RETENTION OF NITROGEN FROM ARTIFICIAL URINE USING PINE BARK AND POTATO WASTE BIOCHAR AND ASH AND THE FERTILISER VALUE OF THE PRODUCTS

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PREFACE

The research contained in this thesis was completed by Samukelisiwe Pinky Vilakazi in KwaZulu-Natal, Pietermaritzburg, Republic of South Africa. Experimental work was carried out while registered in the School of Agricultural, Earth and Environmental Sciences, University of KwaZulu-Natal, Pietermaritzburg, under the supervision of Professor Pardon Muchaonyerwa.

This thesis, submitted for the degree of Doctor of Philosophy in the College of Agriculture, Engineering and Science, University of Kwa-Zulu, School of Agricultural, Earth and Environmental Sciences, Pietermaritzburg campus, represents original work by the author and has not otherwise been submitted in any form for any degree or diploma to any other University. Where use has been made of the work of others, it is duly acknowledged in the text.

[redacted]

Signed: Samukelisiwe P. Vilakazi

Date: 14/07/2025

I certify that the above statement is correct and as the candidate's supervisor I have approved this thesis for submission.

[redacted]

Signed: Prof Pardon Muchaonyerwa (*Supervisor*)

Date: 14th July 2025

[redacted]

Signed: Prof A.O Odindo (Co-*Supervisor*)

Date: 14th July 2025

DECLARATION 1: PLAGIARISM

I, Samukelisiwe P. Vilakazi, declare that

1. The research reported in this thesis, except where otherwise indicated, is my original research.

2. This thesis has not been submitted for any degree or examination at any other university.

3. This thesis does not contain other persons' data, pictures, graphs or other information, unless specifically acknowledged as being sourced from other persons.

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DECLARATION 2: PUBLICATIONS/PRESENTATIONS

One manuscript has been published in the journal of Scientific Report. Manuscripts from other chapters are "under review"

Chapter 5

Nitrogen and phosphorus release from dehydration products of urine mixed with contrasting feedstocks, biochar types and ash. Published in **Journal of Scientific Report**, https://doi.org/10.1038/s41598-025-23287-2

Chapter 2

Effect of Crop Residue Feedstock, Biochar, and ash on Nitrogen and Phosphorous Recovery, and Availability: A systematic review. (Submitted to the Journal of Biomass Conversion and Biorefinery, manuscript ID: BCAB-D-25-01871).

Chapter 3

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[redacted]

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ABSTRACT

Inadequate sanitation in rural and peri-urban areas contributes to environmental pollution, while soil nutrient mining poses a serious threat to food security for resource-poor farmers. The situation is becoming increasingly severe, with over 90% of human excreta and wastewater being discharged into the environment with little or no treatment. Human urine, rich in nitrogen (N), phosphorus (P), and potassium (K), is a promising fertiliser alternative, but its high-water content and alkalinity hinder direct use. The increasing need for sustainable waste management and nutrient recycling has prompted interest in the reuse of human urine as a nutrient source in agriculture. Organic materials like potato waste and pine bark are abundant in South Africa and they offer a potential solution for stabilising urine nutrients. However, there is limited information on the use of these organic wastes and their biochar and ash types. The aim of this study was to determine the recovery of nitrogen by potato waste and pine bark and their biochar and ash types and the potential of the urine-based solid fertilisers to release the nutrients and improve crop productivity.

The feedstock, ash and biochar types pyrolysed at 350 °C and 650 °C from cull potatoes (CP) and pine bark (PB) were used for recovery of nitrogen (N) from artificial human urine (AHU). The AHU was added to each medium (50 g) six times at 29 and 33 ml and dehydrated each time at 45 and 60 °C, respectively. Therefore, cumulative urine additions at days 0, 14, 21, and 28 were 0, 58, 116 and 174 ml for 45 °C (4.5 L/m²) and 0, 66, 132 and 198 ml for 60 °C (5.1 L/m²), with dehydration in between. In addition, pine bark feedstock, kiln biochar and ash were also treated with the AHU and dehydrated at the glasshouse (Min temp: 9 and max. temp: 57 °C). Samples of the urine-products were collected before each urine addition, and analysed for pH, total carbon (C), and nitrogen (N) and total N recovered from AHU was calculated.

The final urine-based products were applied to soil from the 0-20 cm depth of a Westleigh (Acrisols) to study nutrient release patterns in the laboratory and crop growth under glasshouse conditions. A field trial was also carried out on the same soil. The urine-products were applied to soils at triple N (300 kg ha^{-1}) and at recommended N (100 kg ha^{-1}) rate for the incubation and spinach experiments. Destructive sampling was done at days 0, 7, 14, 21, 28, 42, and 56, with analysis of pH, ammonium- and nitrate-N and extractable P. The urine- products were applied to 2 kg of soil (0-20 cm depth) for spinach growth under a glasshouse. The pot trial ran for 84 days with plant sampling at 6 and 12 weeks and analysis for dry matter, nutrient uptake and tissue micronutrients. The residual soil samples were analysed for pH and extractable P.

For the field trial, the PB- urine products were applied at the recommended N rate for spinach in the first season. The PB feedstock, biochar and ash that were not pre-treated with urine were also included as reference materials. In the second season, the plots were split into two, with half treated with the same treatment at the same rate (cumulative effects), while the other half did not receive additional treatment (residual effects). Spinach dry matter yield, nutrient uptake, and tissue micronutrients were determined.

The different media from PB recovered more N than those from CP. The 60 °C temperature generally promoted rapid dehydration and consumed less energy than 45 °C. The most N was recovered on PB biochar pyrolysed at 650 °C with dehydration at 60 °C. The CP feedstock also recovered more N than CP ash and CP biochar, which showed negative N recovery. Results demonstrated that urine-PB ash treatment enhanced nitrate-N, while CP ash maintained high extractable phosphorus and pH.

In glasshouse trials, PB-based products significantly improved spinach dry matter yield and nitrogen uptake, with performance often comparable to synthetic fertilisers. However, elevated sodium (Na) and manganese (Mn) concentrations in plant tissue were observed, necessitating monitoring to prevent potential toxicity. Field trial confirmed the agronomic viability of PB urine-based fertilisers, The pine bark urine treatments had higher dry matter yield and N uptake relative to the Pine bark un-enriched products, during the first season. Cumulative additions led to lower dry matter and nitrogen uptake in PB biochar-enriched urine (PB biochar-U), which was similar to the negative control and lower than PB-U. However, PB biochar-U showed higher residual effects than the negative control. Notably, repeated application did not exacerbate soil salinity due to effective sodium uptake by the initial crop. This research highlights the potential of urine-based fertilisers derived from PB and CP materials as sustainable alternatives to synthetic nitrogen inputs, offering a circular solution to nutrient management. These findings imply that urine-based products derived from pine bark and cull potatoes, particularly biochar and ash from pine bark, are effective in recovering nitrogen, releasing nutrients in the soil, and improving crop productivity. The potential of these products to show similar results to synthetic fertiliser in spinach yield and nutrient uptake under glasshouse and field conditions shows their potential as sustainable nitrogen sources. Moreover, less effects on soil salinity and the observed residual benefits suggest that these material can support long-term soil fertility in resource limited settings. This research offers practical solutions for nutrient recycling, closing sanitation-agriculture loops, and enhancing food security.

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Chapter 1: Introduction

1.1 Background

Sustainable sanitation and agricultural productivity are interconnected challenges, particularly in rural and peri-urban areas where proper waste management and nutrient recovery are critical. In many parts of the world, improper sanitation leads to environmental contamination, loss of valuable nutrients, and public health concerns (Giusti, 2009). An estimated 80% of human excreta and wastewater is discharged into the environment with little or no treatment (van den Broek et al., 2024), contributing to pollution of water, soil, and air. Given the rapid population growth and increasing waste generation, finding sustainable strategies for nutrient recovery from the wastes and recycling is of paramount importance.

Ecological sanitation (ECOSAN) has emerged as an effective approach to address sanitation challenges while promoting nutrient recovery from human waste (Junghanns and Beery, 2020). The ECOSAN technologies, such as urine-diverting dry toilets, enable source separation of urine and faeces, facilitating the recycling of nutrients for agricultural purposes. Literature shows human urine as having higher nutrients, which are in ionic relative to faeces (Senecal and Vinnerås, 2017). Human urine, which contains high levels of essential nutrients like nitrogen (N), phosphorus (P), and potassium (K), has been recognized as a valuable fertiliser resource (Mnkeni et al., 2008; Senecal and Vinnerås, 2017).

Studies have shown that urine application improves nitrogen uptake, enhances soil fertility, and increases crop yields in various agricultural systems (Pradhan et al., 2009; Rumeau et al., 2024). Similarly, urine application in maize, spinach, and cucumber cultivation has been associated with significant yield improvements (Kassa et al., 2018; Heinonen-Tanski et al., 2007). In this regard, the urine-based fertilisers have demonstrated comparable, and in some cases superior, agronomic performance relative to synthetic fertilisers (Magid et al., 2007). However, its high-water content (approximately 97%) poses significant challenges for transportation, storage, and application, making it less competitive than commercial fertilisers (Ganesapillai and Simha, 2016). Moreover, urea in urine is highly prone to hydrolysis and ammonia volatilisation due to alkaline pH, leading to nitrogen losses (Geinzer, 2017). Therefore, strategies for stabilising the activity of urease enzyme activity and concentrating

urine nutrients into solid fertilisers could be beneficial, as there will be better control of nutrient storage, transport and application rates.

Various studies have explored methods to recover nutrients from urine while reducing water content, including NH_3 stripping (Antonini et al., 2012), nitrification (Udert et al., 2015), electrolysis (Udert et al., 2015) and struvite formations (Etter et al., 2011). However, high technological and equipment requirements and costs, associated with such methods, limit their practical application, especially at small-scale. Also, they focus on one element to be recovered, such only P for struvite production. Among these, the addition of alkaline or acidic substrates to modify urine pH and prevent urease activity has shown promising results, which may be cost effective, environmentally friendly and sustainable. The alternative cost-effective strategies that remove excess water, while concentrating nutrients, are therefore, of paramount importance.

Senecal and Vinnerås (2017) suggested that pre-treatment of the urine to limit urease activity may allow dehydration to be done. The urease can be inhibited by pH and temperature (Hellström et al., 1999; Hotta and Funamizu, 2008). Literature shows that the activity of the urease enzyme is inhibited at pH <3.5 or >8.5 (Rektorschek et al., 1998). Alkaline amendments such as Birch wood ash (pH >12.5) have been found to stabilize urine, minimize nitrogen losses, and allow for effective dehydration (Senecal and Vinnerås, 2017). While urease inhibition at alkaline pH has been widely studied as a strategy to stabilise urine and prevent urea hydrolysis, recent research has shown that acidification can be a more effective alternative. Acidifying fresh urine to pH < 4 inhibits enzymatic ureolysis, thereby retaining nitrogen in the form of urea (Simha et al., 2023; Simha et al., 2025). Simha et al. (2023) demonstrated that adding fresh urine with readily soluble organic acids such as citric or oxalic acid effectively maintained pH < 3, producing concentrated, nutrient-rich solid fertilisers containing up to 21% nitrogen.

Similarly, biochar derived from various feedstocks, such as agricultural residues, has gained attention for its ability to adsorb nutrients, improve soil fertility, and sequester carbon (Jeffery et al., 2015). However, no literature could be accessible on the use of biochar from pine bark material and food waste on the recovery on urine N and on the potential for the products as N fertilisers. A combination of reduction of urease activity from urine and biochar, from locally available organic materials, could be helpful in improving soil health, the supply of the retained

nutrients and soil organic carbon storage. Pine bark and potato waste are among the most common organic wastes in South Africa.

Potato waste biochar, produced through pyrolysis at different temperatures, exhibits alkaline properties (pH 10-12), which may contribute to urease inhibition and nitrogen retention (Vilakazi et al., 2023). Conversely, pine bark, with a naturally acidic pH (4-5), presents an alternative medium that may also enhance nutrient stabilization and recovery. However, the effectiveness of these materials in urine treatment and fertiliser production has not yet been thoroughly investigated. While the co-application of urine and wood ash resulted in a more than threefold increase in tomato yield compared to untreated soils (Pradhan et al., 2009), the treatment was not a solid dehydration product of ash and urine. Conversely, although wood ash has been found to retain N upon dehydration (Senecal and Vinnerås, 2017), the effects of addition of the product to the soil on N release were not studied. Despite the promising findings from urine as a fertiliser material, there remains limited research on the effectiveness of feedstocks, biochars and even ash from locally available organic materials on their ability to retain nutrients, such as N, from urine. Studies that have used various materials through alkalinisation or acidification approaches to recover nitrogen from urine have been acknowledged throughout this thesis. The novelty of the present study lies in the use of pine bark, which is naturally acidic, and cull potato materials as media for nitrogen recovery from urine. Unlike the thesis by Prithvi, which focused on strongly alkaline materials such as bran ash, wood chip ash, and combinations with alkaline substrates including KOH, $Mg(OH)_2$, and $Ca(OH)_2$, this study investigates pine bark (PB) and cull potato (CP) in their feedstock, biochar, and ash forms. These materials differ considerably from those used in previous studies (e.g., Simha's PhD work) in both surface characteristics and chemical composition, factors that may influence their ability to retain nitrogen and the associated transformation during urine dehydration. In addition, the fertiliser value of the urine-based solid products derived from different materials derived from organic materials, such as potato waste and pine bark, has not yet been established. The effectiveness of these fertilisers in real-world agricultural settings, where environmental conditions vary, therefore needs investigation to be established.

1.2 Objectives of the study

This study evaluated the potential of feedstock, biochar and ash from potato waste and pine bark for nitrogen retention from urine, and the nutrient release from the urine-based solid fertilisers products and their effects on crop productivity. The specific objectives were to:

i. Investigate the effect of different feedstocks, biochar and ash from potato waste and pine bark and dehydration conditions (oven at 45 and 60°C and glasshouse) on the nitrogen retention from artificial human urine.
ii. Assess the impact of urine-based fertilisers from cull potato on nitrogen and phosphorus release in soil and assess their effect on spinach growth and yield under controlled (glasshouse) conditions.
iii. Assess the impact of urine-based fertilisers from pine bark on nitrogen and phosphorus release in soil and assess their effect on spinach growth and yield under controlled (glasshouse) conditions.
iv. Evaluate the agronomic effectiveness of urine-based fertilisers from pine bark on spinach growth and yield (dry matter, N and P uptake and micronutrients) , and residual soil chemical composition (pH, extractable P and mineral-N) under uncontrolled (field) conditions.

1.3 Significance of the study

The findings of this research will contribute to the development of sustainable and cost-effective fertiliser alternatives, particularly in regions where synthetic fertilisers are expensive or inaccessible. By repurposing human urine and organic waste materials, this study aligns with global efforts to close nutrient loss loops, promote circular economy principles, and enhance food security. Additionally, the study will provide insights into the feasibility of large-scale implementation of urine-based solid fertilisers in sustainable agricultural systems. Ultimately, this research has the potential to address both environmental and agronomic challenges by reducing nutrient losses, improving sanitation, and enhancing soil fertility for increased crop productivity. The successful application of urine-based fertilisers could play a pivotal role in advancing sustainable agriculture and ecological waste management strategies worldwide. It also aligns with key Sustainable Development Goals (SDGs). It contributes to SDG 6 (Clean Water and Sanitation) by improving nutrient management and reducing water pollution, supports SDG 12 (Responsible Consumption and Production) through sustainable recycling of nutrients from human urine, and addresses SDG 13 (Climate Action) by minimising greenhouse gas emissions associated with conventional fertilisers.

Chapter 2: Effect of crop residue feedstock, biochar, and ash on nitrogen and phosphorous recovery, and availability: A systematic review

Abstract

With the increasing global demand for these essential nutrients (nitrogen (N) and phosphorus (P), has resulted in the exploration of organic and waste-derived amendments has gained traction as a viable alternative to synthetic fertilisers. Nutrient recovery from liquid waste (urine and wastewater) is an emerging strategy for environmental conservation and sustainable agricultural productivity. This systematic review examines the role of plant-based feedstocks, biochar, and ash in the recovery and availability of nitrogen (N) and phosphorus (P) from human urine and wastewater. The various materials reviewed including in this study include biochar derived from crop residues and wood waste, alkaline-stabilized ash, and industrial by-products such as coal fly ash, highlighting their efficiency in N and P retention. Biochar, particularly when modified or produced under specific pyrolysis conditions, demonstrated a high capacity for nutrient adsorption and slow-release fertilisation, when the products are used for crop fertilisation. Alkaline amendments, including wood ash, effectively stabilized urine-derived nutrients by inhibiting urease activity, reducing nitrogen losses, and facilitating phosphorus precipitation. Despite the promising applications of these materials, challenges such as salinity, media clogging, and ammonia volatilisation remain, requiring that the methods be further optimised. The review also identifies a research gap in developing countries, where limited studies have been conducted on nutrient recovery technologies despite their potential benefits for soil fertility and waste management. By integrating biochar, ash, and plant-based materials into nutrient recovery systems, this review underscores their role in advancing circular economy principles and sustainable agricultural practices by integrating biochar, ash, and plant-based materials into nutrient recovery systems. Future research should focus on the agronomic performance of urine-based fertilisers, biochar modifications, and large-scale implementation strategies to enhance economic feasibility and environmental sustainability.

Keywords: wastewater, nutrient recovery, biochar, ash, sustainable

2.1 Introduction

Global challenges such as nutrient depletion, environmental pollution and inadequate sanitation systems necessitate innovative and sustainable solutions as the world population grows. The demand for agricultural inputs like nitrogen (N) and phosphorus (P) intensifies, putting immense pressure on natural resources (Cassman et al., 2003; Maja and Ayano, 2021). Conventional synthetic fertilisers, known for their energy-intensive production and adverse environmental impacts when mismanaged, contribute significantly to greenhouse gas (GHG) emissions and soil degradation (Lal, 2015). In addition, untreated human excreta and wastewater exacerbate these environmental challenges, leading to nutrient losses and water pollution (Giusti, 2009; Werner et al., 2004). Human urine contains >75-90% of all N and in human excreta and is a valuable yet underutilized resource for nutrient recycling for agricultural productivity enhancement (Mnkeni and Austin, 2009; Mchunu et al., 2018; Masrura et al., 2021). Despite its nutrient richness, >80% of human excreta and wastewater globally remain untreated, causing environmental threats (eutrophication and greenhouse gas emissions) (van den Broek et al., 2024). The nutrient composition (0.74% N, 0.029% P, and 1.62% potassium (K)) of urine closely aligns with the nutrient requirements of many crops, making it a balanced and sustainable source of fertiliser.

In many countries, human urine is directly applied as a method for recycling nutrients to support agricultural production (Heinonen-Tanski et al., 2007; Mnkeni et al., 2008; Masrura et al., 2021). Its application has been successfully used as a fertiliser in maize (Kassa et al., 2018), tomato (Pradhan et al., 2009), cucumber (Heinonen-Tanski et al., 2007) and spinach (Rumeau et al., 2024). In maize, it increased plant height, stem diameter and leaf length while increasing soil PO_4^{3-} levels and salinity (Kassa et al., 2018). Urine fertilisation of tomatoes produced yields comparable to mineral fertilisers, which were 4.2 times higher than the control (Pradhan et al., 2009). Similarly, cucumbers fertilised with urine performed as well or slightly better than those treated with mineral fertilisers (Heinonen-Tanski et al., 2007). Urine also increased shoot biomass in spinach compared to the control (Rumeau et al., 2024). Despite its potential as a fertiliser, direct use of urine as fertiliser poses several challenges.

The high-water content (≈ 97%) and alkaline pH of urine (stored) present challenges for its direct use as a fertiliser, complicating storage, transportation, application logistics and costs (Geinzer, 2017; Patel et al., 2020; Senecal and Vinnerås, 2017). To address these challenges, potentially sustainable strategies have emerged for nutrient recovery and reuse (Junghanns and

Beery, 2020). These approaches aim to transform human urine into a concentrated nutrient source, mitigating environmental pollution.

On the other hand, large quantities of wastewater could also contribute to significant environmental pollution (Lu et al., 2018). Approximately 1,000 km^3 of anthropogenic wastewater is generated globally each year, with nearly 30% coming from municipal sources and more than 60% from industrial sources (Li et al., 2015; Lu et al., 2018; Lu et al., 2021). wastewater generated contains 300-600 grams of carbon, 40-60 grams of nitrogen, 5-20 grams of phosphorous per 1 cubic meter (Li et al., 2015). While conventional wastewater treatment plants are crucial for sanitation, they consume about 3% of global electricity, making them large energy consumers and GHG emitters (Li et al., 2015; McCarty et al., 2011). As a result, there is an urgent need for more sustainable water treatment technologies, especially for communities with limited access to electricity, and without access to centralised wastewater treatment plants.

Various technologies and materials (alkaline treatments, biochar, and ash) have been investigated for their ability to recover and stabilize nutrients, particularly N and P, from human urine (Randall et al., 2016; Senecal and Vinnerås, 2017; Simha et al., 2020a; Simha et al., 2021), and these could also be applicable for other wastewaters. Using $Ca(OH)_2$ has been shown to effectively raise the pH above 12, inhibiting urease activity (Randall et al., 2016). Different media also alkaline media such as wheat bran, wood biochar, wood ash mixed with $Ca(OH)_2$, have also been shown to recover > 90% of N, by preventing the enzymatic hydrolysis of urea (Simha et al., 2020a). Crop residues offer cost-effective and sustainable options for nutrient recovery through adsorption and dehydration processes (Divyabharathi et al., 2024). Ash derived from combustion of organic material is alkaline and nutrient-rich and provides additional benefits for soil fertility. The crop residues and ash offer scalable approaches to nutrient recycling, especially in resource-limited settings. Conversely, biochar's high porosity and adsorption capacity derived from pyrolysis of organic biomass at different temperatures makes it suitable for stabilising N and enhancing soil P retention (Pathy et al., 2021).

Biochar has gained increasing attention due to its potential to improve soil organic carbon storage, and overall fertility, while reducing GHG emissions (Pathy et al., 2021). Its use for nutrient recovery is an emerging initiative with limited research reported. Its properties (surface area and ion exchange capacity) vary depending on the feedstock and pyrolysis conditions, which could influence its effectiveness in nutrient recovery (Cao et al., 2013). Depending on

biomass type and pyrolysis temperature, biochar develops various functional groups (carboxyl, hydroxyl, carbonyl and phenolic hydroxyl) (Harvey et al., 2011). These functional groups contribute to ion-exchange reactions and adsorption processes with targeted molecules. This systematic review explores the role of plant-based feedstocks, biochar and ash in recovering N and/or P from human urine and wastewater and their potential to enhance crop productivity. It aims to provide a comprehensive overview of current research, identify and highlight knowledge gaps, which provide opportunities for future investigations to advance ecological sanitation and sustainable agriculture.

2.2 Materials and methods

A systematic review approach was used to map the literature supporting the topic's broad research question. The systematic review methodology was based on the framework outlined by Koutsos et al. (2019). The review included the following six steps (Figure 2.1).

i. Scoping
ii. Planning
iii. Identification/ search
iv. Screening
v. Eligibility/ assessment and
vi. Presentation/ interpretation

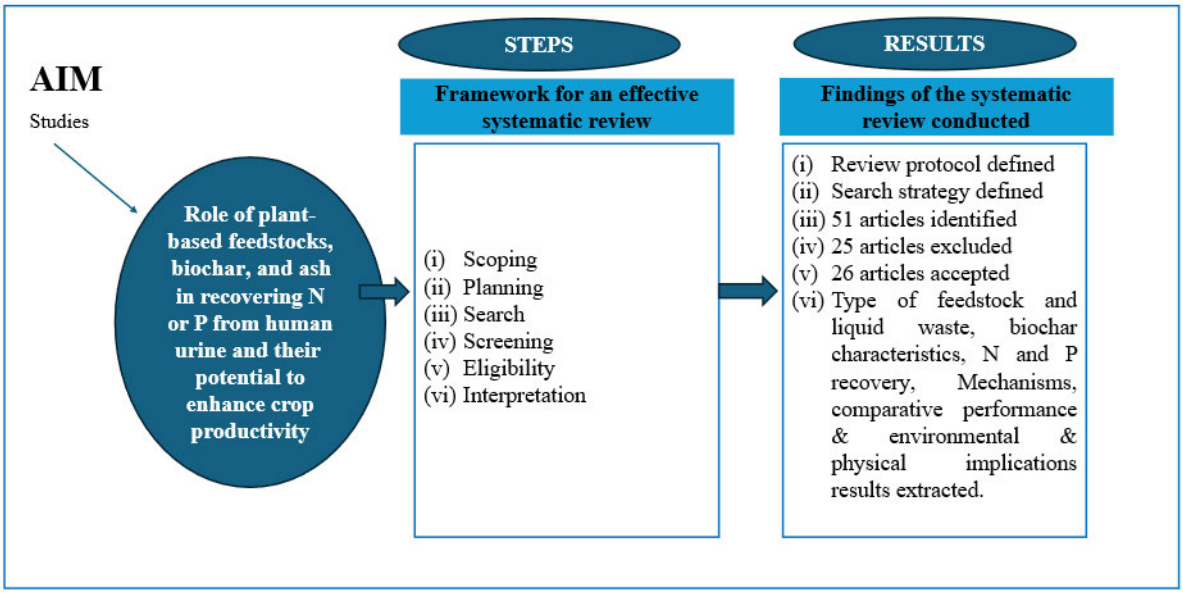


Figure 2.1: The framework used to perform a systematic review for current study

2.2.1 Research question

The following questions guided the review: (i) What is the role of plant-based feedstock, biochar, and ash in nitrogen and phosphorous recovery from urine and wastewater? and (ii) What is the N release and availability of N/P for plant uptake by the urine-based products?

2.2.2 Data sources and search strategy

The search was implemented in five electronic databases:

1. Scopus (www.scopus.com) (accessed on 21 January 2025)
2. Web of Science (www.webofknowledge.com) (accessed on 21 January 2025)
3. Google scholar (scholar.google.com) (accessed on 23 January 2025)
4. Science Direct (www.sciencedirect.com) (accessed on 24 January 2025)
5. Science.gov (www.science.gov) (accessed on 24 January 2025)

These databases were selected to be inclusive and cover discipline in agriculture and science. The data range limitation was chosen to focus on contemporary literature on different materials used for urine dehydration to recover nitrogen/phosphorous and its ability to be released back into the soil. The search strategy employed broad search terms (Table 2.1) to ensure that some publications were not overlooked.

Table 2.1: Search strategy with Boolean operators for each database to identify peer-reviewed articles examining nitrogen and phosphorous recovery from urine and wastewater.

Database (s)	Primary terms (s)	Expanded term (s)
Scopus, Web of Science, Science Direct, Science.gov and Google scholar	Biochar, ash, waste material	plant-based material*" OR "plant biomass" OR "agricultural residue*" OR "organic material*" OR "plant-derived feedstock*" OR "biochar" OR "crop residue*" OR "agro-waste" OR "ash*" AND
	Biochar characteristics	"characteristics" OR "properties" OR "composition" OR "traits" OR "qualities" OR "attributes" AND
	Nutrient recovery	"nitrogen recovery" OR "N recovery" OR "phosphorous recovery" OR "nutrient recovery" OR "ammonia recovery" OR "fertiliser production" OR "resource recovery" OR "waste valorisation" AND
	Urine and wastewater	"urine" OR "human urine" OR "liquid waste"

2.2.3 Citation management

Citations were imported into the DistillerSR (Evidence Partners Incorporated, Ottawa ON, Canada) web-based application, and duplication citations were removed using the duplicated removal function of DistillerSR. Subsequently, the title and abstract relevance screening and data characterisation of complete articles were carried out using DistillerSR.

2.2.4 Relevance screening and eligibility criteria

A two-step screening relevance technique was employed. For the first step, the titles and abstracts of the articles were examined for relevance. Next, all citations considered relevant after the title and abstract screening went through a full-text review. Studies were eligible for inclusion if they were original articles on any plant-based feedstock, biochar or ash on N/P recovery from urine or wastewater and availability of the nutrients in growth media.

2.2.5 Data charting

The data collection categories included author, title, type of feedstock used, biochar characteristics and treatment conditions, type of liquid waste, N and P recovery efficiency, comparative performance, environmental and physical implication and country of study.

2.2.6 Summarising and reporting

A narrative synthesis approach was used to provide an overview of the existing literature. Firstly, a summary of the study was combined, considering variations that may affect the N/P recovery and release. Then, the study results were organised into categories.

2.3 Results

2.3.1 Study area locations

Figure 2.2 shows the distribution of the screened journal articles across various countries and provides insights into the geographic focus of the research. China and the USA stood out with four occurrences, indicating that more studies were conducted in these countries than in others. Other countries such as Sweden, Canada, Finland, and the UK also had notable representations.

Some countries appeared with one study highlighting their minimal engagement with the subject, while there were no records from most countries in South America, Europe, Asia, Australia/New Zealand, and Africa. The N and P recovery using various materials is being researched in multiple countries across continents, highlighting the global significance of this topic. It is important to acknowledge that North America and Asia have made substantial contributions, whereas Africa's representation appears relatively limited.

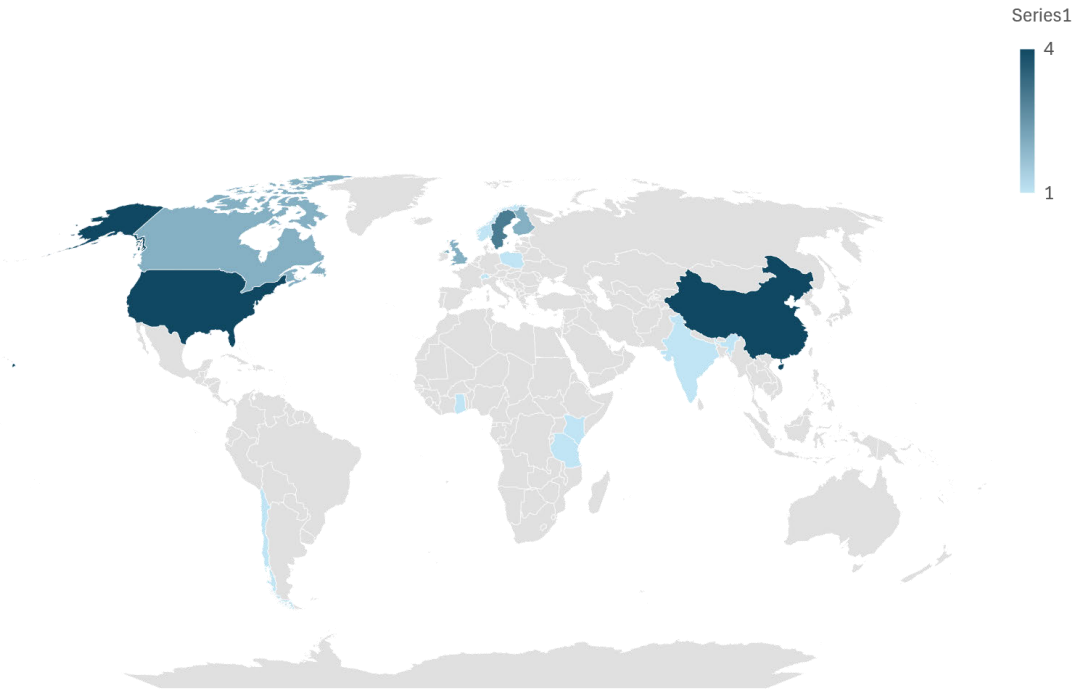


Figure 2.2:The breakdown of countries showing the relative distribution of the 26 publications that use different materials to recover N and P from liquid waste.

2.3.2 Frequency analysis

Figure 2.3 presents the distribution of publications over the years. No articles published between 2010 and 2011 met the inclusion criteria. Between 2012 and 2016, the number of publications remained consistently low, with one publication each year, while a slight increase was observed in 2017, with two publications, followed by a significant increase in 2018 (4 publications). In the year 2019, there were two publications, which increased and peaked in 2020-2021, and peaked in 2023, with three publications. This was followed by a decrease in 2024 and 2025, with zero and one (by January 2025) publication, respectively. These trends of publications suggest that, both in Africa and globally, limited work is done on the N and P

recovery from liquid waste using different organic amendments (plant-based feedstocks, biochar and ash).

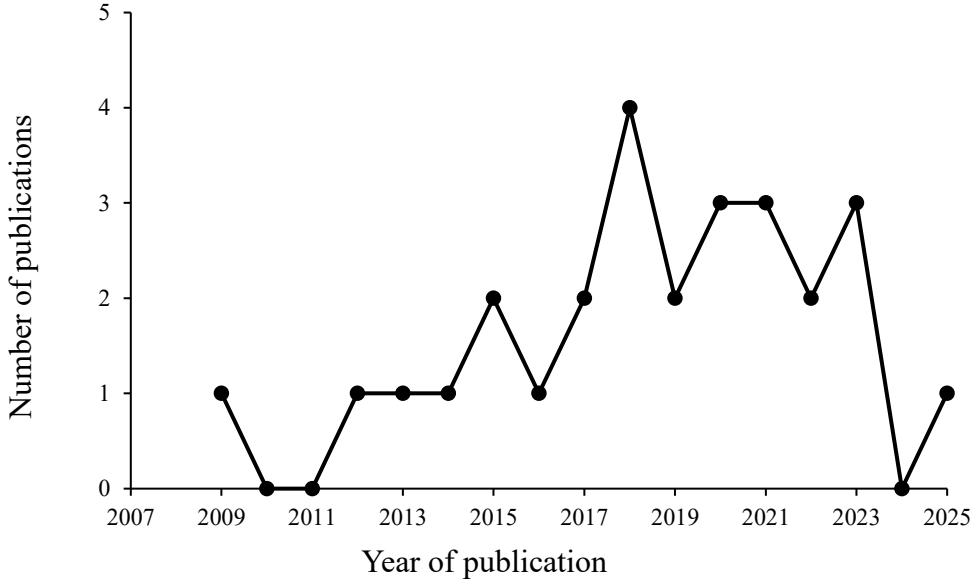


Figure 2.3:Trend of publications for the N and P recovery from different liquid waste.

2.3.3 Overview of studies identified

The review examined N and P recovery from liquid waste (urine and wastewater) using various materials, including plant-based feedstocks, biochar, and ash. Table 2 summarizes the 26 studies identified from the past 16 years (2009-2025) investigating nutrient adsorption mechanisms and material effectiveness. The research also explored environmental and physical implications, assessing feasibility. Only two studies (Pradhan et al., 2009; Schmidt et al., 2015) evaluated effects of direct urine application with organic material on crop productivity.

Table 2.2:Summary of the studies on N and P recovery from liquid waste using different materials (plant-based feedstocks, biochar and ash), including the environmental and physical implications of the end-products.

Author	Title	TOF	BCTC	TOLW	Nitrogen and phosphorous recovery efficiency	Mechanism(s) of nitrogen and phosphorous recovery	Comparative performance	Environmental and physical implication	Country of study
(Ganesapillai et al., 2021)	Sustainable recovery of plant essential nitrogen and phosphorus from human urine using industrial coal fly ash	Industrial coal fly ash	The coal fly ash used had the following characteristics: SiO_2 (57.6%), Al_2O_3 (31.6%), Fe_2O_3 (2.88%), surface area 5.59 m^2/g, particle size between 74–88 µm	Synthetic human urine	Nitrogen (Urea–N) Removal: Enhanced adsorption with increased initial urine concentration from 20% to 60%, raising adsorption potential from 10.26 mg/g to 29.59 mg/g. Phosphorus Recovery: Approximately 83% of phosphorus recovered via struvite precipitation.	Nitrogen: Adsorption onto fly ash through surface interactions. Phosphorus: Precipitation as struvite ($Mg.KNH_4PO_4{\cdot}6H_2O$) by adjusting pH to 8.5 and adding MgO, following urea adsorption	The study highlights coal fly ash as a promising low-cost alternative for nutrient recovery, particularly effective compared to conventional methods like chemical precipitation alone.	Environmental Impact: Utilizes industrial by-products (fly ash) for nutrient recovery, reducing waste disposal issues. Practical Implications: Provides a sustainable method for nutrient recovery from human urine, potentially applicable in sanitation and agricultural sustainability.	India
(He et al., 2022)	Waste-derived biochar for water pollution control and sustainable development	Lignin- and cellulose-rich crop residues, wood waste, Animal manure, sewage sludge	Production Methods: Slow pyrolysis, fast pyrolysis, gasification, microwave-assisted pyrolysis. Temperature Range: 300–900 °C, with slow pyrolysis being the most economically feasible. Characteristics: Surface area, porosity, ash content, and surface functionality can be engineered based on feedstock and pyrolysis conditions.	Municipal and industrial wastewater.	**Nutrient Recovery:** Biochar recovers N and P through adsorption and can be used as a slow-release fertiliser. Fe-rich sludge-derived biochar enhances phosphorus recovery, while MgO-supported biochar facilitates simultaneous N and P recovery via struvite precipitation.	**Mechanisms:** Adsorption, precipitation (e.g., struvite formation), and catalytic degradation processes are involved. Surface functional groups and mineral components in biochar aid nutrient binding and recovery.	Biochar offers comparable adsorption capacity at a significantly lower cost (51–381 USD/ton vs. 800–2,500 USD/ton for activated carbon), with additional carbon sequestration and waste valorisation benefits.	Environmental Impact: Reduces GHG emissions through carbon sequestration, improves wastewater treatment efficiency, and supports sustainable development goals (SDGs). Practical Applications: Enhances sludge management, odour mitigation, nutrient recovery, and stormwater management.	China
(Hunter and Deshusses, 2020)	Resources recovery from high-strength human waste anaerobic digestate using simple nitrification and denitrification filters	Biochar (pine and coconut shell), granular activated carbon (GAC), coconut husks, bamboo, rice husks, sunflower seeds, and zeolite	Pine Biochar: Sourced from Biomass Controls, Coconut Shell Biochar: Sourced from Buy Activated Charcoal.com	anaerobic digestate (faecal and urine waste).	69% of total nitrogen IN biochar and 89% of phosphorus (PO_4-P) were removed from the waste stream.	Nitrogen: Removed via nitrification and denitrification. Phosphorus: Recovered through struvite precipitation (ammonium magnesium phosphate hydrate) in denitrification filters.	Biochar vs. GAC: GAC achieved higher COD removal (84%) than biochar (72%). Zeolite: Provided the highest TN removal in Phase 1 (69%). Coconut Husk Filters: Effective for denitrification but showed reduced phosphate removal over time due to adsorption saturation.	Sanitation Improvement: The system suits regions lacking improved sanitation, supporting resource recovery and water reuse. Agricultural Benefits: Nutrients recovered as fertilisers, enhancing local agricultural productivity. Challenges: Media clogging and organic leaching issues.	USA
(Jiang et al., 2023)	Treatment of Pyrolytic Eco-toilet Waste: Characterization of Faeces-Based Biochar Produced from Different Temperatures and Their Effects on Urine Properties and Fractions	Human faeces biochar.	Pyrolysis Temperatures: 300°C, 400°C, 500 °C, 600 °C. Residence Time: 60 minutes at target temperature. Surface Area: 7.45 to 17.51 m^2/g. Fixed Carbon: Increased from 29.9% to 44.6%. pH: Increased with temperature from 11.28 to 12.4.	Human urine	TN and $NH4^+$-N Removal: Effective adsorption by faeces-based biochar, mainly FB400, shows the best adsorption effect. TP Recovery: Faeces-based biochar at 600 °C (FB600) effectively adsorbed phosphorus in urine.	Nitrogen: Promoted urea hydrolysis and adsorption of ammonium. Phosphorus: Stabilization during biochar preparation at lower pyrolysis temperatures and adsorption at higher temperatures.	Faeces-based biochar prepared at high temperatures (500 °C and 600 °C) demonstrated superior nutrient recovery performance compared to lower temperature biochars, with better stabilization effects on urine properties.	Sanitation Improvement: Provides valuable information for developing eco-toilets, promoting nutrient recovery and reducing environmental pollution from untreated excreta. Practical Use: It enhances urine stabilization and is suitable for resource recovery systems.	China
(Kakaei Lafdani et al., 2021)	Nitrogen Recovery from Clear-Cut Forest Runoff Using Biochar: Adsorption–Desorption Dynamics Affected by Water Nitrogen Concentration	Spruce and birch wood biochar.	Pyrolysis Temperature: 600 °C, Specific Surface Area: Spruce biochar: 320 m^2/g, Birch biochar: 260 m^2/g, pH: Spruce: 9.25, Birch: 9.75, Nitrogen Content: Spruce: 1.19%, Birch: 1.39%.	Clear-cut forest runoff (water containing nitrogen).	Total Nitrogen (TN) Adsorption: Birch biochar adsorbed up to 0.3 mg TN/g biochar, spruce biochar up to 0.2 mg TN/g biochar. Nitrate-Nitrogen (NO_3^--N) Adsorption: Birch biochar showed linear adsorption up to 0.15 mg NO_3^--N/g biochar, while spruce biochar's adsorption was unaffected by initial concentration.	Nitrogen Adsorption: Influenced by biochar surface area, porosity, and functional groups. Birch biochar exhibited better adsorption capacity than spruce due to different surface properties. Desorption: Minimal for both biochars, indicating strong adsorption capacity suitable for water purification	Birch biochar had higher nitrogen adsorption and desorption capacity than spruce biochar despite spruce having a higher specific surface area. This indicates factors beyond surface area influence nutrient recovery efficiency.	Water Purification: Biochar effectively reduces nitrogen in clear-cut forest runoff, particularly in peatland areas where traditional methods are inefficient during non-growing seasons. Sustainability: Provides a complementary, eco-friendly method for water protection in boreal forests.	Finland
(Koulouri et al., 2023)	Source separation of human excreta: Effect on resource recovery via pyrolysis	Human faeces biochar (SSF) and urine+ faeces (MUF)	Pyrolysis temperatures: 450 °C, 550°C, 650°C. Characteristics: SSF biochar had higher calorific value and lower ash content; MUF biochar had higher porosity and nutrient (P, K) content	Human excreta (faeces and urine).	Nitrogen Recovery: Urine diversion avoided 70% of total nitrogen losses during thermal treatment. Phosphorus Recovery: Both SSF and MUF biochars had high phosphorus recovery potential, with MUF biochars richer in phosphorus and potassium	Nitrogen: Retention enhanced by source separation, reducing volatilisation during pyrolysis. Phosphorus: High availability up to 550 °C, primarily present as phosphate compounds confirmed by XRD and FTIR.	MUF Biochar: Higher nutrient content (P and K), cation exchange capacity, and salinity may limit agricultural use.	Sanitation Improvement: Supports circular faecal sludge management system design, promoting safe sanitation and resource recovery. Agricultural Benefits: Provides biochar with high fertiliser value, especially MUF biochar, though salinity concerns must be managed.	United Kingdom
(Krause and Rotter, 2018)	Recycling Improves Soil Fertility Management in Smallholdings in Tanzania	Compost and microgas stoves biochar	Source: Biochar recovered from cooking with microgasifier stoves and sanitation processes. Application: Used as part of "CaSa-compost", which includes biochar and sanitized human excreta	Human urine	Nitrogen (N) and Phosphorus (P) Recovery: Biogas slurry and composts supply sufficient phosphorus, while urine supplements nitrogen effectively. CaSa-compost contributes significantly to total nitrogen and phosphorus recovery.	Nitrogen: Supplied through urine and biogas slurry. Phosphorus: Provided via composts and biochar, with biochar contributing to total carbon and phosphorus content in the compost.	CaSa-compost (biochar + sanitized excreta): More effective for soil nutrient replenishment than standard compost or biogas slurry alone. Standard Compost: Provides some nutrient recovery but less effective compared to CaSa-compost.	Soil Fertility: Recycling residues from cooking and sanitation mitigates phosphorus deficiency and soil acidity, restoring organic matter. Environmental Impact: Reduces environmental emissions and improves nutrient cycling in smallholder farms.	Tanzania
(de Kraker et al., 2019)	Decentralized Valorization of Residual Flows as an Alternative to the Traditional Urban Waste Management System: The Case of Peñalolén in Santiago de Chile	Kitchen waste (KW) and garden residues (GR).	The study does not specifically focus on biochar but examines composting, vermicomposting, anaerobic digestion, and struvite precipitation for nutrient recovery.	Human urine	Anaerobic Digestion (AD): Recovers 12% nitrogen and 24% phosphorus from kitchen and garden waste. Vermicomposting: Recovers 6% nitrogen and 24% phosphorus.	Urine: nutrients conserved during storage; struvite precipitation recovers 85% phosphorus. Composting/Vermicomposting: Nutrient retention through organic matter stabilization, though nitrogen losses can occur. Anaerobic Digestion: Converts organic nitrogen to ammonium, retaining phosphorus in digestate.	Vermicomposting and anaerobic digestion are compared. Vermicomposting is more cost-effective at the household level, while anaerobic digestion provides higher nutrient recovery rates at the municipal level.	Environmental impact: reduces landfill use and transportation emissions, enhances urban agriculture, and promotes nutrient recycling. Economic Benefits: Vermicomposting shows significant cost savings and profits; anaerobic digestion offers high nutrient recovery but has higher initial investment costs.	Chile
(Fang et al., 2014)	Application of Magnesium Modified Corn Biochar for Phosphorus Removal and Recovery from Swine Wastewater	Corn biochar modified with magnesium.	Modification: soaked in $MgCl_2$ solution before pyrolysis. Pyrolysis Temperatures: 300°C, 450°C, 600 °C in nitrogen atmosphere for 3 hours. Surface Area: Increased with temperature, up to 494.93 m^2/g for B600 and 490.29 m^2/g for Mg/B600.	Swine wastewater	Phosphorus Adsorption: Mg/biochar adsorbed up to 239 mg/g P from swine wastewater, outperforming regular biochar (225 mg/g). Efficiency: 90% of equilibrium P adsorbed within 30 minutes. P Release: Post-adsorption Mg/biochar released 3.3% to 4.4% of total adsorbed P per interval.	Phosphorus: adsorption primarily driven by chemical interactions due to magnesium nanoparticles, enhancing P retention and selectivity.	Mg/biochar vs. Regular Biochar: Mg/biochar showed higher P adsorption capacity and better resistance to pH changes and coexisting ions. Selectivity: Mg/biochar was more selective in P adsorption, with minimal interference from competing ions than regular biochar.	Wastewater treatment: Mg/biochar is an effective, selective, and sustainable method for phosphorus recovery from swine wastewater, reducing eutrophication risks. Agricultural Use: Recovered P from Mg/biochar is suitable as a slow-release fertiliser, promoting nutrient recycling.	China
(Krishnamoorthy et al., 2023)	Biochar Seeding Properties Affect Struvite Crystallization for Soil Application	Lignocellulosic Biomass: Canola straw, wheat straw, sawdust of white spruce. Non-Lignocellulosic Biomass: Manure pellets	Pyrolysis method: microwave-assisted pyrolysis. Pyrolysis temperatures: 300 °C, 400 °C, 500 °C. Surface Area: Ranged from 1.5 m^2/g (CS300) to 43 m^2/g (SD500). Hydrophobicity: Increased with temperature, highest in wheat straw biochar at 500 °C.	Synthetic urine	Ammonium recovery: up to 87.1% with SD500. Phosphate Recovery: Up to 97.9% with sawdust biochar (SD500). Struvite Yield: 7.91 g/L using SD500, higher than non-seeded treatments.	Seeding mechanism: biochar acts as a nucleation site for struvite crystallization, enhancing crystal growth due to its surface area and hydrophobicity. Surface Characteristics: High surface area and pore volume facilitate nutrient retention and slow release.	Sawdust biochar (SD500): provided the highest struvite yield and nutrient recovery. Manure Pellet Biochar (MP500): Comparable performance to SD500 due to higher surface area and hydrophobicity. Wheat Straw Biochar (WS500): Superhydrophobic nature limited its efficiency in nutrient recovery.	Sustainability: enhances phosphorus recovery from urine and wastewater, reducing eutrophication risks. Agricultural Benefits: Recovered struvite-biochar composites can be used as slow-release fertilisers, improving soil health and crop yield.	Canada
(Marcińczyk et al., 2022)	From Waste to Fertiliser: Nutrient Recovery from Wastewater by Pristine and Engineered Biochars	Rice straw, digested sludge, conocarpus waste	Pyrolysis method: slow pyrolysis. Pyrolysis Temperatures: Typically, 300–700 °C. Engineered biochars: Modified with metals (Mg, Fe, Al, Zn) to enhance adsorption properties. Properties: Large specific surface area, pH buffering, and ion exchange ability.	Municipality sludge	Phosphorus recovery: up to 97% efficiency with engineered biochars (metal-modified). Nitrogen adsorption: Biochar selectively adsorbs $NH4^+$ and NO_3^-, with efficiency influenced by pH and biochar type. Slow Release: Biochar-based fertilisers show gradual nutrient release over time.	Nitrogen adsorption: electrostatic interaction, ion exchange, and hydrogen bonding. Phosphorus Adsorption: Surface precipitation, ligand exchange, and formation of struvite with metal modifications. pH Dependence: Adsorption and desorption influenced by solution pH.	Pristine vs. Engineered Biochar: engineered biochars exhibit selective adsorption and enhanced nutrient recovery. Biochar vs. Traditional Fertilisers: Biochar-based fertilisers are cost-effective and promote sustainable agriculture compared to synthetic fertilisers.	Sustainability: supports circular economy principles by recycling nutrients from wastewater. Soil health: improves soil fertility and structure while reducing nutrient leaching. Economic Benefits: Reduces reliance on synthetic fertilisers, potentially lowering agricultural production costs.	Poland
(Oarga-Mulec et al., 2019)	A Comparison of Various Bulking Materials as a Supporting Matrix in Composting Blackwater Solids from Vacuum Toilets	Oat husks, wheat bran, bark, peat, sawdust, food waste	This study focuses on composting rather than biochar production. Various bulking materials were mixed with blackwater solids (faeces, urine, and toilet paper) from vacuum toilets and composted over 60 days without stirring, with controlled aeration.	Blackwater solids from vacuum toilets	Total Nitrogen Losses: As low as 7.5% with oat husks, wheat bran, and bark mixture. Total Phosphorus Losses: 3.8% in the same mixture. Highest phosphorus accumulation: 76.4% in the mixture with oat husks and wheat bran.	Nitrogen: peat and food waste improved ammonia sorption, reducing nitrogen losses. Phosphorus: accumulation due to reduced nutrient leaching and better stabilization from specific bulking material combinations.	The combination of oat husks, wheat bran, and bark performed best in terms of nutrient retention and pathogen reduction. Peat and food waste enhanced ammonia retention but had higher methane emissions. Sawdust combinations maintained higher composting temperatures.	Sanitation improvement: composting blackwater solids with natural bulking materials offers a low-cost, efficient sanitation method, especially suitable for remote and sensitive areas. Environmental Impact: Reduces nutrient losses, minimizes greenhouse gas emissions, and improves compost quality.	Sloveria and Norway
(Simha et al., 2018)	Alkaline dehydration of anion-exchanged human urine	alkaline media (Chopped willow wood tree ash or alkalized biochar)	Biochar feedstock: chopped willow trees. Pyrolysis temperature: 450°C. Alkalization: Biochar was treated with KOH to increase pH to ~12.5.	Human urine	N retention: >70% nitrogen retained during dehydration. Phosphorus and Potassium Recovery: 100% recovery of phosphorus and potassium.	Nitrogen: stabilization of urea by raising urine pH above 10 using anion exchange resin, followed by dehydration which minimizes nitrogen loss. Phosphorus: Complete recovery through dehydration with alkaline media, possibly precipitating as struvite and other phosphate minerals.	Wood ash vs. alkalized biochar: wood ash achieved a higher dehydration rate (11.9 L/day/m^2 at 50 °C) than biochar (6.4 L/day/m^2 at 45°C). Nutrient Retention: Both media retained >70% nitrogen and 100% phosphorus and potassium, but ash showed better dehydration performance.	Volume reduction: achieved >90% urine volume reduction, significantly reducing transportation and storage costs. Nutrient valorization: Provides a sustainable method for nutrient recovery, offering potential for decentralized sanitation systems and reducing reliance on chemical fertilisers.	Sweden
(Tarpeh et al., 2017)	Comparing Ion Exchange Adsorbents for Nitrogen Recovery	wood husk biochar	Pyrolysis temperature: 950 °C for 2 hours. The biochar contained carboxyl functional groups, which contributed to its ion exchange capacity.	Human and synthetic urine	Ammonium adsorption capacity: 1.91 mmol N/g for biochar. Regeneration efficiency: over 100% for biochar, suggesting potential desorption of pre-loaded ammonium or slight decomposition.	Nitrogen recovery: through ion exchange, specifically targeting ammonium ions from source-separated urine.	Biochar vs. other adsorbents: Dowex Mac 3 (4.23 mmol N/g) and Dowex 50 (2.74 mmol N/g) had higher ammonium adsorption capacities than biochar (1.91 mmol N/g). However, biochar and clinoptilolite (2.00 mmol N/g) were more cost-effective due to lower material costs	Cost-efficiency: Biochar is a cost-effective option for nitrogen recovery despite its lower adsorption capacity, especially when considering direct application as fertiliser. Practical use: Biochar could be applied directly as fertiliser after saturation, reducing the need for regeneration and processing.	USA, Switzerland

(Wu et al., 2025)	Nutrient recovery from urine using biochar and Na-chabazite	Wood biochar integrated with Na-chabazite	Pyrolysis temperature: 600–800 °C for 15 minutes under atmospheric pressure. Surface area: 308 m^2/g (increased to 546 m^2/g after steam activation). Modifications: Steam activation and H_2O_2 oxidation, with steam activation improving urea adsorption capacity by 15.6%.	human urine	Urea adsorption capacity: 25.4 mg/g (pristine biochar), improved by 15.6% with steam activation. Total N recovery: 93% from source-separated urine. Total P recovery: 98% through struvite precipitation.	Nitrogen recovery: urease inhibition using Na-chabazite slows urea hydrolysis, allowing biochar to adsorb urea. Urea hydrolysis post-adsorption increases pH for subsequent ammonium recovery via stripping. Phosphorus recovery is achieved through struvite precipitation after urea hydrolysis increases pH.	Biochar vs. Modified Biochar: Steam-activated biochar exhibited superior adsorption capacity compared to H_2O_2-modified biochar. Na-chabazite: dual function as a urease inhibitor and potassium adsorbent, efficiently recovering K+ while slowing urea hydrolysis.	Sustainability: the integrated process enables efficient nutrient recovery (N, P, K) from urine, reducing energy use and greenhouse gas emissions in wastewater treatment plants. Agricultural use: urea-enriched biochar and K-enriched Na-chabazite function as slow-release fertilisers, reducing nutrient leaching.	Canada
(Adhikari et al., 2015)	Use of duckweed-based constructed	Duckweed	The study focuses on duckweed-based constructed wetlands rather than biochar.	Dairy wastewater	Total Nitrogen (TN) Recovery: 22.4 g N/m^2/year on average by harvesting duckweed. Total Phosphorus (TP) Recovery: 7.4 g P/m^2/year on average by harvesting duckweed. Nutrient removal rates: 194.9 g TN/m^2/year and 13.0 g TP/m^2/year in primary wetlands; 104.1 g TN/m^2/year and 9.3 g TP/m^2/year in secondary wetlands.	Nitrogen recovery: uptake by duckweed plants and microbial processes in constructed wetlands (nitrification, denitrification). Phosphorus recovery: plant uptake and sorption/precipitation processes within the wetland substrate.	-	Water quality improvement efficiently treats dairy wastewater by removing nutrients and pollutants. Sustainability: provides a dual function of wastewater treatment and nutrient recovery through duckweed biomass harvesting, which can be used as fertiliser or animal feed.	USA
(Bai et al., 2018)	Recovery of Ammonium in Urine by Biochar Derived from Faecal Sludge	Faecal sludge biochar	Pyrolysis temperature: 600 °C for 70 minutes in a tube furnace. Surface area: 690.8 m^2/g. pH: 10.4. Composition: 84.37% carbon, 2.40% hydrogen, 0.77% nitrogen, 3.13% ash.	human urine	Ammonium removal efficiency: 87.5% under optimal conditions (60 min reaction time, 5 g/L dosage, 1–2 mm particle size). Ammonium adsorption capacity: Up to 19.6 mg/g depending on temperature and particle size.	Nitrogen recovery: achieved through ammonium adsorption.	Biochar demonstrated higher ammonium adsorption than natural zeolite and hardwood biochar but similar to microwave-treated zeolite. The adsorption performance varied with particle size and biochar dosage.	Soil conditioning: application of ammonium-saturated biochar increased soil cation exchange capacity from 8.4 to 13.6 cmol/kg and raised soil pH by 0.9 units over 140 days. Sustainability: Biochar from faecal sludge offers a dual benefit of waste treatment and soil improvement.	China
(Dutta and Vinnerås, 2016)	Fertiliser from dried human urine added to ash and lime	wood ash and slaked lime as drying agents.	-	Human urine	Nitrogen retention: 74% nitrogen retention at 35 °C and 54% at 6 0°C in the final dried fertiliser product. Phosphorus and Potassium: complete recovery due to the retention properties of the ash and lime mixture.	Nitrogen recovery is achieved by maintaining a high pH (>10) using ash and lime, inhibiting urease activity and preventing urea hydrolysis. Phosphorus and Potassium Recovery: Retained in the drying agent during the dehydration process	Ash and lime vs. other methods: urine drying at 35 °C achieved higher nitrogen retention than solar thermal evaporation or chemical precipitation methods. This method also requires less energy and resources, making it more suitable for low- and middle-income regions.	Sustainability: reduces environmental pollution from human waste and minimizes the need for chemical fertilisers. Practical implementation: suitable for eco-sanitation systems, especially in rural or tropical regions, due to the simplicity and low-cost of materials (ash and lime).	Sweden
(Moure Abelenda et al., 2021)	Effects of Wood ash-Based Alkaline Treatment on Nitrogen, Carbon, and Phosphorus Availability in Food Waste and Agro-Industrial Waste Digestates	Wood ash	-	Food waste digestate	Nitrogen recovery: the main route of nitrogen loss was ammonia volatilisation, with rates reaching ~50 mg NH_3-N/kg blend/hour by the end of incubation. The study observed substantial ammoniacal nitrogen retention due to alkaline pH stabilization. Phosphorus recovery: Adsorption of orthophosphate onto ashes was significant.	Nitrogen recovery: hydrolysis at alkaline pH converted organic nitrogen to ammoniacal nitrogen, with high pH (>10) preventing CO_2 release and stabilizing nitrogen in ammonia. Phosphorus recovery: orthophosphate adsorption onto ashes, with desorption inversely related to ash concentration.	The study compares different blending ratios of wood ash and digestates. Higher ash content improved chemical stabilization but increased ammonia volatilisation. The blends acted as slow-release fertilisers, offering an alternative to conventional fertilisers.	Sustainability: utilization of food waste and agro-industrial waste digestates combined with wood ashes enhances nutrient recycling and reduces waste disposal costs. Soil health: the blended materials offer slow-release fertiliser properties, contributing to soil nutrient balance and carbon sequestration.	UK
(Otieno et al., 2021)	Pineapple Peel Biochar and Lateritic Soil as Adsorbents for Recovery of Ammonium Nitrogen	Pineapple peel biochar (PPB) and lateritic soils (LS)	Pyrolysis temperature: 400 °C under vacuum for 4 hours. Surface area: 140 m^2/g. Pore diameter: 23 nm, indicating microporous nature.	Human urine	Ammonium adsorption capacity: 13.40 mg/g for PPB and 10.73 mg/g for LS. Adsorption process: physical adsorption characterized by weak electrostatic forces.	Nitrogen recovery: physical adsorption of ammonium nitrogen (NH_4^+-N) from human urine onto PPB and LS, driven by electrostatic attraction between negatively charged surfaces and NH_4^+ ions.	PPB vs. LS: PPB showed higher adsorption capacity due to its larger surface area and porosity. Comparison with Other Biochars: The adsorption capacity of PPB (13.40 mg/g) is comparable to other biochars like spent mushroom substrate biochar (12.6 mg/g) but lower than rice husk biochar (44.64 mg/g).	Waste management: conversion of pineapple peel waste into biochar offers an eco-friendly way to manage fruit peel residues, reducing methane emissions from landfills and toxic discharges from incineration. Agricultural Use: Enrichment of PPB with NH_4^+-N creates slow-release fertilisers, enhancing soil fertility.	Kenya
(Pradhan et al., 2009)	Stored Human Urine Supplemented with Wood ash as Fertiliser in Tomato Cultivation and Its Impacts on Fruit Yield and Quality	birch wood ash	Pyrolysis temperature: 400 °C under vacuum for 4 hours. Surface area: 140 m^2/g. Pore Diameter: 23 nm, indicating microporous nature	Human urine	Nitrogen recovery: The urine + ash treatment provided 135 kg N/ha and 90 kg P/ha, comparable to mineral fertilisers in yield outcomes. Nitrogen retention was higher in urine-only fertilisation than urine + ash due to potential nitrogen loss from increased soil pH.	Nitrogen recovery: stabilized in the soil through pH regulation and plant nutrient uptake. The urine-only treatment had lower nitrate concentrations, suggesting more efficient nitrogen uptake or reduced volatilisation than urine + ash. Phosphorus recovery: phosphorus was supplemented from wood ash, ensuring sufficient P availability for plant growth.	Urine + ash vs. Mineral Fertiliser: Tomato yields in urine + ash and mineral fertilisation were similar, while urine-only fertilisation produced slightly higher yields than no fertilisation but lower than mineral fertiliser. Nutrient Content: Urine-fertilized tomatoes had higher β-carotene content but lower nitrate levels than mineral and urine + ash treatments.	Sustainability: using urine and wood ash as fertilisers reduces reliance on synthetic fertilisers and promotes nutrient recycling without microbial risks. This method effectively increases crop yields and enhances fruit nutrient content while minimising environmental pollution from chemical fertilisers.	Finland
(Sarkhot et al., 2012)	Impact of Biochar Enriched with Dairy Manure Effluent on Carbon and Nitrogen Dynamics	Hard wood shaving (maple, aspen, choke cherry, and alder) biochar	Pyrolysis temperature: 300 °C (slow pyrolysis). Enrichment: biochar was enriched by shaking with dairy manure effluent for 24 hours, increasing carbon and nitrogen concentrations by 9.3% and 8.3%, respectively	Dairy manure effluent	Nitrogen: enriched biochar reduced net nitrification by 75% and net ammonification by 229% compared to untreated soil.	Nitrogen recovery: reduction in nitrogen losses due to adsorption of ammonium and nitrate, coupled with immobilization facilitated by increased bioavailable carbon in enriched biochar.	Biochar vs. enriched biochar: enriched biochar provided slightly better nutrient retention and reduced greenhouse gas than pristine biochar. Both significantly reduced CO_2 and N_2O emissions compared to untreated soils.	Carbon sequestration: enriched biochar contributes to soil carbon storage, reduces greenhouse gas emissions, and enhances soil physicochemical properties. Nutrient management: reduces nutrient leaching, improves long-term soil fertility, and reduces environmental pollution from manure effluents.	USA
(Senecal and Vinnerås, 2017)	Urea Stabilisation and Concentration for Urine-Diverting Dry Toilets: Urine Dehydration in ash	Wood (birch and wood pellets) ash	-	Human urine	Nitrogen retention: up to 90% nitrogen retention at 35 °C and 66% at 65 °C. Phosphorus and Potassium recovery: The end product contained 2.5% P and 10.9% K, indicating high nutrient retention during dehydration.	Nitrogen recovery: increased urine pH (>10.5) with wood ash inhibits urease activity and prevents urea hydrolysis to ammonia. Phosphorus Recovery: Phosphorus retained in the ash through precipitation of metal phosphates like struvite and hydroxyapatite.	Wood ash vs. other methods: the ash-based dehydration method retained more nitrogen than solar drying and ash/lime bed treatments. The end product is a dry powder, more concentrated and easier to transport and apply than slurry products from other systems.	Sustainability: The process eliminates the need for liquid disposal, reduces storage and transport costs, and provides a concentrated fertiliser comparable to commercial products. Sanitation Benefits: Simplifies urine management in dry toilets, making it a practical solution for decentralized sanitation systems.	Sweden
(Neina and Dowuona, 2013)	Short-term effects of human urine fertiliser and wood ash on soil pH and electrical conductivity	Wood (wood waste and construction timber) ash	-	Human urine	Nitrogen recovery: urine contained 3.93 g/L total nitrogen. The application increased soil pH initially, but nitrification led to acidification over time, reducing pH and possibly affecting nitrogen retention. Phosphorus recovery: wood ash contributed significantly to phosphorus availability in soil.	Nitrogen recovery: Initially increased soil pH due to ammonium deposition, followed by acidification from nitrification of NH_4^+ to NO_3^- over 12 weeks. Ammonia volatilisation was minimal due to soil incorporation of urine and high soil moisture content. Phosphorus Recovery: Enhanced by adding wood ash, which supplied basic cations and stabilized soil pH.	Wood ash vs. Urine Alone: Wood ash doubled the cation content (Ca, Mg, K, Na) in soil compared to urine alone. Combining urine and ash improved nutrient retention and soil pH stability better than urine alone.	Sustainability: using human urine and wood ash as fertilisers reduces dependency on synthetic fertilisers and promotes nutrient recycling in tropical soils. Wood ash mitigates soil salinization risks associated with urine application. Soil Health: Improved pH and cation exchange capacity benefit long-term soil fertility.	Ghana
(Simha et al., 2020a)	Alkaline dehydration of source-separated fresh human urine: Preliminary insights into using different dehydration temperature and media	Various alkaline media: wood ash, calcium hydroxide (lime), wheat bran, biochar (from willow), and desert soil	Pyrolysis temperature: 450 °C. Treatment: mixed with calcium hydroxide in a 75:25 ratio for urine dehydration experiments.	Human urine	Nitrogen Recovery: Over 90% nitrogen recovery was achieved across most media at 60 °C. For example, Char-Lime at 60 °C recovered 98% nitrogen; at 50 °C, it only recovered 11%. Phosphorus recovery: Complete phosphorus retention was observed, with final products containing ~1.5% P.	Nitrogen recovery: inhibited enzymatic hydrolysis of urea due to elevated pH (>12) in the drying media and reduced chemical hydrolysis by efficient dehydration at high temperatures. Phosphorus Recovery: Phosphorus retained in non-volatile forms in the alkaline media.	Biochar (Char-Lime) vs. Other Media: Char-Lime showed excellent nitrogen recovery at 60 °C but poor performance at 50 °C due to pH drop. Wood ash (ash) consistently provided high nitrogen recovery (~98%) at both temperatures. Lime-based media maintained better. alkalinity, enhancing nutrient retention.	Sustainability: this method reduces the volume of human waste and recovers nutrients for agricultural use, promoting eco-friendly sanitation systems. The dry, nutrient-rich fertilisers reduce the need for synthetic fertilisers and minimize environmental pollution from wastewater.	Sweden
(Schmidt et al., 2015)	Fourfold Increase in Pumpkin Yield in Response to Low-Dosage Root Zone Application of Urine-Enhanced Biochar to a Fertile Tropical Soil	*Eupatorium adenophorum* (an invasive forest shrub)	Produced in Kon-Tiki flame curtain kiln; pyrolysis temperature 680–700 °C; pH 9.8; C = 72%; H/Corg = 0.2; BET surface area 215 m^2/g; application rate 750 $kg \cdot ha^{-1}$; quenched with water; used <10 mm particle size biochar	Fresh cow urine	-	Organic coating of biochar surfaces by urine constituents' formation of a hybrid material with improved nutrient capture and slow release; reduction of leaching losses; reversible sorption of nitrate, phosphate, ammonium via water bridges and H-bonds	Urine-biochar: 82.6 $t \cdot ha^{-1}$ pumpkin yield +306% vs. urine-only, +85% vs. biochar-only treatment; biochar-only also increased yield (+119%) vs. urine-only	Reduced N leaching; increased nutrient use efficiency; improved soil water retention; possible enhancement of microbial activity, CEC; low application rate effective (<1 $t \cdot ha^{-1}$) more sustainable practice; potential climate-positive impact.	Nepal

TOF: type of feedstock used, **BCTC**: biochar characteristics and treatment conditions, **TOLW**: type of liquid waste, **N/P RE**: nitrogen and phosphorus recovery efficiency

Figure 2.4 illustrates the materials used for N and P recovery from liquid waste. Biochar was the most commonly used material, appearing in 35% of studies, emphasizing its effectiveness. It was derived from various feedstocks, including pine shells, coconut husks, bamboo, rice husks, sunflower seeds, human faeces, and multiple wood and crop residues. Crop residues, such as oat husks, wheat bran, bark, sawdust, and food waste, were used in 25% of studies, while ash (from construction timber, birch wood, and willow wood) accounted for 17.5%. Sewage sludge and coal fly ash were used in only 2.5% of studies, indicating limited N and P recovery through these materials.

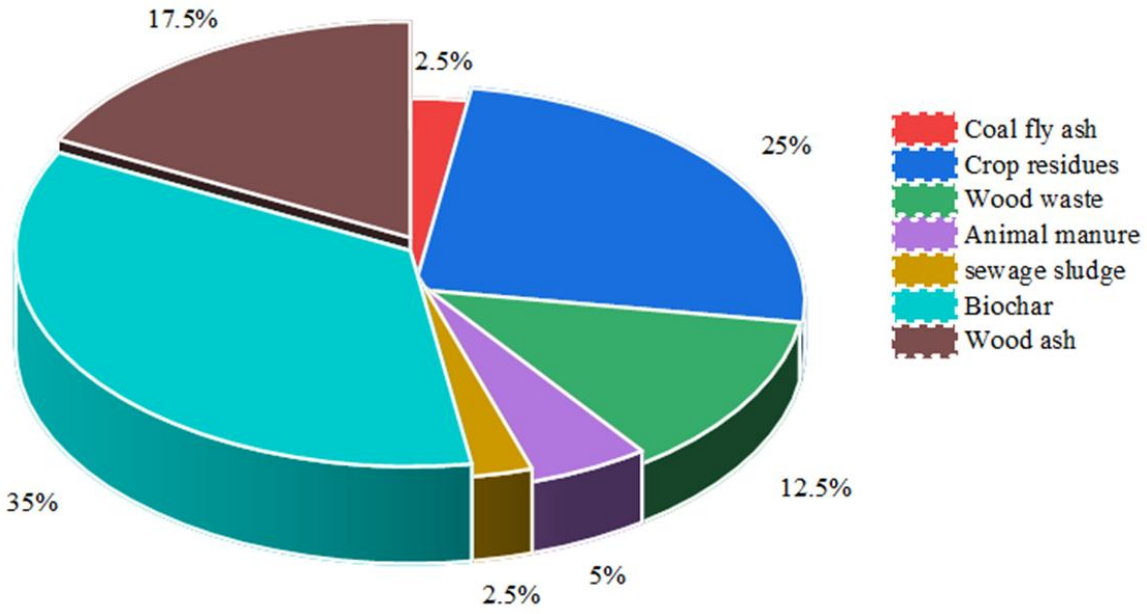


Figure 2.4: Pie chart summarizing 40 materials used to recover N and P from different liquid wastes

2.4 Discussion

This systematic review identified 26 studies focusing on materials used to recover nitrogen (N) and phosphorus (P) from urine and other liquid waste sources. A significant research gap exists in nutrient recovery from diverse liquid waste sources, particularly in developing countries. Only a few studies were based on dehydration experiments (4) of the mixtures. They were more based on adsorption and precipitation experiments. The review found that, per country, a maximum of four publications addressed N and P recovery, with most studies conducted in developed nations.

Most research on N and P recovery from liquid waste originates from North America and a part of Asia (China), while African countries have limited studies despite facing significant waste management challenges. Rapid urbanisation in these regions exacerbates environmental degradation and human health risks, highlighting the urgent need for research and implementation of sustainable nutrient recovery solutions (Nasim et al., 2022). Chianu et al. (2012) emphasized that smallholder farmers in Sub-Saharan Africa (SSA) often suffer from soil nutrient depletion, and utilising municipal waste as fertiliser could help enhance soil fertility. However, the lack of ecological sanitation systems and inadequate waste management infrastructure may contribute to the scarcity of research in developing countries (Hu et al., 2016; Mnkeni et al., 2008; Seetharam, 2015). The decline shown in Figure 2.2 from 2018 may be linked to community resistance and ethical concerns regarding the use of urine as fertiliser in some communities.

2.4.1 Human urine as a nutrient source

Human urine has been widely studied as a liquid waste source for N and P recovery, with 15 studies highlighting its potential as a sustainable nutrient resource. Urine offers a unique alternative to conventional fertilisers due to its ionic nutrient composition, making nutrients readily available for plant uptake (Masrura et al., 2021). On average, urine contains 0.74% nitrogen, 1.62% potassium, 0.029% phosphorus, and 0.90% sodium (Masrura et al., 2021; Mchunu et al., 2018; Mnkeni and Austin, 2009). The nitrogen in urine is primarily in the form of urea (75–90%), while phosphorus is available as soluble phosphate ions, and potassium remains highly soluble (Martin et al., 2022). Other macronutrients, such as calcium and magnesium, further contribute to soil fertility. The nutrient composition of urine aligns well with crop nutrient requirements, making it an effective fertiliser (Heinonen-Tanski et al., 2007; Masrura et al., 2021; Mnkeni et al., 2008).

Direct application of human urine has been successfully tested on crops such as maize (Kassa et al., 2018), tomato (Pradhan et al., 2009), cucumber (Heinonen-Tanski et al., 2007) and spinach (Rumeau et al., 2024). For maize, urine application increased plant height, stem diameter, and leaf length, while also enhancing soil phosphate levels and salinity (Kassa et al., 2018). In tomatoes, urine-fertilized plants produced a fruit yield comparable to those treated with mineral fertilisers and 4.2 times higher than the control (Pradhan et al., 2009). Similarly, cucumber plants fertilized with urine yielded results that were equal to or slightly better than

those grown with commercial mineral fertilisers (Heinonen-Tanski et al., 2007). Schmidt et al., 2015, also observed fourfold increase in pumpkin yield in response to urine-enhanced biochar. Additionally, urine application led to increased shoot biomass in spinach compared to the control (Rumeau et al., 2024). Despite its potential as a fertiliser, urine application poses challenges, including high water content (97%), nitrogen losses through volatilisation, and odour issues (Patel et al., 2020; Senecal and Vinnerås, 2017). The high-water content increases transportation and storage costs, while ammonia volatilisation during storage and application reduces nitrogen composition and contributes to environmental pollution. Addressing these challenges requires innovative treatment and storage solutions to optimize urine use as a nutrient source.

The potential of nutrients in urine has led to various approaches to concentrate nutrients and many has been reviewed in literature (Maurer et al., 2006) including NH_3 stripping (Antonini et al., 2012), nitrification (Udert et al., 2015), electrolysis (Udert et al., 2015) and struvite formations (Etter et al., 2011). However, the practical implication of such treatments is associated with high costs, which may not be applicable in resource poor communities. Also, it focuses on one element to be recovered, e.g., struvite only focuses on P. Innovative methods were adopted to stabilize urea by using acidification or alkalization to inhibit the activity of the urease enzyme (Martin et al., 2022; Senecal and Vinnerås, 2017). Among the numerous technologies for urea removal, adsorption stands out as one of the most thoroughly investigated approaches. It relies on directing a urea-containing solution through a bed of adsorbent material (Weerakoon et al., 2023). Adsorption can be categorised as physical (physisorption) and chemical (chemisorption) sorption. However, some nutrients remain in the effluent after the adsorption process, and strategies that use dehydration could be more effective.

2.4.2 Materials for nitrogen and phosphorous recovery

Various materials have been explored for nutrient recovery, including plant-based feedstocks, biochar, and ash. The choice of material depends on factors such as physicochemical properties, economic feasibility, and environmental benefits. Biochar, in particular, has been extensively studied, with 35% of the reviewed studies highlighting its use for N and P recovery through adsorption. Biochar is produced through the thermal degradation of biomass under low oxygen conditions, resulting in a highly porous material with a large surface area and functional groups that enhance nutrient adsorption (Bista et al., 2019; Kloss et al., 2012; Mohan et al., 2014).

The efficiency of biochar in nutrient recovery depends on feedstock type and pyrolysis conditions (Lehmann and Joseph, 2015). Studies have explored biochar derived from lignocellulosic feedstocks such as crop residues, wood waste, animal manure, and sewage sludge. Findings indicate that biochar adsorbs nutrients and functions as a slow-release fertiliser (He et al., 2022; Hunter and Deshusses, 2020). This agrees with literature by Pathy et al. (2021), who characterised biochars as promising biosorbents because of their high surface area and porous structure. However, surface area alone does not determine nutrient recovery efficiency (Kakaei Lafdani et al., 2021). Slow pyrolysis at 300–900 °C enhances biochar's adsorption capacity through the formation of functional groups such as carbonyl, hydroxyl, and phenolic hydroxyl, which facilitate ion exchange (Harvey et al., 2011; Parida et al., 2006). Biochar generated at lower temperatures (200-300 °C) shows an almost negligible surface charge, leading to the absence of electrostatic attraction with phosphate ions, this reduces its capacity for phosphate adsorption (Yin et al., 2017).

Research by Bai et al. (2018) showed that faecal sludge biochar (produced at 600 °C) has higher ammonium (N) adsorption compared to woody biochar. Faeces are rich in organic matter, making them ideal for biochar production, which effectively adsorbs nitrogen and phosphorus due to high ash content (Asai et al., 2009; Chan et al., 2008). Jiang et al. (2023) found that faecal biochar produced at higher temperatures (500-600 °C) demonstrates superior nutrient recovery due to increased surface functional groups and surface area (Yin et al., 2017). The total phosphorus increased from 66.17 mg L^{-1} (300 °C) to 125 mg L^{-1} (500 °C). This aligns with Alsewaileh et al. (2019) who also noted the suitability of biochar produced at 500-600 °C for N adsorption. Additionally, a mixture of faeces and urine biochar has been shown to enhance N and P retention (Koulouri et al., 2023), but its use in soil can increase salinity, potentially limiting its agricultural applications. However, it is vital to note that nutrient recovery through adsorption and precipitation involves binding of nutrients onto surfaces or forming insoluble compounds, whereas dehydration primary removes moisture (water), thereby concentrating nutrients but not actively binding them. The challenge of salinity is similar to issues raised in studies on urine applications (Simons and Clemens, 2003; Jönsson et al., 2004; Mnkeni et al., 2008), likely due to the high sodium concentration in human urine (Beler-Baykal et al., 2011; Martin et al., 2022). The effects of this increased salinity may vary depending on soil characteristics. For young soils with 2:1 expanding clays, the potential for dispersion is increased when the exchange sites are dominated by sodium (Na), potentially leading to structural degradation. This could be the case in the studies conducted by Mnkeni

et al. (2008) on a young soil (Oakleaf). In contrast, highly weathered soils, stabilised by oxides and containing kaolinite-type clays, tend to be more resistant to dispersion, but the elevated sodium levels can still affect soil fertility. Therefore, the impact of urine biochar on soil salinity must be carefully considered, depending on the soil type.

Ongoing research is focused on developing new structures and altering the surface properties of biochar to enhance its ability to adsorb nitrogen (N) and phosphorus (P) (Pathy et al., 2021). By modifying biochar, it is possible to introduce more acidic and basic groups on its surface, which has been shown to improve its adsorption capacity (Wang et al., 2015). Modified biochar has been found to increase N and P adsorption in comparison to pristine wood biochar (Sarkhot et al., 2012; Fang et al., 2014; Marcińczyk et al., 2022; Wu et al., 2025). These studies highlight the role of biochar modifications in improving nutrient recovery efficiency. This could further optimize nutrient recovery while reducing energy consumption and N and P runoff into water bodies. These engineered biochars enhance adsorption capabilities, pH buffering, and ion-exchange capacity, improving nitrogen retention and enabling slow nutrient release. Currently, there are limited studies in South Africa on recovering nitrogen (N) and phosphorus (P) using biochar made from food waste and other materials, such as pine bark, which are significant waste products in the country.

Ganesapillai et al. (2021) demonstrated that coal fly ash effectively adsorbs nitrogen from synthetic human urine, with phosphorus recovery occurring through struvite precipitation. The study highlighted that higher initial urine concentrations enhance adsorption potential. Utilising coal fly ash for nutrient recovery addresses waste disposal challenges and offers a low-cost alternative to conventional chemical precipitation methods. However, concerns such as potential heavy metal contamination and the long-term stability of recovered nutrients must be addressed to ensure safe application.

Wood waste, particularly from hardwood and softwood species, has been extensively studied for its nutrient recovery potential. The high lignin and cellulose content in wood contributes to biochar stability, enhancing nutrient retention capacity (Bird et al., 1999). Kitchen waste, a heterogeneous biomass source rich in organic matter, has been explored for its ability to recover nutrients through composting, anaerobic digestion, and struvite precipitation. de Kraker et al. (2019) investigated kitchen waste with garden residues for anaerobic digestion and vermicomposting. Anaerobic digestion recovered 12% of nitrogen and 24% of phosphorus, while vermicomposting was more efficient at the household level. These findings suggest that

decentralized organic waste valorisation could enhance nutrient cycling while reducing landfill dependency. Additionally, composting with bulking materials like oat husks, wheat bran, bark, peat, sawdust, and food waste improves phosphorus accumulation and nitrogen retention, making the end-product a safe and effective soil amendment (Oarga-Mulec et al., 2019).

The efficiency of different materials in nitrogen and phosphorus recovery varies based on their physicochemical properties and the mechanisms involved (Pathy et al., 2021). Coal fly ash, for example, provides a cost-effective solution for nutrient recovery while addressing waste disposal concerns (Ganesapillai et al., 2021). However, it is less sustainable than plant-based biochar due to potential environmental concerns associated with heavy metal leaching. Biochar derived from crop residues, wood waste, and kitchen waste exhibits superior adsorption properties and can be engineered to optimise nutrient recovery (Pathy et al., 2021). Mg-modified biochar demonstrates higher phosphorus adsorption capacity than unmodified biochar, making it a viable alternative to synthetic fertilisers (Marcińczyk et al., 2022). Additionally, lignocellulosic biomass-based biochar, such as wheat straw and sawdust, enhances structural crystallisation performance, thus improving phosphorus recovery (Krishnamoorthy et al., 2023).

Although many studies have been conducted on the recovery of N and P from wastewater and urine using biochar, these studies focus on nutrient recovery through adsorption and precipitation. However, ash-based materials, including wood ash and alkaline media, have been used for urine nutrient stabilization to preserve N as urea from urine in order to remove excess water by dehydration (Senecal and Vinnerås, 2017).), which has shown some potential. This is achieved by inhibiting urease enzyme activity, which is inhibited by temperature and pH (Hellström et al., 1999; Hotta and Funamizu, 2008; Senecal and Vinnerås, 2017). The literature presents some discrepancies regarding the pH range for urease activity. Rektorschek et al. (1998) report that urease can be inhibited at pH values below 3.5 or above 8.5, whereas Chipako and Randall (2020) suggest inhibition occurs at pH levels below 6.5 or above 11. Such discrepancies highlight the need for further research to precisely determine the pH sensitivity of urease under different environmental conditions. Simha et al. (2018) examined urine dehydration using chopped willow tree ash and alkalized biochar. They recovered >70% of N at all dehydration temperatures (40, 45 and 50 °C), and the recovery was due to inhibition of urease enzyme high as a result of high pH >12 and chemical hydrolysis was reduced because of high temperatures. An exception was for alkaline biochar, which recovered 11% at 50 °C

due to pH drop activating the urease activity. Similarly, Dutta and Vinnerås (2016) demonstrated that Birch wood ash and slaked lime effectively prevented nitrogen loss by maintaining a high pH (>10).), with low dehydration temperature (35°C) recovering more N than at 60 °C. The low recovery in higher temperatures could be attributed to chemical hydrolysis since the treatments had a pH of >10. Under such alkaline conditions, urea can spontaneously hydrolyse into ammonia and carbon dioxide without the need for urease enzyme activity. This non-enzymatic hydrolysis is driven by the high pH and temperature, which accelerate the breakdown of urea. Senecal and Vinnerås (2017) also obtained similar results on wood (birch and wood pellets) ash. This cost-effective approach is particularly suitable for low- and middle-income regions. Nitrogen retention was highest at 35 °C, making this method viable for eco-sanitation applications in low-resource settings The drying process took 46 days at 35 °C (3.8 L/m^2) and 41 days at 65 °C (8.9 L/m^2), with urine added four times a week. Ash was added either once (Static, 50 g) or with each application (Dose, 5% w/w). Although this extended duration may be a consideration for practical implementation, the high nutrient retention makes it a promising method for eco-sanitation applications in low resource settings. The potential scalability of alkaline-based nutrient recovery methods warrants further exploration for large-scale agricultural and waste management applications. This also opens a room for other waste products to be tested for the ability to inhibit urease enzymes. Currently, there are few to no studies on N and P recovery using acidic materials. This gap presents an opportunity for further research on the recovery of both nitrogen (N) and phosphorus (P) using acidic materials, including pine bark, which is a major waste of the forestry industry, in sub-Saharan Africa.

Despite the advantages of nutrient recovery technologies, challenges remain. Studies such as those by Senecal and Vinnerås (2017) showed that while nitrogen losses during urine dehydration were generally low (<5% of total nitrogen), these losses, mostly as free ammonia, could become more pronounced under certain conditions such as high temperatures. Additionally, concerns over biochar salinity and its impact on agricultural applications, as noted by Koulouri et al. (2023), require further investigation. Other waste products, such as pine bark and potato waste, should also be tested for nutrient recovery, as they are produced in large quantities in South Africa. Additionally, research on this topic in the country is still limited, highlighting the need for further studies. Addressing these challenges through improved material engineering, and process optimisation will be essential for scaling nutrient recovery technologies.

Furthermore, urine-treated biochar could be a valuable soil amendment that improves soil structure and nutrient availability (Bai et al., 2018; Pradhan et al., 2009). While biochar exhibits numerous advantages, some challenges remain, such as media clogging, organic leaching, and the need for modifications to improve selectivity (Huang et al., 2019). For instance, biochar derived from coconut husks demonstrated effective denitrification but suffered from reduced phosphate removal over time due to adsorption saturation (Hunter and Deshusses, 2020). Similarly, some biochars exhibited salinity issues that may limit their direct agricultural use.

Beyond nutrient recovery efficiency, these materials offer significant environmental benefits. Biochar derived from lignin-rich crop residues and wood waste contributes to carbon sequestration, mitigating greenhouse gas emissions (He et al., 2022). Utilizing industrial by-products like fly ash for nutrient recovery reduces waste disposal concerns and enhances resource circularity (Ganesapillai et al., 2021). Furthermore, applying recovered nutrients in agriculture as slow-release fertilisers decreases reliance on synthetic fertilisers, lowering costs and minimising environmental pollution. Optimising these nutrient recovery strategies can support sustainable agricultural practices and contribute to circular economy solutions.

2.4.3 Agronomic implications of nutrient recovery

Using plant-based feedstocks, biochar, and ash for nutrient recovery presents numerous environmental and practical benefits. These materials support waste valorisation by converting organic and industrial waste into valuable fertilisers, promoting circular economy principles. Biochar production from agricultural residues enhances nutrient recovery and contributes to carbon sequestration, mitigating greenhouse gas emissions (He et al., 2022). Nutrient-enriched biochar-based fertilisers improve soil health, and water retention in agricultural applications (Bai et al., 2018). The slow-release nature of nutrients recovered via biochar-based products reduces nutrient leaching and enhances crop productivity (Marcińczyk et al., 2022). Additionally, decentralized urine-diverting sanitation systems with biochar and wood ash-based treatment offer a practical resource recovery solution in rural and peri-urban areas.

Integrating various materials in nutrient recovery from liquid waste represents a promising approach to sustainable resource management. While further optimisation and scalability studies are needed, the potential benefits of biochar, plant-based feedstocks, and ash in nutrient recycling make them valuable tools in addressing global challenges related to wastewater

treatment, agricultural sustainability, and environmental conservation. The practical implications of nutrient recovery strategies extend beyond wastewater treatment. The use of biochar and ash-based fertilisers has significant benefits for agriculture. Despite the development of various urine-based products, there has been limited testing of their agronomic effectiveness, as found in one study. Tomato yields from co-application of urine with birch wood ash was comparable to mineral fertilisers. However, urine-only fertilisation resulted in slightly higher yields compared to no fertilisation, yet lower than those from mineral fertilisers (Pradhan et al., 2009). The effects of application of solid urine-based fertiliser materials is yet to be evaluated.

Bai et al. (2018) found that ammonium-saturated biochar increased soil cation exchange capacity and improved soil pH, contributing to long-term soil fertility. This demonstrates the potential for integrating nutrient recovery methods into agricultural systems for improved soil health and crop productivity. Future research should focus on the effects of plant-based feedstocks, biochar and ash-based fertilisers on nutrient cycling to ensure sustained benefits and test other waste materials for N recovery and their potential as soil amendments. Economic and environmental sustainability also play a crucial role in nutrient recovery applications. Studies such as that by Krause and Rotter (2018) in Tanzania highlight the benefits of using biochar and compost from cooking and sanitation processes to improve soil fertility.

Similarly, de Kraker et al. (2019) analysed decentralized waste valorisation systems in Chile, emphasizing the cost savings and environmental advantages of vermicomposting and anaerobic digestion. These approaches reduce landfill use, lower transportation emissions, and enhance urban agriculture. Expanding such initiatives on a global scale, including many countries in Europe, Asia, and Africa could provide localized, sustainable solutions for waste management and nutrient recovery, contributing to circular economy principles. Using lignocellulosic and non-lignocellulosic biomass for nutrient recovery has far-reaching environmental and agricultural benefits.

Firstly, it reduces reliance on synthetic fertilisers, which are energy-intensive to produce and contribute to environmental pollution through leaching and runoff. Secondly, biochar-based nutrient recovery supports carbon sequestration by stabilizing organic matter in the soil, thereby mitigating climate change (He et al., 2022; Moure Abelenda et al., 2021; Sarkhot et al., 2012) and improves soil fertility (Bai et al., 2018). Despite these advantages, challenges remain in optimizing the performance of biochar-based nutrient recovery systems. Factors such

as feedstock type, pyrolysis temperature, surface modification, and nutrient binding efficiency influence the overall effectiveness of these materials. Future research should focus on fine-tuning biochar production processes to maximise nutrient recovery while minimising production costs and environmental footprints.

Previous studies show that potato waste has an alkaline pH while pine bark has an acidic pH (Vilakazi et al., 2023). Pine bark is rich in lignin and cellulose content and can show similar results to those noted by (He et al., 2022), and the PB acidity can further contribute to N and P adsorption (Wang et al., 2015). This implies that these waste products could be used as materials for N and P recovery in liquid waste. South Africa produces 1.09 million tons of potato waste annually (Potato South Africa, 2010); of these, cull potatoes and potato peels are commonly discarded at landfills (dumpsites) or other disposal sites, while other portions are converted into livestock feed and compost (Charmley et al., 2006). While data on quantities of pine bark production in South Africa could not be found in the literature, an estimated annual production of softwood bark was about 1.5 million m^3 in 1985 (Smith, 1985). As such, pine bark is a major waste of the timber industry in South Africa and contains high carbon content. In contrast, no published research on recovering N and P from liquid waste could be accessed in the literature, especially in South Africa.

2.5 Conclusion

This systematic review highlights the potential of plant-based feedstocks, biochar, and ash for N and P recovery from liquid waste, particularly human urine. The findings indicate that these materials are crucial in sustainable nutrient management, reducing environmental pollution, and improving agricultural productivity. Biochar, in particular, has been extensively studied, demonstrating superior adsorption properties and slow-release nutrient capabilities, making it an effective amendment for soil fertility improvement. However, its efficiency varies based on feedstock type, pyrolysis conditions, and surface modifications.

Wood ash and alkaline-treated biochar have shown promise in stabilizing urine nutrients by preventing nitrogen losses through urea hydrolysis inhibition, offering a cost-effective and scalable solution for nutrient recycling. Coal fly ash and other industrial by-products also exhibit potential for nutrient recovery, though concerns regarding heavy metal contamination and long-term soil health implications require further investigation. Despite these advances,

significant research gaps remain, particularly in developing countries where nutrient recovery technologies could significantly enhance agricultural sustainability and waste management. the only studies that stabilised N, by reducing urease activity, followed by dehydration have been limited to wood ash, with no accessible literature in this regard of the direct use of the feedstocks and biochar. Even though there are studies using ash to inactivate urease, the ash used was from birch would, with none reported on ash from major organic wastes in South Africa such as pine bark and potato wastes. The limited studies on the agronomic benefits of ash-based fertilisers suggest, reported in the literature, and those of dehydration products from locally available feedstocks and biochar types suggest need further trials to validate their effectiveness across different soil types and cropping systems. Integrating biochar, ash, and plant-based feedstocks into nutrient recovery systems may present an environmentally sustainable approach to addressing global challenges in wastewater management and soil fertility enhancement

Chapter 3: Nitrogen recovery from synthetic human urine with feedstock and biochar from cull potato and pine bark due to contrasting dehydration conditions

Abstract

Inadequate sanitation facilities prevalent mostly in informal and unplanned urban settlements often leads to poor waste disposal and consequently environmental pollution and increases risks to human health. The introduction of onsite innovative sanitation technologies that allow for the recovery of nutrients could provide solutions to the challenges of human waste management. Human urine contains essential plant nutrients such as nitrogen (N) and phosphorus (P), with up to 97% of the urine being water, and strategies to remove excess water and concentrating nutrients are needed. This study investigated the effects of feed stocks, ash and biochar types from cull potatoes (CP) and pine bark (PB) for recovery of N from artificial human urine (AHU) at contrasting dehydration conditions, including at two oven temperatures and in a glasshouse. The biochar from CP and PB were pyrolysed at 350 and 650 °C for 2 hours. Additionally, another PB biochar was produced using a kiln., while ash from the different two feedstocks were produced by heating in a muffle furnace at 750 °C for 6 hours. The AHU was added to each medium (50 g) at 4.5 L/m² and 5.1 L/m² and dehydrated in the oven at 45 and 60 °C, respectively. For the glasshouse dehydration the urine was added at 5.1 L/m^2. The temperature in the glasshouse ranged from a minimum of 9 °C to a maximum of 57 °C. The urine was added six times at 29 and 33 ml each time and kept at dehydration temperatures of 45 and 60 °C, respectively, throughout the experiment. Therefore, cumulative urine additions at days 0, 14, 21, and 28 were 0, 58, 116 and 174 ml kept at 45 °C and 0, 66, 132 and 198 ml at 60 °C, with dehydration in between. Samples of the urine-products were collected before each urine addition, and analysed for pH, total carbon (C), and nitrogen (N) and total N recovered from AHU was calculated. The different media from PB recovered more N than those from CP. The 60 °C temperature promoted rapid dehydration than 45 °C. The most N was recovered on PB biochar pyrolysed at 650 °C with dehydration at 60 °C. The CP feedstock also recovered more N than corresponding ash and biochar from CP, which showed negative N recovery. These findings suggest that feedstock, biochar and ash from PB and the feedstock of CP can be effectively used to recover N from human urine, particularly when dehydrated at 60 °C.

Keywords: Dehydration; biochar, recovery, artificial urine, alkalinisation; acidification

3.1 Introduction

Problems caused by lack of proper sanitation facilities worldwide are becoming increasingly severe, with over 80% of human excreta and wastewater being discharged to the environment with little or no treatment (van den Broek et al., 2024). The exponential increase in population will inevitably result in more waste being discharged into the environment, leading to contamination of water, soil, and the atmosphere, and significantly impacting public health (van den Broek et al., 2024). Ecological sanitation is an important initiative to address these challenges, and focuses on closing the nutrient loop, through recovering nutrients and organic matter (Junghanns and Beery, 2020). With this approach, human excreta are viewed as a valuable resource and not a waste. Consequently, there is an increasing interest in the fertiliser value of human waste.

Urine contains considerably more nutrients compared to feaces (Senecal and Vinnerås, 2017). On average, urine contains 0.74% nitrogen (N), 1.62% potassium (K), 0.029% phosphorus (P), and 0.90% sodium (Na) (Mnkeni and Austin, 2009; Mchunu et al., 2018), in ionic form, with N mainly in the form of urea (Mnkeni and Austin, 2009). About 75-90% of N in human urine is in the form of urea, similar to nitrogen-rich fertiliser. The alkaline pH of stored urine makes itself sterilising but encourages the loss of N through ammonification (Geinzer, 2017). The high amount of water (approximately 97%) in human urine makes it bulky, with nutrients occurring at low concentration (Senecal and Vinnerås, 2017). The bulkiness increases logistics and costs of storage, transportation and application, which limits its competitiveness against commercial fertilisers. While approaches to concentrate the nutrients, including NH_3 stripping, nitrification, electrolysis, struvite formation, and reverse osmosis have been successful in the recovery of the nutrients, the high cost makes their practical application limited, especially in resource poor communities. Alternative cost-effective strategies that remove the excess water, while concentrating nutrients, are of paramount importance. However, urea in the urine is prone to hydrolysis and ammonia volatilisation, resulting in N losses, during dehydration.

Following excretion, the urea is immediately hydrolysed to ammonia by the urease enzyme (Senecal and Vinnerås, 2017). Pre-treatment of the urine to limit urease activity in order to allow for dehydration has been suggested (Senecal and Vinnerås 2017). The urease can be inhibited by pH adjustment and temperature (Hellström et al., 1999; Hotta and Funamizu, 2008), and also using hydrogen peroxide (Arve et al., 2024). Literature shows that the activity of the urease enzyme is limited by pH <3.5 or >8.5 (Rektorschek et al., 1998). In the humid

northern temperate zones, alkalinisation has been the center of attention for stabilising urine as there are plenty of available strong bases (FAO 1986). A previous study reported that wood ash with pH >12.5 minimised urease activity, retained N and allowed for dehydration (Dutta and Vinneras, 2016; Senecal and Vinnerås, 2017). Although many nutrient recovery technologies remain at the laboratory stage, urine dehydration has advanced to pilot scales. For example, an alkaline dehydration (wood ash+lime) treating ~30 L/day of source-separated urine was field-tested in Finland, yielding end-product with 1.4% N, 0.9% P, and 8.3& K in solid fertiliser form under real-use conditions (Simha et al., 2020b). Other materials with either alkaline or acidic pH may also allow for successful dehydration with recovery of urea, hence producing a valuable by-product.

Ashes and biochar from various organic wastes may have the desirable high pH, for recovery of N from human urine. In addition to pH effects, surface characteristics including the functional groups of biochar have been shown to retain N from urine through adsorption (Harvey et al., 2011). The application of human urine to biochar, a carbon-rich substrate, may help recover the N while the dehydration product may have potential to improve soil fertility and sequester carbon (C) (Jeffery et al., 2015). However, the effectiveness of the ash and biochar could depend on the characteristics of the feedstock and pyrolysis conditions (biochar) used to produce them and the temperature used for dehydration. Studies indicate that cull potatoes have a relatively high pH of 8.12 (with 39% C), while their biochars pyrolyzed at 350 and 650 °C exhibit even higher pH values of 10.3 and 11.6 (with 66.1% and 71.5% C), respectively (Vilakazi et al., 2023). In contrast, pine bark has a lower pH of 4 (with 50.9% C), with its biochars pyrolyzed at 350 and 650 °C showing pH levels of 4.78 and 9.10 and higher C of 70.3% and 90.1%, respectively (Vilakazi et al., 2023).

The addition of feedstock these feedstocks, ash and biochar types from cull potato (CP) and pine bark (PB) to urine, followed by dehydration could remove the excess water and produce a nutrient-rich product. The nutrient-rich solid products could make it easier to store, transport and apply the N, (from products with consistent concentrations), as fertiliser than liquid urine, for which the nutrient concentration can be variable. While this appears to be theoretically feasible, the effectiveness of these materials has not been tested for recovering nutrients from human urine, according to the available literature. Therefore, the objective of this study was to evaluate the effects of products from potato and pine bark and contrasting dehydration temperatures on retention of nitrogen from artificial human urine.

3.2 Methods and materials

3.2.1 Preparation of solid media

The solid materials (feedstock, ash and biochar) used in this study were produced from cull potato (CP) and pine bark (PB) materials. The cull potatoes (CP) were collected from the Pietermaritzburg Fresh Produce Market located at Mkondeni (29°38'30" S 30°24'46" E), Pietermaritzburg., South Africa. The pine bark (PB) was collected from a private forestry by-product factory located at Cramond (29°24'50" S 30°25'42" E), Pietermaritzburg., South Africa. The biochars were produced by pyrolysis at 350 and 650 °C in a muffle furnace (Labcon, Type RM4), where the temperature was raised to set levels at a rate of 10 °C /min (Enders et al., 2012). The materials were carbonized for 2 hours per pyrolysis temperature (Koetlisi and Muchaonyerwa, 2018). The ash was produced by combustion for 6 hours in a muffle furnace set at a temperature of 750 °C. Another PB biochar was produced using a two-container kiln at Ukulinga Research Farm, which was fabricated by the School of Engineering, at the University of KwaZulu-Natal. The kiln was a drum of 860 mm diameter and was made of mild steel plate of 3 mm thickness. While the furnace drum had 1260 mm diameter and was made of 2 mm galvanise steel sheet. The lid for sealing the kiln drum was 730 mm in diameter and was made of 3 mm mild steel sheet. For closing the furnace drum, a ring of 860 mm internal diameter and 1260 external diameter was made of a 2 mm thick galvanised sheet. The kiln was designed to reach a temperature of 400 °C. The operational conditions were careful controlled to ensure consistency throughout the production process.

The original feedstocks, biochar and the ash were analysed for ultimate composition, and surface functional groups as described by Koetlisi andand Muchaonyerwa (2018). The ultimate C and N compositions were analysed by the dry combustion using the Leco Trumac (CNS) autoanalyser instrument (Leco corporation, 3000, Lakeview, Ave, ST, Joseph, MI, USA). The surface functional groups were analysed using the Fourier Transformed Infrared (FTIR) spectra using Perklin Elmer FTIR spectrometer (FTIR-100) in the region of 400-4000 cm^{-1} (Koetlisi and Muchaonyerwa, 2018).

3.2.2 Artificial human urine

The artificial human urine (AHU) was prepared according to the protocol described by Chutipongtanate and Thongboonkerd (2010). The use of artificial human urine in this study

was primarily aimed at ensuring experimental consistency, and control of nutrient composition across treatments. Although artificial urine does not fully replicate the microbial and organic complexity of real human urine (Simha et al., 2024), it remains a valuable research tool under controlled experimental conditions. Its use allows standardisation of nutrient concentrations, minimises variability associated with donor diet and storage. This approach ensured that observed differences in nutrient recovery and plant response could be attributed to treatment effects rather than inconsistencies in urine composition. The components in AHU are within the physiological range of normal human urine and are summarized in Table 3.1. All the chemicals used were of analytical grade (AR) and the solution was prepared with deionised water (18.2 MΩ cm) obtained from an Aqua MAXTM ultra 370 series. The amount of each chemical used to make up 1L of artificial human urine (AHU) were detailed in Table 3.1.

Table 3.1:The chemical composition and amount of each chemical used to make up the artificial human urine (AHU)

Chemical composition	**g/L**
Urea	12.135
Uric acid	0.17
Creatinine	0.45
Tri-sodium citrate dihydrate	1.485
Sodium Chloride	3.17
Potassium Chloride	2.25
Ammonium Chloride	0.805
Calcium Chloride	0.445
Magnesium Sulphate	0.5
Sodium bicarbonate	0.17
Sodium Oxalate	0.015
Disodium Sulphate	1.29
Monosodium phosphate	0.5
Disodium phosphate	0.55

3.2.3 Experimental set-up

The first experiment was conducted with CP based media (feedstock, biochars and ash) in ovens set at of 45 and 60 °C, with each treatment tested in triplicates. A sample of 50 g of each medium was initially added to storage glass container, followed by AHU application at a rate of 4.5 (45 °C) and 5.1 (60 °C) L/m^2, and after each urine application, the mixtures were stirred for 5 s (Senecal and Vinnerås, 2017) and then allowed to dehydrate in ovens set at 45 and 60

°C. The experiment was run for 28 days with cumulative AHU addition of 0, 58, 116 and 174 ml for 45 °C and 0, 66, 132 and 198 ml for 60 °C on days 0, 14, 21, and 28, with sampling just before urine addition. The products were analysed for pH and total C and N. The experiment was repeated with PB based media (feedstock, biochars and ash). The organic materials used in this study were not sterilised. Therefore, urease may originate from the materials used.

The same experimental set up was repeated with PB feedstock, PB kiln biochar and PB ash and dehydration was conducted under glasshouse conditions (G), with the cooling system switched off and no temperature control, resulting in temperatures ranging from a minimum of 9 °C to a maximum of 57 °C. This was included with view of the need to produce materials on a large-scale. A sample of 7 kg of each medium was added initially to plastic basins, followed by AHU application at a rate of 5.1 L/m^2, and after each application the mixtures were stirred and allowed to dehydrate. The selection of the pine bark materials was based on the overall superior performance of all PB treatments, in terms of recovering N from urine, compared to the cull potato waste treatments, at 45 and 60 °C.

3.2.4 Analysis

The changes in pH of the dehydration media were determined in fresh urine at a ratio of 1:5 (Simha et al., 2021). For these analyses, 5 g of the dried medium+AHU from the glass container were mixed with 25 ml of fresh AHU and left to sit for 1 hour in sealed centrifuge tubes before pH measurement. After the measurement, the samples (mixture) were returned to the glass containers. Total C and N were analysed, on 0.2 g sample (<250 µm), by dry combustion using the Leco Trumac (CNS) auto-analyser (Leco corporation, 3000, Lakeview, Ave, ST, Joseph, MI, USA). The total recovered nitrogen was calculated as the difference between the final nitrogen content in the urine-based products after the dehydration experiment and the initial nitrogen content in the medium [total N recovered (%) = N in the final urine-product (%) – Initial N in the medium (%)].

3.2.5 Statistical analysis

The results of each of the parameters studied were subjected to the two-way analysis of variance (ANOVA), separately for different sampling days, for each dehydration condition, using GenStat 18th edition (VSN International 2015). The statistical analysis for the glasshouse dehydration study was conducted separately using one-way analysis because it focused solely

on PB products. Means separation between treatment was done using least significant difference (LSD) and Tukey test at $p < 0.05$.

3.3 Results

3.3.1 Characteristics of media

The selected characteristics of the media are presented on Table 3.2 and show highly significant ($p<0.001$) differences between the materials. The pH was significantly higher in CP ash (12.43) while the PB treatment (3.02) had lower compared to all other treatments. The PB ash and PB650 biochar had higher pH than the rest of the other treatments, followed by CP650 biochar, and then CP and CP350 biochar, with PB350 biochar having higher pH than only the PB feedstock. The CP ash treatment had significantly higher total P (1.52%) and total K (3.82%) compared to all the treatments, while the CP biochars had higher K than CP feedstock and all material from PB. For all the PB media (except PB ash) the concentration of P was below detection. The PB biochar (350 and 650 °C) and kiln biochar media had low calcium carbonate equivalent (CCE), while CP ash had the highest CCE (57.1%). Calcium carbonate equivalent for the different media ranged from 8.01% in the PB 350 to 57.1% in the CP ash treatment, and for the CP materials CCE was in the order CP ash > CP650 >CP 350>CP. Total C, N and H were significantly lower for both the ash media, while PB 650, CP 350 and CP media had the highest total C, total N, and total H; respectively. Total C ranged from 2.22% in the PB ash to 90.1% in the PB 650 treatment. For total N, it ranged from below detection in the CP and PB ash to 2.44% in the CP 350 treatment.

Table 3.2: Characteristics of the media

Media	pH	P	K	CCE	C	N	H	C/N
		%						
CP	7.97c	0.07[a]	0.570[a]	11.3[c]	38[b]	1.13[c]	6.20[f]	34.2[a]
CP 350	8.18[c]	0.946[b]	2.75[b]	15.4[d]	62.2[d]	2.44[e]	4.38[d]	25.5[a]
CP 650	8.65[d]	0.958[b]	3.00[bc]	23.7[e]	68.3[e]	1.84[d]	1.52[b]	37.1[a]
CP ash	12.4[g]	1.52[c]	3.82[c]	57.1[f]	2.45[a]	0.00[a]	0.730[a]	-
PB	3.02[a]	0.00[a]	0.218[a]	11.8[bc]	49,8[c]	0.295[b]	5.29[e]	169[b]
PB 350	4.78[b]	0.00[a]	0.351[a]	8.01[a]	70.3[f]	0.346[b]	3.78[d]	203[c]
PB 650	9.10[e]	0.00[a]	0.267[a]	9.43[ab]	90.1[g]	0.367[b]	2.19[c]	246[d]
PB ash	10.1[e]	0.176[a]	0.642[a]	18[d]	2.22[a]	0.00[a]	0.221[a]	-

*Values on the same column with different letters indicate significant differences ($p<0.05$). CCE= Calcium carbonate equivalent; C = total carbon content; N = total nitrogen content; H = total nitrogen content; CP= Cull potato waste; PB= pine bark waste.

The chemical functional groups for the media are shown in Table 3.3. All the media showed a band at 680-900 cm^{-1}, which are assigned to C-H bending of aromatic. The O-H stretching (3200-3570 cm^{-1}), N-H secondary amine (1550-1650 cm^{-1}) and C-O stretching (1050-1150 cm^{-1}) were visible in the raw CP and PB media. Heterocyclic amine and sulfate ions were present in CP 350 biochar and stretching aldehyde was only visible in CP 650 biochar. Subjecting the CP to pyrolysis led to the formation of O-H stretching of carboxylic acid (2500-3000 cm^{-1}) and carbonates (1410-1490 cm^{-1}) functional group. Aliphatic C-H stretch (2850-2950 cm^{-1}) were observed in both the raw substrates while subjecting the CP to pyrolysis at 350 °C decreased the band as shown by the decrease in frequency, wavenumber (cm^{-1}) and pyrolysis at 650 °C wiped out the band. For the CP media, the alkyne, metal carbonyl (1800-2100 cm^{-1}), and C=C bending of aromatic C (1500- 1700 cm^{-1}) were visible except in the CP ash. The C-H bending of aromatic C, O-H bending of phenol functional groups were visible throughout all CP media.

The raw PB media had metal carbonyl, stretching aldehyde, C=C bending of aromatic C, carbonates ions, ether bonds, phosphate ions and C-H bending of aromatic C bands, and subjecting the PB to high temperature led to a total disappearance of stretching aldehyde, C=C bending of aromatic C, while some bands were increased (phosphate ions, ether bonds, carbonates, metal carbonyl and C-H bending or aromatic C). The carbonyl functional group band increased with pyrolysis temperature for all biochars. An increase in temperature for PB to ash gave rise to bands including aromatic phosphate and alkyne.

Table 3.3:Fourier Transform Infrared Spectroscopy (FTIR) Spectra visible for media from potato waste and pine bark.

Frequency, Wavenumber (cm^{-1})	Functional group	CP				PB				
		0 ˚C	350 ˚C	650 ˚C	ash	0 ˚C	350 ˚C	650 ˚C	ash	Kiln biochar
3570-3200	O-H stretching	3265.08				3285.59				
3490–3430	Heterocyclic amine		3436.44							
3000-2500	O-H stretching of carboxylic acid		2595.38	2649.308	2997.57		2659.71			2666.12
2950-2850	aliphatic C-H stretch	2922.29	2919.42			2917.14	2918.08			
2260-2100	Alkyne	2175.69	2107.99	2111.726			2106.62	2149.17	2197.69	2112.49
2100 - 1800	Metal carbonyl	1876.04	1947.52	2038.011		1881.16		2086.94	2014.30	2004.33
2000 -1900	C=C-C stretching			1992.611			1911.56	1989.9		1933.5
2000 -1750	Aromatic							1797.53		
1740-1690	Stretching aldehyde			1720.777		1730	1710			
1700-1500	C=C bending of aromatic C	1633.1	1564.06	1544.955		1557.12	1589.07	1563.74		1572.23
1650 - 1550	N-H Secondary amine	1556.81				1510.98				
1490-1410	Carbonate ions		1424.97	1442.518	1444.54	1440.89	1433.69		1419.83	1425.22
1410-1310	O-H bending of phenol	1403.82	1370.71	1359.074	1367.69					
1350 - 1330	C-H bending	1336.67								
1300 - 1000	ether bonds -C-O-C	1243.02	1250.27		1121.68	1263.07			1034.93	
1150 - 1050	C-O stretch	1147.912				1149.98		1148.78		1125.14
1130 - 1080	Sulfate ions		1116.03							
1100-1000	Phosphate ions	1082.29				1026.04		1009.71	1047.71	
995 - 850	Aromatic phosphate	961.258			982.48				984.34	
900-680	C-H bending of aromatic C	806.492	722.613	840	881.64	811.869	809.035	799.303	771.46	775.36

*CP= cull potato waste; PP= peel potato waste; PB= pine bark

3.3.2 Changes in pH

At the beginning of the experiment, all the treatments were basic (pH > 8), except for PB (pH 3.98) and PB 350 (pH 4.78), which were acidic. The pH changes in the media were similar at both dehydrating temperatures (Figure 3.1a and b). The addition of the first volume of urine resulted in pH increase for the PB and PB 350 and decrease for PB 650 and PB ash, with no further change with addition of more urine, irrespective of dehydrating temperature (Figure 3.1a and b). While pH of CP and CP ash did not change with urine addition, that of CP 350 and CP 650 was increased by the addition of the first two volumes of urine with no further increase with third and final volume, irrespective of dehydrating temperature (Figure 3.1a and b). The CP ash had significantly higher pH than the rest of the treatments throughout the dehydration experiments, reaching a maximum of pH 12 (Figure 3.1a and b). At the end of the experiment, pH under the contrasting dehydration temperatures had a similar trend, i.e. CP ash > CP350 =CP650 >PB ash > PB650 =PB350=PB (Figure 1a and b). The PB products dehydrated at the glasshouse (PB, kiln biochar and ash) (Figure 3.1c) followed a similar trend to the corresponding PB products (PB, PB 350 and PB ash) dehydrated at the 45 and 60 °C (Figure 3.1a and b).

3.3.3 Changes in total nitrogen

The cull potato feedstock and biochars, produced at 350 and 650°C, initially had higher total N content compared to those from pine bark and the ashes (Figure 3.2a and b). In the dehydration temperature experiments (45 and 60 °C), the N content in the cull potato biochars decreased with the addition of the first volume (58 ml) of urine, while the N content in the other treatments increased. Further volumes of urine increased N in all the treatments, with only PB 350 and PB 650 biochars approaching a maximum (Figure 3.2a and b). The ash from both pine bark and cull potato had a significantly lower N concentration compared to other treatments throughout the dehydration experiment. Urine addition under glasshouse dehydration increased total N in all treatments, with only PB and PB ash approaching a maximum (Figure 3.2c).

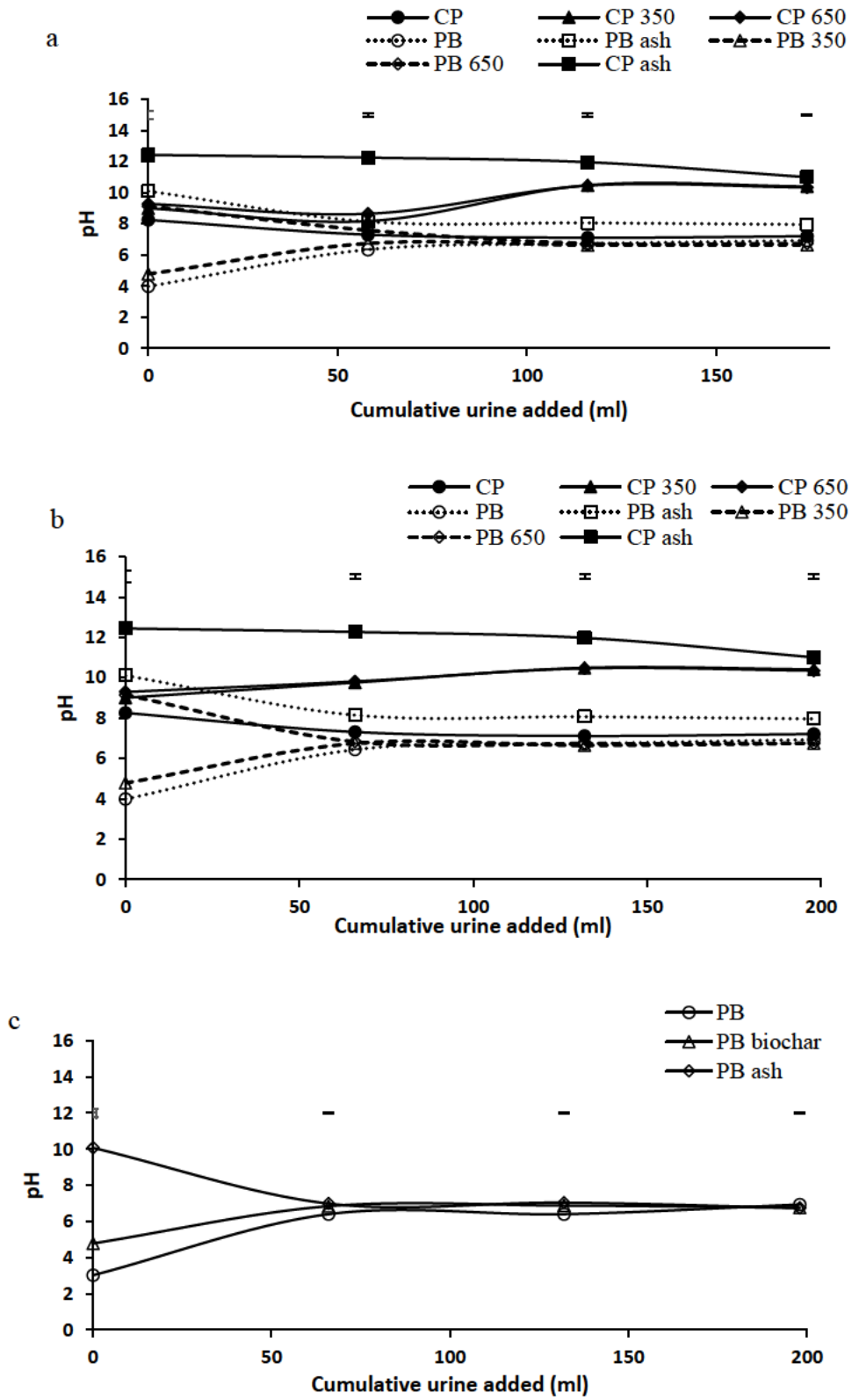


Figure 3.1: The pH during dehydration at a) 45 °C, b) 60 °C and c) glasshouse (G) of different media amended with urine. CP = cull potato; PB = Pine bark; CP 350 = Cull potato biochar at 350 °C; CP 650 = Cull potato biochar at 650 °C. The vertical error indicates LSD ($P<0.05$). The sampling points for cumulative urine added (in ml) at 45 °C were 58, 116, and 174, while at 60 °C they were 66, 132, and 198. The dehydration process took 4 days at 45 °C and 2 days at 60 °C following the addition of urine.

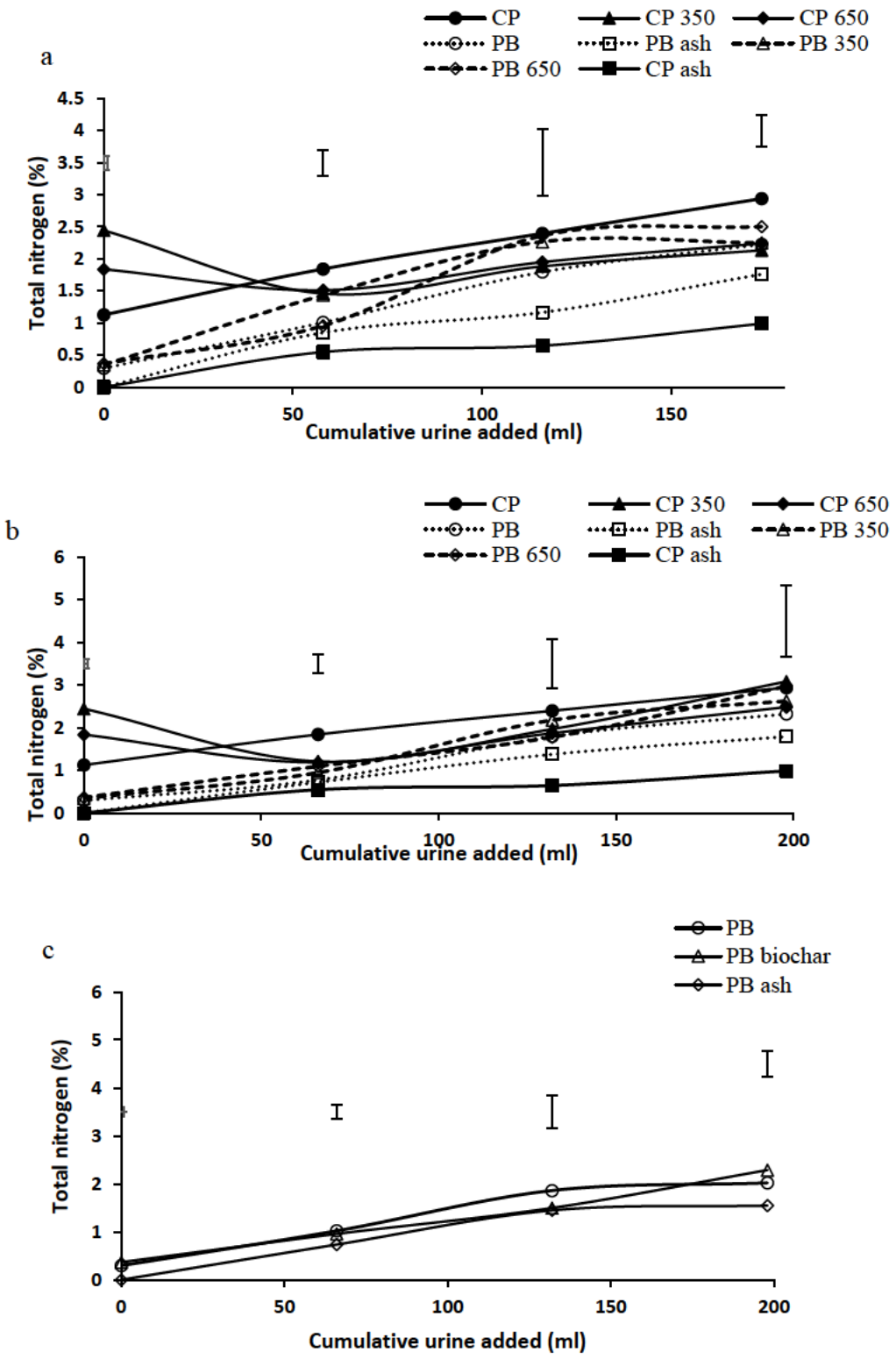


Figure 3.2: The total nitrogen (N) concentration (%) during dehydration at a) 45 °C, b) 60 °C and c) glasshouse (G) of different media amended with urine. CP = cull potato; PB = Pine bark; CP 350 = Cull potato biochar at 350 °C; CP 650 = Cull potato biochar at 650 °C. The vertical error indicates LSD ($P<0.05$). The sampling points for cumulative urine added (in ml) at 45 °C were 58, 116, and 174, while at 60 °C they were 66, 132, and 198. The dehydration process took 4 days at 45 °C and 2 days at 60 °C following the addition of urine.

3.3.4 Total carbon, nitrogen and carbon-to-nitrogen ratio at the end of the experiment

At the end of the experiment, at a dehydration temperature of 45°C, CP had significantly higher total nitrogen (N) compared to ash from both cull potatoes and pine bark, which had the lowest N levels across all treatments (Table 3.4). However, biochars from cull potatoes and pine bark, as well as PB feedstock, showed no significant differences in total N.

At 60 °C, CP 350 and PB 650 biochars had significantly higher N levels than CP ash, which had the lowest N. Additionally, there were no significant differences in total N between the CP materials (feedstock and biochars) and pine bark (feedstock, biochar, and ash). Total carbon and C:N ratio were significantly higher for PB 650 biochar at both 45 °C (62.5% and 27.9) and 60 °C (80% and 27.1) dehydration temperatures. In contrast, CP ash had the lowest C at 45 °C, while both CP and PB ashes had significantly lower C at 60 °C.

At 45 °C, CP 650 and CP feedstock had higher C than CP 350 biochar. However, at 60 °C, the CP biochars had similar C levels which were significantly lower than CP feedstock. For PB materials, the trend remained consistent at both dehydration temperatures, with PB 650 > PB 350 >PB > PB ash. Cull potato ash treatment at both dehydration temperatures had the lowest C:N ratio. The CP materials (except CP ash) had similar C:N ratio at both dehydration temperatures, which was significantly lower than PB materials (PB feedstock and biochars) For the products dehydrated in the glasshouse, there were no significant differences in total N. However, both PB and PB biochar had higher C content relative to PB ash. The PB ash had the lowest C:N ratio (4.98), while PB had a significantly higher C:N ratio of 18.8.

Table 3.4: Total nitrogen and carbon and carbon-to-nitrogen ratio of the end-products at both 45 and 60 °C.

Urine Products	N (%)	C (%)	C: N
45 °C Dehydration temp			
CP	2.85^{c}	29.7^{d}	10.3^{c}
CP 350	2.13^{b}	21.3^{c}	9.99^{c}
CP 650	2.24^{b}	27.1^{d}	12.1^{c}
CP ash	1.24^{a}	2.8^{a}	1.26^{a}
PB	2.23^{b}	38.8^{e}	17.4^{d}
PB 350	2.50^{bc}	43.2^{f}	17.3^{d}
PB 650	2.25^{b}	62.5^{g}	27.9^{e}
PB ash	1.76^{a}	7.97^{b}	4.56^{b}
60 °C Dehydration temp			
CP	2.94^{bc}	32.8^{c}	11.2^{c}
CP 350	3.08^{c}	20.7^{b}	6.93^{bc}
CP 650	2.48^{abc}	21.9^{b}	10.8^{c}
CP ash	1.76^{a}	2.38^{a}	1.38^{a}
PB	2.32^{abc}	40.6^{c}	17.5^{d}
PB 350	2.63^{abc}	53.6^{d}	20.5^{d}
PB 650	2.97^{c}	80^{e}	27.1^{e}
PB ash	1.79^{ab}	7.3^{a}	4.12^{ab}
Glasshouse dehydration			
PB	2.02^{a}	36^{b}	18.8^{b}
PB biochar	2.29^{a}	37.2^{b}	16.3^{ab}
PB ash	1.55^{a}	7.13a	4.98^{a}

Values on the same column with different letters indicate significant differences ($p<0.05$). CP= Cull potato waste; PB= pine bark waste.

3.3.5 Changes in total recovered N

The pine bark treatments had higher total recovered N compared to cull potato throughout the urine dehydration experiments (Figure 3.3a and b). The PB 650 had the highest recovered N, reaching 2.29% at 45 °C and 2.61% at 60 °C. The CP as showed similar N recovery trend across the two dehydration temperatures. Biochar from cull potato had the lowest recovered N, initially showing a negative recovered N after the addition of the first volume of urine (Figure 3.3a and b). With increasing urine addition, the CP 650 began to recover some N, while CP 350 continued to experience negative recovered N (N losses) even after the addition of the second volume of urine (Figure 3.3a). At a dehydration temperature of 60 °C, the recovered N with CP biochars increased with more urine addition but remained lower compared to other treatments (Figure 3.3b). There were no significant differences at each sampling time, but kiln biochar showed increasing N recovery with urine addition. (Figure 3.3c).

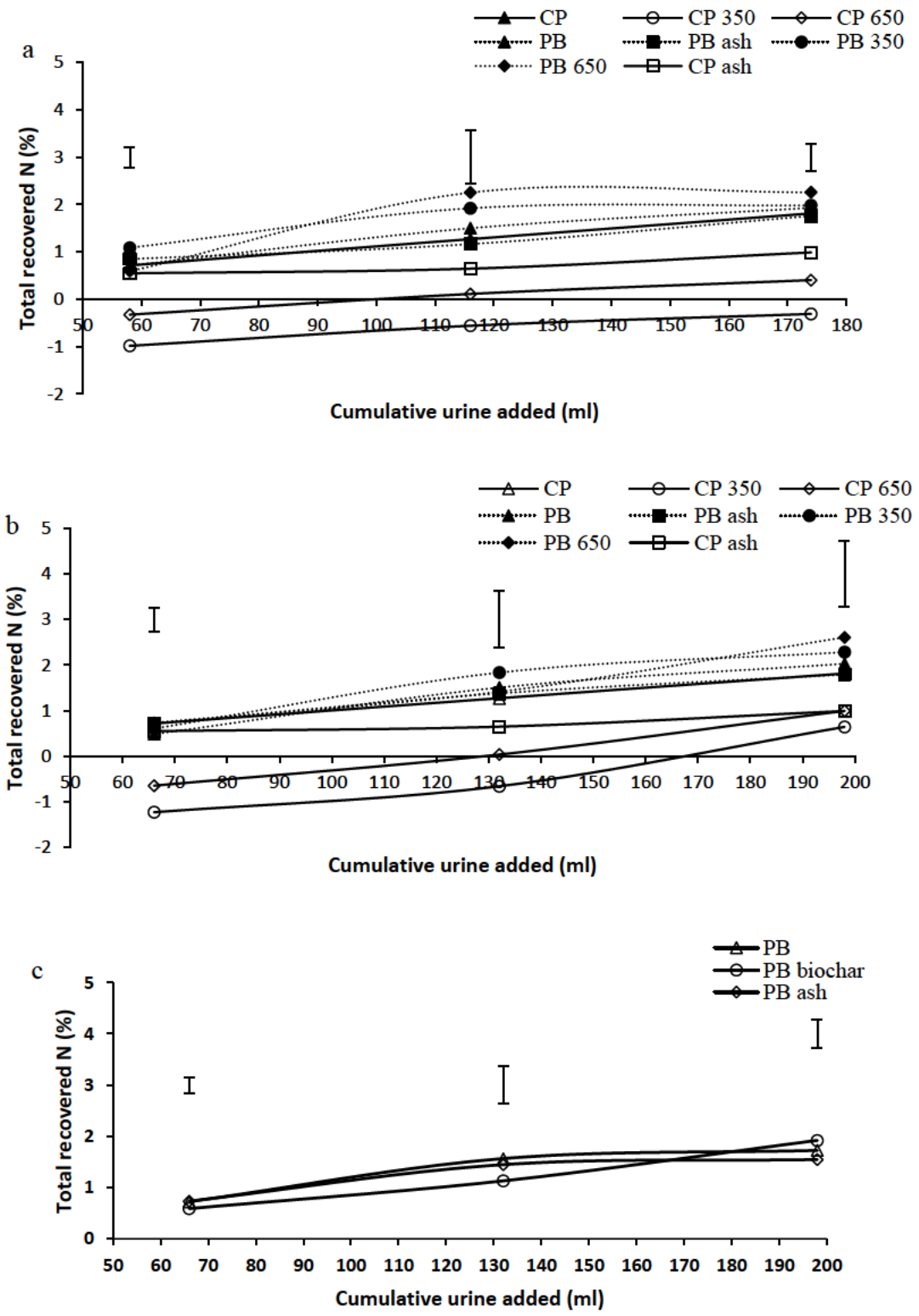
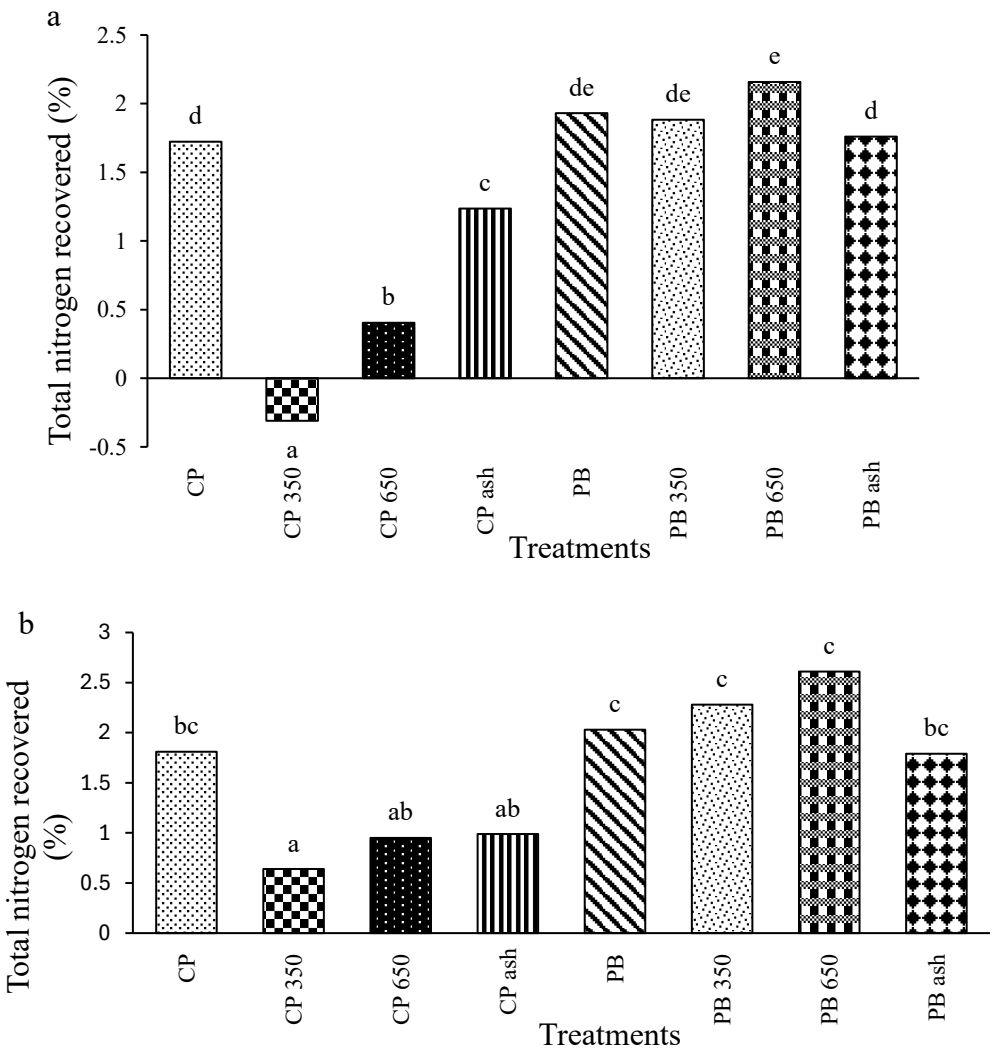


Figure 3.3: Recovered Nitrogen (N) during dehydration at a) 45 °C, b) 60 °C and c) glasshouse (G) of different media amended with urine. CP = cull potato; PB = Pine bark; CP 350 = Cull potato biochar at 350 °C; CP 650 = Cull potato biochar at 650 °C. The vertical error indicates LSD ($P<0.05$). The sampling points for cumulative urine added (in ml) at 45 °C were 58, 116, and 174, while at 60 °C they were 66, 132, and 198. The dehydration process took 4 days at 45 °C and 2 days at 60 °C following the addition of urine.

3.3.6 The total nitrogen recovered in the final urine products

At the end of the experiments, the PB 650 had significantly higher total N recovered (2.16%) while CP 350 recovered the least (negative recovered/ N loss) of N (-0.31%). The CP biochars and ash showed the lowest recovered N, which were lower compared to the PB treatments (feedstock, biochar, and ash). However, the CP feedstock medium had similar recovered N with the PB treatments, except that it had lower than for PB 650 (Figure 4.4 a). The PB treatments (feedstock and ash) had significantly higher recovered N in comparison to CP 350, which had the lowest (0.64%) (Figure 4.4 b). The PB products dehydrated at the glasshouse showed similar total N recovered (Figure 4.4c).



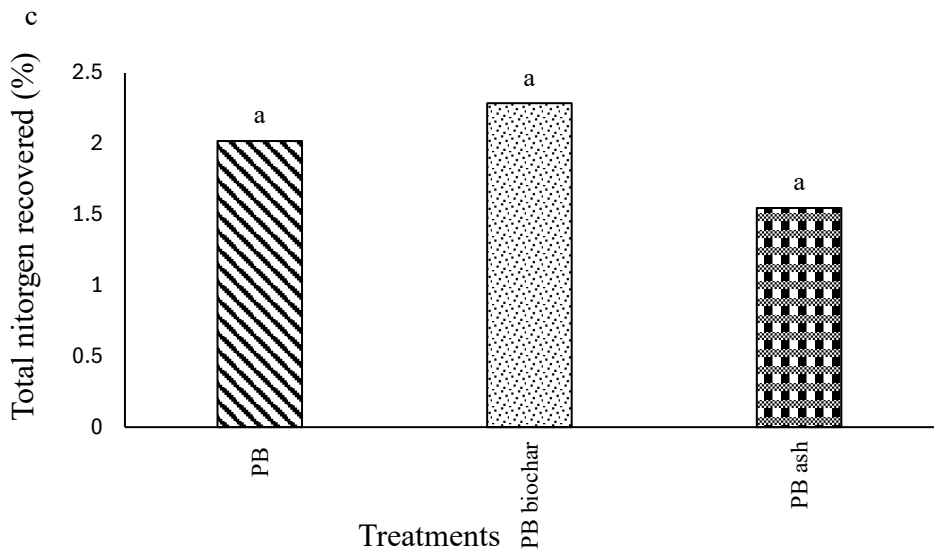


Figure 3.4: The total nitrogen recovered (%) by the urine-based products from pine bark and cull potato media dried at temperatures of a) 45 °C, b) 60 °C and c) glasshouse (G) after treatment with artificial human urine. The dehydration process took 4 days at 45 °C and 2 days at 60 °C following the addition of urine.

As a percentage of the urine N added, the CP biochar (350 °C) dehydrated at 45 °C recovered significantly lower N than CP feedstock and all the materials from PB (feedstock, biochar, ash), which had no significant differences irrespective of dehydration temperature (Table 3.5).

The PB ash dehydrated in the glasshouse showed a significantly higher pH compared to all other treatments. The percentage of urine nitrogen (N) recovered, the PB biochar dehydrated in the glasshouse showed significantly higher N recovery than the other materials, while PB ash had the lowest recovery.

Table 3.5: Recovered N of end-products as a percentage of the urine N added at 45 and 60 °C.

Dehydration temp (°C)	Urine Products	pH	Recovered N as % of N in the urine added
45	CP	8.52^{f}	82.6^{de}
	CP 350	10.4^{hi}	-19^{a}
	CP 650	10.3^{hi}	19.4^{b}
	CP ash	10.5^{i}	59.2^{cd}
	PB	6.93^{cd}	92.6^{e}
	PB 350	6.64^{abc}	90.3^{de}
	PB 650	6.74^{bc}	$*103^{e}$
	PB ash	7.96^{e}	90.5^{de}
60	CP	7.21^{d}	81.9^{de}
	CP 350	9.95^{g}	27^{bc}
	CP 650	10.1^{gh}	39.4^{bc}
	CP ash	11^{j}	37^{bc}
	PB	6.37^{a}	85.4^{de}
	PB 350	6.64^{abc}	88.9^{de}
	PB 650	6.74^{bc}	$*110^{e}$
	PB ash	7.92^{e}	81^{de}
Glasshouse	PB	6.40^{a}	72.9^{b}
	PB biochar	6.88^{b}	81.5^{c}
	PB ash	7.05^{c}	65.5^{a}

Values on the same column with different letters indicate significant differences ($p<0.05$). CP= Cull potato waste; PB= pine bark waste. * The percentage exceeding 100% suggests that the instrument may have overestimated the results.

3.3.7 The FTIR for urea detection in the final urine products

Figures 3.5 and 3.6 display the FTIR spectra of the various organic materials enriched with urine. The presence of urea encapsulation in the media is confirmed by observing specific amine functional groups in the FTIR spectra. In each stacked graph, the first FTIR spectrum represents the reference organic material without urine enrichment, serving as a baseline for comparison with the enriched samples. Prominent adsorption peaks indicating primary amines include N-H stretch (3500-3300 cm-1), N-H bend (1650-1580 cm^{-1}), C-N stretch (1335-1250 cm^{-1}), and C-N aliphatic mine (1250-1020 cm^{-1}), and urine addition led to prominent amine peaks which were intensifying. The CP biochar (350 and 650 °C) and ash enriched with urine showed intensified N–H stretching peaks, indicating the presence of nitrogen. However, the reference material also exhibited similar amine peaks, suggesting that urine addition did not result in significant urea incorporation into the biochars. Furthermore, both the CP enriched,

and CP ash samples exhibited an additional N-H bend in their FTIR spectra, further confirming the presence of urea.

All PB urine products displayed the N-H bend primary amine adsorption peak, while PB ash also showed the C-N stretch amine peak (Figure 3.6). The PB urine-based products also differed, with only PB feedstock and PB ash urine products showing the N-H stretching amine. The FTIR spectra confirmed the successful adsorption of urea from urine in the urine-enriched products (CP and PB), indicating effective urea retention.

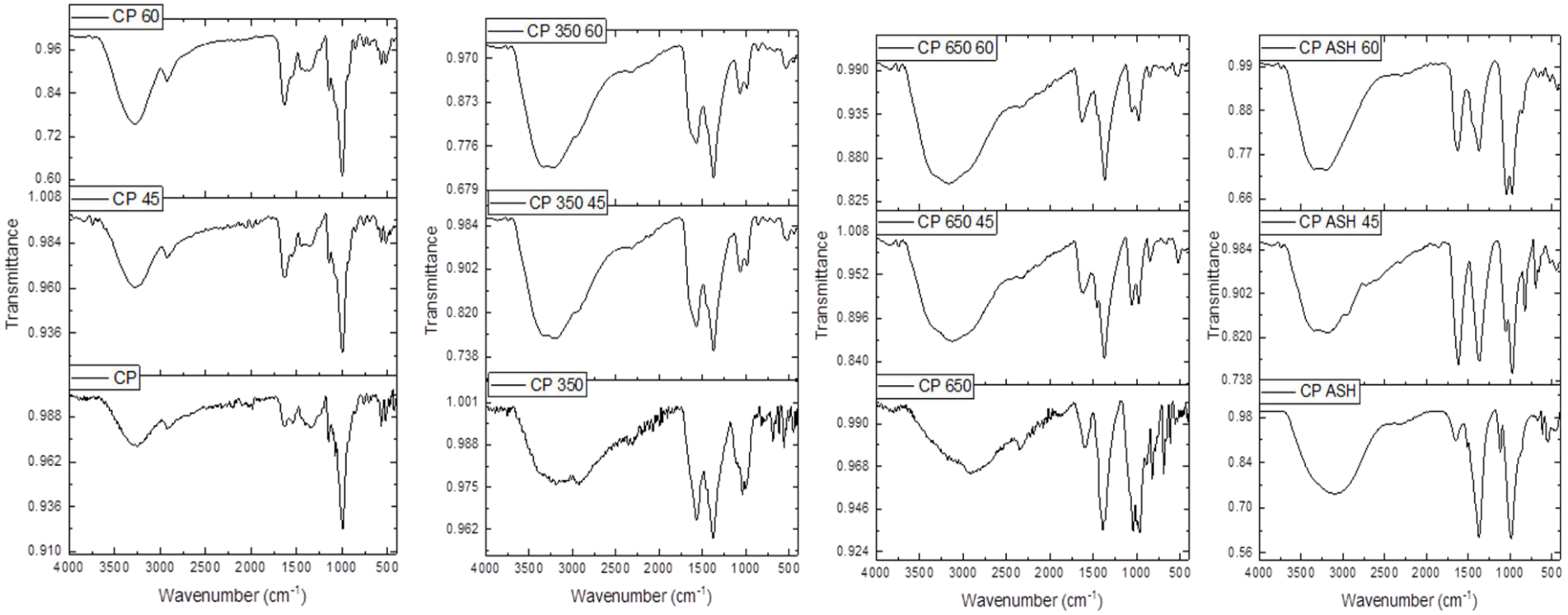


Figure 3.5: Stacked FTIR spectra of CP products (feedstock, biochar, ash) enriched with artificial human urine.

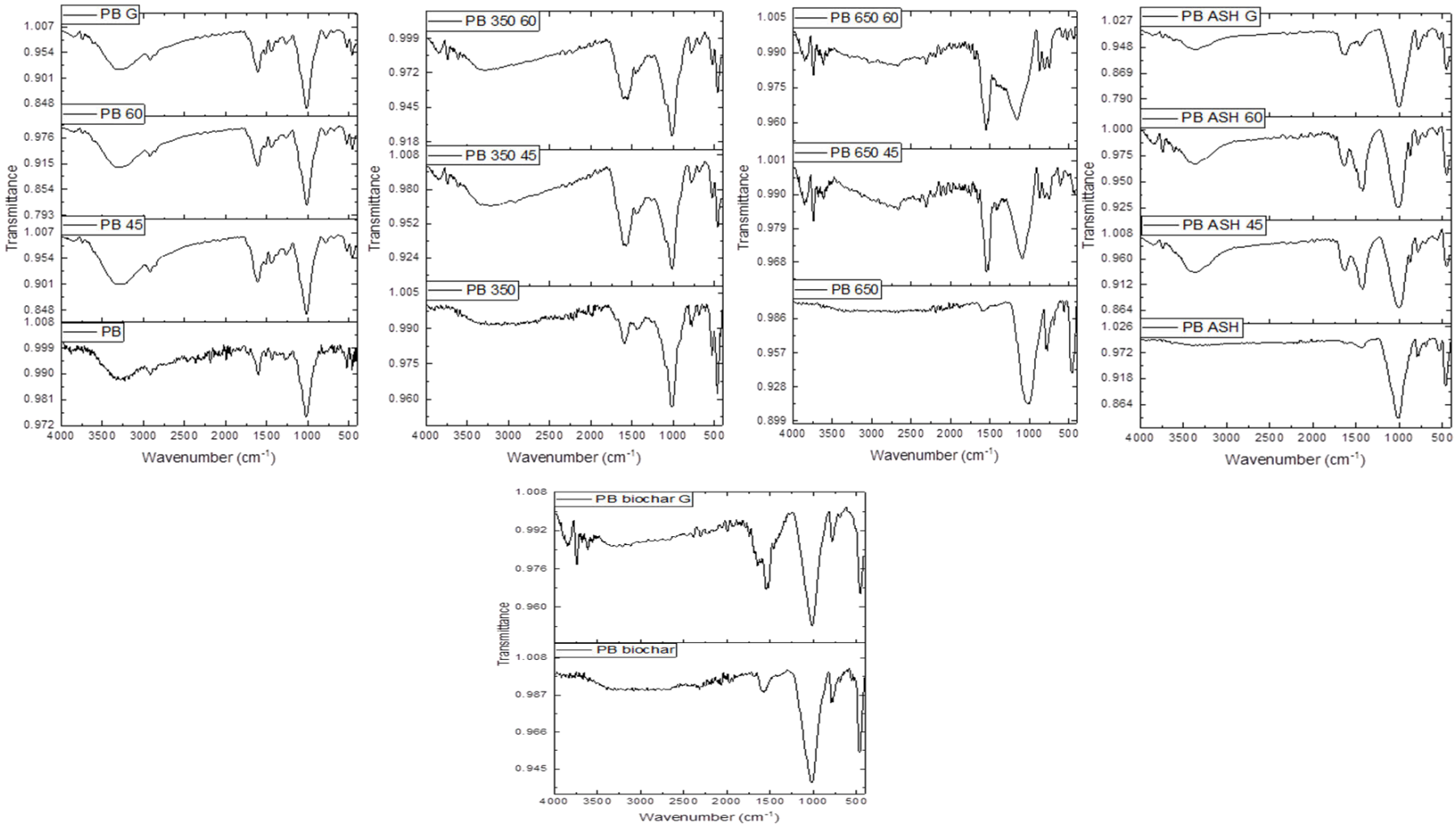


Figure 3.6: Stacked FTIR spectra of PB products (feedstock, biochar, ash) enriched with artificial human urine.

3.4 Discussion

The generally higher N after dehydrating urine at 60 °C, compared to 45 °C, across all dehydration media (Figure 3.3 a, b), could be a result of lower urine N added (174 ml at 45 than 198 ml at 60 °C). However, the shorter period required at 60 °C (2 days) than for 45 °C (4 days) makes it better to produce a larger quantity of product in a shorter time. The dehydration at 60 °C could also be related to energy efficiency, where setting the oven at 60 °C would consume 96 kWh in two days, while 192 kWh would be consumed at 45 °C in 4 days, which shows greater energy saving at the higher dehydration temperature. The 60 °C temperature promotes rapid dehydration (2 days at 60 °C versus 4 days at 45 °C) lowering N losses (Simha et al., 2021). In addition to rapid drying, higher temperatures also reduce the solubility of alkaline chemicals like $Mg(OH)_2$ and $Ca(OH)_2$, which slows chemical hydrolysis of urea (Simha et al., 2022). The findings align with those of Simha et al. (2020a), who reported increased N recovery at 60 °C using wood ash as a medium. Conversely, Warner (1942) reported that higher temperatures resulted in urea losses as a result of chemical hydrolysis. The differences may be attributed to the use of sealable tubes and alkaline media, potentially leading to overestimations of urea chemical hydrolysis at high temperatures, as noted by Simha et al. (2021). Dehydration in the glasshouse produced results comparable to those achieved in the ovens, suggesting that this method could be used for large-scale production with minimal energy consumption. In this current study, CP ash had a pH greater than 11, which suggests that urease activity would be minimal. The artificial urine does not contain urease. However, the organic materials used were not sterilised. The urease might have occurred from those materials. Therefore, any loss observed could likely be attributed to chemical hydrolysis. Higher temperatures did not generally increase chemical hydrolysis across all media, except for CP ash. This indicates that conclusions regarding chemical hydrolysis at elevated temperatures are influenced by different media, particularly those with a pH greater than 12. Considering that urea hydrolysis could have been limited in most of the treatments due to either high or too low pH, the retained N could likely remain in the form of urea, either adsorbed onto surfaces or freely present in the medium as supported by the FTIR (Figure 3.5 and 3.6).

The literature presents some discrepancies regarding the pH range for urease activity. Rektorschek et al. (1998) report that urease can be inhibited at pH values below 3.5 or above 8.5, whereas Chipako and Randall (2020) suggest inhibition occurs at pH levels below 7 or above 11. Given these differing thresholds, the selection of pH control strategies in our study

was guided by the broader inhibitory range (pH <6.5 or >11) to ensure complete suppression of urease activity during urine stabilization. In the current study, only the CP (feedstock) and PB ash had pH between 7 and 11, throughout the study, while PB650 and CP350 and CP650 biochars had the same range only after addition of two volumes of urine, and large N losses, through volatilisation, would be expected in these treatments. However, the N recovered showed that little (CP and PB ash) to no (PB650) N was lost as a percentage of amount added through urine (Table 3.5). The increase in pH for acidic media (4 to 6.7) was within the range of urease enzyme activity inhibition, which coincided with the increasing trend for total N (Figure 3.2 a, b and c) and total recovered N (Figure 3.3a, b and c). The potential inhibition of urease activity and the presence of acidic functional groups, which could have favoured N adsorption, could explain the high nitrogen recovered. The increase in pH following urine addition in acidic treatments (PB, PB 350 and PB kiln biochar) (Figure 3.1 a, b and c) could be possible due to urea hydrolysis releasing ammonia, which could have reacted with the H^+ ions leading to ammonium ion in the solution (Udert et al., 2006). The decrease in pH for some materials might be associated with the adsorption of carbon dioxide from the atmosphere (Senecal and Vinnerås, 2017). As the CO_2 gets absorbed and dissolved it results in the increase in H^+ concentrations. Similar findings were observed by Simha et al. (2018) in alkaline media where the pH dropped following urine addition. The CP ash maintained its alkalinity throughout; this could be assigned to its high calcium carbonates equivalence (57.1%) (Table 3.2).

The PB ash initially had an alkaline pH (10) and decreased reaching pH of 7.96 (Figure 3.1 a, b) 6.98 (Figure 3.1c), during the dehydration, which was within the range of enzymatic urea hydrolysis. However, the total N and total recovered N depicted an increasing trend, and total recovered N was approximately 1.7% (Figure 3.4 a, b) and 1.55% (Figure 3.4c), which shows that there was little effect of urea degradation. This could be explained by the presence of functional groups within the media, which might have adsorbed the N lowering it's susceptibility to urease enzyme. The PB 650 recovered more N, which could be due to the absence of urea degradation as a response to pH as it maintained a pH below 7. Biochar is widely known for its ability to adsorb N from solution, due to surface area and functional groups (Pathy et al., 2021). This is further supported by highest N in total recovered N (Figure 3.3 and 3.4 a, b) and total recovered N as a percentage of the N added as urine (Table 3.5). The total N and N recovered by PB 350 biochar appeared to have plateaued at the end of the experiment, which could be explained by saturation of the sorption sites. Both PB ash and PB

dehydrated in the glasshouse also plateaued at the end, which may indicate saturation of the sorption sites. Based on the results, it appears that materials (CP and PB) cannot retain nitrogen (N) once it reaches a threshold of 2.5-3%. The nitrogen that is not retained may then undergo hydrolysis, which pose a need for further investigation. The CP also showed a similar trend to that of PB 650; but the recovery (80%) was lower. This could plausibly be explained by more negatively charged surface functional groups and surface area in PB 650 than CP which adsorb the urea (Vilakazi et al., 2023), which aid in urea retention. It further highlights that N recovery in the materials used in the current study is less dependent on pH and more influenced by other media characteristics such as aromatic structure, pores, and surface area (Pathy et al., 2021).

The CP biochars were initially alkaline (pH 8) and increased following urine addition and later stabilised with final pH 10, which might have activated the urease enzyme activity and can plausibly explain the poor N recovery observed. The result for total N further supports the findings as the total N concentration decreased with the first urine addition, as the pH was increasing towards > 11. This further proves that there is a relationship between pH and nitrogen recovery, but this effect is dependent on the materials used (Senecal and Vinnerås, 2017). The CP biochars lacked pores (Vilakazi et al., 2023), indicating a low number of active sites for nitrogen (N) adsorption at exchange sites (Yin et al., 2017). This also suggests that the materials may have reached their nitrogen threshold, causing any additional nitrogen to be lost. The CP biochars showed low N recovery (Figure 3.3 a, b) while CP 350 showed a negative recovery of N (Figure 3.4a), which signifies that even the N in the original materials was lost, possibly due to the activity of micro-organisms especially at 45 °C where drying took longer (4 days) after having added N rich solution (more like an incubation). The CP ash maintained its alkalinity with pH reaching 12 yet it recorded lowest N recovery. This could be attributed to the presence of $CaCO_3$, which is known to stimulate ammonia volatilisation (Freney et al., 1981; Powlson and Dawson, 2022). This was further advocated by the pungent smell that were detected during the dehydration experiment. Another potential explanation could be the chemical hydrolysis leading to losses of N which is highly favoured in media with pH> 12 (Senecal and Vinnerås, 2017; Simha et al., 2021), catalysed by OH^- ions forming cyanic acid and ammonia (Alizadeh et al., 2017).

During the dehydration experiment, the CP treatments (CP feedstock and CP biochars) had high total N within the first volume of urine addition (Figure 3.2 a and b), and this higher N could be explained by the N content of the original material (cull potatoes) (1.13%) compared

to 0.295% N in pine bark. Even though the CP treatments maintained a relatively high total N than other treatments during the experiment, only a small proportion originated from the urine, while the rest originated from the original material. This is further shown by the results in Figure 3.3, where recovered N on the PB treatments (PB feedstock, biochar and ash) were relatively higher than CP treatments. The low total N in both the ash treatments (Figure 3.2 a and b) (CP and PB) appeared to result in some significant N recovered (Figure 3.3 a and b) but CP ash had lower N recovered compared to the feedstock (CP). The initial N (Table 3.2) for the ash was below detection and any total N recorded originated from the urine. These findings suggest that, for purposes of N recovery from urine, pine bark-based materials are more ideal than those from CP, while the CP feedstock would be more ideal than the corresponding ash and biochar from the same material. This suggests that, if material from CP is to be used to retain N from urine through dehydration, the feedstock would be more ideal than the ash and biochars. The more N recovered by CP feedstock than CP biochar and ash, demonstrated better energy use efficiency since the CP feedstock treatment requires only the energy for dehydration, while the others also require energy to produce the biochar and ash.

The higher N recovered by PB 650 (2.15% and 2.61% recovered N at 45 °C and 60 °C; respectively) media compared to the CP feedstock, even though they had similar pH trend, demonstrated that more N in urine can be recovered by using PB materials which minimizes chemical hydrolysis, which is exacerbated by high pH (Warner, 1942). Treatments PB 350 and PB ash followed a similar trend to that of PB 650. While Rektorschek et al. (1998) concluded that pH is the primary variable in controlling urea degradation, the finding with PB biochars highlights that N recovery in PB media was less dependent on pH and more influenced by media characteristics such as aromatic structure, pores, and surface area. The high surface area of PB media might promotes faster dehydration (Vilakazi et al., 2023). Additionally, the presence of basic and acidic functional groups facilitates urea adsorption, thereby inhibiting urease enzyme activity by not availing the urea (Pathy et al., 2021). Zhang and Wang (2016) concluded that a larger surface area enhances urea adsorption. Thus, PB biochar is a more suitable medium for dehydrating urine, producing a dry product with high N concentration, and is lighter to transport and potentially easier to apply than liquid urine, especially considering that the N concentration can be controlled. Moreover, pine bark is an easily available waste product that can be utilized by people to dry urine for use in their fields.

The dehydration process with PB treatments effectively eliminates the need for liquid disposal of urine while retaining most of the nitrogen (N). Dehydrating urine produces a urine-based material that may have fertiliser value for agriculture. Producing at a large scale using the glasshouse is also a promising approach as the results were generally similar to those produced at the ovens. The N concentration in the final products is higher than that in manures (0.4%) (Brown, 2008; Penhallegon, 2003) and fresh human urine (0.7%) (Vinnerås et al., 2006) but still lower than that in synthetic fertilisers (Maguire et al., 2009).

While the bulk of the N in CP products came from the original materials (Table 3.2), the change in C/N of these products from around 30 to around 10 as a result of urine addition, suggestion the drastic decrease in carbon concentrations (Table 3.4), suggested that the addition of urine might have led to the loss of some functional groups, which may include N-containing organic substances that degrade into ammonia (NH_3), which could have been lost as gas. This could also explain the decline of original N in the media (CP biochars) and the low N recovery for these treatments. Conversely, decreased C/N from over 100 (Table 3.2) to less than 30 for the PB products due to urine addition and dehydration, could have been as a result of N recovered with minimum losses. The final C/N of the PB products suggested that, during the initial stages of incorporation in soil, the N may be immobilised until a significant decline C/N to less than 25 occurs, where mineralisation will increase, and the fertiliser value can be observed. Upon addition of this materials some of the urea maybe free and not adsorbed to any surfaces.

3.5 Conclusion

The study showed that a dehydration temperature of 60 °C generally promoted rapid dehydration than at 45 °C. Dehydrating in a glasshouse offers a promising approach to resource constrained smallholder farmers. The choice of media used for dehydrating urine significantly impacts the concentration of N recovered from the urine, with greater benefits from PB feedstock, biochar and ash than those from cull potato. These findings suggest that, for purposes of N recovery from urine, pine bark-based materials are more ideal than those from CP. Pine bark products recovered more nitrogen (N) at both dehydration temperatures compared to potato waste products, with final pH less than the neutral. However, where CP products are to be used, the CP feedstock, which requires no additional energy for its production, would be more ideal than the corresponding ash and biochar. The use of pine bark products to treat urine and produce a nitrogen-rich fertiliser could be an innovative method that

reduces waste (urine and woody waste) and provides a valuable product, with an additional benefit of storing more C in the soil, especially with the biochar. The fertiliser value of the final products needs to be studied, to understand the effects of the different products on N availability when added to soil. Therefore, further research should focus on understanding how much of the N recovered in the materials will be readily available when applied to soil and the response of plants when the products are utilized as sole N sources.

Chapter 4: Nitrogen release from dehydration products of urine mixed with cull potato based materials and nitrogen fertiliser value of products: A glasshouse study with spinach

Abstract

The recycling of human urine has gained significant attention due to its high nitrogen (N) content. Nutrient recovery from human urine, using solid products, could be a sustainable approach to minimise nutrient losses and contribute to a more closed nutrient loop, with valuable products for soil fertility improvement. This study evaluated effects of various products derived from dehydration of urine, mixed with solid materials from cull potatoes (CP) on mineral nitrogen (N) in soil. The effects of various products derived from dehydration of urine mixed with various materials from cull potato (CP) on spinach growth, nutrient uptake and micronutrients concentrations under glasshouse conditions. The CP materials included feedstock, biochar (pyrolysed at 350 and 650 °C) and ash. The mixtures of urine and these materials were dehydrated at 45 and 60 °C. The dehydration products were applied to soils at triple (300 kg ha^{-1}) the recommended N rate (100 kg ha^{-1}) for spinach. The soils were incubated at field capacity moisture and 25 °C temperature. Destructive sampling was done at days 0, 7, 14, 21, 28, 42, and 56, with analysis of pH, ammonium- and nitrate-N and extractable P. The same materials and application rate used in the incubation experiment were also used in the glasshouse pot experiment. The products were applied to 2kg of soil (0-20 cm depth). Two spinach seedlings were then planted and the pot trial run for 84 days with plant sampling at 6 and 12 weeks and analysis for dry matter, nutrient uptake and tissue micronutrients, while pH and extractable phosphorus (P) were analysed in the residual soils. In the incubation experiment, nitrate-N increased with decrease in ammonium-N, reaching the highest of 89.4 mg kg^{-1}. Extractable P and pH were higher in urine-CP ash dehydrated at 45 and 60 °C, than other treatments indicating its liming effects. In the glasshouse study, biochar-based treatments enhanced spinach dry matter and N uptake, though tissue sodium (Na) levels were elevated across all urine-based treatments. These findings demonstrates that cull potato biochars, can be used as an alternative to synthetic N fertilisers for smallholder farmers. However, tissue Na and Mn in the tissue need to be monitored.

Keywords: Ash; dehydration; dry matter; mineral nitrogen; nitrogen recovery; spinach.

4.1 Introduction

Globally, over 80% of wastewater, which includes human urine, is discharged to the environment with little or no treatment (van den Broek et al., 2024). Large amount of nutrients such as nitrogen (N), phosphorous (P) and potassium (K) in the wastewater negatively impact the environment (Sene et al., 2019). The high concentration of total N (0.74%), K (1.62%), and P (0.02%) in human urine suggest that, if its source-separated through ecological sanitation approaches (Guest et al., 2009; Larsen et al., 2009), its application could have a significant contribution to soil fertility. However, the nitrogen could be lost to the atmosphere through ammonia volatilisation, while practical challenges associated with transportation costs, due to a huge proportion of human urine being water, makes it complicated to benefit from fertilisation with urine. Recovery of N by using different substrates has, therefore, become an emerging research focus. Adding a substrate that will increase or lower pH, could inactivate the urease enzyme responsible for hydrolysis of urea to ammonia and result in absorption of the N, which allow for dehydration of the excess water, producing a potentially valuable product.

Different urine-based products have been produced by modifying urine pH followed by dehydration, resulting into products with high nutrient concentrations and monetary value (Simha et al., 2021; Simha et al., 2020a). Modifying urine with $Ca(OH)_2$ followed by drying resulted in a product with 9.3% N, 0.7% P and 3.9% K (Simha et al., 2020a). Furthermore, a urine-based fertiliser from wood ash produced a nutrient-rich end product with concentration of nutrients (9.7% N, 1.5% P and 8.4% K) than in the original fresh urine (Simha et al., 2020a). However, it is worth noting that the elemental composition and functional groups of the substrate/medium could affect the nutrient composition of the final fertiliser material. The use of biochar may therefore produce a carbon-rich fertiliser material that can promotes N retention and improve soil fertility (Nguyen et al., 2018). Having different solid media for dehydration would produce a variety of urine-based fertiliser materials which differ in their contributions to available nutrients in the soil. Understanding nutrient release patterns of the different urine-based products in the soil is paramount for evaluating their fertiliser value.

Cull potato, an underutilised waste, can serve as organic amendment for biochar or ash and may improve the N recovery potential from urine. Preliminary results have shown that application of CP feedstock and dehydrating at 45 and 60 °C recovers more N (1.72 and 1.81%, respectively) than potato waste (biochar and ash) which showed lower nitrogen recovery rates, particularly with CP 350 showing negative nitrogen recovery (-0.31%). These differences

indicate that, although potato waste and its biochar have higher pH (inactivation of urease enzyme), they contribute less to the recovery of N from urine. However, potato wastes and their corresponding biochars have higher N (from original materials), which will also be mineralised, potentially contributing to N release in addition to that recovered from urine. The contributions of the urine-potato waste products to nutrient release could be affected by the rate of degradation of the different substrates, which depends on the C/N ratio, lignin content (Bird et al., 1999) and low water-soluble carbohydrates (Charmley et al., 2006). Furthermore, the solubility of potentially free (not adsorbed) urea could depend on the characteristics of the products together with those of the soil. However, no evidence could be accessed in the literature, on the effects of dehydrated urine-enriched potato waste products on nutrient release patterns in soil and there is limited literature on their effects on crop productivity. Panigrahi (2013) reported that urine-enriched coconut shell biochar resulted in a better French bean growth and yield, reflecting and confirming the nutrient availability in soils. No previous research has investigated the nutrient release pattern of urine-enrichment products of raw potato waste, biochar and ash from potato waste. There is a need to understand the contribution of urine-enriched products from contrasting materials from potato waste on availability of N, in order to understand their contribution to soil fertility. The objective of this study was to determine the effect urine-based products of feedstock, biochar and ash from potato waste, when dehydrated at 45 and 60 °C on mineral -N, available P and spinach productivity in amended soil.

4.2 Materials and methods

4.2.1 Experimental soil

The incubation study was conducted at the University of KwaZulu Natal, Pietermaritzburg campus (29° 37’ 33.9’’ S; 30° 24’ 14’E), South Africa. The soil used in this study was collected under natural vegetation at the Ukulinga research farm of the University of KwaZulu-Natal, South Africa. The soil was loam in texture and classified as Westleigh (Soil classification working Group, 2018), which was translated to Plinthic Acrisols (IUSS Working Group WRB, 2014). Bulk soil samples were collected from the 0–20 cm depth, mixed, homogenised, air-dried, and sieved (<2 mm), before analysis. The clay content of the samples was estimated using the near infrared reflectance (HTS-XT, Bruker, Germany), while total carbon (C) and nitrogen (N) were measured using the LECO Trumac CNS Autoanalyser (Leco corporation,

3000, Lakeview, Ave, ST, Joseph, MI, USA)). Phosphorus was extracted with the AMBIC-2 method (Manson et al., 2012) and analysed with a UV/VIS spectrophotometer following the molybdenum blue method (Murphy and Riley, 1962). Soil pH, exchangeable bases and acidity were analysed using standard methods (Non-Affiliated Soil Analysis Work Committee, 1990). The characteristics of the soils are shown in Table 4.1.

Table 4.1: Characteristics of the soil used in the study.

Property	Acrisols
Clay (%)	29
Bulk density (g cm^{-3})	1.24
$pH_{(KCl)}$	5.08
Total Carbon (%)	1.95
Total Nitrogen (%)	0.165
C/N	11.8
Extractable phosphorus (mg/kg)	3.86
Exchangeable potassium (cmolc/kg)	0.397
Exchangeable calcium (cmolc/kg)	8.79
Exchangeable magnesium (cmolc/kg)	7.48
Exchangeable acidity (cmolc/kg)	0.05
Acid saturation (%)	0

4.2.2 Urine-based product production and characterisation

The urine-based products used in this study were produced from cull potatoes (CP) and pine bark (PB). The methods for producing urine-based products and their characterisations are as described in chapter 3, sub-section 3.2.3.

4.2.3 Incubation experiment

The incubation experiment was set up in a completely randomised design with urine-based products, with three replicates. The urine-based products were from CP feedstock, CP biochars (350 °C and 650 °C), CP ash, all dehydrated at 45 and 60 °C. The products were applied to 100 g of soil at triple the recommended N rate for spinach (equivalent to 300 kg ha^{-1}). The triple N rate was used to ensure measurable nutrient release from the urine-based fertiliser products during the incubation and pot trial. This was particularly important because some products contained N primarily derived from the feedstock rather than urine, and lower application rates might have produced nutrient concentrations below detection limits. Therefore, the higher rate was selected to enhance experimental sensitivity and not to represent agronomic field conditions and also considering potential interactions with soil particles. Unamended soil (no urine-based product) was also included as a control. The soil + urine-based product mixtures

were placed in 500 ml plastic containers, which were tightly closed with lids and had four holes drilled below the rim to allow gas exchange. The soils were maintained at 100% water holding capacity, and the moisture was corrected weekly throughout the incubation based on weight loss. The soils were incubated in a constant temperature room at 25˚C with destructive sampling at 0, 7, 14, 21, 28, 42, and 56. The samples were analysed for ammonium- and nitrate-N, extractable P and soil pH.

4.2.3.1 Soil sample analysis

Ammonium- and nitrate-N were determined using the Gallery Discrete Analyzer (Thermofisher scientific, Waltham, Massachusetts, USA), after extraction with 2 M KCl solution. For this, soil (2 g), suspended in 20 mL of 2 M KCl solution, was shaken on a reciprocal shaker (Model E5850 Thomas Scientific, Swedesboro, NJ, USA) at 180 cycles per minute for 30 min, followed by filtration using Whatman No. 1 filter paper into storage bottles. Extractable P was determined calorimetrically following AMBIC – 2 extraction (Manson et al., 2012). Soil (2.5 g) was weighed into a 100 cm3 centrifuge tube and 25 ml of AMBIC-2 solution was added, and the suspension was shaken at 180 cycles per minutes for 30 minutes using a reciprocal shaker (Model E5850 Thomas Scientific, Swedesboro, NJ, USA). The supernatant was filtered using a Whatman No. 41 filter paper into storage bottles. An aliquot of the extract (2 ml) was diluted with 8 ml of distilled water, followed by slow addition of the colour reagent (10 ml) while mixing to allow for even distribution. The mixture was allowed to stand for 45 min prior to analysis using the UV/VIS spectrophotometer at a wavelength of 670 ηm (Murphy and Riley, 1962).

4.2.4 Pot trial experiment

The experiment was conducted under glasshouse conditions at about 25 °C for 84 days, following a randomized complete block design with four replicates. The same soil and treatments used in the incubation experiment (same urine-based products) were also used in the pot trial. The characteristics of the CP based urine products are provided in Chapter 3 sub-section 3.2.3. Treatments included urine-based products derived from CP feedstock, CP biochars (produced at 350 °C and 650 °C), CP ash, all dehydrated at 45 °C and 60 °C. Negative (no urine products) and positive (mineral N) controls were also included. The treatments were applied to 2 kg of soil, in pots, at a rate equivalent to 300 kg N ha^{-1} (triple the recommended N rate for spinach). This was to allow for sufficient N to be available from mineralisation of some

of the treatments for which most of the N is from the original feedstock (not from urine). The mineral N in the positive control was applied as ammonium nitrate (NH_4NO_3) to achieve a rate equivalent to 100 kg N ha^{-1} for optimum yield. Equivalent amounts of 124 kg P ha^{-1} and 117 kg K ha^{-1} were applied as NaH_2PO_4 and KCl, respectively (Manson et al., 2012). The pots had saucers underneath to trap any leachate, which would then be returned to the pots. The amended soils were moistened to 100% water holding capacity (WHC). Two spinach seedlings were planted to each pot and allowed to grow for 84 days. Correcting for moisture replenished (based on weight loss) distilled water was used. After 6 weeks (42 days), the spinach was harvested by cutting at 1 cm above the soil surface and leaving two younger leaves allowed to grow. The harvested shoots were washed to remove soil particles, and bloated with a paper towel, before weighing for fresh weight. The biomass was then dried at 60˚C for three days and weighed to determine dry weight. The same was done after 12 weeks, when all the spinach leaves were harvested. The dried plant tissue samples harvested at 6 and 12 weeks were combined to form a composite sample, ground (<1mm) using a blender and analysed for total N, P, K, calcium (Ca), magnesium (Mg), sodium (Na), zinc (Zn), copper (Cu), manganese (Mn) and iron (Fe). The N and P uptake were calculated from their tissue concentrations and the dry matter yield. After harvesting at 12 weeks the soils were analysed for pH and extractable P. The soils were oven dried over night at 105 °C for moisture correction of the soil P results.

4.2.4.1 Plant tissue analysis

Tissue N was analysed using the Leco Trauma CNS Autoanalyser (Leco corporation, 3000, Lakeview, Ave, ST, Joseph, MI, USA). For other elements, the oven- dry and ground spinach sample (0.5 g) was placed into a crucible, treated with 10 ml of concentrated HNO_3 and HCl (1:3) (Gupta and Sharma, 2014), and digested for one hour using a MARS CEMS microwave digester. Thereafter, the samples in the digest were analysed for P, Ca, Mg K, Fe, Mn, Zn and Cu concentration using an inductively coupled plasma atomic emission spectrometry (Varian 720-ES ICP-AES). The N and P uptake were calculated using equation 5.1.

N/P uptake (g $plot^{-1}$) = dry matter yield (g $plot^{-1}$) $*\frac{(N\ /\ P\ concentration)}{100}$ Equation 4-1

4.2.4.2 Residual soil analysis (pH, mineral-N and P)

Soil pH and extractable P were analysed in the residual soils. Soil pH was measured in KCl at a ratio of 1:5 using 1M KCl. Extractable P was read using the molybdenum-blue method (Murphy and Riley, 1962), after extraction with AMBIC-2 (Manson et al., 2012).

4.2.5 Statistical analysis

The incubation results of soil pH, mineral-N (ammonium- and nitrate-N) and extractable P were analysed using GenStat 18th edition (VSN International 2015) by subjecting them to the two-way analysis of variance (ANOVA). The results were analysed separately for different sampling days. Separation between treatment means was done using least significant difference (LSD) at $p < 0.05$. The Tukey-Kramer test was also used to separate treatment means at $p < 0.05$.

All data of the pot experiment were analysed using GenStat 18th edition by subjecting them to the one-way analysis of variance (ANOVA). One-way ANOVA was used because the controls did not have the same number of levels as the feedstocks and dehydration condition combinations. Separation between treatment means was done using least significant difference (LSD) at $p < 0.05$. The Tukey HSD test was also used to separate treatment means at $p < 0.05$.

4.3 Results

4.3.1 Changes in Ammonium-N

There was a sharp increase in ammonium-N concentration within the first 7 days of the incubation for all the treatments (Figure 4.1), except the control and CP ash-60, which were below detection until day 84 (Figure 4.1a). A general decrease occurred from day 7-56, except and CP 650-45 (day 56).

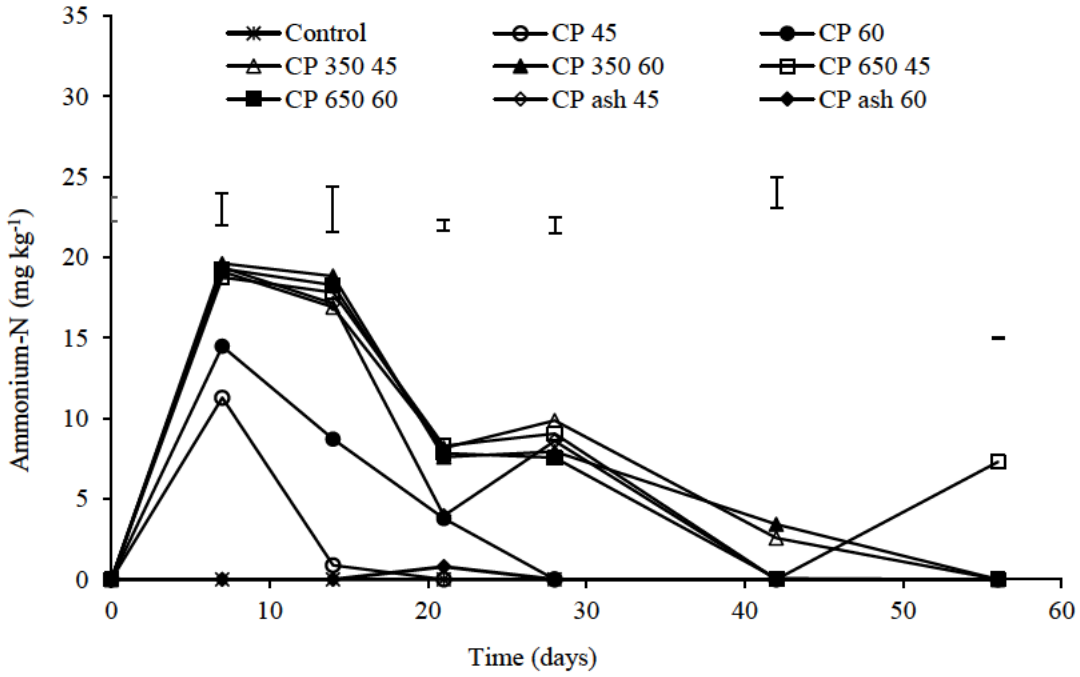


Figure 4.1: Concentration of Ammonium-N released during the incubation of the products with soil. CP 45 and CP 60- Cull potato dehydrated at 45 and 60 °C, respectively; CP 350 45 and CP 350 60- product of cull potato biochar at 350 °C dehydrated at 45 and 60 °C, respectively; CP 650 45 and CP 650 60- Cull potato biochar at 650oC dehydrated at 45 and 60 °C, respectively; CP ash 45 and CP ash 60- Cull potato ash products dehydrated at 45 and 60 °C, respectively. The vertical error bars indicate LSD ($p<0.05$).

4.3.2 Changes in Nitrate-N

Application of urine-based products significantly increased nitrate-N concentration (Figure 4.2). The CP treatments (CP-60> CP-45> CP ash-60) which were higher than the control but lower than other treatments. The nitrate-N from the CP biochar products dehydrated at 45 and 60 °C increased up to day 42, followed by a slight decrease on day 56, except for CP 350-45 which increased until day 56 (Figure 4.2).

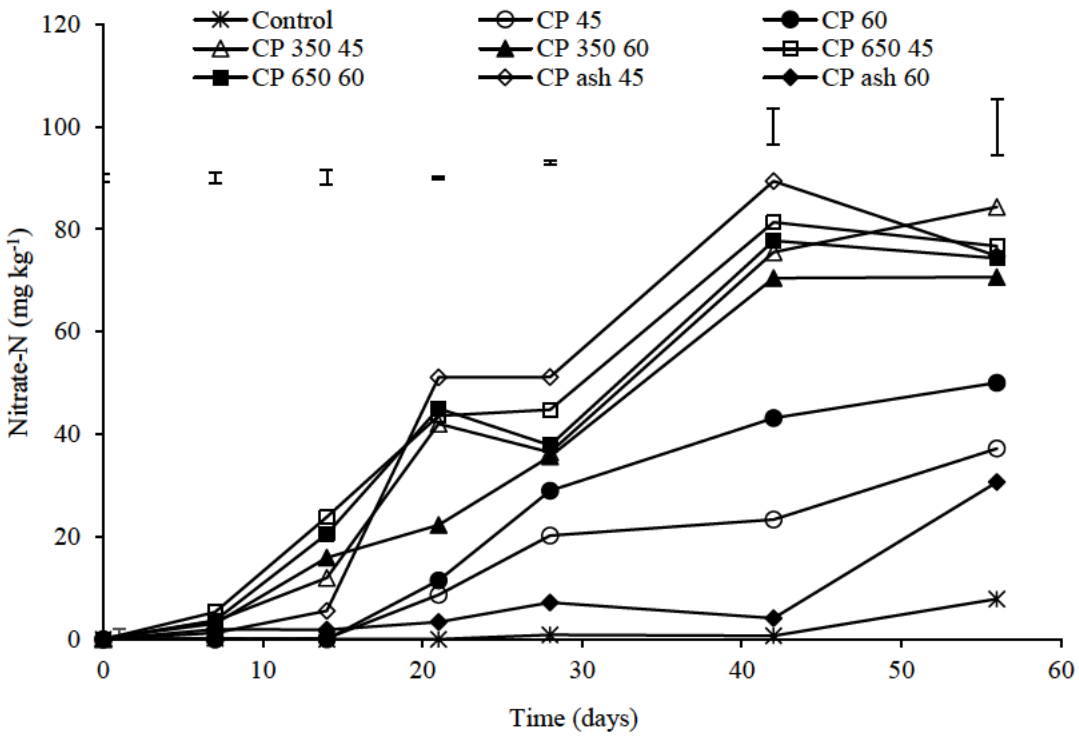


Figure 4.2: Concentration of nitrate-N released during the incubation of the products with soil. CP 45 and CP 60- Cull potato dehydrated at 45 and 60 °C, respectively; CP 350 45 and CP 350 60- product of cull potato biochar at 350 °C dehydrated at 45 and 60°C, respectively; CP 650 45 and CP 650 60- Cull potato biochar at 650ºC dehydrated at 45 and 60°C, respectively; CP ash 45 and CP ash 60- Cull potato ash products dehydrated at 45 and 60°C, respectively. The vertical error bars indicate LSD ($p<0.05$).

4.3.3 Changes in extractable P

Addition of CP ash products, dehydrated at 45 and 60 °C significantly increased soil extractable P in comparison to other treatments (Figure 4.3). The CP ash-60 treatment was 100% higher than all the other treatments on days 0 to 56, except for CP ash-45, which was 91% lower than the CP ash-60 treatment.

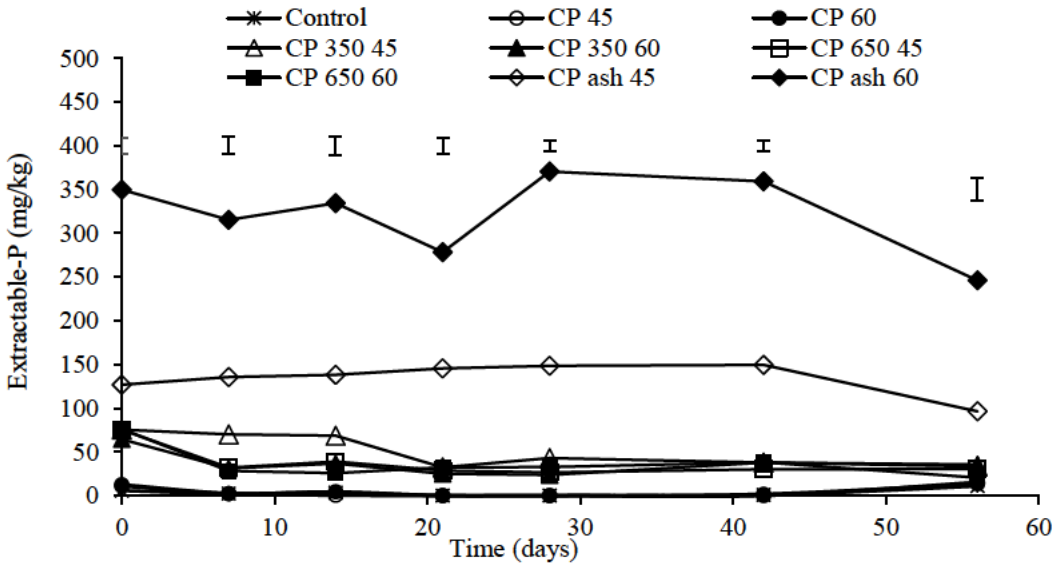


Figure 4.3: Concentration of extractable-P released during the incubation of the products with soil. CP 45 and CP 60- Cull potato dehydrated at 45 and 60 °C, respectively; CP 350 45 and CP 350 60- product of cull potato biochar at 350 °C dehydrated at 45 and 60 °C, respectively; CP 650 45 and CP 650 60- Cull potato biochar at 650oC dehydrated at 45 and 60 °C, respectively; CP ash 45 and CP ash 60- Cull potato ash products dehydrated at 45 and 60 °C, respectively. The vertical error bars indicate LSD ($p<0.05$).

4.3.4 Changes in soil pH

There were significant differences in soil pH throughout the incubation (Figure 4.4). The soil pH was significantly higher in the CP ash-60 than other treatments, and this was followed by CP ash-45, except for day 14, where they were not significantly different from each other (Figure 4.4).

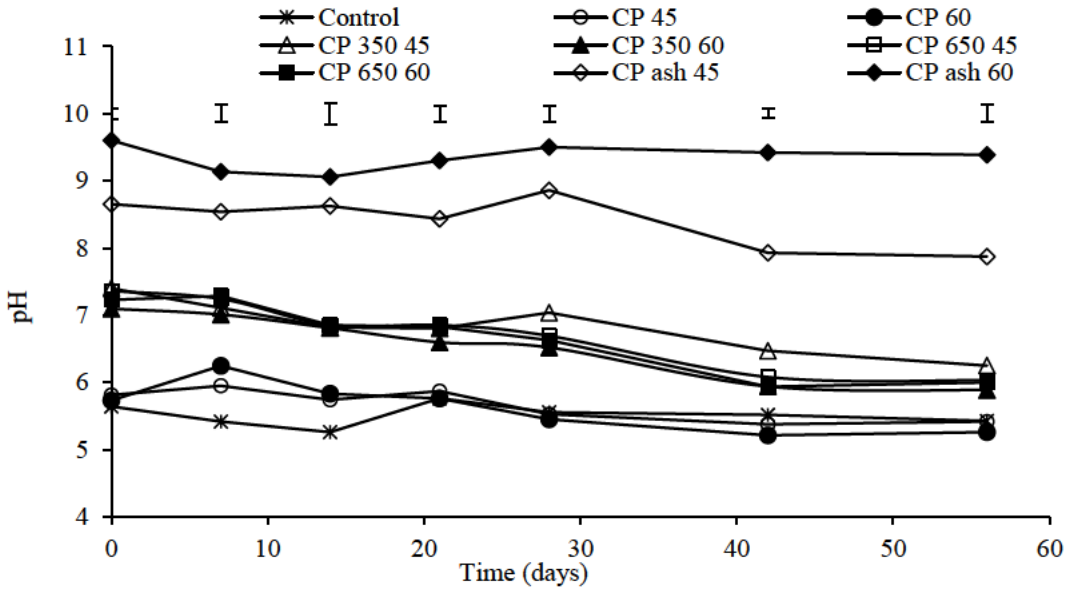


Figure 4.4: Changes in soil pH during the incubation of the products with soil. CP 45 and CP 60- Cull potato dehydrated at 45 and 60 °C, respectively. CP 350 45 and CP 350 60- product of cull potato biochar at 350 °C dehydrated at 45 and 60 °C, respectively; CP 650 45 and CP 650 60- Cull potato biochar at 650 °C dehydrated at 45 and 60 °C, respectively; CP ash 45 and CP ash 60- Cull potato ash products dehydrated at 45 and 60 °C, respectively. The vertical error bars indicate LSD ($p<0.05$).

4.3.5 Dry weight

Application of urine-based products increased the dry matter yield of spinach (Figures 4.5). There were no major differences between dehydration temperatures (45 and 60 °C) for each product type. All the urine-based products, except those from CP-45, had significantly higher spinach dry matter than the negative control. The products from CP biochars had the higher dry matter yield than the positive control (Figure 4.5), with CP 650-45 being significantly higher. The CP feedstock (CP-60) and CP ash treatments had similar levels as the positive control (Figure 4.5).

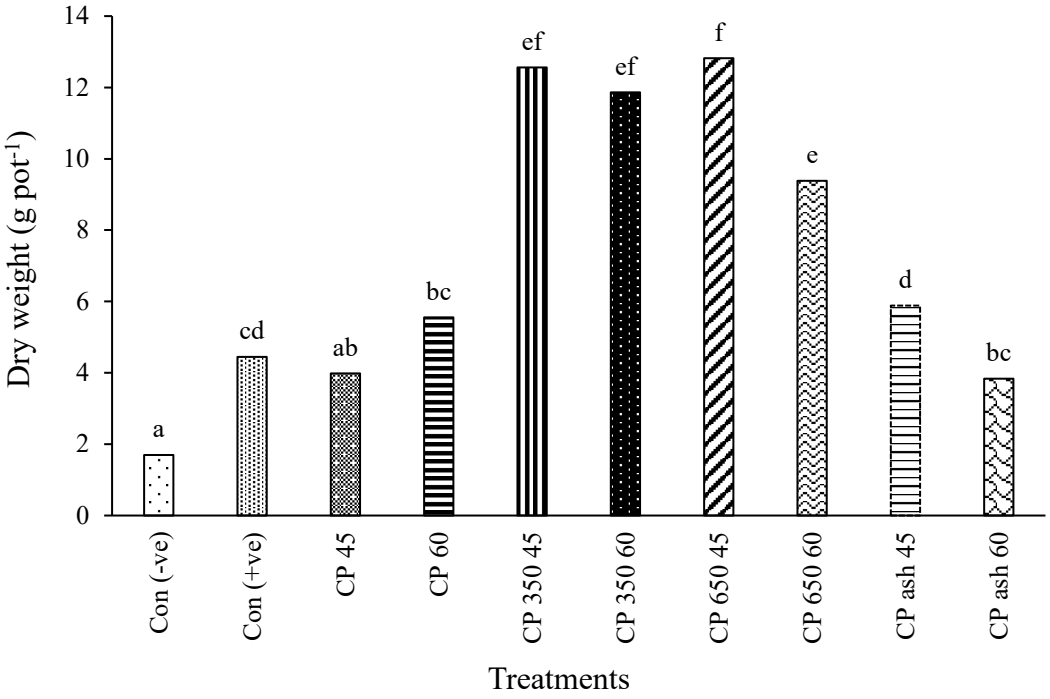


Figure 4.5: Dry matter yield of spinach as affected by different urine-based products. CP 45 and CP 60- Cull potato dehydrated at 45 and 60 °C, respectively; CP 350 45 and CP 350 60- product of cull potato biochar at 350 °C dehydrated at 45 and 60 °C, respectively; CP 650 45 and CP 650 60- Cull potato biochar at 650oC dehydrated at 45 and 60 °C, respectively; CP ash 45 and CP ash 60- Cull potato ash products dehydrated at 45 and 60 °C, respectively.

4.3.6 Nutrient Uptake

In the first experiment, the products from CP biochars (both 350 and 650), had significantly higher N uptake than both controls, while those from the CP feedstock and CP ash were not different from the controls (Figure 4.6).

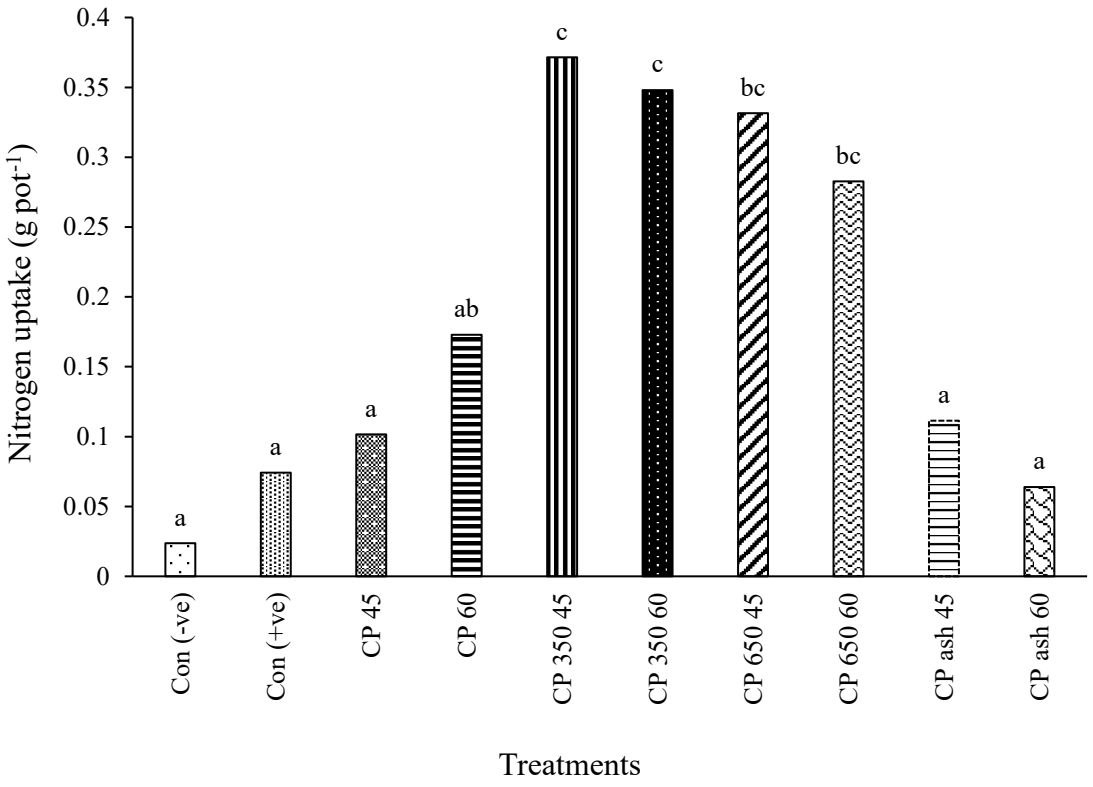


Figure 4.6: The nitrogen uptake of spinach fertilised with different urine-based products. CP 45 and CP 60- Cull potato dehydrated at 45 and 60 °C, respectively; CP 350 45 and CP 350 60- product of cull potato biochar at 350 °C dehydrated at 45 and 60 °C, respectively; CP 650 45 and CP 650 60- Cull potato biochar at 650 °C dehydrated at 45 and 60 °C, respectively; CP ash 45 and CP ash 60- Cull potato ash products dehydrated at 45 and 60 °C, respectively.

4.3.7 Uptake of P and K and tissue concentrations of Ca, Mg and Na

The uptake and mineral concentration in spinach showed varying results across the different treatments. The P uptake was significantly higher at CP 650-45 and was similar to CP 350-60, CP ash-45, which showed significantly higher P uptake than both the positive and negative controls. The CP biochars produced at 350 and 650 °C exhibited significantly higher K uptake compared to the controls. Treatments with CP feedstock and CP ash-45 had significantly higher Ca concentration than the positive control but were comparable to other treatments. Magnesium concentration was significantly higher in CP-60, CP 350-60 and CP ash-45 than in both the controls. The CP products had higher Na concentration compared to the negative control (Table 4.2).

Table 4.2: Effect of application of triple N recommendation rate using urine-based products dehydrated at 45 and 60 °C on the uptake of P and K and Ca, Mg and Na concentration in spinach dry matter.

Treatments	Tissue uptake		Tissue concentration		
	P	K	Ca	Mg	Na
	(g pot^{-1})		(%)		
Con (-ve)	0.0108^{a}	0.0756^{a}	0.709^{ab}	0.705^{ab}	0.824^{a}
Con (+ve)	0.0170^{a}	0.152^{ab}	0.510^{a}	0.624^{a}	1.19^{ab}
CP 45	0.0456^{ab}	0.284^{abc}	1.05^{b}	1.24^{bc}	1.71^{b}
CP 60	0.0572^{abc}	0264^{abc}	1.11^{b}	1.35^{c}	1.81^{c}
CP 350 45	0.0787^{abcd}	0.940^{e}	0.754^{ab}	1.27^{bc}	1.79^{bc}
CP 350 60	0.122^{cd}	0.886^{e}	0.831^{ab}	1.40^{c}	1.74^{bc}
CP 650 45	0.134^{d}	0.943^{e}	0.843^{ab}	1.27^{bc}	1.55^{b}
CP 650 60	0.0511^{abc}	0.691^{de}	0.777^{ab}	1.19^{abc}	1.74^{bc}
CP ash 45	0.1047^{bcd}	0.443^{bcd}	1.222^{b}	1.37^{c}	1.20^{ab}
CP ash 60	0.0687^{abcd}	0.341^{abc}	0.934^{ab}	1.18^{abc}	1.52^{b}

*Values on the same column with different letters indicate significant differences ($p<0.05$). CP= Cull potato waste.

4.3.8 Cu, Zn, Fe, and Mn concentration

The concentration of micronutrients (Cu, Zn, Fe, and Mn) in spinach varied across the treatments (Table 4.3). The CP feedstock had significantly higher Cu concentration than the positive control. The Zn concentration was notably higher in CP-60, CP-45 and the positive control, than the negative control. Higher tissue Fe concentrations ($p < 0.05$) were observed in the negative control. Manganese concentration was significantly higher in CP feedstock than both controls and CP 650-60.

Table 4.3: The Cu, Zn, Fe and Mn concentration of spinach when urine-based products dehydrated at 45 and 60 °C are being applied at the triple N recommended rate.

Treatments	Tissue concentration (mg kg^{-1})			
	Cu	Zn	Fe	Mn
Con (-ve)	8.94^{ab}	26.8^{a}	130^{cd}	394^{abcd}
Con (+ve)	6.34^{a}	77^{bcde}	76.5^{ab}	427^{bcd}
CP 45	16.0^{b}	84.0^{bcde}	89.5^{abcd}	742^{g}
CP 60	15.3^{b}	99.6^{e}	118^{abcd}	611^{efg}
CP 350 45	13.8^{ab}	66.6^{abcd}	66.8^{a}	252^{ab}
CP 350 60	13.4^{ab}	65.6^{abcd}	80.3^{abc}	339^{abc}
CP 650 45	13.5^{ab}	66.1^{abcd}	87.1^{abcd}	377^{abcd}
CP 650 60	9.79^{ab}	47.5^{abc}	72.9^{ab}	228^{a}
CP ash 45	13.6^{ab}	63.1^{abcd}	74.5^{ab}	469^{cde}
CP ash 60	11.78^{ab}	43.6^{ab}	109^{abcd}	387^{abcd}

*Values on the same column with different letters indicate significant differences ($p<0.05$). CP= Cull potato waste.

4.3.9 Soil pH and extractable P after spinach harvest

The CP ash treatment resulted in a significantly higher pH compared to the controls (Table 4.4). Extractable P was highest in CP ash-60, followed by CP ash-45, CP 350-45, and CP-45 than the negative control.

Table 4.4: Soil pH and extractable P concentration after spinach harvest as affected by different urine-based products dehydrated at 45 and 60 °C applied at the triple recommended rate.

Treatments	pH $_{(KCl)}$	Extract. P (mg kg^{-1})
Con (-ve)	5.55^{a}	5.17^{a}
Con (+ve)	5.96^{cd}	10.3^{bcd}
CP 45	5.75^{abc}	13.6^{d}
CP 60	5.66^{ab}	8.17^{abc}
CP 350 45	5.77^{abc}	16.2^{e}
CP 350 60	6.16^{de}	6.77^{ab}
CP 650 45	6.24^{ef}	11.7^{cd}
CP 650 60	6.46^{fg}	5.44^{a}
CP ash 45	6.89^{h}	35.7^{f}
CP ash 60	7.09^{h}	43^{g}

*Values on the same column with different letters indicate significant differences ($p<0.05$). CP= Cull potato waste.

4.4 Discussion

The increase in ammonium-N concentration for the first 7 days (Figure 4.1) could be the result of urea hydrolysis due to addition of moisture. Moisture is a significant factor that initiates urea hydrolysis process (Abera et al., 2012). The findings also align with those of (Kissel, 1988), where urea hydrolysis occurred within 4-7 days in warm temperature with optimum moisture content. Hydrolysis in soils is also influenced by pH, with the process generally increasing as pH rises with optimal pH range for urease enzyme activity being between 5.5 and 9 (Motasim et al., 2024). When the pH exceeds 9, hydrolysis slows down (Motasim et al., 2024). At acidic pH (pH < 7) most of NH_3 will be converted to NH_4^+ (Sigurdarson et al., 2018). In the current study, all the treatment had pH that was less than 9 (Figure 4.4), except for CP ash-60 which had a pH above 9 (Figure 4.4). The ammonium-N levels for CP ash-60 remained below detection, and the low nitrate-N levels, throughout the incubation period, indicating that conditions in this treatment did not support hydrolysis and nitrification. The higher pH likely explains the findings, as ammonium-N levels were undetectable in CP ash-60, presumably because the elevated pH inhibited urea hydrolysis. The N in this treatment was not immediately available but could instead be released gradually over time, depending on dynamics of soil pH. Additionally, the product's high calcium carbonate equivalent (CCE) of 57.1% indicates a strong liming potential (Singh et al., 2017), which helps explain the increase in soil pH to 9.5 (Figure 4.4). Although the C/N ratio of 1.38 for CP ash-60 would typically imply rapid nitrogen release, this was not observed in this case. Thus, CP ash-60 may serve more effectively as a liming product than an N fertiliser.

The CP biochars (350 and 650 °C) urine products showed significant mineral-N release, despite low urine-derived N retention. This suggests that mineralisation of organic N within biochar contributed to the observed ammonium and nitrate-N levels. In contrast, the CP feedstocks urine products had lower N release, despite the materials being applied at similar N rate and low C/N, which is likely to promote microbial activity and N mineralisation. The low ammonium could be a result of the presence of low water-soluble carbohydrates, hindering the decomposition of the organic material (Charmley et al., 2006). The CP 650-45 had the highest peak ammonium-N at day 56 (Figure 4.1), signifying its accumulation, which was attributed to slower rate of nitrification, as shown by the lower nitrate-N results (Figure 4.2). This could be assigned to decreasing pH values at between day 42-56. This concurred well with the findings of (Tian et al., 2013) that lower pH favours accumulation of ammonium ions. The decrease in

pH reduces the microorganisms responsible for nitrification, which consequently prevents the transformation of ammonium to nitrate (Camargo and Alonso, 2006). The rapid decline in ammonium-N for all treatments from day 7 (Figure 4.1), was attributed to nitrification, which was supported by the increase in nitrate-N. Furthermore, the increase in nitrate-N explains the decrease in pH (Figure 4.4), as nitrification releases H^+ ions, which lower soil pH as observed by (Sahrawat, 2008). This aligns with the findings of (Sigurdarson et al., 2018), who observed a decrease in pH following urea application, attributed to nitrification.

Application of CP ash products dehydrated at 45 and 60 $^{\circ}$C, significantly increased extractable P and maintained it up to 56 days (Figure 4.3). The increase was attributed to the high phosphorus concentration (1.52%) in the original CP products, which enhanced phosphorus availability, along with the phosphorus recovered from urine, although it was not measured in this study. Similar results were observed by (Panigrahi, 2013) on a coconut shell enriched with human urine. Additionally, increasing soil pH as a result of CCE to optimum levels increases the negative surface charges which causes less adsorption of P increasing its availability (Cui et al., 2011; Masto et al., 2013). Similar observations have been reported where P availability increased in acidic soils following application of biochar with liming effects (Singh et al., 2010; Xu et al., 2014). This implies that CP ash could be used as P fertilisers even in acidic soils, with potentially beneficial effects. The maximum extractable P released from CP ash-45 (370 mg kg^{-1}) would be sufficient for most crops without any need to supplement of inorganic P fertilisers. These results further advocate the CP ash as a P rich, liming material for acidic soils.

The increased dry matter yield and N uptake in spinach with CP biochar treatments (especially CP 650-45) than the control (Figure 4.5), could be explained by the significantly higher N applied (3 times higher than the positive control), which resulted in greater N uptake. This view was supported by the results of N uptake which followed the same trend (Figure 4.6). Results from incubation experiments (Figure 4.2) indicated that the N in the urine-based products was rapidly released in mineral form when applied to soil. This rapid release increased N availability and uptake by the spinach resulting in greater growth, and yield. Biochar is also known to improve soil aeration and microbial activity which might improve yield (Liao et al., 2020). This shows the agronomic potential for these products. The higher N uptake and dry matter yield in CP biochar-based products, even though they retained lower urine N, than the CP feedstock and CP ash, suggest that the pyrolysis of potato waste significantly increases the release of organic N in these wastes. This view was supported by the incubation experiment results, which showed high release on nitrate-N in CP biochars relative to CP products

(feedstock and ash). The lower dry matter yield in CP feedstock and CP ash treatments than the CP biochars (same N rate added), could be explained by the lower N released from the CP feedstock and CP ash, as shown in the preliminary incubation experiment, resulting in lower N uptake and biomass production (yield). For the ash-based products the low N uptake and shoot yield could be a result of N being lost due to high pH in CP ash. Similar findings of N losses were observed when urea was added to a soil treated with plant ash (Martikainen, 1985; Raison and McGarity, 1978). For the feedstock-based products, the lower N uptake and dry matter yield was due to slower N release from the CP product due to the presence of low water-soluble carbohydrates hindering the decomposition of the organic material (Charmley et al., 2006).

The negative control consistently showed lower dry matter than other treatments (Figure 4.5), which can be attributed to limited N uptake (Figure 4.6). This resulted from the insufficient soil N content (0.165%) and a lower pH (5.08). These findings were further supported by low pH values presented in Tables 4.4. The higher K uptake in the CP biochar (350 and 650 °C) treatments, even though all the treatments received the same amount of K (KCl), could be attributed by the high K concentration in the original feedstocks in comparison to that of pine park (Vilakazi et al., 2023), this could also explain the high dry matter. The higher Na concentration in spinach tissue fertilised with urine-based products (at 3 time the recommended N rate) may be due to over application of these products than the control, introducing more Na, which the plants likely absorbed. Human urine has high Na concentration (Beler-Baykal et al., 2011; Martin et al., 2022). However, it is well documented that high Na affects plant growth and yield (Friedler et al., 2013; Martin et al., 2022). Despite this, spinach did not show negative effects, possibly because, it is considered a moderately salt-tolerant crop (Ors and Suarez, 2016; Xu and Mou, 2016). This suggests that urine-based products should only be used to fertilize crops that are tolerant to high Na concentrations and their continuous application need to be monitored to prevent soil salinity especially in arid areas.

The plant micronutrients (including Cu, Fe, and Zn) found in spinach were all in the permissible limit (Kabata-Pendias, 2000). The high Mn concentration (Table 4.3) after fertilisation with CP feedstock products was above the 500 ppm tissue threshold (Adriano, 2001; Kabata-Pendias, 2000). Excess available soil Mn leads to bio-accumulation in plants (Patnaik, 2003) and inhibits chlorophyll biosynthesis, photosynthesis rate (Shi et al., 2006), CO_2 assimilation and stomatal conductance (Santos et al., 2017). It further changes the uptake, redistribution and use of nutrients as N, P, Ca, Mg, and Fe, which are essential for normal plant growth (Huang et al., 2016). Similar findings were reported where spinach yield was not affected by Mn toxicity

following application of wastewater (Nawaz et al., 2019; Pathak et al., 1999). However, CP feedstock had low dry matter yield, and this could be explained by minimal release of nitrate-N in combination with the Mn solubility. These concentrations accumulated in the spinach was above the permissible limit (Kabata-Pendias, 2000). Although higher than the normal sufficiency range, the Mn levels did not reach phytotoxic thresholds, suggesting that the urine-based fertilisers supplied essential micronutrients without compromising crop or food safety (Kabata-Pendias, 2000).

At the end of the experiment, CP ash showed high soil pH due to its high liming potential, these also increased extractable P a resulted of decrease in soil acidity, which improved P availability (Singh et al., 2010; Xu et al., 2014).

4.5 Conclusion

Urine-enriched cull potato ash (CP ash), significantly improved soil pH and extractable P, showing its strong liming potential and phosphorus availability. However, it released limited mineral N, suggesting its limited use as a nitrogen fertiliser. In contrast, CP-derived biochars increased spinach dry matter yield and nitrogen uptake, showing their potential as N fertiliser substitutes for smallholder farmers. Despite lower N retention during dehydration, these biochars depicted significant nitrate-N release, likely due to mineralisation of the original N in the products. Applying the treatments at triple the recommended resulted in succulent spinach leaves with dark green colour, which implies excess N application. Future research should explore the effects of urine-based products on crop growth in both controlled and field settings when applied at the recommended rate.

Chapter 5: Nitrogen release from dehydration products of urine mixed with pine bark-based materials and nitrogen fertiliser value of products: a glasshouse study with spinach

Abstract

The recycling of human urine has gained significant attention due to its high nitrogen (N) content. Nutrient recovery from human urine, using solid products, could be a sustainable approach that closes nutrient loss loop, with valuable products for soil fertility improvement. This study evaluated effects of various products derived from dehydration of urine, mixed with solid materials from pine bark (PB) on mineral nitrogen (N) and phosphorus (P) in soil. This study evaluated effects of various products derived from dehydration of urine mixed with various materials from pine bark (PB) on spinach growth, nutrient uptake and micronutrients concentrations under glasshouse conditions. The PB materials included feedstock, biochar (pyrolysed at 350 and 650 °C) and ash. The mixtures of urine and these materials were dehydrated at 45 and 60 °C. Others from PB were also dehydrated in a glasshouse, with biochar produced from a field kiln. The dehydration products were applied to soils at the recommended N (100 kg ha^{-1}) rate for spinach, in both experiments, respectively. The soils were incubated at field capacity moisture and 25 °C temperature. Destructive sampling was done at days 0, 7, 14, 21, 28, 42, and 56 for both experiments, with analysis of pH, ammonium- and nitrate-N and extractable P. The PB based products used in the incubation experiments were also used for the glasshouse pot experiment. In the first experiment, the products dehydrated at 45 and 60 °C were applied to 2kg of soil (0-20 cm depth) at recommended N rate for spinach. A second experiment was done with PB materials dried in the glasshouse, with biochar produced from the field kiln. The pot trials ran for 84 days with plant sampling at 6 and 12 weeks and analysis for dry matter, nutrient uptake and tissue micronutrients. The residual soil samples were analysed for pH and extractable phosphorus (P). In the incubation experiment, nitrate-N increased with decrease in ammonium-N, reaching the highest 37.2 mg kg^{-1} in PB ash-45 and 38.7 mg kg^{-1} in PB ash dehydrated at the glasshouse. Urine-based products increased dry matter and N uptake of spinach, with high tissue sodium (Na) concentration. In both glasshouse experiments, the PB products had high dry matter, which was comparable to the positive control. Also, the high manganese toxicity will need to be monitored in PB feedstock and PB biochar. These findings demonstrates that Pine bark (PB) urine-based products, increase

nitrate-N, in soil and can be used as an alternative to synthetic N fertilisers for smallholder farmers but Na and Mn in the tissue need to be monitored.

Keywords: biochar, dehydration, dry matter; spinach; mineral nitrogen, urine-based products

5.1 Introduction

Globally, Sustainable sanitation poses a significant challenge in rural and peri-urban areas (Nasim et al., 2022). over 80% of wastewater, which includes human urine, is discharged to the environment with little or no treatment (van den Broek et al., 2024). Large amount of nutrients such as nitrogen (N), phosphorous (P) and potassium (K) in the wastewater contribute negatively to the environment (Sene et al., 2019). The high concentration of total N (0.74%), K (1.62%), and P (0.02%) in human urine suggest that, if its source-separated through ecological sanitation approaches (Guest et al., 2009; Larsen et al., 2009), its application could have a significant contribution to soil fertility. However, the nitrogen could be lost to the atmosphere through ammonia volatilisation, while practical challenges associated with transportation costs, due to a huge proportion of human urine being water, makes it complicated to benefit from fertilisation with urine. Recovery of N by using different substrates has, therefore, become an emerging research focus. Adding a substrate that will increase or lower pH, could inactivate the urease enzyme responsible for hydrolysis of urea to ammonia and result in absorption of the N, which allow for dehydration of the excess water, producing a potentially valuable product.

Different urine-based products have been produced by modifying urine pH followed by dehydration, resulting products with high nutrient concentrations and monetary value (Simha et al., 2021; Simha et al., 2020a). Modifying urine with $Ca(OH)_2$ followed by drying resulted in a product with 9.3% N, 0.7% P and 3.9% K (Simha et al., 2020a). Furthermore, a urine-based fertiliser from with wood ash produced a nutrient-rich end product with concentration of nutrients (9.7% N, 1.5% P and 8.4% K) than in the original fresh urine (Simha et al., 2020a). The composition of the substrate used in dehydration plays a vital role in determining the final end product. Biochar, for example, can yield carbon-rich fertiliser products, enhancing both soil fertility and carbon sequestration (Nguyen et al., 2018). Having different solid media for dehydration would produce a variety of urine-based fertiliser materials which differ in their contributions to stored carbon and available nutrients in the soil. Understanding the nutrient release patterns of these products in soil is essential to assess their fertiliser value. Preliminary

results indicate that using acidic pine bark as a substrate and dehydrating at 45 °C and 60 °C resulted in nitrogen recovery rates of 2.23% and 2.96%, respectively. However, the effectiveness of these products may depend on factors such as the degradation rate of the substrates driven by C/N ratio, lignin content (Bird et al., 1999), and overall carbon content as well as the solubility of potentially free (unadsorbed urea), which is influenced by both product and soil characteristics.

Currently, there is limited evidence on how urine-enriched pine bark products affect soil nutrient release, and their agronomic effectiveness remains largely limited. Nitrogen is a critical macronutrient and often a limiting factor for crop yield. Human urine, due to its high nitrogen content (Friedler et al., 2013)., has been successfully utilised in agriculture, enhancing the productivity of crops such as maize (Kassa et al., 2018), tomato (Pradhan et al., 2009), cucumber (Heinonen-Tanski et al., 2007), and spinach (Rumeau et al., 2024). For instance, Pradhan et al. (2009) demonstrated that combining urine with wood ash led to more than a three-fold increase in tomato yield compared to control treatments. Panigrahi (2013) reported that urine-enriched coconut shell biochar resulted in a better French bean growth and yield, reflecting and confirming the nutrient availability in soils. These findings highlight the potential of urine-based products as viable fertiliser sources.

Preliminary experiments in our study showed that pine bark, its biochar, and ash are effective in recovering nitrogen from synthetic human urine, suggesting that they may also perform well in soils. However, the nutrient release pattern and crop productivity effects of these products have not been tested. Therefore, the objective of this study was to determine the effect urine-based products of feedstock, biochar and ash from pine bark, when dehydrated at 45 and 60 °C and under glasshouse conditions, on nutrient release patterns and spinach productivity of amended soils.

5.2 Materials and methods

5.2.1 Experimental soil

The experimental soil used in this study was collected under natural vegetation at the Ukulinga research farm of the University of KwaZulu-Natal, South Africa, as detailed in Chapter 4 (Section 4.2). The characteristics of the soil are also detailed in Chapter 4 (Section4.2.1) Table 4.1).

5.2.2 Urine-based product production and characterisation

The urine-based products used in this study were produced from pine bark (PB) and the methods for producing urine-based products and their characterisations are as described in chapter 3, sub-section 3.2.3.

5.2.3 Experimental design

The incubation experiments were conducted consecutively, with the findings from one experiment guiding the design of the next.

5.2.4 Incubation experiments

The first incubation experiment was set up in a completely randomised design with urine-based products, with three replicates. The urine-based products were from PB feedstock, and PB ash, all dehydrated at 45 and 60 °C. The products were applied to 100 g of soil at the recommended N rate for spinach (equivalent to 100 kg ha^{-1}). Unamended soil (no urine-based product) was also included as a control. The incubation procedure, management, sampling and analyses were as detailed in Chapter 4 (Section 4.2.3). The soils were incubated with destructive sampling at 0, 7, 14, 21, 28, 42, and 56 days, and analysed for ammonium- and nitrate-N, extractable P and soil pH.

Experiment as experiment 1 was repeated with urine-based products from PB feedstock, biochar (from field kiln) (Experiment 2) and ash dehydrated in the glasshouse (PB G, PB biochar G and PB ash G), with all other materials, management and measurements being the same. The details of analysis of ammonium- and nitrate-N and AMBIC-2 extractable P are as detailed in Chapter 4 (Section 4.2.3.1).

The same soil and urine-based products used in the incubation experiments were also used in the glasshouse pot experiments. The details of the pot trial are as described in Chapter 4, except that the materials were added to the soil at rates equivalent to 100 kg N ha^{-1} (recommended for spinach). In the first pot trial, products of dehydration at 45 and 60 °C were used. For the second experiment, products of dehydration under glasshouse conditions were used (with the biochar produced using a field kiln). The parameters measured and the details of measurement are as described in Chapter 4.

Tissue composition of N, P, Ca, Mg K, Fe, Mn, Zn and Cu were measured, and the N and P uptake were calculated using equation 5.1.

$$\text{N/P uptake (g plot}^{-1}\text{)} = \text{dry matter yield (g plot}^{-1}\text{)} * \frac{(N \,/\, P\ concentration)}{100} \qquad \text{Equation 5-1}$$

Soil pH and extractable P in the residual soils were analysed.

5.2.5 Statistical analysis

The results of soil pH, mineral-N (ammonium- and nitrate-N) and extractable P were analysed using GenStat 18th edition (VSN International 2015) by subjecting them to the two-way analysis of variance (ANOVA). The results were analysed separately for different sampling days for each experiment. All data for the pot experiments were also analysed using one-way ANOVA. One-way ANOVA was used because the controls did not have the same number of levels as the feedstocks and dehydration condition combinations. Separation between treatment means was done using least significant difference (LSD) at $p < 0.05$. The Tukey HSD test was also used to separate treatment means at $p < 0.05$.

5.3 Results

5.3.1 Changes in Ammonium-N

There was a sharp increase in ammonium-N concentration within the first 7 days of the incubation for all the treatments (Figure 5.1a, b). A general decrease occurred from day 7-56, where all the treatments were below detection at day 56 (Figure 5.1a). The ammonium-N concentration decreased throughout the incubation period from day 7 up to 42 (Figure 5.1a, b) and stabilised between days 42-56.

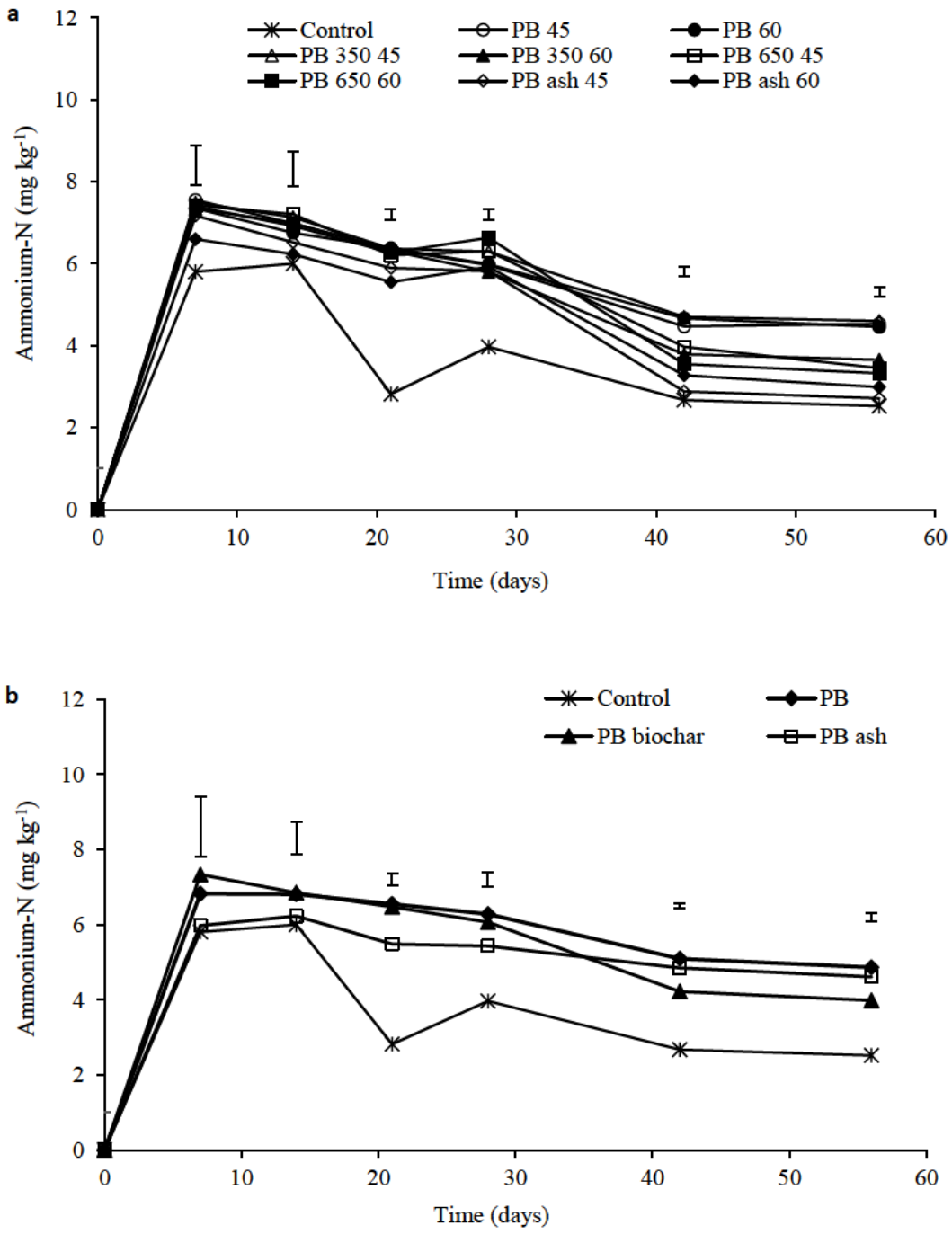
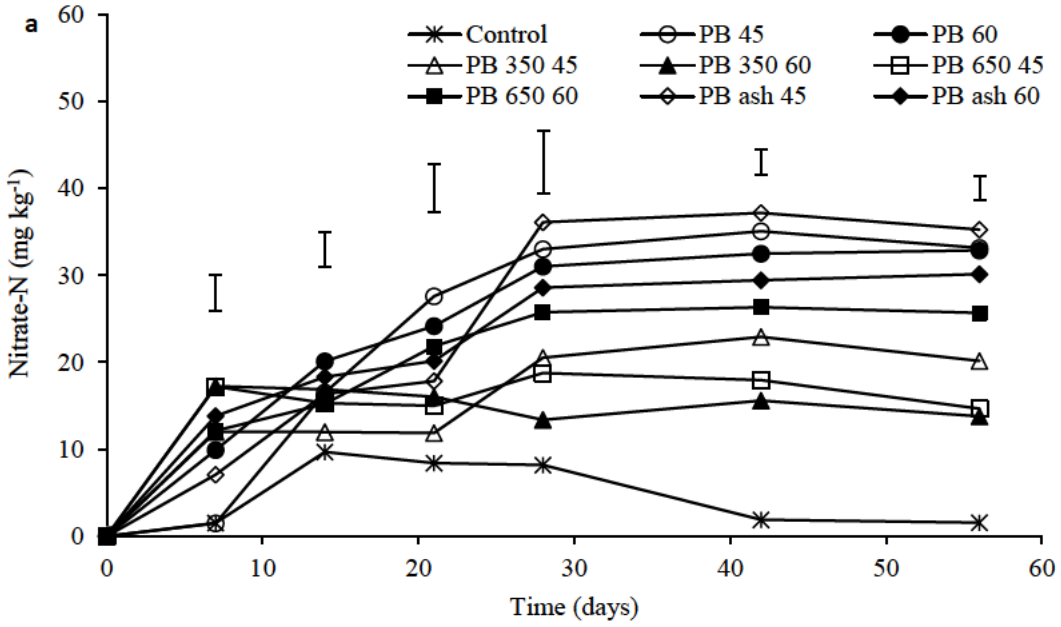


Figure 5.1: Concentration of Ammonium-N released during incubation of the products to soil in (a) experiment 1 (b) experiment 2. PB ash 45 and PB ash 60- pine bark ash products dehydrated at 45 and 60 °C, respectively; PB 350 45 and PB 350 60- product of pine bark biochar at 350 ˚C dehydrated at 45 and 60 °C, respectively; PB 650 45 and PB 650 60- pine bark biochar at 650 ˚C dehydrated at 45 and 60 °C, respectively; PB G, PB biochar G and PB ash G – products of pine bark, pine bark biochar from the kiln and pine bark ash, respectively, dehydrated in the glasshouse. The vertical error bars indicate LSD ($p<0.05$).

5.3.2 Changes in Nitrate-N

Application of urine-based products significantly increased nitrate-N concentration (Figure 5.2a, b). In the experiment, the maximum nitrate-N was 37.159 mg kg^{-1} (PB ash-45) at day 28 (Figure 5.2a). All treatments showed an increase from day 0, stabilizing between day 28 and day 56, following a similar trend. The order of effectiveness was as follows: PB ash-45 > PB-45 = PB-60 > PB ash-60 > PB 650-60 < PB 350-45 > PB 650-45 > PB 350-60 > Control. Of the products dehydrated in the glasshouse, the one from PB ash showed the higher increase in nitrate-N than those from raw PB and PB biochar (not significantly different), especially from day 14, reaching a maximum of 38.7 mg kg^{-1} and stabilizing between day 28 and day 56 (Figure 5.2b). The control had lower nitrate N that all other treatments throughout the incubation period (Figure 5.2b).



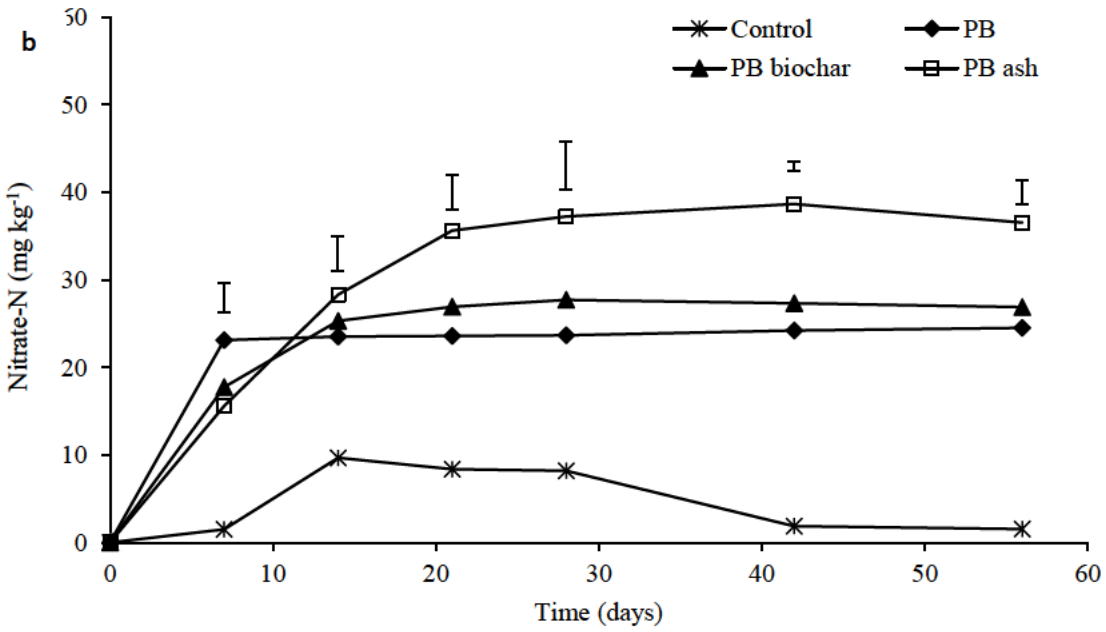


Figure 5.2: Concentration of nitrate-N released during incubation of the products to soil in (a) experiment 1 (b) experiment 2. PB 45 and PB 60- pine bark product dehydrated at 45 and 60 °C, respectively; PB ash 45 and PB ash 60- pine bark ash products dehydrated at 45 and 60 °C, respectively; PB 350 45 and PB 350 60- product of pine bark biochar at 350 ˚C dehydrated at 45 and 60 °C, respectively; PB 650 45 and PB 650 60- pine bark biochar at 650 ˚C dehydrated at 45 and 60 °C, respectively; PB G, PB biochar G and PB ash G – products of pine bark, pine bark biochar from the kiln and pine bark ash, respectively, dehydrated in the glasshouse. The vertical error bars indicate LSD ($p<0.05$).

5.3.3 Changes in extractable P

Extractable-P decreased in all treatments until day 28 (Figure 5.3a, b), except for PB ash-60, which increased and peaked at day 21, reaching 29.8 mg kg-1(Figure 5.3a), followed by decrease until day 28. Following the decrease, all the treatments increased between days 28-42, then stabilised until day 56 (Figure 5.3a, b). Extractable-P increased for both PB biochar and PB ash, reaching a maximum of 44.2 mg kg^{-1} on day 21, followed by a decrease until day 28 (Figure 5.3b). In contrast, PB showed a decrease in extractable P from day 0 until day 28.

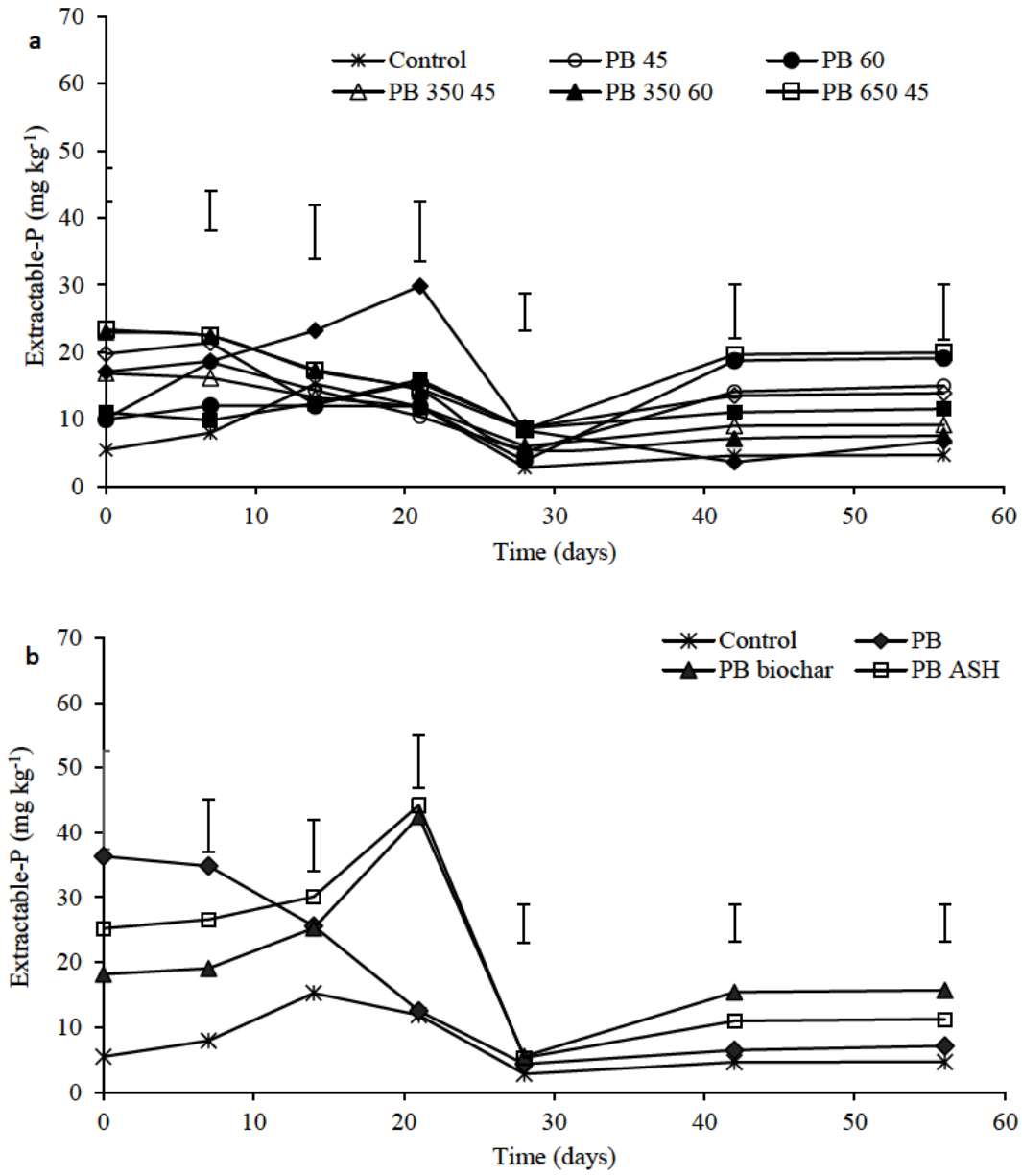


Figure 5.3: Concentration of extractable-P released during incubation of the products to soil in (a) experiment 1 (b) experiment 2. PB 45 and PB 60- pine bark product dehydrated at 45 and 60 °C, respectively; PB ash 45 and PB ash 60- pine bark ash products dehydrated at 45 and 60°C, respectively; PB 350 45 and PB 350 60- product of pine bark biochar at 350 °C dehydrated at 45 and 60 °C, respectively; PB 650 45 and PB 650 60- pine bark biochar at 650 °C dehydrated at 45 and 60 °C, respectively; PB G, PB biochar G and PB ash G – products of pine bark, pine bark biochar from the kiln and pine bark ash, respectively, dehydrated in the glasshouse. The vertical error bars indicate LSD ($p<0.05$).

5.3.4 Changes in soil pH

There were significant differences in soil pH throughout the incubation (Figure 5.4a, b). Soil pH decreased throughout the incubation with PB ash-60 and PB ash-45 being significantly higher than other treatments (Figure 5.4a). At day 42 treatments, PB-60, PB biochars (350 and

650) dehydrated at 45 and 60, were below the control up to day 56. Soil pH generally decreased throughout the incubation period, with PB ash being significantly higher than other treatments (Figure 5.4b).

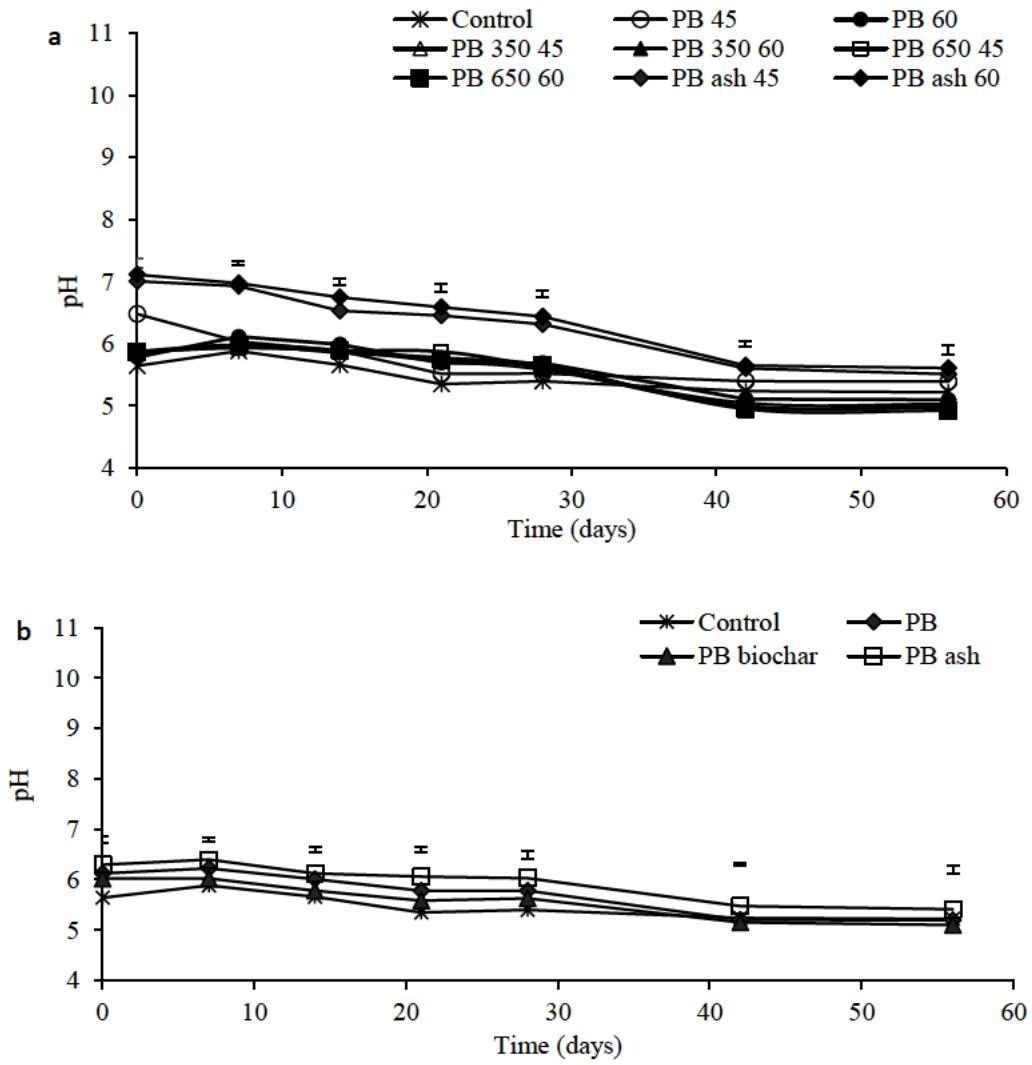


Figure 5.4: Changes in soil pH during incubation of the products to soil in (a) experiment 1 (b) experiment 2. PB 45 and PB 60- pine bark product dehydrated at 45 and 60 °C, respectively; PB ash 45 and PB ash 60- pine bark ash products dehydrated at 45 and 60 °C, respectively; PB 350 45 and PB 350 60- product of pine bark biochar at 350 ˚C dehydrated at 45 and 60 °C, respectively; PB 650 45 and PB 650 60- pine bark biochar at 650 ˚C dehydrated at 45 and 60 °C, respectively; PB G, PB biochar G and PB ash G – products of pine bark, pine bark biochar from the kiln and pine bark ash, respectively, dehydrated in the glasshouse. The vertical error bars indicate LSD ($p<0.05$).

5.3.5 Dry weight

Application of urine-based products increased the dry matter yield of spinach (Figures 5.5a, b). There were no major differences between dehydration temperatures (45 and 60 °C) for each product type. In the experiment (products from PB materials applied at 100 kg N ha-1), the positive control yielded (5.9 g pot-1) the highest dry matter, comparable to products PB ash and PB biochar 650, which were all higher than the negative control (Figure 5.5a). Dry matter yield for PB biochar 350 and PB-45 was similar to the negative control (3 g pot-1). In the second experiment, all the products from PB materials had similar spinach dry matter to the positive control and were significantly higher that the negative control (3 g pot^{-1}) (Figure 5.5b). The PB ash (6.7 g pot^{-1}) resulted in higher dry matter yield than that from PB feedstock product.

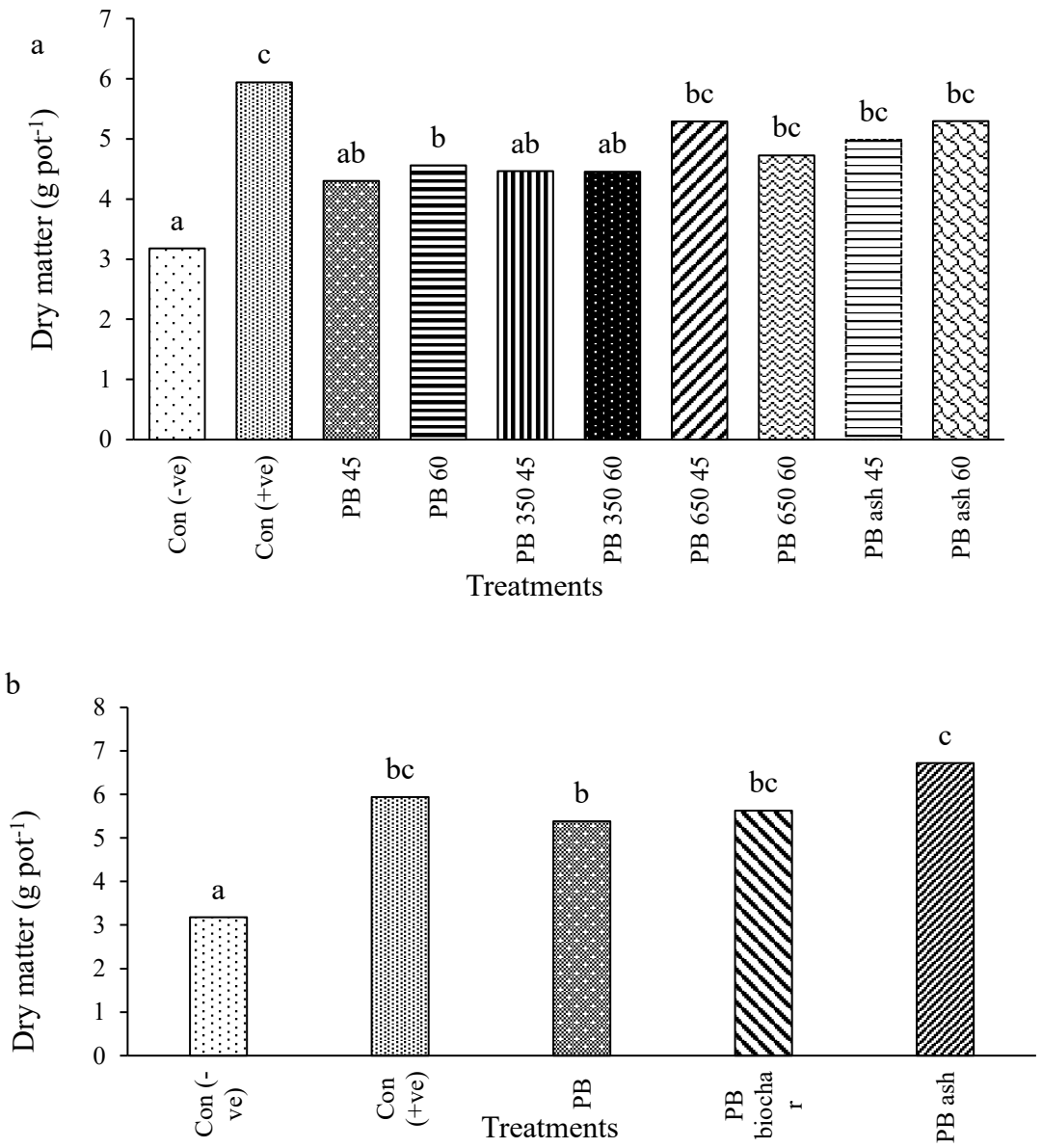


Figure 5.5: Dry matter yield of spinach as affected by different urine-based products (a) experiment 1, (b) experiment 2. PB 45 and PB 60- pine bark product dehydrated at 45 and 60 °C, respectively; PB ash 45 and PB ash 60- pine bark ash products dehydrated at 45 and 60 °C, respectively; PB 350 45 and PB 350 60- product of pine bark biochar at 350 °C dehydrated at 45 and 60 °C, respectively; PB 650 45 and PB 650 60- pine bark biochar at 650oC dehydrated at 45 and 60 °C, respectively; PB G, PB biochar G and PB ash G – products of pine bark, pine bark biochar from the kiln and pine bark ash, respectively, dehydrated in the glasshouse. The vertical error bars indicate LSD ($p<0.05$).

5.3.6 Nitrogen uptake

In the first experiment, the PB biochars (350 and 650 °C) had significantly higher N uptake than the negative control, while the PB feedstock and PB ash were not different from both the control. In experiment 2, PB ash treatment had the highest N uptake among all treatments,

following a trend of PB ash> PB biochar= PB feedstock= positive control> negative control (Figure 5.6b).

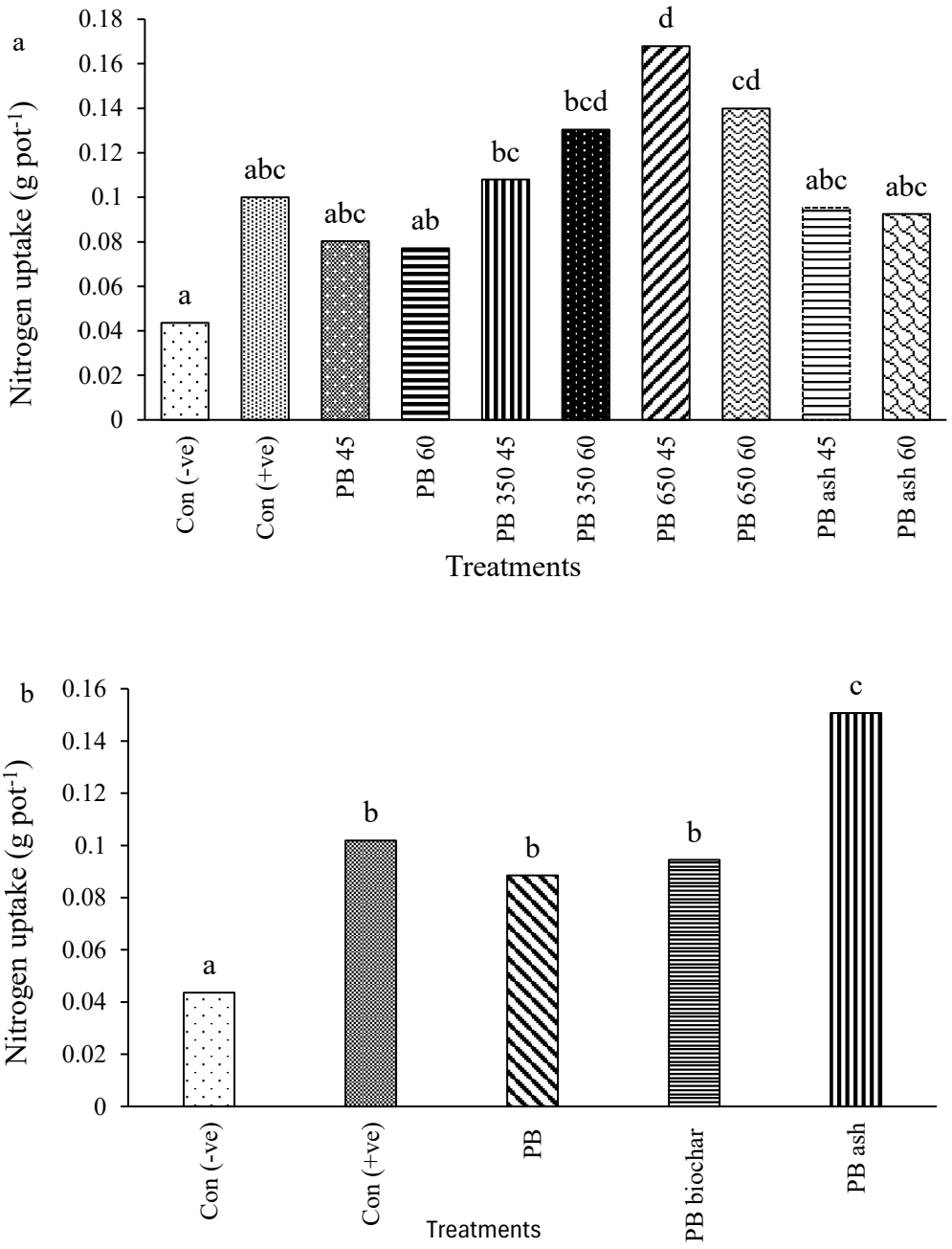


Figure 5.6: The nitrogen uptake of spinach fertilised with different urine-based products. (a) experiment 1, (b) experiment 2. PB 45 and PB 60- pine bark product dehydrated at 45 and 60 °C, respectively; PB ash 45 and PB ash 60- pine bark ash products dehydrated at 45 and 60 °C, respectively; PB 350 45 and PB 350 60- product of pine bark biochar at 350 °C dehydrated at 45 and 60 °C, respectively; PB 650 45 and PB 650 60- pine bark biochar at 650 °C dehydrated at 45 and 60 °C, respectively; PB G, PB biochar G and PB ash G – products of pine bark, pine bark biochar from the kiln and pine bark ash, respectively, dehydrated in the glasshouse. The vertical error bars indicate LSD ($p<0.05$).

5.3.7 Uptake of P and K and tissue concentrations of Ca, Mg and Na

In Experiment 1, there were no significant differences in P and K uptake and tissue Ca and Mg concentration for the PB products in the second experiment (Table 5.1). The concentration of Na was significantly lower in the negative control compared to all the other treatments which were similar to each other.

Table 5.1: The uptake of P and K and Ca, Mg and Na concentration of spinach when urine-based products dehydrated at 45 and 60 °C are being applied at the recommended rate.

Treatments	Tissue uptake		Tissue concentration		
	P	K	Ca	Mg	Na
	(g pot^{-1})		(%)		
Con (-ve)	0.0195^{a}	0.138^{a}	0.709^{a}	0.705^{a}	0.646^{a}
Con (+ve)	0.0209^{a}	0.212^{a}	0.510^{a}	0.624^{a}	1.19^{b}
PB 45	0.0226^{a}	0.173^{a}	0.695^{a}	0.694^{a}	1.29^{b}
PB 60	0.0203^{a}	0.166^{a}	0.684^{a}	0.752^{a}	1.45^{b}
PB 350 45	0.0170^{a}	0.162^{a}	0.625^{a}	0.729^{a}	1.36^{b}
PB 350 60	0.0183^{a}	0.187^{a}	0.533^{a}	0.691^{a}	1.37^{b}
PB 650 45	0.0211^{a}	0.212^{a}	0.603^{a}	0.765^{a}	1.59^{b}
PB 650 60	0.0193^{a}	0.170^{a}	0.693^{a}	0.678^{a}	1.29^{b}
PB ash 45	0.0219^{a}	0.179^{a}	0.651^{a}	0.715^{a}	1.50^{b}
PB ash 60	0.0192^{a}	0.173^{a}	0.651^{a}	0.767^{a}	1.27^{b}

*Values on the same column with different letters indicate significant differences ($p<0.05$). PB= Pine bark waste.

The P uptake in Experiment 2 was significantly higher in PB biochar by at least 87% than the PB ash (Table 5.2). The positive control, PB and PB biochar was significantly higher than the negative control and PB ash. There were no significant differences in the tissue Ca concentration, while PB ash had significantly higher Mg concentration than both the controls and PB feedstock. The Na concentration was significantly higher in PB ash than other treatments. The positive control, PB feedstock, PB biochar had similar Na which was lower than PB ash and higher than the negative control.

Table 5.2: The uptake of P and K and Ca, Mg and Na concentration of spinach when urine-based products dehydrated at the glasshouse (G) are being applied at the recommended rate.

Treatments	Tissue uptake		Tissue concentration		
	P	K	Ca	Mg	Na
	(g pot^{-1})		(%)		
Con (-ve)	0.0195^{ab}	0.138^{b}	0.709^{a}	0.705^{a}	0.781^{a}
Con (+ve)	0.0197^{ab}	0.194^{c}	0.510^{a}	0.624^{a}	1.19^{b}
PB	0.0233^{ab}	0.175^{bc}	0.535^{a}	0.610^{a}	1.40^{b}
PB biochar	0.0293^{b}	0.211^{c}	0.608^{a}	0.786^{ab}	1.43^{b}
PB ash	0.0157^{a}	0.092^{a}	0.631^{a}	1.06^{b}	2.35^{c}

*Values on the same column with different letters indicate significant differences ($p<0.05$). PB= Pine bark waste.

5.3.8 Cu, Zn, Fe, and Mn concentration

In Experiment 1, the Cu concentration did not vary significantly across the treatments, while the Zn concentration was significantly higher in PB feedstock, PB 350-60, PB 650-60 and PB ash-60 than the negative control (Table 5.3). Tissue Fe concentration was highest in PB ash-45 which was significantly greater than the concentration in the positive control. The PB feedstock had higher Mn concentration than other treatments, except for PB 650-60.

Table 5.3: The Cu, Zn, Fe and Mn concentration of spinach when urine-based products dehydrated at 45 and 60 °C are being applied at the recommended rate.

Treatments	Tissue concentration (mg kg^{-1})			
	Cu	Zn	Fe	Mn
Con (-ve)	8.94^{a}	32.4^{a}	144^{ab}	396^{abc}
Con (+ve)	6.34^{a}	80.9^{bc}	89.7^{a}	429^{abc}
PB 45	9.45^{a}	68.2^{b}	207^{a}	692^{e}
PB 60	9.50^{a}	68.3^{b}	110^{a}	653de
PB 350 45	7.46^{a}	61.5^{ab}	145^{ab}	498^{bc}
PB 350 60	7.91^{a}	110^{c}	157^{ab}	346^{a}
PB 650 45	7.40^{a}	58.5^{ab}	69.8^{a}	451^{abc}
PB 650 60	7.38^{a}	68.8^{b}	133^{ab}	521^{cd}
PB ash 45	5.65^{a}	59.5^{ab}	244^{b}	395^{abc}
PB ash 60	5.04^{a}	73.2^{b}	211^{ab}	384^{ab}

*Values on the same column with different letters indicate significant differences ($p<0.05$). PB= Pine bark waste.

Both Cu and Zn concentration in Experiment 2 were significantly higher in the PB ash treatment than all other treatments, which had low concentrations (Table 5.4). The PB biochar treatment exhibited a higher Fe concentration than all other treatments. The PB biochar treatment also had the highest Mn concentration among treatments, except for PB ash, which showed a similar Mn concentration.

Table 5.4: Concentration of spinach when urine-based products dehydrated at the glasshouse (G) are being applied at the recommended rate.

Treatments	Tissue concentration (mg kg^{-1})			
	Cu	Zn	Fe	Mn
Con (-ve)	8.94^{a}	41.2^{a}	257^{a}	348^{ab}
Con (+ve)	6.34^{a}	87.3^{a}	217^{a}	355^{ab}
PB	6.68^{a}	54.6^{a}	234^{a}	256^{a}
PB biochar	8.74^{a}	59.1^{a}	197^{a}	787^{c}
PB ash	18.2^{b}	157.9^{b}	198^{a}	558^{bc}

*Values on the same column with different letters indicate significant differences ($p<0.05$). PB= Pine bark waste

5.3.9 Soil pH and extractable P after spinach harvest

In Experiment 1, there was no significant difference in soil pH across the treatments (Table 5.5). Extractable P was significantly higher in the PB feedstock and PB ash treatments compared to the negative control, PB biochar at 350 °C, and PB 650-45.

Table 5.5: Soil pH and extractable P concentration after spinach harvest as affected by different urine-based products dehydrated at 45 and 60 °C applied at the recommended rate.

Treatments	pH $_{(KCl)}$	Extract. P (mg kg^{-1})
Con (-ve)	5.69^{a}	2.13^{a}
Con (+ve)	5.60^{a}	10.22^{abc}
PB 45	5.62^{a}	17.4^{bcd}
PB 60	5.57^{a}	22.3^{cd}
PB 350 45	5.50^{a}	10.4^{abc}
PB 350 60	5.8^{a}	7.60^{ab}
PB 650 45	5.84^{a}	3.99^{a}
PB 650 60	5.88^{a}	9.34^{abc}
PB ash 45	5.98^{a}	18.88^{bcd}
PB ash 60	5.85^{a}	26.7^{d}

*Values on the same column with different letters indicate significant differences ($p<0.05$). PB= Pine bark waste.

In Experiment 2, the pH was significantly higher in the PB feedstock treatment (5.87) by 0.4 units compared to PB ash (Table 5.6). The positive control had a significantly higher level of extractable P than both the feedstocks and the negative control.

Table 5.6: Soil pH and extractable P concentration after spinach harvest as affected by different urine-based products dehydrated at the glasshouse (G) and applied at the recommended rate.

Treatments	pH $_{(KCl)}$	Extract. P (mg kg^{-1})
Con (-ve)	5.69^{ab}	2.13^{a}
Con (+ve)	5.60^{ab}	10.2^{b}
PB	5.87^{b}	2.08^{a}
PB biochar	5.74^{ab}	5.17^{ab}
PB ash	5.47^{a}	3.36^{ab}

*Values on the same column with different letters indicate significant differences ($p<0.05$). PB= Pine bark waste.

5.4 Discussion

The increase in ammonium-N concentration for the first 7 days (Figure 5.1 a, b) could be the result of urea hydrolysis due to addition of moisture (Abera et al., 2012). The findings also align with those of (Kissel, 1988), where urea hydrolysis occurred within 4-7 days in warm temperature with optimum moisture content. Other factors may also have influenced the nutrient release and uptake. The physicochemical properties of the pine bark materials (pH, surface area, and cation exchange capacity), together with soil pH and moisture conditions. Hydrolysis in soils is also influenced by pH, with the process generally increasing as pH rises with optimal pH range for urease enzyme activity being between 5.5 and 9 (Motasim et al., 2024). When the pH exceeds 9, hydrolysis slows down (Motasim et al., 2024). At acidic pH ($pH < 7$) most of NH_3 will be converted to NH_4^+ (Sigurdarson et al., 2018). In the current study, all the treatment had pH that was less than 9 (Figure 5.4a, b).

The rapid decline in ammonium-N for all treatments from day 7 (Figure 5.1 a, b), was attributed to nitrification, which coincides with the increase in nitrate-N (Figure 5.2 a, b). Furthermore, the increase in nitrate-N explains the decrease in pH (Figure 5.4 a, b), as nitrification releases H^+ ions, which lower soil pH as observed by (Sahrawat, 2008). This aligns with the findings of (Sigurdarson et al., 2018), who observed a decrease in pH following urea application, attributed to nitrification. Nitrate -N was high in PB ash-45 (Figure 5.2a), while the PB ash initially had no N (below detection) (Table 3.2) and it had 91% recovery of N from AHU (Table 3.5), which suggests that dissolution (Wahl et al., 2006) and hydrolysis of urea occurred and all the ammonium-N was nitrified to nitrate-N. The urea might have remained free and ash is known to release all constituent nutrients to the soil upon application and stimulate microbial activities (Demeyer et al., 2001).

At the N recommended rate, the highest nitrate-N in the PB ash-45 (Figure 5.2a) and PB ash-G (Figure 5.2b) treatments, which reached 37.2 and 38.7 mg kg^{-1} respectively, could supply an equivalent of 89.3 and 92.9 kg N ha^{-1} and less synthetic N will be required for optimum yield in Spinach since it requires 100 kg N ha^{-1}. However, under field conditions the nitrate-N released could be lower due to fluctuations of soil moisture and temperature conditions. The PB biochar (350 and 650 °C) products, dehydrated at 45 and 60 °C had significantly low nitrate-N than other treatments, although they had a C/N less than 30, which would potentially support N mineralisation. The low nitrate-N may be due to the presence of stable carbon structures,

such as aromatic rings, within the biochars, which could trap the urea, making it less accessible to urease enzyme activity (Crombie et al., 2013; Uzoma et al., 2011).

All the treatments had a C/N which is less than 30 (Table 3.4), and it is expected that mineralisation would be high in all the treatments. However, the mineralisation rate varied between treatments, indicating the differences in the ability of each urine product to release N into the soil. These differences can be attributed to variations in their functional groups, pore distribution, surface area, and chemical properties (Pathy et al., 2021). The decrease in extractable-P at the beginning of the incubation until day 28, could be attributed to P fixation to the acidic sites (Figure 5.3 a, b). During the first 28 days, rapid nitrification reduced soil pH, creating positively charged sites/colloids that increased phosphorus fixation. Pine bark materials (PB) are acidic in nature and its decomposition releases organic acids which might lower soil pH and adsorb the available P (Bolan et al., 2023; Johan et al., 2021). The decline of available P coincided with the declining pH (Figure 5.4 a, b). The sharp decline at day 28 could also be due to microbial immobilisation considering high N mineralisation in soil low in available P (3.86 mg kg^{-1}). The increase from day 28–42 could be explained by the release of the previously adsorbed P, and potential mineralisation of microbial P, thus causing an increase until day 42 and stabilise until day 56.

The higher dry matter yield after application of urine-based products, compared to the negative control, can be attributed to the greater availability and uptake of N from these nitrogen-rich products. In the glasshouse experiment, while N uptake could have contributed to biomass production, other factors could have also contributed. This is so because only products from PB biochars (both 350 and 650 °C) resulted in greater N uptake than the negative control. The results may not be explained by the uptake of other macronutrients either (Table 5.1). The observed effects suggest that the biochars may have had more positive effects on soil moisture than the other treatments, resulting in greater biomass production. It is also well noted that biochars are characterised as having high water retention (Bao et al., 2021; Lin et al., 2016). However, soil moisture was not measured in this study. The similar dry matter yield in the PB products dehydrated at the glasshouse to the positive control, could be explained by N uptake (Figure 5.6b), and to some extent K uptake but not P uptake (Table 5.1). When applied at the same N rate these urine-based products dehydrated in the glasshouse had similar effects as the mineral fertiliser, indicating that the release of the urine N is as rapid as mineral N. This could be because the N in the products is in the form of urea, which rapidly got hydrolysed when the products were added to the moist soil. This coincides with the incubation results which showed

PB ash had the highest mineral N release (equivalent to 93 kg N ha^{-1}), which was sufficient for growing spinach for optimum yield, since plant preferentially takes up the nitrate-N form (Song et al., 2015). This suggests that these treatments could serve as alternatives to synthetic N fertilisers. This approach would be beneficial for smallholder farmers, as they often have limited access to synthetic nitrogen fertilisers. Similar findings were observed in tomatoes, where application wood ash+ human urine significantly increased yield (Pradhan et al., 2009). Other studies have also reported yield increase in maize (Kassa et al., 2018; Morgan, 2003), carrot, beetroot (Mnkeni et al., 2008), and cucumber (Heinonen-Tanski et al., 2007) following the application of human urine. These yield improvements were generally attributed to the higher availability of N provided by urine.

The negative control consistently showed lower dry matter than other treatments (Figure 5.5a, b), which can be attributed to limited N uptake (Figure 5.6a, b). This resulted from the insufficient soil N content (0.165%) and a lower pH (5.08). These findings were further supported by low pH values presented in Tables 5.5 and 5.6. The pine bark (PB) products are characterised as having low nutrient concentration (P and K), which could likely explain similar P and K uptake in all the PB treatments (Table 5.2). The PB ash dehydrated at the glasshouse also showed high Na concentration, plausibly because when ash is being applied to the soil it releases all the nutrients. However, it is well documented that high Na affects plant growth and yield (Friedler et al., 2013; Martin et al., 2022). Despite this, spinach did not show negative effects, possibly because, it is considered a moderately salt-tolerant crop (Ors and Suarez, 2016; Xu and Mou, 2016). This suggests that urine-based products should only be used to fertilize crops that are tolerant to high Na concentrations and their continuous application need to be monitored to prevent soil salinity especially in arid areas.

The plant micronutrients (including Cu, Fe, and Zn) found in spinach were all in the permissible limit (Kabata-Pendias, 2000). The high Mn concentration (Table 5.3 and 5.4) after fertilisation with PB feedstock products and PB biochar dehydrated at the glasshouse (Table 5.3 and Table 5.4), was above the 500 ppm tissue threshold (Adriano, 2001; Kabata-Pendias, 2000). The high tissue Mn was attributed to the acidifying effect of PB feedstock due to presence of acidic functional groups (El-Jaoual and Cox, 1998), while for PB biochar it could be caused by the nitrification process, which increases the solubility of Mn (El-Jaoual and Cox, 1998). Excess available soil Mn leads to bio-accumulation in plants (Patnaik, 2003) and inhibits chlorophyll biosynthesis, photosynthesis rate (Shi et al., 2006), CO_2 assimilation and stomatal conductance (Santos et al., 2017). It further changes the uptake, redistribution and use of nutrients as N, P,

Ca, Mg, and Fe, which are essential for normal plant growth (Huang et al., 2016). Thus, Mn toxicity decreases the biomass yield. Spinach is considered a hyper-accumulator of heavy metal and toxic ions (Nawaz et al., 2019), which explains the observed high yield irrespective of Mn solubility. Similar findings were reported where spinach yield was not affected by Mn toxicity following application of wastewater (Nawaz et al., 2019; Pathak et al., 1999). Although higher than the normal sufficiency range, the Mn levels did not reach phytotoxic thresholds (Kabata-Pendias, 2000). The concentrations of micronutrients in spinach tissue following application of urine-based products were within permissible limits for human consumption, as defined by WHO/FAO guidelines (Rahim et al., 2024).

The residual soil treated with PB feedstock and PB ash had high extractable P, this implies that the remaining nutrient (P) can be used for the subsequent crop. To achieve optimal results, it is recommended that urine-derived fertilisers be carefully prepared to provide a balanced supply of essential nutrients, applied based on crop requirements, and used with consideration of soil pH to ensure maximum nutrient availability. Additionally, applying these fertilisers which are rich in soil C storage and associated physicochemical properties.

5.4 Conclusion

The study showed that at a similar nitrogen rate, both PB ash-45 and PB ash-G resulted in higher nitrate-N levels. This suggests that PB ash enriched with urine could serve as a nitrogen source, potentially reducing the need for synthetic nitrogen fertilisers. However, no differences in ammonium and nitrate-N were observed among the pine bark biochar-urine products in this study. Overall, the findings suggest that these products could enhance soil fertility. Application of the urine-based products, particularly those from PB material, which retained more urine-N, increased nitrogen uptake and dry matter yield of spinach and soil pH and available P compared to the unamended control. When the urine-PB products are applied at the recommended N rate, they provide a positive impact on spinach yields, which will be beneficial for smallholder farmers, who do not usually apply sufficient mineral N fertiliser due to high cost. However, using urine-based products increases Na and Mn levels in plant tissues, so care must be taken to apply them only to salt-tolerant crops and there is need for monitoring these two elements in plants and soil where these products are used. Similar studies should be conducted in field conditions to observe the effects in an uncontrolled environment.

Chapter 6: Nitrogen fertiliser value of products from dehydration of pine bark- human urine mixtures under field conditions

Abstract

There is limited information on the agronomic performance of fertiliser products from dehydration of mixtures of urine and solid materials, such as organic materials and their biochar and ash, under field conditions. This study examined the effects of products derived from dehydration of mixtures of urine with pine bark (PB) biochar (B) and ash (A) as sources of nitrogen for spinach under field conditions. The products were achieved by mixing artificial human urine with raw PB (PB-U), PB-based biochar (biochar-U) and ash (PB ash-U) from PB, followed by dehydration under the glasshouse conditions. A field trial was conducted at the Ukulinga Research Farm to evaluate the effect of these urine-based products on spinach growth over a two-season period. The products were applied at the recommended N rate for spinach (100 kg N ha^{-1}) in the first season (04/06/2024 – 04/09/2024). The PB, biochar and ash that were not pre-treated with urine were included as reference materials, while plots without N and with urea, were included as negative and positive controls, respectively. In the second season (12/09/2024 – 04/12/2024), the plots were split into two, with half treated with the same treatment at the same rate (cumulative effects), while the other half did not receive additional treatment (residual effects). In each season the field trial lasted 84 days. Spinach dry matter yield, nutrient uptake, and tissue micronutrients, and soil pH, extractable phosphorus (P), and mineral N, were determined. The pine bark urine treatments had higher dry matter yield and N uptake relative to the Pine bark un-enriched products, during the first season. Cumulative additions led to lower dry matter and nitrogen uptake in PB biochar-enriched urine (PB biochar-U), which was similar to the negative control and lower than PB-U. However, PB biochar-U showed higher residual effects than the negative control. Continuous application of urine-based fertilisers reduced the risk of soil salinity, as the initial crop absorbed most of the sodium. These findings suggest that PB urine-based (PB biochar-U) fertilisers are a viable and sustainable alternative to synthetic nitrogen fertilisers, which supports the philosophy of circular economy through resource efficiency, as promising option for smallholder farmers, although Na concentrations need to be monitored.

Keywords: Amendment; biomass; biochar; nitrogen uptake; residual effects; spinach

6.1 Introduction

Sustainable agriculture that aims at reducing the reliance on synthetic fertilisers to minimize their adverse environmental impacts is an emerging trend. Nitrogen (N) is a vital nutrient for plant growth and development, playing a key role in chlorophyll synthesis, protein formation, and overall crop productivity (Fathi, 2022). However, the efficiency of nitrogen fertilisers in agricultural systems is often limited by significant nutrient losses and associated environmental pollution (Martínez-Dalmau et al., 2021). Furthermore, the high cost of synthetic fertilisers poses an additional challenge, particularly for smallholder farmers (Chianu et al., 2012). Therefore, there is a pressing need to explore sustainable strategies which are affordable options for nitrogen fertilization to optimize crop productivity. These options could be combined with addressing the challenge of sanitation provision, where urine and faeces can be separated at source for future use as N fertiliser. Urine-based products have gained attention as innovative amendments to improve soil fertility and nutrient cycling.

The effectiveness of urine as a fertiliser has been shown in several field studies in the literature. The application of human urine has been shown to enhance cereal production in field trial (Richert Stintzing et al., 2001). Similarly, Morgan (2003) reported higher vegetable yields in urine-treated plots compared to unfertilised ones in Zimbabwe. Field trials on wheat also demonstrated increased yields with urine application compared to unfertilised plots (Akpan-Idiok et al., 2012). However, improper handling of urine can lead to pollution due to its high pH, which promotes nitrogen loss through ammonification (Geinzer, 2017). Additionally, the high transportation costs, caused by the large water content (97%) in human urine, makes it challenging to effectively use urine for fertilization. As a result, recovering nitrogen using various substrates has become an emerging research focus. Dehydrating urine with ash and other products has shown promise in retaining nitrogen

When properly treated, these products offer a readily available source of nitrogen, improving nutrient availability to crops while transforming human and organic waste into sustainable agricultural inputs. This research aligns with global efforts to promote circular economy principles, reduce waste, and transition toward more sustainable agricultural practices. Preliminary experiments demonstrated that pine bark-based materials (raw, biochar, and ash) effectively recovered N from artificial human urine (Chapter 3), rapidly released it when applied to the soil and enhanced dry matter yield of spinach in a glasshouse study (Chapters 5). The design of this field experiment was based on the results from the glasshouse study

(Chapter 5), where pine bark-based urine products, showed superior performance in nitrogen release and spinach dry matter yield. These findings provided the rationale for focusing on pine bark-based products in the field experiment, to prove their agronomic potential under realistic conditions that account for environmental variability (wetting and drying). The field trial therefore aimed to assess whether the positive responses observed under controlled conditions could be replicated or sustained in the field.

This study investigated the N fertiliser value of products of dehydration of mixture of urine with raw PB, biochar, or ash for spinach production under field conditions. Spinach is an important vegetable crop that thrives in cool, temperate climates (Kangjam et al., 2024; Shinde, 2017; Zhang et al., 2023). In warmer southern regions, spinach is grown during winter, while in cooler northern regions, it is sown in spring (Correll et al., 1994), which makes it an ideal crop for testing the fertiliser value of the urine-based fertilisers. The objective of this study was to determine the cumulative and residual effects of urine-based products derived from dehydration of mixtures with raw PB, biochar and ash, tested, on spinach yield, nutrient composition and soil available N and P under field conditions.

6.2 Methods and materials

6.2.1 Experimental site and design

6.2.1.1 Experimental site

The field experiment was conducted at the Ukulinga Research Farm of the University of KwaZulu-Natal (29° 39′ 33.9″ S; 30° 24′ 14″E). This area has warm to hot summers with a mean monthly maximum temperature of 28 °C, and mild winters with a mean monthly minimum temperature of 8.7 °C, and with annual rainfall of 717 mm. The soil is loam in texture and classified as Westleigh (Soil Classification Working Group, 2018), which was translated to Plinthic Acrisols (IUSS Working Group WRB, 2014).

6.2.1.2 Soil and urine-based products

The soil used for this study was sampled from a depth of 0–20 cm at the Ukulinga Research Farm. The soil characteristics are as described in Chapter 4. The soil had a $pH_{(KCl)}$ of 5.08 and contained 29% clay, 1.95% total carbon (C), 0.165% total nitrogen (N), and 3.86 mg kg^{-1}

extractable phosphorus (P). The pine bark urine-based products, which were dehydrated in the glasshouse and used in the second incubation experiment in Chapter 5, as well as in the second pot trial in Chapter 5, were applied in this field trial. The urine-based products were PB-U, biochar-U, ash-U and their characteristics are provided in Chapter 3.

The field trial lasted 84 days (04 June2024 – 04 September2024 for season 1 and 12 September2024 – 04 December2024 for season 2). The first planting season (04 June to 04 September 2024) occurred during the winter period, characterised by lower temperatures and limited rainfall. In contrast, the second planting season (12 September to 04 December 2024) took place during the spring to early summer period, which experienced higher temperatures and increased rainfall. During the study period the rainfall (TE525, Texas Instruments, USA) and air temperature (HMP60, Vaisala, Sweden) were measured and recorded on a datalogger (CR3000, Campbell Scientific, Utah, Logan, USA) every 60 s and total rainfall and average air temperature were calculated daily. The rainfall and temperature data for both seasons are shown in Figure 6.1.

6.2.1.3 Field trial experiment 1 (Season 1)

The experiment was set up as a randomised complete block design with treatments replicated four times. Treatments included urine-based products derived from dehydration of mixtures of artificial urine with raw PB, PB biochar and PB ash (PB-U, biochar-U and ash-U). Negative (no N) and positive (mineral N) controls were also included. The products were applied at the recommended N rate for spinach, equivalent to 100 kg ha^{-1}. The mineral N in the positive control was applied as Urea at 100 kg N ha^{-1} for optimum yield. Equivalent amounts of 124 kg P ha^{-1} and 117 kg K ha^{-1} were applied to all plots (all treatments) as monocalcium phosphate and KCl, respectively (Manson et al., 2012). The same materials (PB, biochar, ash), without urine treatment were included as reference materials on adjacent plots in the first season.

The field layout of treatments is shown in Figure 6.2. In total there were 32 plots, of which 20 plots had urine-based products and 12 was for the treatments that were not enriched with urine. The 20 plots were 2m x 5m, with 52 plants per plot. The other plots, with the reference materials were 2m x 2.5m in size, with 24 plants per plot. The spinach seedlings were planted on the 12th of June 2024 with harvesting done after 6 weeks and harvested on 4th of September 2024 at the end of the trial. Supplemental irrigation was added to make sure that water was not a major limiting factor. After 6 weeks (42 days), the spinach was harvested by cutting at 1 cm above

the soil surface and leaving younger leaves allowed to grow, until after 12 weeks where all the spinach was harvested. The biomass was then dried at 60˚C for three days to determine dry weight. The plant tissue samples harvested at 6 and 12 weeks were combined to form a composite sample and then ground (<1mm) using a blender before analysis of total N, P, Ca, Mg, Na, K, Zn, Mn, Cu, and Fe. The N and P uptake were also calculated as the product of concentrations and dry matter yield. At the end of the experiment (12 weeks) soil samples were collected from the 0-20cm depth and analysed for pH and extractable P. The soils were oven dried over night at 108 °C to correct mineral-N and available P results for moisture.

6.2.1.4 Experiment 2 (Season 2)

The second trial was set-up the same way as that described in section 6.2.1.3 in terms of management and measurements. However, plots from the first season were split in half with one part receiving the same treatment as the previous season (cumulative effects) and the other without additional treatment (residual effects). The reference materials were excluded. The field layout is shown in Figure 6.3. Each plot was 2m x 2m in size and had 24 plants. The spinach seedlings were planted on the 12th of September 2024 and harvested on the 4th of December 2024.

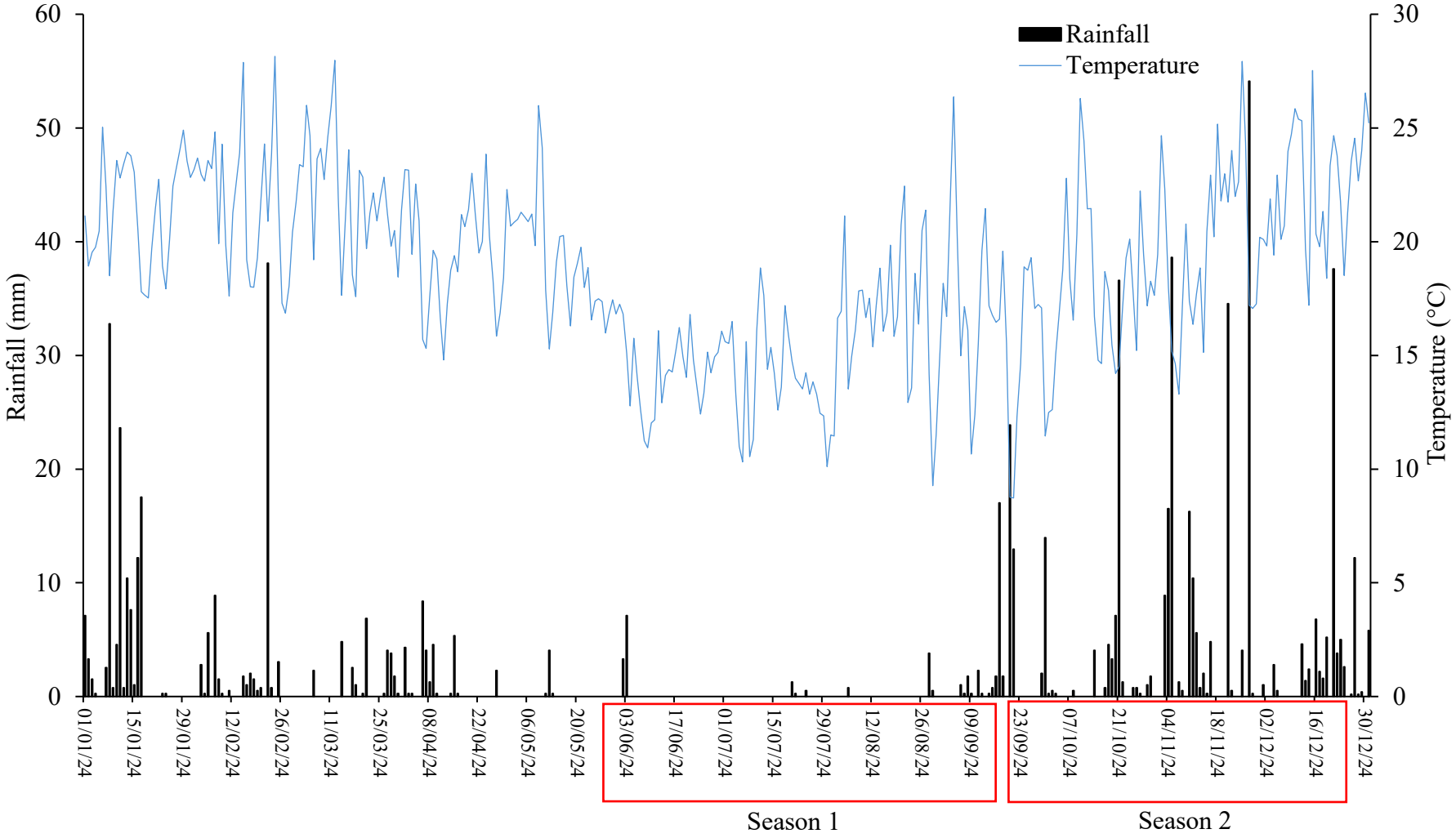


Figure 6.1: Rainfall and temperature for Ukulinga Research farm

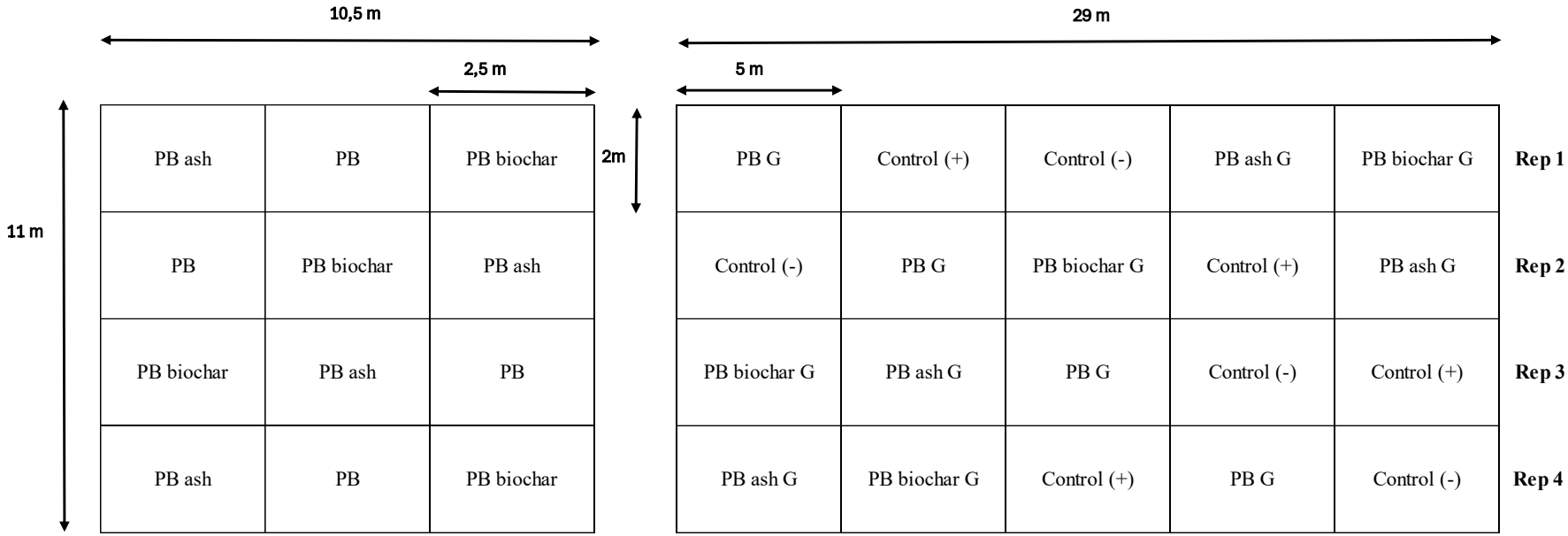


Figure 6.2: Experimental layout for the first planting season

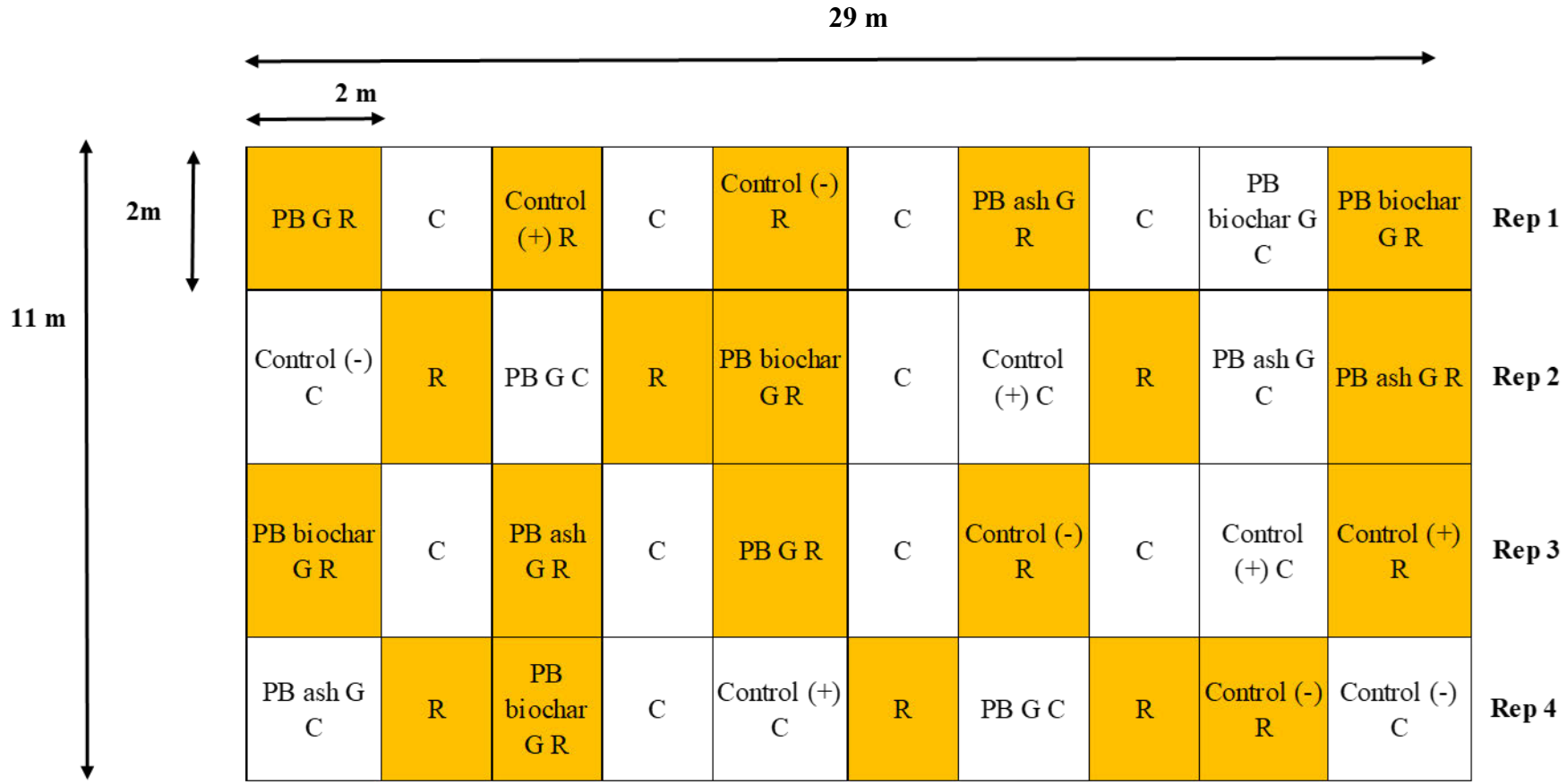


Figure 6.3:Experimental layout for the second planting season

6.2.2 Analyses

Total N was analysed using the Leco Trumac CNS Autoanalyser (Leco corporation, 3000, Lakeview, Ave, ST, Joseph, MI, USA). For other analyses, the ground spinach samples (0.5 g) were placed into crucibles, treated with 10 ml of concentrated HNO_3 and HCl (1:3) (Gupta and Sharma, 2014), and digested for one hour using a MARS CEMS microwave digester, before analysis of phosphorous (P), calcium (Ca), magnesium (Mg), potassium (K), Iron (Fe), manganese (Mn), zinc (Zn), and copper (Cu) concentration using an inductively coupled plasma atomic emission spectrometry (Varian 720-ES ICP-AES). The N and P uptake were calculated using Equation 6.1.

$$\text{N/P uptake (g plot}^{-1}) = \text{dry matter yield (g plot}^{-1}) * \frac{(N \,/\, P\ concentration)}{100} \qquad \text{Equation 6-1}$$

6.2.2.1 Soil pH, mineral-N and available P analysis

Soil pH and extractable P were analysed in the residual soils at the end of each season. Mineral-N (NH_4- and NO_3-N) was only analysed at the end of the second season. Soil pH was measured in KCl at a ratio of 1:5 using 1M KCl. Ammonium- and nitrate- N were extracted using 2M KCl and measured using Gallery auto analyser (Thermofisher scientific, Waltham, Massachusetts, USA). Extractable P was analysed using the molybdenum-blue method (Murphy and Riley, 1962), after extraction with AMBIC-2 (Manson et al., 2012).

6.2.3 Statistical analysis

All data of spinach yield, macronutrient uptake and micronutrient concentration and soil pH, mineral N and available P, were subjected to the one-way analysis of variance (ANOVA) using GenStat 18th edition. Separation of treatment means was done using least significant difference (LSD) at $p < 0.05$. The Tukey HSD test was also used to separate treatment means at $p < 0.05$.

6.3 Results

6.3.1 Dry matter yield and nitrogen uptake

Application of all the urine-based products significantly increased dry matter yield (Figure 6.4a) and N uptake (Figure 6.5a) of spinach, relative to the control, in the first season. The positive control had the highest dry matter yield (767 g $plot^{-1}$) and N uptake (25.6 g pot^{-1}), which were comparable to biochar-U, but was significantly higher than PB-U and ash-U treatments. There were no significant differences in dry matter (Figure 6.4a) and N uptake (Figure 6.5a) between all treatments enriched with urine. There were significantly higher dry matter (Figure 6.4a) and N uptake (Figure 6.5a) in plots amended with the PB products enriched with urine than corresponding ones without urine treatment, which were not different from the negative control (first season).

In the second season, cumulative application of the products, resulted in similar trend of dry matter (Figure 6.4b) and N uptake (Figure 6.5b) results compared to the first season, except that the dry matter in the biochar-U was lower than the PB-U treatment, and not different from the control. Dry matter yield (Figure 6.4b) and N uptake (Figure 6.5b) of all other treatments were significantly lower than the positive control, and significantly higher than the negative control. However, the results of the cumulative effect were numerically lower than those of the first season. The dry matter yield and N uptake results of the experiment on the cumulative effects were generally higher than those of the residual effects. For the residual effects, only the biochar-U treatment had higher dry matter yield (Figure 6.4c) and N uptake (Figure 6.5c) than both the controls. While the N uptake was not significantly different, dry matter yield in the biochar-U treatment was higher than in the PB-U treatment (Figure 6.4c).

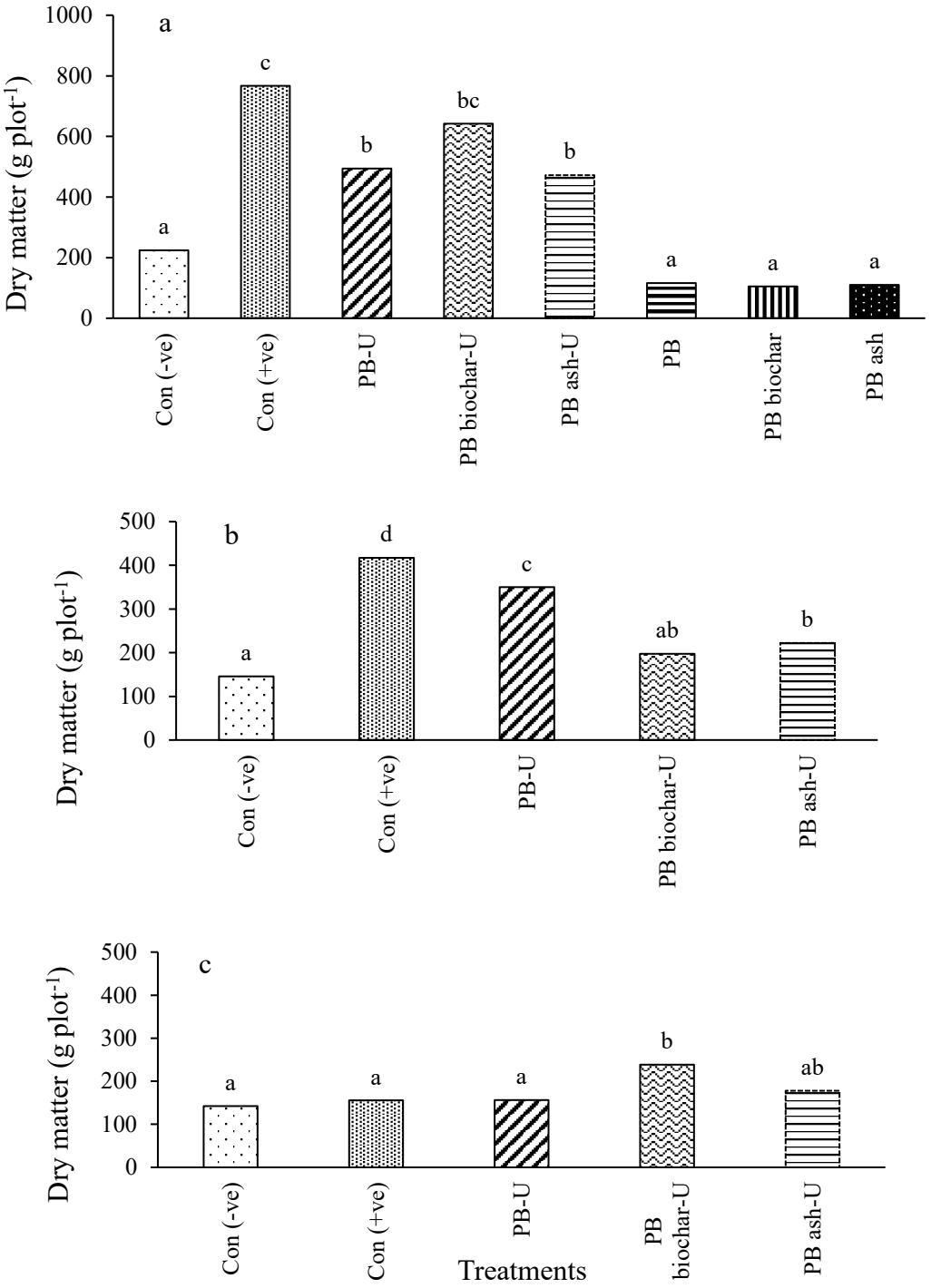


Figure 6.4: Dry matter yield of spinach as affected by different PB-urine based products and seasons (a) experiment 1, (b) experiment 2, (c) experiment 3. PB-U, PB biochar-U and PB ash-U- products of pine bark, pine bark biochar from the kiln and pine bark ash, respectively, dehydrated in the glasshouse. PB, PB biochar and PB ash- products of pine bark, pine bark biochar from the kiln and pine bark ash, respectively, with no urine enrichment.

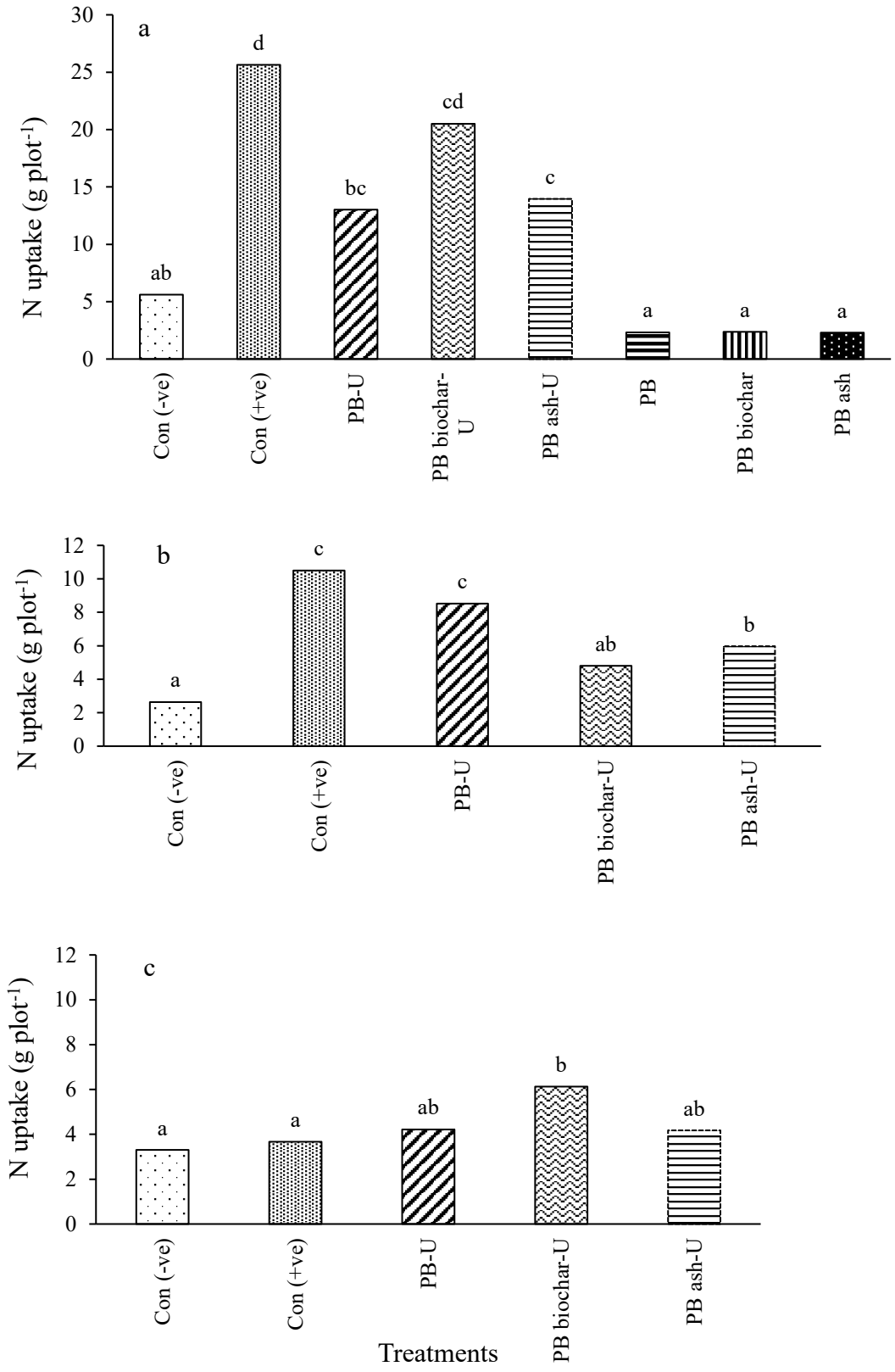


Figure 6.5: The nitrogen uptake of spinach as affected by different PB-urine based products and seasons (a) experiment 1, (b) experiment 2, (c) experiment 3. PB-U, PB biochar-U and PB ash-U- products of pine bark, pine bark biochar from the kiln and pine bark ash, respectively, dehydrated in the glasshouse. PB, PB biochar and PB ash- products of pine bark, pine bark biochar from the kiln and pine bark ash, respectively, with no urine enrichment.

6.3.2 Uptake of P and K and tissue concentrations of Ca, Mg and Na

The uptake and mineral concentration in spinach showed varying results across different treatments (Table 6.1). The biochar-U, PB-U and the positive control showed significantly higher P uptake compared to PB products without urine enrichment. All products with urine enrichment were not different and only the biochar-U had higher P uptake than the negative control (Table 6.1). There were no treatment responses of K uptake and Ca concentration when compared with the negative control (Table 6.1). However, the products without urine enrichment, had lower K uptake than those with urine enrichment and the positive control. The Mg concentration higher in the positive control and lower in the ash without urine enrichment, when compared with the negative control (Table 6.1). The PB-urine products had significantly higher Na concentration than all other treatments, which were not significantly different.

Table 6.1: The uptake of P and K and Ca, Mg, and Na concentration of spinach when PB- non enriched and enriched with urine were applied during the first planting season.

Treatments	Tissue uptake (g plot^{-1})		Tissue concentration (%)		
	P	K	Ca	Mg	Na
Con (-ve)	1.51^{ab}	8.74^{ab}	1.16^{a}	1.45^{b}	1.84^{a}
Con (+ve)	3.51^{bc}	20.9^{b}	1.06^{a}	1.88^{c}	1.61^{a}
PB	0.934^{a}	3.27^{a}	1.51^{a}	1.28^{ab}	1.51^{a}
PB biochar	0.933^{a}	3.41^{a}	1.31^{a}	1.28^{ab}	1.45^{a}
PB ash	0.625^{a}	2.79^{a}	0.919^{a}	0.976^{a}	1.43^{a}
PB-U	3.50^{bc}	21.0^{b}	0.941^{a}	1.09^{ab}	2.47^{b}
PB biochar-U	3.95^{c}	23.2^{b}	1.05^{a}	1.35^{ab}	2.92^{b}
PB ash-U	2.53^{abc}	18.5^{b}	0.949^{a}	1.20^{ab}	2.45^{b}

6.3.3 Uptake of P and K and tissue concentrations of Ca, Mg and Na for second season cumulative and residual

In the experiment to determine cumulative effects, P uptake was significantly higher in the negative control compared to the PB biochar-U (Table 6.2), while K uptake, and tissue concentration of Ca, Mg and Na were not significantly affected by the different treatments.
In the experiment to determine residual effects, K uptake was higher in the biochar-U treatment than the negative control, while tissue Mg concentration in the same treatment (biochar-U) was higher than the positive control (Table 6.2). There were no significant differences in the treatments for P uptake, tissue Ca and Na (Table 6.2).

Table 6.2: The uptake of P and K and Ca, Mg, and Na concentration of spinach when PB enriched with urine were applied during the second planting season as a residual or cumulative treatment.

Treatments	Tissue uptake (g plot^{-1})		Tissue concentration (%)		
	P	K	Ca	Mg	Na
Cumulative					
Con (-ve)	1.64^{b}	7.51^{a}	0.502^{a}	0.528^{a}	1.58^{a}
Con (+ve)	1.41^{ab}	6.89^{a}	0.703^{a}	0.723^{a}	1.55^{a}
PB-U	1.47^{ab}	6.87^{a}	0.775^{a}	0.746^{a}	1.87^{a}
PB biochar-U	0.856^{a}	4.48^{a}	0.701^{a}	0.642^{a}	1.01^{a}
PB ash-U	0.933^{ab}	5.09^{a}	0.852^{a}	0.686^{a}	1.99^{a}
Residual					
Con (-ve)	0.829^{a}	3.27^{a}	0.770^{a}	0.70^{ab}	1.73^{a}
Con (+ve)	0.867^{a}	3.79^{ab}	0.711^{a}	0.595^{a}	1.60^{a}
PB-U	0.722^{a}	3.68^{ab}	0.792^{a}	0.735^{ab}	1.88^{a}
PB biochar-U	1.18^{a}	5.92^{b}	0.805^{a}	0.785^{b}	1.51^{a}
PB ash-U	1.01^{a}	5.43^{ab}	0.743^{a}	0.691^{ab}	1.86^{a}

6.3.4 Cu, Zn, Fe, and Mn concentration for the first season

There were no significant differences in Copper (Cu), zinc (Zn), and iron (Fe) concentrations in spinach plant tissues across treatments (Table 6.3). However, PB treatment had significantly higher manganese (Mn) concentrations compared to PB biochar-U.

Table 6.3: The Cu, Zn, Fe and Mn concentration of spinach when PB non-enriched and enriched with urine were applied during the first planting season.

Treatments	Tissue concentration mg kg^{-1})			
	Cu	Zn	Fe	Mn
Con (-ve)	15.9[a]	247[a]	101[a]	1371[ab]
Con (+ve)	15.1[a]	199[a]	126[a]	1292[ab]
PB	18.9[a]	176[a]	129[a]	1712[b]
PB biochar	16.3[a]	259[a]	156[a]	1451[ab]
PB ash	16.3[a]	266[a]	143[a]	1629[ab]
PB-U	19.2[a]	176[a]	92.6[a]	1110[ab]
PB biochar-U	11.3[a]	203[a]	159[a]	926[a]
PB ash-U	11.1[a]	191[a]	158[a]	1171[ab]

The optimum concentrations: Cu= 5-26 ppm; Zn= 25-301 ppm; Fe= 60-201 ppm and Mn= 30-251 ppm (Kabata-Pendias, 2000)

6.3.5 Cu, Zn, Fe, and Mn concentration for the second season

In the cumulative and residual experiment, the tissue concentration of Cu, Zn, Fe and Mn in spinach plant tissues followed a similar trend, where there were no significant differences within the treatments (Table 6.4).

Table 6.4: The Cu, Zn, Fe and Mn concentration of spinach when PB- enriched with urine were applied during the second planting season as a residual or cumulative treatment.

Treatments	Tissue concentration mg kg^{-1})			
	Cu	Zn	Fe	Mn
Cumulative				
Con (-ve)	12.7[a]	104[a]	575[a]	638[a]
Con (+ve)	13.1[a]	81[a]	556[a]	618[a]
PB-U	13.1[a]	79[a]	426[a]	618[a]
PB biochar-U	13.6[a]	99[a]	478[a]	651[a]
PB ash-U	15.5[a]	101[a]	332[a]	646[a]
Residual				
Con (-ve)	15.0[a]	105[a]	383[a]	639[a]
Con (+ve)	15.5[a]	105[a]	671[a]	698[a]
PB-U	16.3[a]	101[a]	563[a]	812[a]
PB biochar-U	16.0[a]	124[a]	670[a]	744[a]
PB ash-U	13.4[a]	107[a]	451[a]	720[a]

The optimum concentrations: Cu= 5-26 ppm; Zn= 25-301 ppm; Fe= 60-201 ppm and Mn= 30-251 ppm (Kabata-Pendias, 2000)

6.3.6 Soil pH and extractable P after spinach harvest

In the first season, the soil pH in the ash-U treatment was higher than all other treatments except ash, biochar-U and biochar. There were no significant differences between all other treatments (Table 6.5). The pH in the treatments with ash and biochar, with or without modification with urine, did not significantly differ. Similarly, extractable P was significantly similar for all treatment with urine, with ash-U and biochar-U having higher than all other treatments, except PB-U.

Table 6.5: Soil pH and extractable P concentration after spinach harvest as affected by application of PB non enriched and enriched with urine during the first planting season.

Treatments	pH $_{(KCl)}$	Extract. P (mg kg^{-1})
Con (-ve)	4.75^{a}	137^{a}
Con (+ve)	4.57^{a}	129^{a}
PB	4.71^{a}	81.9^{a}
PB biochar	4.96^{ab}	111^{a}
PB ash	4.84^{ab}	148^{a}
PB-U	4.68^{a}	153^{ab}
PB biochar-U	4.95^{ab}	228^{b}
PB ash-U	5.30^{b}	226^{b}

6.3.7 Residual soil characteristics after the second season

Within the cumulative experiment, the ash-U treatment resulted in a significantly higher pH compared to all other treatments, except biochar-U (Table 6.6). There were no significant differences between treatments on extractable P, ammonium-N, nitrate-N and mineral-N (Table 6.6). In the experiment to determine the residual effects, the soil pH in the ash-U treatment was higher than the PB-U treatment, while there were no significant treatment differences on extractable P, ammonium-N, nitrate-N and mineral-N concentrations (Table 6.6).

Table 6.6: Soil pH and extractable P concentration after spinach harvest as affected by application of PB-enriched with urine during the second planting season as a residual or cumulative treatment.

Treatments	pH $_{(KCl)}$	Extract. P	Ammon. -N	Nitrate-N	Mineral-N
Cumulative			mg kg^{-1}		
Con (-ve)	4.65^{a}	193^{a}	0.493^{a}	3.88^{a}	3.37^{a}
Con (+ve)	4.50^{a}	137^{a}	0.249^{a}	3.39^{a}	3.63^{a}
PB-U	4.53^{a}	156^{a}	0.299^{a}	3.51^{a}	3.81^{a}
PB biochar-U	4.71^{ab}	153^{a}	0.280^{a}	3.48^{a}	3.76^{a}
PB ash-U	4.97^{b}	136^{a}	0.192^{a}	4.41^{a}	4.60^{a}
Residual					
Con (-ve)	4.64^{ab}	158^{a}	0.136^{a}	4.09^{a}	4.43^{a}
Con (+ve)	4.65^{ab}	198^{a}	0.158^{a}	5.51^{a}	5.87^{a}
PB-U	4.61^{a}	180^{a}	0.199^{a}	4.18^{a}	4.38^{a}
PB biochar-U	4.69^{ab}	124^{a}	0.129^{a}	3.87^{a}	4.00^{a}
PB ash-U	4.97^{b}	117^{a}	0.155^{a}	3.46^{a}	3.62^{a}

6.4 Discussion

The higher dry matter content in treatments with PB (feedstock, biochar and ash)-urine-based products, compared to the negative control and products that are not enriched with N, in the first season could be attributed to greater availability and uptake of N, P and K. This is supported by the N (Figure 6.5a), P and K uptake results (Table 6.1), which followed similar trends. The N supplied by urine during the enrichment and dehydration could have been in the form of urea, which hydrolysed following the application of urine-rich products to moist soil (Abera et al., 2012), increasing availability as mineral N. These findings align with Schmidt et al. (2015), who reported a four-fold increase in pumpkin yield when using urine-impregnated *eupatorium adenophorum* biochar on a silt loam compared to the control. The similarity in dry matter yield and N uptake in PB biochar-U with the positive control suggested that, when applied at the same rate of N, this treatment has the potential to be used in place of urea, without a loss of yield. In contrast, the lower dry matter yield in PB products without urine enrichment despite having the same added N rate as their corresponding PB-urine treatments can be attributed to low levels of N in a form that may have been unavailable, and the high C: N, indicate that the N could not mineralise rapidly enough, resulting in insufficient N availability.

Considering that P and K were added at the same recommended rate for all the treatments, the higher P and K uptake (Table 6.1) in the treatments with urine-based products and the positive control, could be because of the higher biomass production as a result of increased N, increasing the amounts of P and K taken up from the soil, than the negative control and products that are not enriched with urine. In addition, the urine-based products also had P and K added as part of the urine before dehydration, considering that the artificial urine used had 5 g monosodium phosphate L^{-1}, and 5.5 g disodium phosphate L^{-1} and also 2.25 g KCl L^{-1} (Chapter 3).

The numerically lower dry matter yield in the experiment to determine cumulative application of PB-enriched treatments (Figure 6.4b) than the first season (Figure 6.4a) could be explained by climatic conditions, which were warmer for the cool season crop (Figure 6.1). Similarly, Lester et al. (2013) observed an increase in leaf dry matter of spinach during the winter solstice in the subtropic compared to summer solstice grown spinach. The decline in dry matter yield in the PB biochar-U treatment, that was similar to the negative control, and lower than PB-U may be attributed to lower N uptake in that treatment, which was comparable to the negative control. The low N uptake could have been a result of lower available N in the soil as a result a significant accumulation of stable soil carbon storage (from biochar) after two applications of biochar, with the constituent N of the first application being taken up by the plants in the first season, leading to an imbalance in the soil carbon-to-nitrogen (C/N) ratio. As a result, this could have resulted in competition between microorganisms and plants for N, reducing soil N availability (immobilisation), which caused N deficiency in spinach plants, and ultimately inhibiting plant growth (Ali et al., 2020; Amoah-Antwi et al., 2020). Similar observations were made by Yang et al. (2024), who found that applying high rates of biochar derived from flue-cured tobacco stems led to a decrease in tobacco yield. The similarity between the treatments in terms of P and K uptake suggests that these nutrients did not contribute to this difference in dry matter yield.

The higher residual effects of PB biochar-U on dry matter yield in spinach, particularly when compared to negative control, could be attributed to increased N uptake (Figure 6.5c) and K uptake and Mg concentration (Table 6.2). Biochar-based fertilisers are known for their gradual nutrient release over time (Krishnamoorthy et al., 2023; Marcińczyk et al., 2022). This explains the continued increase in spinach growth, even without the application of additional N (PB biochar-U) prior to planting. The higher Na concentration in spinach tissue fertilised with PB

urine-enriched treatments was due to the application of Na-rich products, which were absorbed by the plants. Human urine naturally contains high levels of Na (0.9%) (Beler-Baykal et al., 2011; Martin et al., 2022), while artificial urine used in this study contains 0.22%). However, this elevated Na concentration did not negatively impact yield, as spinach is moderately salt-tolerant (Ors and Suarez, 2016; Xu and Mou, 2016). Interestingly, the cumulative application of these treatments did not lead to an increase in Na concentration in plant tissue (Table 6.2). Additionally, the residual effects of these treatments did not result in Na accumulation in plants, implying that most of the Na was absorbed during the first growing season, while the remainder would have been held tightly on the surfaces of the materials, especially biochar. Therefore, these products can be a sustainable N source for smallholder farmers, as their continued use did not increase soil sodicity risks, after two applications. While these products may not appear to pose a risk of soil sodicity, there is need to monitor the tissue and soil Na levels where these products are repeatedly used.

The Ca concentration in plant tissue did not differ between the treatments, which signifies that little was being contributed by the urine-based products. The lack of differences in tissue Cu, Zn, Fe in both seasons and Mn in the second season, suggested that these treatments did not significantly contribute to the available forms of these elements. The copper (Cu) content in spinach remained within the permissible limit, while Zn levels exceeded the limit during the first season, and iron (Fe) levels were above the limit in the second season (Kabata-Pendias, 2000). Additionally, manganese (Mn) levels were consistently above the permissible limit in all planting seasons. After treatment application, Mn concentrations (Table 6.3 and 6.4) exceeded the tissue threshold of 500 ppm (Adriano, 2001; Kabata-Pendias, 2000). The higher Mn levels in PB-treated spinach than the PB-biochar-U (Table 6.3) were likely due to the acidifying effect of PB (El-Jaoual and Cox, 1998), while the biochar contributes to liming (Vilakazi et la., 2023). In PB-urine products, elevated Mn levels may have resulted from the nitrification process, which acidified the soil, making Mn more soluble (El-Jaoual and Cox, 1998). Excess soil Mn, Zn, and Fe, which were not significantly different between the treatments, means that these elements were high in the original soil before treatment. These high levels can lead to bioaccumulation in plants (Patnaik, 2003) and negatively impact plant functions by inhibiting chlorophyll biosynthesis, reducing photosynthesis rates (Shi et al., 2006), and impairing CO_2 assimilation and stomatal conductance (Santos et al., 2017). High Mn levels can also disrupt the uptake, redistribution, and utilization of essential nutrients such as N, P, Ca, Mg, and Fe, all of which are critical for normal plant growth (Huang et al., 2016).

However, these effects were not evident in this study. Spinach is known as a hyperaccumulator of heavy metals and toxic ions (Nawaz et al., 2019), which may explain the high yield observed despite the increased solubility of Mn, Zn, and Fe. Similar findings have been reported, showing that spinach yield was not affected by Mn toxicity following wastewater application (Nawaz et al., 2019; Pathak et al., 1999).

The higher soil pH in the PB ash-U at the end of the experiments (both seasons) (Table 6.5 and 6.6), highlighted the liming potential of ash. The increase in pH also led to higher extractable phosphorus (P), during the first season, demonstrating its ability to enhance P availability (Singh et al., 2010; Xu et al., 2014), which contributed to increased P uptake. The higher extractable P remaining in the soil with the PB-biochar-U treatment, after harvesting in the first season, may have contributed to increasing P uptake and dry matter yield observed in the residual experiment during the second season.

6.5 Conclusion

The application of pine bark (PB) urine-based products increased nitrogen uptake, dry matter yield in spinach, sodium concentration soil pH, and available phosphorus (P) relative to the negative control and the Pine bark products with no urine enrichment. Cumulative addition resulted in low dry matter and N uptake for PB biochar-U which was similar to the negative control and lower than the PB-U. However, PB biochar-U showed higher residual effects than the negative control. This suggests that PB biochar enriched with urine positively impacts spinach yields, making it a promising option for smallholder farmers due to its slow nutrient release properties and reduces need for annual synthetic N inputs. For promising results, smallholders need to apply pine bark urine-based fertilisers at the appropriate recommended rates and time, consider soil pH, to enhance nutrient availability and crop performance.

Additionally, applying PB ash-U raises soil pH in both seasons and increased extractable P availability in the first season. This indicates that PB ash-U can improve acidic soils by acting as a liming agent while enhancing P availability. However, using pine bark urine-based products can lead to an increase in sodium (Na) concentration in spinach tissue. Fortunately, with continuous application, this impact diminishes, as most of the Na accumulates in the leaves of the first harvests, thereby reducing the risk of soil salinity over time. The findings imply that dehydration production of urine, especially with PB biochar can be used as an

alternative to urea fertiliser with similar levels of yield, although continuous use would require that Na in the tissue and in the soil need to be monitored.

Chapter 7: General discussion, conclusion and recommendations

7.1 General Discussion

A lack of proper sanitation facilities remains a major challenge in rural and peri-urban areas, where soil nutrient mining poses a serious threat to resource-poor farmers. The situation is becoming increasingly severe, with over 80% of human excreta and wastewater being discharged into the environment with little or no treatment (van den Broek et al., 2024). This improper disposal can lead to excess nutrients entering waterways, resulting in eutrophication, algal blooms, and significant consequences for both human and ecosystem health (Katz, 2020). Urine recycling has gained attention among researchers due to its nutrient content of approximately 0.74% nitrogen (N), 0.029% phosphorus (P), and 1.62% potassium (K), which closely aligns with the nutrient requirements of many crops (Mchunu et al., 2018; Mnkeni and Austin, 2009). However, the high-water content (≈ 97%) and alkaline pH of urine present challenges for its direct use as a fertiliser, complicating storage, transportation, application logistics, and associated costs (Geinzer, 2017; Patel et al., 2020; Senecal and Vinnerås, 2017). One alternative solution is the use of organic waste materials such as potato waste and pine bark to mitigate these challenges while retaining the valuable nutrients in urine. Although various studies have examined N recovery using ash and biochar derived from wood waste, limited research has explored the use of potato waste and pine bark, or their corresponding biochar and ash. The effectiveness of these materials may depend on factors such as the properties of the feedstock and the pyrolysis temperature used during biochar production. These urine-based amendments could support crop growth by serving as a N source in agricultural soils, while biochar treatments also offer the added benefit of carbon sequestration. The overall objective of this research was to determine the potential of plant-based materials (feedstock, biochar and ash) derived from potato waste and pine bark on nutrient retention, especially N, from artificial human urine through dehydration, and nutrient release of the products in soil and their effects on spinach growth.

The generally higher N retention after dehydrating mixtures of urine with all treatment media at 60 °C compared to dehydration at 45 °C, was likely due to the shorter dehydration time (2 days versus 4 days) with an added benefit of greater energy efficiency at the higher temperature. These findings are consistent with Simha et al. (2020a), who also reported increased N recovery at 60 °C. Higher temperatures promoted rapid dehydration and minimised N losses, although the extent of N retention was more significantly influenced by the characteristics of the media.

While the cull potato (CP) treatments had higher total N content, the N recovery was lower than those of pine bark (PB) treatments. Although the same amount of N was added to both CP and PB materials, the CP products showed lower N retention, which suggests that some of the urea was lost. Even though the pH during dehydration was sufficiently high to inhibit urease activity (>11), the losses observed indicate that chemical hydrolysis and volatilisation likely occurred, especially in CP ash where the pH exceeded 12. It is known that urea can undergo non-enzymatic hydrolysis at pH >10, leading to ammonia volatilisation (Senecal and Vinnerås, 2017; Simha et al., 2021). In addition, the CP biochars had poor porosity and fewer active sites, which limited their ability to adsorb and retain N (Vilakazi et al., 2023; Yin et al., 2017). The results also suggest that there is a threshold of around 2.5–3% total N in these materials. Once this level was reached, the products could no longer retain additional N, resulting in further losses through hydrolysis and volatilisation. Although there is limited literature that specifically addresses whether high total N concentrations reduce the effectiveness of urease inhibition through pH adjustment, the trends observed in this study suggest that once the adsorption capacity is reached, chemical losses can occur even when urease activity is suppressed. In contrast, PB materials had better porosity and more functional groups, which improved N adsorption and retention. This highlights the importance of media characteristics such as aromatic structure, porosity, surface area, and the presence of functional groups (Pathy et al., 2021; Zhang and Wang, 2016).

The literature presents some discrepancies regarding the pH range for urease activity. Rektorschek et al. (1998) report that urease can be inhibited at pH values below 3.5 or above 8.5, whereas Chipako and Randall (2020) suggest inhibition occurs at pH levels below 7 or above 11. Given these different thresholds, I used the broader inhibitory range (pH <6.5 or >11) in this study to ensure that urease activity was fully suppressed during urine stabilisation. The artificial urine does not contain urease; any urease present may have come from the materials used, as they were not sterilised. The CP biochars had pH values within the active range for urease, resulting in poor N recovery (Figure 3.1a, b). With CP 350 exhibiting negative N recovery, which signifies that even the N in the original materials was lost, possibly due to the activity of micro-organisms especially at 45 °C where drying took longer (4 days) after having added N rich solution (more like an incubation). On the other hand, PB biochars showed high N recovery, likely due to the presence of negatively charged functional groups and greater surface area, with pH playing a lesser role in these treatments. However, PB biochar reached a saturation point following the addition of 174 (45 °C) and 196 mL (60 °C) of urine, suggesting

that a threshold was reached and could not retain any N. The CP biochar also lacked sufficient porosity, limiting the number of active sites available for N adsorption (Yin et al., 2017), also the N was initially high and any added N might be lost due to hydrolysis. The ash treatments (CP ash and PB ash) also showed differences in N recovery. The CP ash maintained a high pH (>12) due to its liming effect but resulted in low N recovery. This may be attributed to the presence of calcium carbonate ($CaCO_3$), which promotes ammonia volatilisation (Freney et al., 1981; Powlson and Dawson, 2022), and chemical hydrolysis processes that become prevalent at pH levels above 12 (Senecal and Vinnerås, 2017; Simha et al., 2021). The mechanism through which this occurs is that $CaCO_3$ raises the pH, and at pH above 12, urea can chemically hydrolyse even without urease enzyme activity, producing ammonia and carbon dioxide.

The high pH also shifts the ammonia/ammonium equilibrium towards volatile NH_3 gas, which can escape to the atmosphere (Senecal and Vinnerås, 2017). This condition was prevalent in CP ash, which maintained a pH greater than 12 throughout the study, driven by its high calcium carbonate equivalence (57.1%). The combination of high pH and strong liming effect created conditions favourable for both chemical hydrolysis and volatilisation. In contrast, PB ash did not maintain such high pH and had more micropores and functional groups, which likely retained nitrogen and reduced its susceptibility to volatilisation, despite being within the urease activation range. This discrepancy could be explained by the presence of micropores and surface area in PB ash, which may have adsorbed N and reduced its susceptibility to enzymatic degradation. Wood ash has high adsorption capacity of N because of its surface and porosity (Andersson et al., 2011; Hamidi et al., 2021). Pine bark feedstock is acidic and was within the urease enzyme activation range. Despite being in the urease activation range, the acidic nature of the original pine bark (4.13) feedstock may have contributed to improved N retention. These findings suggest that surface area and functional groups are key drivers of N recovery. Interestingly, some CP feedstock treatments recovered more N than their corresponding CP biochars and ash, indicating that CP feedstock products may be more effective for N recovery than others within the same feedstock group.

Pine bark materials generally recovered more N than the CP products, which suggests that high nitrate-N can be the result of urea from synthetic urine dissolution and hydrolysis (Wahl et al., 2006). All the treatments had a C: N which is less than 30: 1 (Table 2), and it is expected that mineralisation would be high in all the treatments. The N in the PB products was already in urea form (from the urine) which only needed hydrolysis to produce ammonia, ammonium and nitrate, while the CP products had low urine N (urea) and the N from the original material

needed mineralisation, before nitrification could take place. The C: N ratio of any materials is commonly used to access it mineralisation or immobilisation rates (Hadas et al., 2004). A low C: N ratio (< 30) generally promotes mineralisation, while a high C: N ratio tends to lead to immobilisation. However, the mineralisation rate varied between treatments, indicating the differences in the ability of each urine product to release N into the soil. These differences can be attributed to variations in their functional groups, pore distribution, surface area, and chemical properties (Pathy et al., 2021).

Nitrogen release patterns differed between the products derived from cull potato (CP) and those from PB. In Chapter 4, the urine-based products derived from CP feedstock, biochars, and ash showed different N release patterns during incubation. The CP ash released limited mineral N due to its high pH, which inhibited urea hydrolysis and subsequent nitrification. While CP biochars, despite lower urine-N retention, showed higher N mineralisation, likely due to the mineralisation of organic N within the biochar. Additionally, the CP feedstock products released lower mineral N, possibly because of the presence of low water-soluble carbohydrates hindering decomposition (Charmley, et al., 2006). In the pot trials, this resulted in relatively lower N uptake and dry matter yield for CP feedstock and ash products, while CP biochar treatments performed better due to their greater N release. The high K uptake could also explain the higher dry matter yields in the CP biochars, which could be attributed to the high K concentration in the original feedstock relative to pine bark (Vilakazi et al., 2023). In addition to the high mineral N availability, extractable P was higher in cull potatoes than pine bark urine-based products during incubation. The differences in the extractable P concentration are explained by the higher extractable P in the cull potatoes. In addition, the higher extractable P in soils amended with CP ash could be attributed to the liming potential (57.1%) of this treatment, which increases the negative surface charges causing less adsorption of P (Cui et al., 2011; Masto et al., 2013). Similar observations have been reported where P availability increased in acidic soils following the application of biochar with liming effects (Singh et al., 2010; Xu et al., 2014), while Panigrahi (2013) observed similar results in soils applied with coconut shell enriched with urine. The limited N availability from CP ash also corresponded to its primary benefit being as a liming and phosphorous source rather than an N fertiliser.

The PB urine-based products showed a consistent N release patterns in both incubation and pot trials (Chapters 5). Treatment PB ash-U released the highest nitrate-N rapidly, whereas PB biochar-U exhibited more gradual N release. The PB-U (feedstock) also supported N release,

although it was less than biochar-U. In the pot trial (Chapter 5), this resulted into significantly improved N uptake and dry matter yield, particularly for PB ash-U and PB biochar-U, which performed comparably to mineral N treatments. This is attributed to plants preferentially taking up the nitrate-N form (Song et al., 2015). The PB products dehydrated in the glasshouse performed exceptionally well across incubation and spinach pot trials: PB ash G exhibited the highest nitrate-N release and significantly raised soil pH. In the pot trial, PB ash G supported high dry matter yields and N uptake comparable to synthetic fertiliser. However, elevated manganese concentrations were detected in spinach tissue from PB G and PB biochar G treatments, highlighting the need for monitoring when applying these products repeatedly or on Mn-sensitive crops. In the field trial (Chapter 6), during the first season, all PB urine-enriched products significantly improved N uptake and dry matter yield relative to unenriched products, with PB biochar-U being similar to the positive control. This confirms that PB urine-based products can serve as effective N fertilisers under field conditions.

In the second season (cumulative application), PB biochar-U showed reduced N uptake and dry matter yield compared to PB-U and was similar to the negative control. This decline was likely caused by biochar-induced N immobilization as the C/N ratio shifted due to cumulative biochar inputs. However, PB biochar-U showed strong residual effects (residual application), sustaining higher dry matter yield and N uptake even without additional application, highlighting its potential for long-term soil fertility improvement as it acts as a slow nutrient releaser. The yields were numerically lower in the second season relative to the first season. This could be explained by the climatic conditions which were warmer for the cool season crop (Figure 6.1). Similarly, Lester et al. (2013) observed an increase in leaf dry matter of spinach during the winter solace in the subtropic compared to summer solstice grown spinach.

Enhanced P and K uptake clearly contributed to the dry matter yield responses observed. In the field experiments, higher P and K uptake in PB ash-U and PB biochar-U treatments improved N uptake, supporting overall spinach biomass accumulation. Micronutrients (Cu, Zn, Fe, Mn) present in the PB urine-based products and CP products were monitored. Across both pot and field experiments, these elements did not substantially change spinach tissue composition in ways that would compromise food safety. However, elevated manganese (Mn) concentrations were observed in spinach tissue following application of PB and CP products. This was linked to the acidifying effect of PB feedstock and the nitrification process increasing Mn solubility (El-Jaoual and Cox, 1998). Applying urine-based products increased sodium (Na) concentration in spinach tissue. The high Na could be due to the high concentration of Na in

urine which could be taken up by the crops (Beler-Baykal et al., 2011; Martin et al., 2022). Despite elevated Mn and Na, spinach yield was unaffected, consistent with its status as a hyperaccumulator (Ors and Suarez, 2016; Xu and Mou, 2016). Other micronutrients (Cu, Zn, Fe) remained within acceptable limits. In the field trials, these trends were consistent; micronutrients did not impact spinach tissue composition adversely. In Overall, PB urine-based fertilisers, particularly PB biochar-U and PB ash-U, provide valuable alternatives to synthetic fertilisers for smallholder farmers. PB biochar-U offers long-term soil fertility benefits, while PB ash-U supplies immediate P and liming effects. Both products, when used appropriately, enhance spinach yield and nutrient uptake while managing soil health risks. However, sodium and manganese levels must be monitored to avoid long-term accumulation, especially in non-tolerant crops.

A comparison between the glasshouse and field trial results reveals important insights into the predictive validity of controlled environment studies. In the glasshouse, pine bark-based urine products consistently enhanced spinach dry matter yield and nitrogen uptake, often matching the performance of synthetic fertilisers. These trends were largely replicated in the field, particularly during the first season, where pine bark-urine treatments outperformed non-enriched controls. However, cumulative applications in the second season led to reduced yield and uptake in some treatments, such as PB biochar-U, highlighting the importance of application rate and timing. These findings suggest that while glasshouse trials are useful for initial screening, field trials are essential for understanding long-term effects and scalability.

This study demonstrated the effectiveness of urine-enriched biochar and ash derived from pine bark as nitrogen fertilisers under both controlled (glasshouse) and field conditions. The results provide strong evidence that locally available organic waste materials can be used to stabilise and concentrate urine nitrogen, producing fertiliser products that deliver agronomic performance comparable to synthetic nitrogen fertilisers. These findings contribute to the global understanding of nutrient recycling from human excreta and offer practical solutions for closing nutrient loops between sanitation and agriculture. The production of urine-enriched biochar and ash fertilisers is feasible at the farm level using simple dehydration techniques and small-scale pyrolysis units, with low capital investment and the use of abundant local organic materials. This process offers a cost-effective alternative to synthetic fertilisers for smallholder farmers while supporting circular economy approaches. For broader adoption, national policy frameworks and sanitation guidelines should incorporate and promote the safe use of stabilised urine-based fertilisers. Furthermore, the inclusion of such products in agricultural extension

services would help overcome adoption barriers, ensure product quality, and support sustainable nutrient recycling in rural and peri-urban agricultural systems.

7.2 Conclusion

The choice of media for dehydrating urine significantly impacts the concentration of N recovered from urine. From the current study, PB feedstock, biochar and ash enriched with urine had greater potential to recover N from urine compared to the corresponding cull potato products. The study demonstrated that dehydration at 60 °C is more energy-efficient and effective than at 45 °C, accelerating moisture loss while retaining significant N content. Among the tested media, pine bark and its derivatives (biochar and ash) consistently outperformed cull potato products in N recovery, making them more suitable carriers for nutrient-rich fertiliser production. Notably, CP feedstock requires no energy input for its preparation and retained more N than its biochar and ash forms, suggesting it may be favourable in low-energy settings. The field experiment demonstrated that PB-based urine-enriched products significantly increased spinach dry matter yield, N uptake, soil pH, and extractable P. The PB biochar-U matched mineral N fertiliser (urea) in enhancing spinach growth, indicating its potential as a viable replacement for the costly synthetic fertilisers, particularly for resource-limited smallholder farmers. The PB ash enriched with urine also showed promising results, acting as liming agent, effectively reducing soil acidity and improving P availability. However, a consistent observation across all treatments was the elevated Na. These are attributed to the naturally high Na content in urine and the acidifying nature of the amendments, which increase Mn solubility.

In Overall, this study demonstrated that locally available organic waste materials such as pine bark can be effectively used to stabilise and concentrate nitrogen from urine through simple dehydration and pyrolysis techniques. The resulting urine-enriched biochar and ash fertilisers produced agronomic performance comparable to that of synthetic N fertilisers, both under controlled and field conditions. These findings contribute to global knowledge on nutrient recycling from human excreta and offer practical, low-cost solutions for smallholder farmers to close nutrient loops and enhance soil fertility.

7.3 Recommendations

Future work should expand on plant response across diverse crop types and soil types, with special attention to nutrient availability, heavy metal accumulation, and long-term soil health under repeated application scenarios. In addition, although spinach tolerated these levels due to its salt-resilient nature, the findings underscore the importance of monitoring Na and Mn in long-term use and recommending salt-tolerant crops during initial application cycles. Given the potential of urine-based fertilisers to contribute to sustainable agriculture, policies should be developed to support their production, regulation, and integration into existing farming systems. Awareness campaigns should also be conducted to promote farmer adoption. In parallel, scaling of these technologies should be supported through the development of simple on-farm production guidelines for urine-enriched biochar and ash fertilisers using locally available organic materials. National agricultural extension programmes should incorporate training modules on safe preparation and effective use of such fertilisers. Regulatory frameworks should clearly include stabilised urine-based fertilisers as recognised nutrient sources, with quality standards and application guidelines to ensure safe and effective use across diverse farming.

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