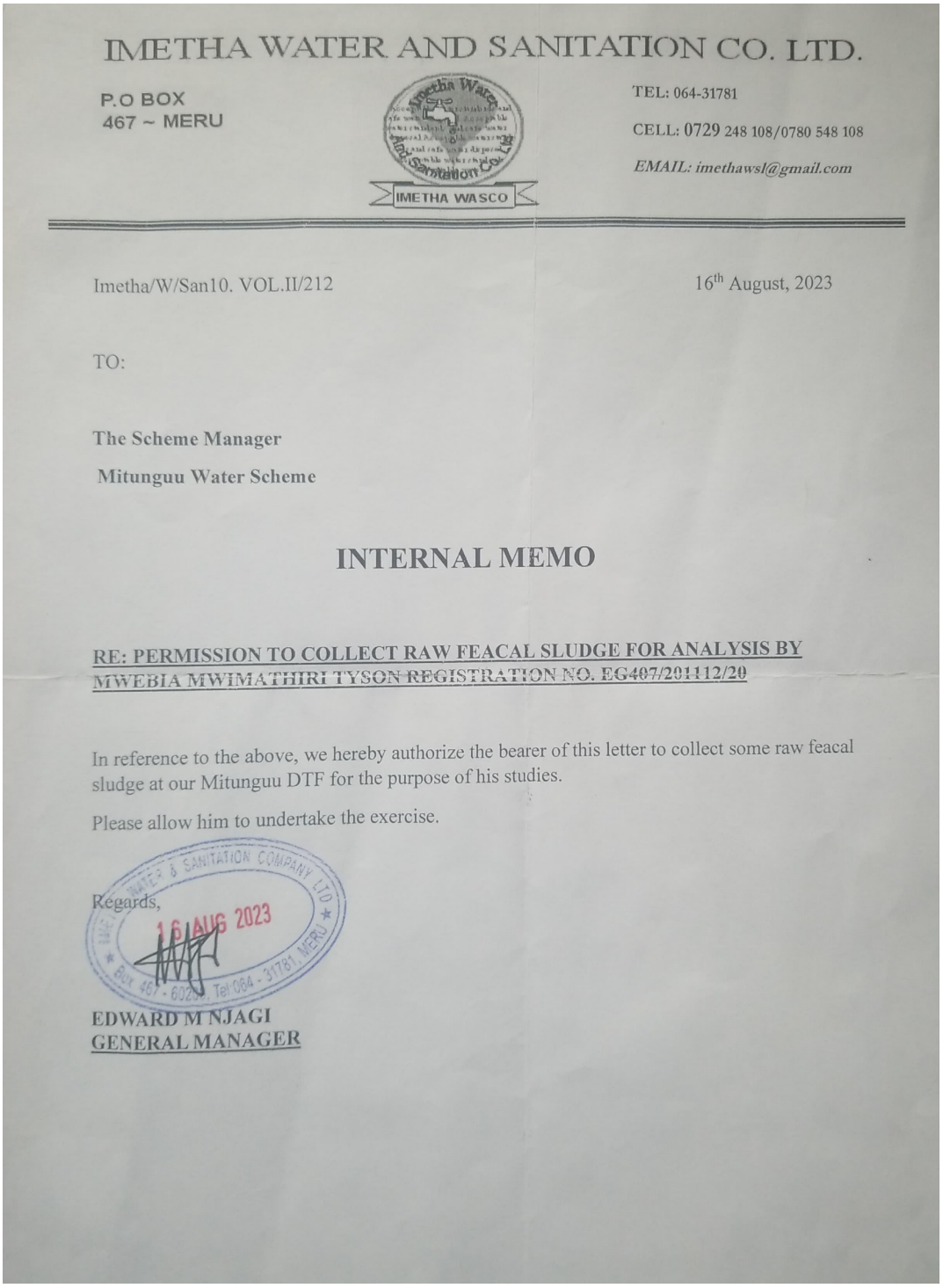

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The Scheme Manager

Mitunguu Water Scheme

INTERNAL MEMO

RE: PERMISSION TO COLLECT RAW FEACAL SLUDGE FOR ANALYSIS BY MWEBIA MWIMATHIRI TYSON REGISTRATION NO. EG407/201112/20

In reference to the above, we hereby authorize the bearer of this letter to collect some raw feacal sludge at our Mitunguu DTF for the purpose of his studies.

Please allow him to undertake the exercise.

Regards,

16 AUG 2023

EDWARD M NJAGI
GENERAL MANAGER

Appendix C: Publication

AFRICAN JOURNAL OF SCIENCE, TECHNOLOGY AND SOCIAL SCIENCES. 4 (2) 2025, PAS 93-103 OPEN ACCESS

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Evaluation of lactic acid from selected vegetable wastes for Faecal sludge treatment

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ABSTRACT

Effective faecal sludge treatment is essential for pathogen reduction and compliance to World Health Organization standards for disposal or reuse. In developing countries, these treatments are frequently absent, ineffective, or immensely expensive. This study used an experimental research design to investigate the efficacy of lactic acid derived from selected vegetable wastes for the treatment of FS collected from an on-site sanitation system. Equal quantities of fresh cabbage, tomato, and carrot wastes were collected, pre-treated, and subjected to lacto-fermentation at 37 °C for six days. Daily monitoring of pH and lactic acid concentrations was performed using an electrode pH meter and a UV-Vis spectrophotometer, respectively.gas chromatography-mass spectrometry (GC-MS) was used to identify lactic acid in the fermented vegetable wastes through a derivatization reaction. FS treatment was performed in four reactors with varying lactic acid ratios (1:1, 1:0.5, 1:0.35), and a control, with *Escherichia coli* (*E. coli*) as the pathogen indicator for faecal contamination. *E. coli*, total solids, volatile solids, and odor levels were monitored over a 16-day treatment period in the four reactors. The fermentation process was deemed successful as indicated by the decrease in pH levels. Successful GC-MS detection of butyl lactate, the derivatized form of lactic acid was observed in the fermented vegetable wastes. Lactic acid concentrations post fermentation were 1.39 ± 0.09 mg/mL, 1.17 ± 0.13 mg/mL, and 1.61 ± 0.34 mg/mL for cabbage, tomato, and carrot wastes respectively. Statistical analysis revealed no significant differences ($p > 0.05$) in lactic acid concentrations among the vegetable wastes on day six of fermentation. Consequently, cabbage waste-derived lactic acid was selected for subsequent experiments in addition to the local abundance of cabbage waste. Total solids and volatile solids decreased across all the reactors over time. From day four, *E. coli* was undetectable in reactor 1:1; which also showed the highest reduction in odor levels. Therefore, reactor 1:1 treatment produced the optimal *E. coli* elimination and odor reduction conditions. This study demonstrates the potential of cabbage waste derived lactic acid for effective FS treatment.

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Appendix D: Plagiarism Result

DrillBit

The Report is Generated by DrillBit Plagiarism Detection Software

Submission Information

Author Name	MWEBIA TYSON MWIMATHIRI
Title	EVALUATION OF LACTIC ACID PRODUCED BY FERMENTATION OF SELECTED VEGETABLE WASTE AS A TREATMENT IN FECAL SLUDGE
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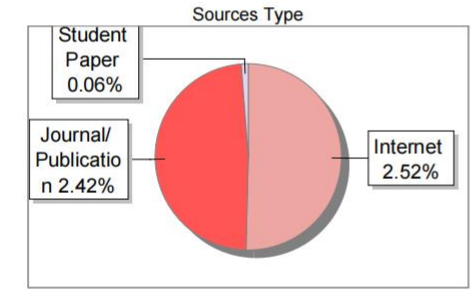

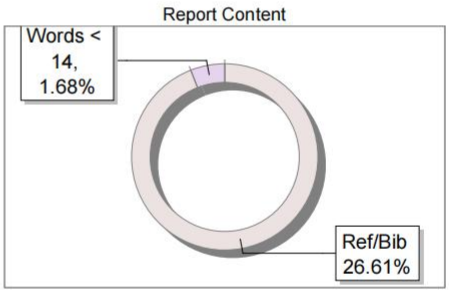

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Appendix E: Supplementary Data

Supplementary Table 1

Total Titrable Lactic Acid in fermented vegetable wastes

Vegetable waste Total Titrable Lactic Acid (mg/ml)

	Carrot		Tomato		Cabbage	
Days	**Mean**	**Std dev**	**Mean**	**Std dev**	**Mean**	**Stad dev**
0	0.92	0.14	1.33	0.12	1.13	0.08
6	8.22	0.49	7.51	0.35	7.95	0.39

Supplementary Figure 1

LAB count across the fermentation period of the vegetable wastes