

**ASSESSING THE POTENTIAL USE OF STRUVITE AND EFFLUENT
FROM DECENTRALIZED WASTEWATER TREATMENT SYSTEMS
(DEWATS) AS PLANT NUTRIENT SOURCES FOR EARLY MAIZE
(*Zea Mays*) GROWTH.**

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DECLARATION

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SUMMARY

The Decentralised Wastewater Treatment System (DEWATS) effluent has been shown to contain considerable concentrations of mineral elements such as nitrogen (N) and phosphorus (P), which are important for plant growth. The use of effluent for agriculture as a sole nutrient source is limiting in terms of macronutrient and micronutrient content supplied to plants. There is little information about the effects of combining the effluent with struvite and commercial fertilizer for crop production. The study aimed to determine the effect of applying struvite and DEWATS effluent as nutrient sources combined or in combination with urea/single superphosphate (SSP) fertilizers on the growth, nutrient uptake, and biomass production of maize. The specific objectives were: (1) to determine N and P release pattern of struvite when applied solely or combined with urea relative to SSP fertilizers combined with urea in a sandy soil, (2) to determine N and P release pattern of DEWATS effluent applied solely or combined with struvite and or SSP fertilizers in a sandy soil, (3) to investigate the effect of applying struvite and DEWATS effluent as nutrient sources combined together or with urea/SSP fertilizers on the growth, nutrient uptake and biomass production of maize. Two soil incubation experiments were set up under controlled room temperature at 25°C and 80% atmospheric humidity to determine the N and P release pattern of human excreta derived materials (HEDMs) (struvite and DEWATS effluent) with supplementary chemical fertilisers urea and SSP. The first experiment was laid out as a single factor analysis with the following treatments: (i) struvite alone, (ii) urea alone, (iii) SSP alone, (iv) struvite + urea, (v) SSP + urea. Each treatment was replicated 3 times to give 15 experimental units (in 5 litre ventilated containers). The second experiment was also laid out as a single factor comprising the following treatments: (i) effluent alone, (ii) struvite + effluent, (iii) effluent + SSP, and (iv) a control, all replicated 3 times to give 12 experimental units (in 5 litre ventilated containers). The fertiliser materials

were applied to achieve an equivalent of 200 kg N/ha and 60 kg P/ha to meet maize nutrient requirements from the Cartref (sandy soil). The effluent in the study was applied as an irrigation source to achieve a 100% soil water holding capacity while supplying nutrients at the same time. Data was collected on the ammonium N, nitrate N, and extractable P release weekly, for 56 days. A pot trial was set up in 20 litre pots in the tunnel at 26°C air temperature and 65% atmospheric humidity to determine the effect of applying struvite and treated effluent from the anaerobic filters (AF) on growth, nutrient uptake, and biomass production of maize. The pot experiment was set up as a 9 x 2 factorial experiment in a completely randomised design (CRD) with the following treatments: fertilizer combinations (8 levels- (i) struvite + urea (recommended rates); (ii)) struvite + urea (half recommended), (iii) struvite + effluent (recommended rates); (iv) struvite + effluent (half recommended); (v) SSP + effluent (recommended rates); (vi) SSP + effluent (half recommended); (vii) SSP + urea (recommended rates); (viii) SSP + urea (half recommended) and the control. The second treatment was maize variety with 2 levels –‘Colorado’ and ‘IMAS’. The treatments were replicated three times. Three maize seeds were planted per pot and were thinned 3 weeks after planting to one plant per pot. The amount of water applied as irrigation was based on Cartref soil water requirements. Soil moisture was maintained at 70-100% field capacity. The soil incubation experiment showed that there were significant ($P < 0.05$) differences among treatments- struvite (S), effluent (E), SSP (P), urea (U), struvite + urea (SU), struvite + effluent (SE), effluent + SSP (PE), SSP + urea (PU) and zero fertilizer. The combination of HEDMs and commercial nutrient sources released higher ammonium-N and nitrate-N than sole applications and when commercial SSP + urea was applied together. Ammonium N declined over time and nitrate N increased rapidly over time. The findings suggested that the fertiliser combination of HEDMs and commercial fertiliser increased nutrient N availability to the soil. Phosphorus did not change over time in all treatments. The pot experiment result showed that there were significant ($P < 0.05$)

differences observed in plant height, leaf number, chlorophyll content, dry matter, N and P uptake, and grain + cob yields among the different fertiliser combinations (SE, SU, PE, PU) at both recommended and half recommended application rates. In conclusion, optimising N and P supply through a combination of the effluent and struvite or with inorganic fertilisers could potentially be considered as a better option for providing a balanced supply of nutrients than when applied separately.

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CHAPTER ONE: GENERAL INTRODUCTION

1.1 INTRODUCTION

The world population is projected to reach 9 billion by the year 2050 (United Nations, 2019), and it is estimated that 70% of the people will be living in the cities (United Nations Population Fund, 2017). Urban migration has significantly increased over the decades, and this is largely driven by the quest for education and employment opportunities. However, with job scarcity and lack of skills, many would end up living in poor urban and peri-urban areas (Hudson, 2011). Such areas lack proper sanitation facilities since they are far from centralized municipal wastewater treatment systems. The provision of proper sanitation to residents living in peri-urban communities in South Africa is a challenge (Ashipala and Armitage, 2011).

Uncontrolled urbanization, unplanned and informal settlements are making it difficult for municipal authorities to connect such settlements to the wastewater treatment system grid, for example, the hilly terrains of KwaZulu-Natal prohibit constructions of sewerage systems in informal settlements in these areas (Foxon, 2009). About 20% of the residents living in informal settlements do not have any form of sanitation (Foxon *et al.*, 2005). The Decentralised Wastewater Treatment System (DEWATS) is being considered as a potential solution to this sanitation challenge in the peri-urban and urban settlements (Calabria, 2014).

The DEWATS was designed by Bremen Overseas Research and Development Association (BORDA) (Sasse, 1998). This type of sanitation technology has been widely accepted in countries such as India, Indonesia, and China as a potential solution due to its low energy requirements and high treatments efficiency (Gutterer *et al.*, 2009a). Locally, the eThekweni Municipality commissioned Community Ablution Blocks (CABs) intending to connect them to DEWATS, as a solution to the sanitation crisis in the informal settlements (Crous *et al.*, 2013). For example, the Banana City in eThekweni Municipality is among the informal

settlements proposed for the provision of DEWATS. The DEWATS is a modular water-borne sanitation system consisting of a settler, Anaerobic Baffled Reactor (ABR) + Anaerobic Filter (AF), and planted gravel filters (Gutterer et al., 2009a). The system's treatment process involves anaerobic degradation of organic matter within the ABR followed by the AF. The AF effluent is further passed to planted gravel filters which consist of a Vertical Flow Constructed Wetland (VFCW) and Horizontal Flow Constructed Wetland (HFCW) for further polishing of the effluent (Tilley *et al.*, 2011). The final effluent must comply with the stringent South African DWA (2013) discharge standards. Any failure to the wetland may lead to the discharge of poorly treated wastewater with associated environmental concerns into water bodies (Foxon, 2009). The National Environmental Management Act (NEMA) of 2008 (DWS, 2016) and the United Nations Sustainable Development Goal number 6.3 (WWAP, 2017) discourages waste discharge into the environment and strongly emphasise reuse. There is a paradigm shift towards the handling of human excreta. Globally, human excreta is currently considered as a rich resource rather than waste (Andersson et al., 2016; Nansubuga et al., 2016; WWAP, 2017).

The use of human excreta in agriculture has been practiced since ancient times globally, including in developed countries like England, Spain, and Greece (Jaramillo and Restrepo, 2017). The rationale behind the use of human excreta in agriculture is the quest to limit the potential pollution of water resources from organic pollutants (Jaramillo and Restrepo, 2017), to see an increase in crop yields through constant supply of nutrients, in conjunction with improved soil properties (Andersson, 2015), and reduced costs of wastewater treatment to meet the world standards for direct discharge into water bodies (Ricart et al., 2019). However, despite excreta being considered as a potential agricultural resource, there are some limitations associated with it. One major concern is its microbial load, which could impact negatively on human health. World Health Organisation (WHO) standard guidelines highlight all pathogen contamination pathways and pathogen loads in human excreta is rated the greatest aspect of

concern (WHO, 2006). Included in the guidelines is the management of agricultural systems in ways that reduce risks through sanitation safety plans for safe reuse (WHO, 2016) and the development of excreta treatment technologies and techniques (Moya et al., 2019). Furthermore, the Food and Agricultural Organisation (FAO) published a training manual on the safe reuse of wastewater in urban and peri-urban farmer field schools in Sub-Saharan Africa (FAO, 2019), information which is critical for future reuse of excreta. Studies showed that perception and acceptance are not major barriers in Africa especially when comprehensively addressed with regards to human safety and pointing out potential economic benefits (Andersson, 2015; Fred et al., 2014; Moya et al., 2019; Ricart et al., 2019).

Human excreta (urine and faeces) contain mineral elements needed for plant growth and development. The effluent from the DEWATS, which contains both urine and faeces, with considerable concentrations of nitrogen (N) and phosphorus(P) are important for plant growth. The N and P concentrations in the effluent are 60 mg/L and 10.5 mg/L, respectively (Musazura *et al.*, 2019). The AF effluent contains mineral nutrients (N and P) that exist in organic and inorganic forms, which must undergo several transformations before becoming bioavailable. For example, the DEWATS effluent has high concentrations of inorganic N (ammonium N), which can be taken up by plants and can undergo nitrification for it to be taken up by plants in the form of nitrates (Yu, 2012).

In South Africa, several onsite sanitation systems have been considered as potential solutions in most poor urban and informal settlements (Andersson et al., 2016). Those sanitation systems that are currently under consideration especially by the eThekweni Municipality include dry technologies such as Urine Diversion Dehydrated Toilets (UDDT) and the Ventilated Improved Pit (VIP) latrines (Andersson et al., 2016). Urine diversion dehydrated toilets separate faecal matter from urine while in VIP toilets, human excreta materials are not separated but combined. Struvite ($\text{NH}_4\text{MgPO}_4 \cdot 6\text{H}_2\text{O}$), a phosphorus fertiliser can be produced from the separated urine

by the addition of magnesium salts (MgO , MgSO_4 , or MgCl_2). The average removal of nitrogen (N) and phosphorus (P) from this process is 10 and 90%, respectively (Etter *et al.*, 2011 and Jimenez *et al.*, 2009). While significant amounts of N remain in the effluent, the struvite has major fertiliser value. Nutrients from these materials can be used to improve the fertility of degraded soils, especially for the underprivileged peri-urban and rural community farmers. On the other hand, poor soils of low organic matter and plant nutrient content are a major contributor to low crop yields and drivers of food insecurity, especially in sub-Saharan Africa (SSA). This challenge is exacerbated by continuous nutrient mining, the high cost of inorganic fertilisers, the lack of circular economies closing a nutrient loop (Sanchez, 2002), and water scarcity, for example, in South Africa where the average annual precipitation value is below 500 mm. Experiences of continuous dry conditions have led to high competition for water between domestic purposes, including safe drinking water, and the need for agricultural use (Rijsberman, 2006). The use of AF effluent could contribute towards reducing the magnitude of the said challenges through improving the water crisis and rejuvenating nutrient-depleted soils. Studies on the use of these organic materials such as struvite and DEWATS effluent for agricultural production have shown to have great potential to supplying essential plant nutrients (Nongqwenga *et al.*, 2017; Magwaza *et al.*, 2020).

(Bame *et al.*, 2013) conducted some experimental studies on the behaviour of DEWATS effluent in different soils of Kwa-Zulu Natal. The findings from these studies showed that addition of DEWATS effluent increases vegetative growth (plant height, dry mass, and leaf area) of maize and banana plants. Magwaza *et al.*, (2020) investigated the effect of DEWATS effluents and Nitrified Urine Concentrate (NUC) on leaf gas exchange, photosynthetic efficiency, and mineral content of hydroponically-grown tomatoes. The study demonstrated that HEDM such as NUC and DEWATS effluent could be an effective source of nutrients of crops in hydroponic systems with results comparable to commercial fertilizer. In other studies,

struvite showed to be as effective as commercial superphosphate, in supplying phosphorus (Uysal *et al.*, 2014). Maize had a higher yield compared to SSP under greenhouse conditions (Nongqwenga *et al.*, 2017). In another study, buckwheat yield had similar P uptake and yield compared to diammonium phosphate (DAP) (Talboys *et al.*, 2016).

Work on DEWATS has been focused mainly on the effluent used for irrigation during which liquid samples were collected from the anaerobic baffled reactors. However, the use of DEWATS effluent in combination with other human excreta-derived materials (HEDMs) such as struvite and or with commercial chemical fertilizers, to control proportions of the nutrients derived from these products in relation to those required by crops is worth investigating. It is still not clear what effect the co-application of DEWATS effluent and P sources has on soil N and P availability and crop productivity. The other question is finding out what effects co-application of struvite and N sources have on N and P availability in soil and crop productivity. Therefore, this study aims at determining the response of crop productivity and soil chemical properties to the combined application of DEWATS effluent and solid P fertilizers such as struvite and chemical N fertilizers. This could concomitantly generate the knowledge needed for sustainable disposal options of excreta from decentralised sanitation systems into the environment, especially in sub-Saharan African countries' low-income peri-urban and rural communities.

1.2 AIMS AND OBJECTIVES

This study aimed to assess the potential use of struvite and DEWATS effluent as plant nutrient sources either individually, as a combination of themselves, or in combination with synthetic fertilizers (urea and single superphosphate).

1.3 RESEARCH QUESTIONS

- What are the nitrogen and phosphorus release patterns of the AF effluent and struvite when applied singly or combined or in combination with urea/SSP fertilizers in a sandy loam soil?
- What is the effect of applying struvite and DEWATS effluent as nutrient sources combined or in combination with urea/SSP fertilizers on the growth, nutrient uptake, and biomass production of maize?

1.4 SPECIFIC OBJECTIVES

1. To determine the nutrient (N and P) release pattern of DEWATS effluent combined with struvite and or SSP fertilizers in a sandy loam soil.
2. To determine the nutrient (N and P) release pattern of struvite combined with urea relative to SSP fertilizers combined with urea in a sandy loam soil.
3. To investigate the effect of applying struvite and DEWATS effluent as nutrient sources combined together or with urea/SSP fertilizers on the growth, nutrient uptake, and biomass production of maize.

1.5 THESIS STRUCTURE

This dissertation comprises five chapters:

CHAPTER 1 provides the background and justification of the research. The chapter highlights key issues with regards to challenges of sanitation provision in South Africa and wastewater reuse in agriculture towards addressing food insecurity challenges through integrated soil fertility management.

CHAPTER 2 reviews in detail the sanitation challenges and potential use of human wastes (urine and faecal matter) in agricultural production. The section also reviews the recovery and re-use of nutrients (N and P) derived from human excreta materials.

CHAPTER 3 is an experimental chapter that reports on soil incubation study observing the pattern and rate of nutrient release from human excreta-derived materials and chemical commercial fertilizers amended sandy soil.

CHAPTER 4 reports on the pot trial aimed at assessing the effect of different combinations of struvite, the AF effluent, and synthetic fertilizers (SSP and urea) on the growth, nutrient uptake, and biomass production of maize.

CHAPTER 5 presents the general discussion and conclusions of the study. The chapter also offers recommendations and suggestions for future work.

CHAPTER TWO: NUTRIENT RECOVERY FROM HUMAN WASTE AND REUSE FOR CROP PRODUCTION

2.1 Introduction

Rapid and unplanned urban migration has led to densely populated informal settlements in cities of many developing countries (Antonini and Clemens, 2010). Most of the residents in these informal settlements are generally poor and unskilled. Staying in these places exposes dwellers to challenges of food insecurity and limited sanitation facilities. For example, such challenges include poor connections of the settlements to centralised wastewater treatment systems resulting in unmanageable waste streams in cities and peri-urban areas. This makes planning difficult and impractical, for example in places like the hilly terrains of KwaZulu-Natal are prohibiting the construction of sewerage systems in these areas (Foxon, 2009).

On the other hand, changes in climate have presented strong challenges to sustainable crop production and food security. The primary challenges are low agricultural productivity and water scarcity in developing countries such as South Africa (Udert and Wachter, 2011). With the global population predicted to be approximately 9 billion by the year 2050, increased demand for food, water, and sanitation is inevitable (UNDP, 2007). There is reassurance for croplands to produce more with limited resources, in that crop productivity has to meet food production that equals to the population growth (Heinonen-Tanski and Wijk-Sijbesma, 2005).

The continued removal of essential mineral elements from farms via food products and their transportation into cities and their disposal as “waste” has led to the problem of nutrient mining and consequently nutrient-depleted soils (Ladha *et al.*, 2011). Apparently, this is a scenario that is affecting agricultural productivity, particularly among resource-constrained small-holder farmers. In addition, the rapid increase in population in urban areas means an increase in food demand. This requires increased use of chemical commercial fertilisers which are on the other

hand expensive and of negative impact to the environment due to pollution if poorly managed (Wiederholt and Johnson, 2017).

The increase in fertilizer use to increase agricultural production to feed the increasing population has resulted in elevated fertilizer prices which in turn are a challenge especially to the resource-limited smallholder farmers (Antonini and Clemens, 2010). Also, mining and processing of the non-renewable phosphate rock for commercial fertiliser production is expensive, and this presents a threat to future fertilizer production as it may run out (Bonvin, 2013). Similarly, the production of nitrogen fertilisers from atmospheric N is also expensive, as the process is associated with large energy costs. Furthermore, the continuous use of inorganic fertilisers is contributing to increased greenhouse gases emissions, which lead to climate change-related problems such as global warming and erratic rainfall patterns, and consequently poor yields (Antonini *et al.*, 2012).

There is therefore the need to find ways to address the twin challenges of waste management and nutrient mining sustainably by recovering nutrients from some alternative sources like human waste to address the soil fertility challenges and achieve food security and sustainable waste management. Therefore, the objective of this paper is to review information on the use of human excreta-derived material (HEDM) in agriculture. The review will firstly, discuss the current context with regards to HEDM use for crop production, secondly, sanitation technologies for nutrients recovery from human urine, and thirdly, recycling the recovered nutrients for agricultural crop production. The review will conclude with a summary of key findings and recommendations for future research.

2.2 Current context concerning HEDM use for crop production

The National Environmental Management Act (NEMA) of 2008 (DWS, 2016) and the United Nations Sustainable Development Goal number 6.3 of the UN World Water Development Report (WWAP, 2017) discourages waste discharge into the environment while reuse is strongly emphasised. There is a paradigm shift in the handling of human excreta. Globally it is considered as a resource rather than waste (Andersson et al., 2016; Nansubuga et al., 2016; WWAP, 2017). Management of human waste is a critical part of daily lives, and it is an important aspect of human health (Esrey *et al.*, 2001). Many studies have demonstrated the suitability and benefits of using human excreta-derived materials as potential nutrients on arable land (AdeOluwa and Cofie, 2012; Anderson, 2015; Krause and Rotter, 2018).

2.2.1 Human urine and its use in agriculture

Human urine has been shown to contain mineral elements that are essential for plant growth and development (Maurer et al., 2006). Nitrogen (N) is the plant nutrient found in urine largely in the form of urea. However, the chemical composition of urine depends mostly on the diet of the donor. Dry urine solids are composed of 14–18% N, 13% C, 3.7% P, and 3.7% K (Strauss, 1985). Normally, a range of urine output per person is 800-2000 millilitres per day. It has been estimated that each person on average secretes 3-7 grams of nitrogen per litre of urine per year (Richert *et al.*, 2010). This is sufficient to fertilize 300-400 m² of the crop to a level of about 50-100 kg N/ha depending on crop type. Between the faecal and urine fractions, urine contains the largest proportion of N (90%), P (50–65%), and K (50–80%) released from the body (Heinonen-Tanski and van Wijk-Sijbesma, 2005).

There is a substantial amount of literature dealing with the treatment and utilization of urine for agricultural purposes (Jöhnsson *et al.*, 2004; Maurer *et al.*, 2006; Niwagaba, 2009; Pradhan *et al.*, 2010; Richert *et al.*, 2010; Wohlsager *et al.*, 2010; Semalulu *et al.*, 2011; Anderson, 2015). It is evident that the knowledge of using urine as a fertilizer has been known long back.

According to the work by Tanski *et al.*, (2005), cucumber collected from households fertilized with urine had better yields than those fertilized with commercial fertilizer. The nutrients in urine occur in ionic form and their plant availability compares well with chemical commercial fertilizer (Simons and Clemens, 2004). The findings agree with those of Tanski *et al.* (2005), and Winker *et al.* (2009), who also recommended urine as a complete fertiliser after some field trials with vegetables revealed that urine can outperform inorganic fertilisers if soil fertility strategies (soil testing, calculation of application rate and method) are practiced well. Furthermore, Guzha *et al.*, (2005), reported that growing maize with the help of toilet compost and urine on poor sandy soils was beneficial in low-income areas. However, the high water in urine makes nutrient management difficult. Concentrating nutrients, for example, making struvite, is potentially a viable agronomic option due to its high and consistent nutrient composition (Uysal *et al.*, 2010; Antonini *et al.*, 2012) as this product comprises of 98% reduction in original urine volume (Tilley *et al.*, 2011), low pathogen and heavy metal content (Decrey *et al.*, 2011).

2.2.2 Human faeces and its use in agriculture

Faecal matter is channelled to wastewater treatment plants, treated, and released as sewage sludge as a by-product. This by-product is being used for agricultural crop production in many countries for example in Vietnam (Jensen *et al.*, 2005) and China (UNHSP, 2008). Research on sewage sludge application on agricultural soils has shown that farmers recognised the importance of using organic substances to improve soil properties as early as 1862 (Etter *et al.*, 2011). The increase in organic matter can make plants more salt-tolerant as shown in Swiss chard and common beans (Smith *et al.*, 2001) and apple trees (Engel *et al.*, 2001). Previous studies have shown faeces being used as a fertilizer and its performance compared to synthetic fertilizer to be similar (Mnkeni *et al.* 2006), smaller (Richert-Stintzing *et al.*, 2001), or even larger (Heinonen-Tanski *et al.*, 2007) inefficiency magnitude.

Faeces contain carbon which contributes to an increase in organic matter in the soil. In soils, organic matter improves the soil structure, thus, making it more resistant to drought and preventing erosion. Faeces account for about 5-7% nitrogen, 3-5.4% phosphorus and 1-2.5% potassium (Rose *et al.*, 2015). The quantity, physical characteristics, and chemical composition of the excreta fractions are likely to be influenced by factors including age, gender, diet, protein, fibre, and calorie intake (Rose *et al.*, 2015).

However, there are some challenges and risks associated with the use of faecal material in crop production. Dickin *et al.* (2016) reported that the problem with the use of human faeces is that it contains many microbes that pose risks to human health. This makes it difficult to reuse without proper treatment processes. Therefore, to gain maximum benefits from faecal matter reuse, pre-treatment and processing may be required before its use in crop production (Duncker *et al.*, 2007).

Furthermore, faeces always contain high numbers of enteric bacteria (e.g. *Campylobacter*, *Salmonella*), and may contain high numbers of viruses (e.g. *Norovirus*, *Rotavirus*), protozoa (e.g. *Cryptosporidium*, *Giardia*), and parasitic worm eggs (e.g. *Ascaris*) (Heinonen-Tanski and Van Wijk-Sijbesma, 2005). The presence of these microbes in the faecal matter reduces the quality and chances of its use in agriculture unless it is subjected to treatment.

2.3 Sanitation technologies for the recovery of nutrients from human excreta

Onsite sanitation systems have been considered as potential sanitation solutions in most urban and informal settlements of South Africa (Andersson *et al.*, 2016). The onsite sanitation technologies that have been considered especially by the eThekweni Municipality include drying technologies such as Urine Diversion Dehydrated Toilets (UDDT) and the Ventilated Improved Pit (VIP) toilets (Andersson *et al.*, 2016). The UDDT separates faecal matter from urine, while in VIP toilets human excreta materials (urine and faecal matter) are not separated.

2.3.1. Nutrient recovery from urine into struvite and its fertiliser value

Studies have shown that there is a potential benefit in the use of urine as a soil amendment and the findings presented enough evidence that this material can be a source of nutrients for crop growth and development (Antonini *et al.*, 2012; Uysal *et al.*, 2014; Mchunu, 2015; Nongqwenga *et al.*, 2017). However, there are some challenges associated with its direct use as a fertiliser. Handling costs (transportation and storage) of human urine remain challenging due to the high amount of water and the possible nutrient losses (Nongqwenga *et al.*, 2017). Nitrogen loss through ammonia volatilization and inconsistent nutrient concentrations of urine are major concerns (Heinonen-Tanski and Wijk-Sijbesma, 2005). To reduce these challenges and concentrate the nutrients, human urine can be processed into struvite, a solid product which then can be used as a soil amendment.

Struvite is formed by the addition of magnesium salts into the urine, and the reaction process removes approximately 90% phosphorous from urine and precipitates it as struvite (Tilley *et al.*, 2012). The resultant product of the process is a white crystalline substance consisting of magnesium, ammonium, and phosphorus (Lind *et al.*, 2000). Struvite may be referred to as magnesium-ammonium phosphate hexahydrate ($\text{MgNH}_4\text{PO}_4 \cdot 6\text{H}_2\text{O}$) with a solubility of 0.2 g/L in water (Rahman *et al.*, 2013). The low solubility of struvite in water makes it an ideal slow-release fertiliser. In its composition, struvite contains 5.7% N, 12.6% P, and 9.9% Mg by mass. Struvite precipitation is known as an annoyance in sewage treatment plants when it forms blockages in pipes (Jaffer *et al.*, 2002). However, with the developing interests in removing phosphorus from waste streams, the recovery of phosphorus from urine as struvite has gained attention (Barak and Stafford, 2006 and Jimenez *et al.*, 2009).

2.3.2. Nutrient recovery from treated effluents

In many studies worldwide, the use of treated sewage effluents as water and nutrient sources in agricultural irrigation has been introduced as a viable alternative for wastewater disposal

options and management in the environment (Zhang & Liu, 2014; Musazura, 2015; Shirly *et al.*, 2020). One of the greatest opportunities for ammonium recovery occurs in wastewater treatment plants due to wastewater containing a large quantity of ammonium ions (Zhang & Liu, 2014). The effluent has high concentrations of inorganic N (ammonium N) (48.1-60.1 mg/L) which may undergo a nitrification process resulting in nitrates that can also be taken up by plants (Yu, 2012).

In addition to the mentioned technologies, the other onsite sanitation facility is a decentralised wastewater treatment system (DEWATS). In this system, raw waste is passed through a series of baffles where it is filtered and polished with the final effluent then discharged to either nearby wetlands or surrounding water bodies (Figure 2.1).

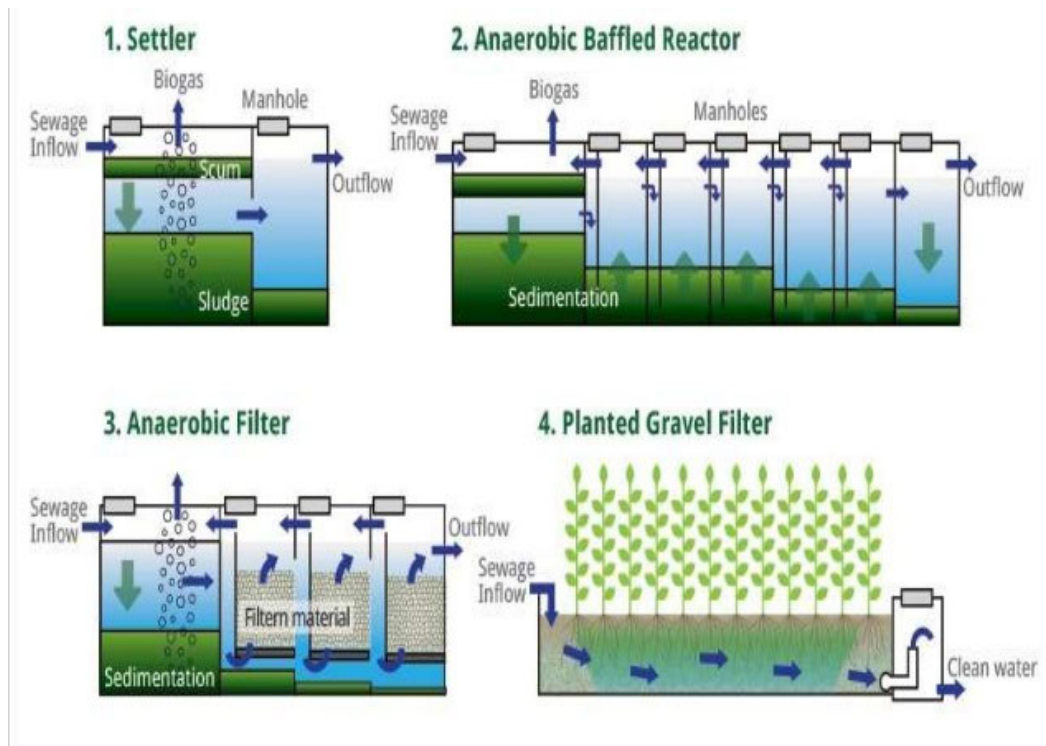


Figure 2.1. A schematic diagram showing the Decentralized Wastewater Treatment System and raw waste from households passing through ABR and anaerobic filters and disposal of effluent onto wetlands (BORDA, 2014).

2.4 Reuse of recycled nutrients in agriculture

The agronomic value of struvite has been evaluated and studies have shown that it can be as effective as commercial superphosphate, or even better under some circumstances (Uysal *et al.*, 2014; Nongqwenga *et al.*, 2017). Uysal *et al.* (2014) have suggested that struvite's effectiveness as a P fertilizer is favoured in acidic soils. Similar observations were made by Cabeze *et al.* (2011), who performed a trial with struvite produced from different sewage treatment plants. They concluded that struvite can be as effective as triple super-phosphate (TSP), even in soils of different properties. The findings suggest that struvite has the potential to complement commercial fertilisers while mitigating the environmental hazard of pollution.

Total N in struvite is found in both organic and inorganic forms; this inorganic N from struvite is not readily available to plants but needs to solubilise first and then be released slowly to the plant (Adler *et al.*, 2005). Richard *et al.* (2001) highlighted that the ammonium N contained in struvite could be as available to plants as in any ammonium N fertilizer. Nelson *et al.* (2003) investigated the effect of struvite particle size (<2mm, 2-3mm, and 4-8mm) on nutrient availability at the North Carolina (NC) State University. Smaller struvite particle sizes released more N than coarser particles and increased N uptake by ryegrass within the first 3 to 6 weeks after planting in a greenhouse trial. When compared to ammonium phosphate fertiliser, struvite released less N during the first 3-6 weeks and more N during the last 9 weeks. These researchers concluded that struvite is indeed a slow-release fertilizer and has most of its nutrients available for plants later upon application. It is therefore important to apply struvite together with a readily available inorganic source that will supply nutrients at the early stages of crop growth while struvite supplies its nutrients at later stages. In addition to the slow-release, struvite has limited N relative to P therefore, for optimum growth, additional N will be required when struvite is used as the source of P. The additional N can be achieved by the application of other waste streams rich in N or through the application of mineral fertilisers.

Various countries worldwide are using either treated or untreated forms of municipal wastewater for agriculture (Hussain *et al.*, 2019). In Pakistan, 26% of national vegetable production is irrigated with wastewater (Ensink *et al.*, 2004). In Hanoi, 80% of vegetable production is from urban and peri-urban areas (Lai, 2000). In Ghana, informal irrigation involving diluted wastewater from rivers and streams occurs on an estimated 11,500 ha, an area larger than the reported extent of formal irrigation in the country (Keraita and Drechsel, 2004). In Mexico, about 260,000 ha are irrigated with wastewater, mostly untreated (Mexico CAN, 2004). In most of these cases, farmers irrigate with diluted, untreated, or partly treated wastewater. Their use has been driven by factors such as management of wastewater volumes, water scarcity, the need for nutrient (N and P) recovery and reducing the effects of disposing nutrient-rich effluents into water bodies (Scott *et al.*, 2004; Mateo-Sagasta *et al.*, 2013). Another factor influencing the use of treated wastewater in agriculture is its impacts on the soil's chemical and physical properties, environmental pollution, effects on plants, irrigation structures, heavy metals, and microbial contamination (Drechsel *et al.*, 2010; Mateo-Sagasta *et al.*, 2013).

2.5 Heavy metals contamination: Effects on crop production, human health, and the environment

2.5.1 Heavy metals

The concentrations of heavy metals in sewage sludge have been considered the most significant restricting factor for agricultural use (Behbahaninia *et al.*, 2009). Cadmium, mercury, and lead are the most hazardous metals to humans, whilst copper, zinc, chromium, and nickel when in high concentrations are particularly poisonous to plants (Naz *et al.*, 2015). Other heavy metals like Co are of little concern since plants cannot take them up in toxic quantities (Chang *et al.*, 2002). Treated wastewater contains lower concentrations of heavy metals as compared to biosolids (Herselman and Snyman, 2009). The concentration of heavy metals in treated effluents is determined by the level of treatment, with high concentrations in raw wastewater

and sewage sludge compared to treated effluents (Behbahaninia *et al.*, 2009). This is due to the presence of organic matter affecting the solubility of heavy metals due to its capacity to form stable complexes with metal ions (organo-metallic complexes) leading to decreasing bioavailability (Levy *et al.*, 2011).

It has been shown that crops and vegetables irrigated with water containing heavy metals may accumulate a greater quantity of heavy metals (Alrawiq *et al.*, 2014). Heavy metal toxicity risks to people are dependent on the use or edible part of the crop (Maksimovic and Ilin, 2012). A study conducted by Jamali *et al.* (2007) demonstrated an increased danger of growing vegetables in soil irrigated with wastewater and sewage sludge. According to Gharbi *et al.* (2005), heavy metals accumulate in lettuce roots as compared to edible parts (leaves). Akbari *et al.* (2012) found that irrigating dry beans with urban wastewater leads to the accumulation of heavy metals in bean roots and leaves as compared to the pods. The main source of heavy metals in sewage sludge is industrial effluent. As such, areas of low industrial activities produce sewage sludge and wastewater with low concentrations of heavy metals. In general, the treatment of wastewater to be used for irrigation purposes requires a low purification level. Some minimum quality standards have been set for agricultural irrigation with wastewater (USEPA, 2012; Alcade-Sanz and Gawlik, 2017) with regards to key pollutants such as chemicals, particles, nutrients, hardly biodegradable organics, heavy metals, and microbes.

2.5.2 Crop production

The use of treated effluents for agricultural irrigation is an old and popular practice (Jimenez and Asano, 2008). Most treated effluents have total N concentrations of between 20 and 85 mg L⁻¹ (Pescod, 1992), making it a good source of N fertiliser. However, its application to the soil as irrigation gets limited by the water holding capacity of specific soil irrigated. This becomes a challenge specially to crops such as maize and tomatoes that require higher N than that can be applied through effluent irrigation. In such instances, supplementary N fertilisers would be

required relative to those crops that require less N. Furthermore, given that the effluent is in liquid form, possible nutrient losses at handling and transportation are inevitable. Loss of nitrogen through ammonia volatilization and inconsistent nutrient concentrations are also of concern. Studies in hydroponic plant production of tomato plants using wastewater solely to supply nutrients have reported that nutrients were deficient such as nitrogen, phosphorus, potassium, and calcium (Roosta and Hamidpour, 2011). However, other studies have reported that supplementation of wastewater with deficient nutrients improved plant growth in hydroponically grown plants. The addition of P and micronutrients particularly, iron increased the shoot biomass of lettuce (Liu *et al.*, 2011).

Magwaza. *et al.* (2020) investigated the combined effect of domestic wastewater and commercial hydroponic fertilizer mix as plant nutrient sources on growth and yield performance and nutrient availability of hydroponically grown tomatoes. The results observed from the study indicated that the fertigation of tomato plants with ABR effluents in a hydroponic system was not sufficient to support plant growth. The low concentration of essential nutrients such as N, P, K, Ca, and Zn in wastewater is the reason for reduced growth and yield performance as compared to plants fed with commercial fertilizer mix. However, plants grown from commercial hydroponic fertilizer mix added to ABR effluent showed increased plant growth, yield performances, and shoot nutrients content. This indicated that the addition of a 50 % dose of commercial hydroponic fertilizer to the wastewater in ABR effluent increased nutrient availability to plants. A similar observation was reported when the effect of foliar application of nutrients such as Mn, Fe, Mg, Zn, Cu, K, Mg was investigated on tomato plants grown in aquaponics (Roosta and Hamidpour, 2011). These studies concluded that optimizing tomato production in hydroponic systems may require the addition of fertilizer to wastewater if used as a nutrient source. Research using the effluent together with struvite and

with supplementary fertiliser for other crops such as maize is important to assess the growth, nutrient uptake, and biomass production under different climatic conditions.

2.5.3 Environment

Irrigation with treated wastewater introduces nitrogen which can be nitrified to nitrates and nitrates are known to be very mobile in soil solution. These cannot be adsorbed by negatively charged soil colloids (Levy *et al.*, 2011) hence they can be easily leached down to underground water resources especially in sandy soils. When nitrates leach through the soil they can contaminate the groundwater, exposing people to risks of diseases (Dickin *et al.*, 2016). Reduced infiltration rates, leads to increased surface runoff, soil erosion, and the deposition of P-rich soil into rivers promote the formation of algal blooms and eutrophication leading to considerable pollution of rivers (Dickin *et al.*, 2016). Groundwater contamination and eutrophication risks can be prevented by proper planning (Gutterer *et al.*, 2009). Barton *et al.*, 2005 reported that soils that have high organic matter content usually have reduced rates of leaching as these can increase soil aggregation as well as the anion exchange capacity and retention of nitrate N (NO_3^-). Factors such as the level of the water table, slope, soil physical characteristics (texture, hydraulic conductivity, bulk density, and matric potentials), and the proximity to water bodies (rivers) must be considered (Pescod, 1992; Helmar and Hespanhol, 1997).

2.5.4 Social acceptance

The safe use of urine and faeces in agriculture has been scientifically proven (Schönning, 2001; Phasha, 2005; WHO 2006), but users' acceptance of such practices has been a concern for policy makers and practitioners in the sanitation sector (Richert *et al.* 2010a). Surveys conducted on the user perceptions of the use of human excreta in agriculture (Roma *et al.* 2013) identified common challenges in the acceptance of such practice. There is poor awareness of the fertilising value of these materials which represents one obstacle in the uptake of such

practice. In a study in Nigeria, Sridhar *et al.* (2005) found that just 7.7% of respondents were in favour of using urine as a fertiliser for vegetable production. Interestingly, after demonstrating the value and potential of urine in agriculture, the research identified a sharp increase in acceptance, with 80% of the respondents showing willingness to use urine in agriculture. Furthermore, concerns for the presence of pathogens in urine and health risks present obstacles in reusing urine in agriculture (Roma *et al.* 2013).

In the eThekweni municipality of KwaZulu-Natal (South Africa), the eThekweni Water and Sanitation Unit (EWS) installed 75,000 Urine Diversion Dehydrated Toilets (UDDTs) in rural areas to address the sanitation backlog and a cholera outbreak in 2000 (Sustainable Sanitation Alliance 2011). The municipality provided households with training on how to use and maintain UDDTs and explored the potential for reusing urine in agriculture, thus transforming UDDTs into 'productive' sanitation technologies, which allows for nutrient recovery from human waste.

A study in the eThekweni municipality by Okem *et al.* (2013) explored UDDT users' perceptions of using urine in agricultural activities. The results of this study demonstrated that gardening is a common activity in the study area with nearly half (46.9%) of participants reporting to own a garden for food and/or flowers. In addition, more than half (79%) of those who do not own a garden would like to have one. Despite the proven value of urine in agriculture, the study observed that only 3.6% of the participants currently use urine as an alternative fertiliser in their gardens and less than 1% (0.4% n=1) of respondents sell the product of their gardens. The limited usage of urine as an alternative fertiliser could be attributed to the lack of knowledge that urine could act as a sustainable and effective replacement of conventional nutrient sources like chemical fertilisers.

2.6 Summary

Urine products have significant nutrient concentrations, as such, these products have a potential for use as fertiliser in crop production. Struvite supplies more P than N and may need supplementary N sources for optimum plant growth. The appropriateness of nutrient materials such as urine and urine-based nutrient sources, as a fertilizer, depends on the solubility of the products and mineralization of the constituent elements, which vary with soil types. Dry matter production increases with time after fertiliser application and declines over time. The use of human excreta (urine and faeces) in agriculture has been studied extensively and scientifically proven to be a viable option to address challenges concerning sanitation and reusing nutrients recycled for agricultural crop production. More interventions, such as the development of appropriate guidelines and standard measures on health effects related to the re-use of urine based on rigorous evidence should be developed and implemented by local authorities.

CHAPTER THREE: NITROGEN AND PHOSPHORUS RELEASE PATTERNS FROM THE CARTREF SOIL AMENDED WITH HUMAN EXCRETA-DERIVED MATERIALS (DEWATS EFFLUENT AND STRUVITE) AND CHEMICAL FERTILIZERS.

3.1 Abstract

Human excreta-derived materials (HEDMs) such as struvite and treated wastewater effluents contain significant concentrations of nutrients important for plant growth and development. Determining nutrient release patterns of HEDMs is vital for optimising application rates for crop production. This study investigated nitrogen and phosphorus release patterns in Cartref soil amended with DEWATS effluent and struvite. Two laboratory incubation experiments were conducted for 56 days. The first study was laid out as a single factor experiment, with the following treatments: (i) struvite alone, (ii) urea alone, (iii) SSP alone, (iv) struvite + urea, (v) SSP + urea, and (vi) control, all replicated 3 times. The second one was also laid as a single factor experiment with the following treatments: (i) effluent alone, (ii) struvite + effluent, (iii) effluent + SSP, and (iv) control, all replicated 3 times. There were significant differences observed amongst the treatments ($P \leq 0.05$). In both experiments, Ammonium-N release was initially high and thereafter declined with time, while nitrate- N increased significantly ($P < 0.05$). The combination of HEDM (struvite or effluent) with commercial (urea or SSP) performed well than when SSP + urea and sole fertilisers were applied in terms of ammonium-N and nitrate- N release. The findings suggest that a combination of struvite and effluent with inorganic fertilisers could potentially be considered as a better option for increasing fertiliser use efficiency and providing a more balanced supply of nutrients than when applied alone.

Keywords: Ammonium N, Nitrate N, Extractable P, Effluent, Struvite

3.2 Introduction

Chemical/synthetic phosphorus fertilizers are becoming more limiting for agriculture use. It is projected that phosphate rock may run out in the future due to excessive mining to improve soil fertility and maximize agricultural productivity (Bonvin, 2013). Similarly, the production of nitrogen fertilizers from atmospheric N is an expensive process (Karaka and Bhattacharyya, 2011). This has resulted in increased prices of commercial fertiliser, making them inaccessible to resource-constrained smallholder farmers in most developing countries. The use of human excreta-derived products, such as struvite and effluents from wastewater treatment systems could be an effective waste management strategy while providing potential alternative nutrient sources to substitute or use in combination with chemical fertiliser for agricultural crop production.

In recent years, significant research has been done on the use of human excreta-derived materials (HEDMs) as sources of mineral nutrients for crop production (Prazares *et al.*, 2017; Carvalho *et al.*, 2018; Gebeyehu *et al.*, 2018). Currently, the focus is increasingly towards nutrient cycling and reuse of waste and wastewater in agriculture. For example, in South Africa, the eThekweni Municipality is assessing the use of alternative sanitation technologies such as the Decentralised Wastewater Treatment System (DEWATS) (Hudson, 2011). The DEWATS is a modular water-borne sanitation system, which consists of the settler, anaerobic baffled reactor (ABR) + anaerobic filter (AF), and planted gravel filters (Gutterer *et al.*, 2009b). The treatment process involves anaerobic degradation of organic matter within the ABR and then AF. The AF effluent is further passed to planted gravel filters, which consist of vertical flow constructed wetland (VFCW) and horizontal flow constructed wetland (HFCW) for further polishing. The AF effluent has been shown to contain high concentrations of mineral elements such as nitrogen (N) and phosphorus (P), which are important for plant growth (Musazura *et al.*, 2019). Instead of water-based sanitation systems, the urine-diversion toilets separate urine

and faeces at the source making their management easier. Generally, most of the nutrients are in urine while carbon is more in the faecal matter. However, a large amount of water in urine makes its use, as a nutrient source very challenging. This has led to the development of technologies that can treat human urine and extract P in concentrated form as struvite (Mihelcic et al., 2011).

Struvite is formed by the addition of magnesium (Mg) to urine, which results in a precipitate (struvite) that can be used as fertilizer. Struvite contains an average of 5.7% N, 12.6% P, and 9.9% Mg, and can be used as a fertilizer to supply mainly P, together with N and Mg (Johnston and Richards, 2003). The suitability of the effluent and struvite as fertilizer depends on the relative compositions of the nutrients and their release to plant-available forms in soil (Murugan and Swarnam, 2013). Waste-based nutrient sources such as struvite and effluent contain nutrients (N and P) that exist in organic and inorganic forms and must undergo some transformations before becoming bioavailable for plant uptake. For example, DEWATS effluent has high concentrations of inorganic N (ammonium N), which can be taken up by plants (Musazura, 2014). Whilst struvite needs to be solubilised to make the nutrients available (Nongwenga *et al.*, 2017).

Mineralisation and immobilization are essential biochemical processes that affect the availability of plant essential nutrients in the soil and are mediated through the activity of microorganisms (Murugan and Swarnam, 2013). Biochemical processes, dissolution, precipitation, and sorption are essential biochemical processes affecting the availability of nutrients. These processes are affected by several factors like temperature, soil moisture, and pH (Murugan and Swarnam, 2013) in addition to clay mineralogy. When these conditions are favourable for microorganisms to metabolize, mineralisation, the microbial conversion of organic nutrients to inorganic forms will take place after which, nitrate, ammonium, and orthophosphates would be available for plant uptake. Immobilisation occurs when these plant-

available forms are taken up by microbes, turning the nutrients into organic forms that are not available to plants. In addition to the soil's physical environment, the processes of mineralisation and immobilisation are also affected by the balance of nutrients in the soil, including those added as organic or inorganic fertilizer materials.

Therefore, differences in HEDMs properties would likely cause differences in nutrient release patterns. Understanding these nutrient dynamics provides a basis for decision-making options on their sustainable application rates to meet crop requirements and while minimising environmental pollution. The higher P than N in struvite suggests that co-application with another source of N is essential, while the higher N than P in the effluent may mean that additional P may be required. However, the implications of co-application of struvite and effluent with other concentrated N and or P sources need to be clearly understood if this approach is to be used to optimise nutrient availability. There is a lack of information on the release patterns of N and P from the combined application of HEDMs or in combination with commercial fertilisers.

Therefore, the objectives of this study were;(i) To determine nitrogen and phosphorus release patterns from struvite, superphosphate (SSP) when used singly or combined with urea in a sandy loam soil, and (ii) To determine the nitrogen and phosphorus release patterns of the effluent when used singly, and in combination with struvite or with SSP in a sandy loam soil.

3.3 Materials and methods

3.3.1 Research materials

3.3.1.1 Soil

The Cartref (Cf) soil form was collected from an arable field in KwadinaBakubo area (Hillcrest, South Africa) (29°46'48"S and 30°45'46"E). Soil samples were taken from a depth range of 0 to 0.3 m using a 3 m soil bucket auger and sent for fertility analysis at the Soil Fertility and Analytical Services in Cedara (KwaZulu-Natal, Department of Agriculture and Rural Development). The samples were air-dried and sieved to pass through a 2 mm sieve before use. The characteristics of the soil used are shown in Table 3.1.

3.3.1.2 Plant nutrient sources investigated

The plant nutrient sources used in this study were obtained from an experimental site at Newlands Mashu, Durban in South Africa (longitude of 30°57'E and latitude of 29°58'S). These consisted of struvite (S) processed from source-separated urine and the effluent (E) from anaerobic filters of the DEWATS. The struvite was processed at a reactor plant at Newlands Mashu and contained 12.6% P and 5.7%N. The inorganic N and P of DEWATS effluent, which was mainly used as a nitrogen source in the study are shown in Table 3.2.

Table 3.1. Chemical and physical properties of the Cartref soil used during the study (Musazura et al., 2019)

Property	Value
Bulky density (kg m ⁻³)	1430
Clay (%)	12
Silt (%)	15
Sand (%)	73
Field capacity (m m ⁻¹)	0.24
Permanent wilting point (m m ⁻¹)	0.12
Organic C (%)	<0.5
Extractable P (mg kg ⁻¹)	0.7
pH (KCl)	4.21
Total cations (cmol _c kg ⁻¹)	1.2
Acid saturation (%)	18
Exchangeable K (mg kg ⁻¹)	0.01
Exchangeable Ca (mg kg ⁻¹)	0.4
Exchangeable Mg (mg kg ⁻¹)	0.4
Exch. acidity (cmol _c kg ⁻¹)	0.18
Extractable Zn (mg kg ⁻¹)	0.1
Extractable Mn (mg kg ⁻¹)	0.7
Extractable Cu (mg kg ⁻¹)	0.2

Table 3.2: Nutrient content inorganic N, total N, and P (mg/L) in the DEWATS effluent used for the study (Musazura *et al.*, 2019).

Nutrient element	NO ₃ ⁻ -N	NH ₄ ⁺ -N	PO ₄ ³⁻	Total N
N	9	9	9	3
Mean ± SE	2.1 ± 0.5	54.8 ± 1.6	10.5 ± 1.5	60.6 ± 2.7
Median	1.8	55.6	8.7	59.8
Range	0.2 - 4.1	48.1 - 60.1	5.9 - 19.5	51.2 - 68.4

3.3.2 Experimental set up

Two laboratory soil incubation studies were conducted at the School of Agricultural, Earth, and Environmental Sciences (SAEES), University of Kwazulu-Natal, Pietermaritzburg Campus, South Africa. The first experiment was laid out as a single factor design with the following treatments: (i) struvite alone, (ii) urea alone, (iii) SSP alone, (iv) struvite + urea, (v) SSP + urea, with each treatment replicated three times to give 15 experimental units, in rectangular 5 litre containers. The containers were drilled on the sides to allow aerobic conditions. Two (2) kg of air-dried soil was placed in each container and treated according to the treatment rates shown in Table 3.3. Treatments rates were applied to achieve an equivalent of 200 kg N/ha and 60 kg P/ha to meet maize nutrient requirements from the Cf soil. Since struvite supplies both N and P, it was applied as a P source to supply 60 kg P/ha, resulting in the application of 27.1 kg N/ha. This experiment was conducted under controlled room temperature conditions maintained at 25°C, 80% relative humidity, and irrigated with distilled water to maintain a 100% field capacity moisture content. To monitor the changes in mineral N and P, sampling from the treatments were carried out on day 0, 1, 3, 7, 14, 21, 28, 35, 42, 49, and 56. The samples were immediately stored in a fridge (0°C) after sampling.

The second experiment was also laid out as a single factor comprising the following treatments: (i) effluent alone, (ii) struvite + effluent, (iii) effluent + SSP, and (iv) a control, all replicated 3 times to give 12 experimental units (in 5 litre containers). The effluent in this incubation study was applied as an irrigation source to achieve a 100% soil field capacity and supply nutrients at the same time, while the treatments without effluent were irrigated with distilled water. The management and sampling were as described in the first experiment.

Table 3.3. Converted application rates (grams/2 kg soil) for fertilizer treatments used.

Treatment	Application rate (g per 2 kg soil)
Struvite	0.254
SSP	0.305
Urea	0.234

SSP = single superphosphate

The effluent was added to meet the nutrient N requirements of maize and when field capacity was reached the extra effluent was added the preceding day.

3.3.3 Extraction and analyses for inorganic N and P.

For each sampling day, ammonium- and nitrate- N were extracted with 2M KCl (potassium chloride). An aliquot of 2 g soil was suspended in 2M KCl at a soil: solution ratio of 1:5. The suspension was shaken using a reciprocal shaker for 30 minutes at 180 rpm, allowed to stand for 30 minutes (Chiyoka, 2011) before filtering into 250 ml volumetric flasks using Whatman® No.42 paper. Sample extracts were immediately analysed for inorganic nitrogen using a discreet analyser (Thermo Gallery: Thermo Scientific).

Orthophosphate-P was extracted using 0.25 M ammonium bicarbonate (NH_4HCO_3), EDTA disodium salt, and 0.01 M ammonium fluoride (AMBIC) solution. The sample aliquot of 2 g was suspended in 20 ml of AMBIC solution in a centrifuge tube and shaken using a reciprocal shaker at 180 rpm for 30 minutes and analysed using the molybdenum blue method, based on absorbance read through the UV/VIS spectrophotometer (Murphy and Riley, 1962).

The pH of the soil treated with fertilizers was also measured at each sampling date to monitor the changes in soil acidity or alkalinity. Two grams of soil were placed in a 30 ml plastic beaker and 20 ml of 1M KCl was added. The mixture was stirred using a magnetic stirrer and left to stand for 30 minutes. pH was then read using a calibrated H198129 pH meter (HANNA Instrument, Romania).

3.3.4 Data analysis

All data were statistically analysed using GenStat® 18th edition (VSN International, United Kingdom). The data were subjected to one-way analysis of variance (ANOVA) and the least significant differences were used to separate the means at the 5% significance level.

3.4 Results

3.4.1 pH, mineral N, and P in the soil treated with solid fertilizers only

3.4.1.1 Soil pH

The initial pH of 4.21 (Table 3.1) was recorded in all treatments carried out on the sandy soil. Significant differences ($P < 0.05$) were observed among the fertilizer treatments. Treatments containing struvite and urea showed a slightly higher pH with a range of 4.38 – 5.12 but were not significantly different from the other treatments on days 3 and 7. On average, soil pH constantly remained in the acidic range throughout the incubation period across all treatments (Fig 3.1).

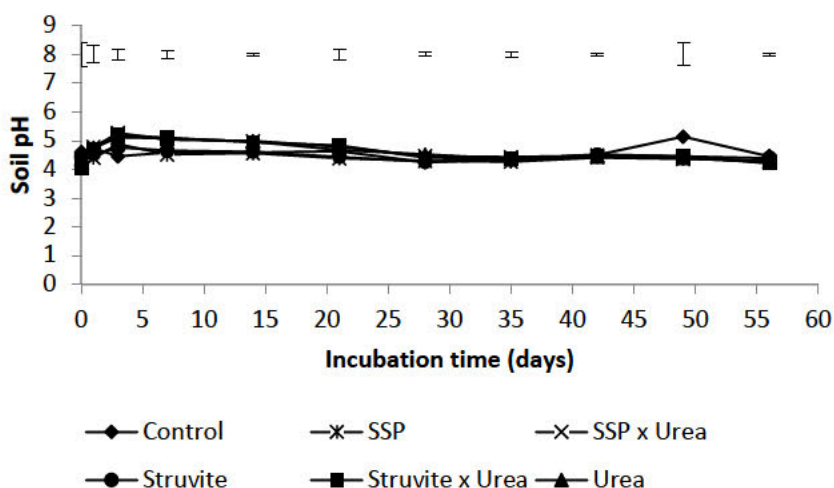


Figure 3.1. Changes in soil pH during incubation time of 56 days from co-applied treatments with struvite relative to single applications. The error bars represent the standard error of means.

3.4.1.2 Ammonium- and nitrate- N

Within the first half of the incubation period, Ammonium-N concentrations were decreasing in all treatments. There were no significant differences noted among treatments up to day 56 of incubation. Generally, the concentrations of ammonium N (mg/kg) were initially higher in the early weeks (0-21 days) of the incubation and then decreased steadily throughout the incubation period (Fig 3.2). Ammonium N was higher in urea added treatments compared to treatments containing struvite only, SSP only, and the control up to day 56 of the incubation (Fig 3.2).

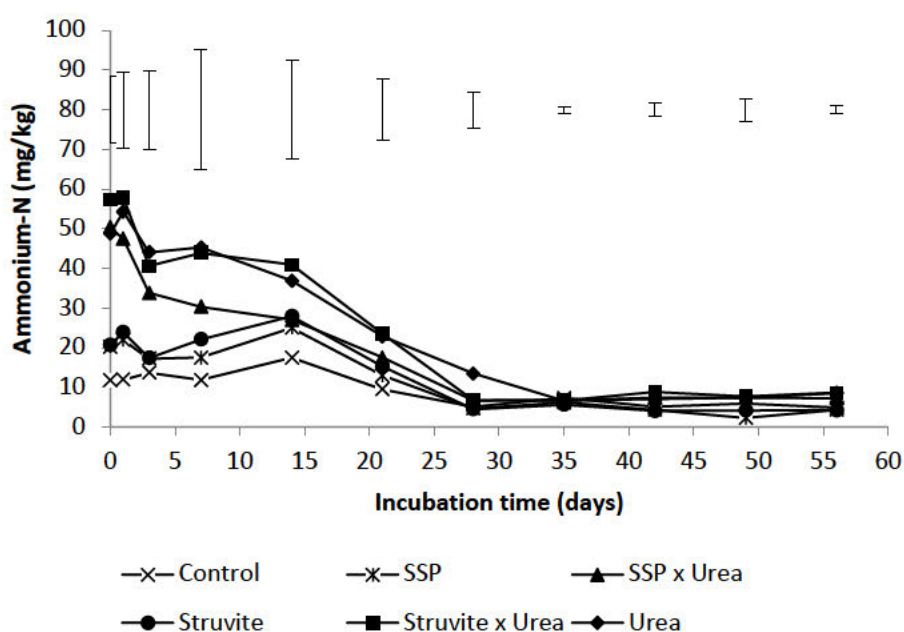


Figure 3.2. Changes in NH₄-N concentration during incubation time of 56 days from co-applied treatments with struvite relative to single applications. The error bars represent the standard error of means.

There were no significant differences observed in nitrate-N release across treatments over the period of 56 days of incubation. The initial nitrate-N concentrations in all treatments were low and in the range of 2.22-2.69 mg/kg between days 0 to 7. However, from day 7, a rapid increase was observed in all treatments particularly between day 14 and 28, with a slower increase thereafter until day 56 of the incubation. Treatments containing urea (urea alone, urea + struvite, and urea + SSP) released significantly ($P < 0.05$) higher nitrate-N quantities compared

to the rest of the treatments (Fig 3.3) whilst the remaining treatments did not differ significantly ($P>0.05$) from each other.

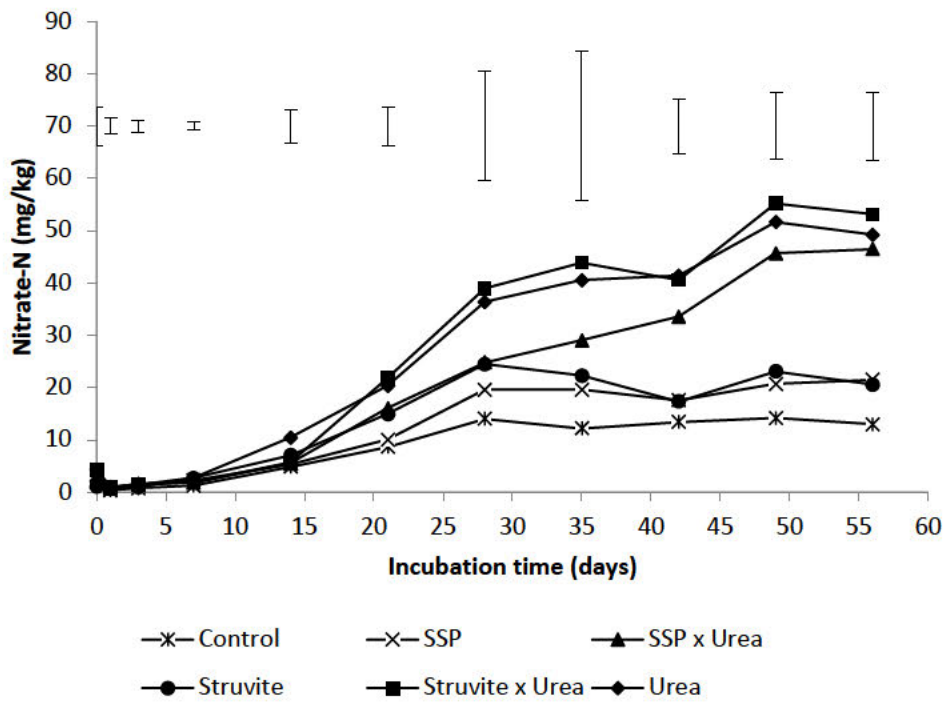


Figure 3.3. Changes in $\text{NO}_3\text{-N}$ concentration during incubation time of 56 days from co-applied treatments with struvite relative to single applications. The error bars represent the standard error of means.

3.4.1.3 Extractable-P

Extractable-P concentration in the Cartref soil treated with different fertilizer treatments (SSP, struvite, urea, SSP + urea, struvite + urea, and the control) showed a constantly fluctuating trend, with P ranging from 10 - 17.34 mg/kg (Fig 3.4). There were no significant differences ($P>0.05$) amongst the treatments throughout the incubation period.

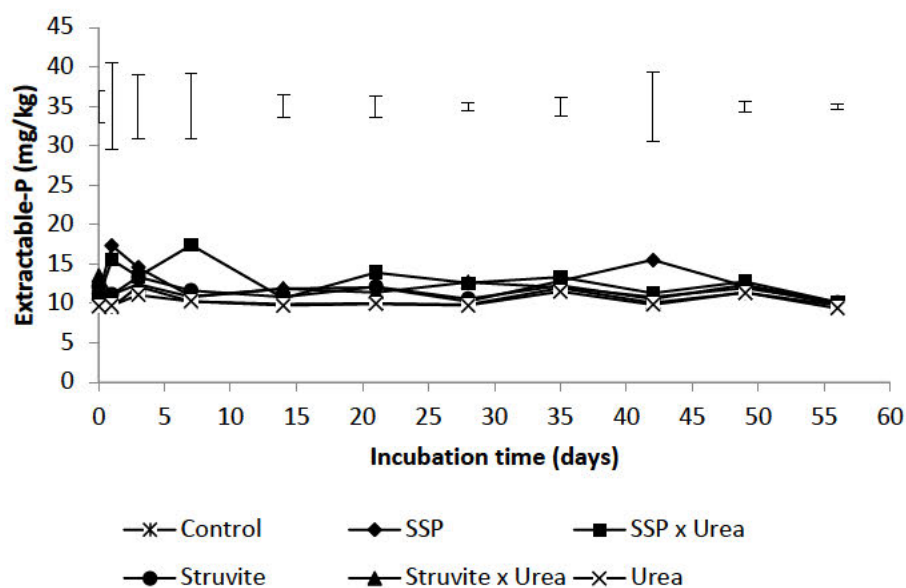


Figure 3.4. Changes in Orthophosphate-P concentration during incubation time of 56 days from co-applied treatments with struvite relative to single applications. The error bars standard error of means.

3.4.2 The pH, mineral N, and P changes in the soil treated with effluent over the incubation period

3.4.2.1 Soil pH

The effluent-containing treatment showed pH trends similar to that of the first incubation experiment (Fig 3.5). The initial pH of all treatments was between 4.3 and 4.6 with effluent + SSP treatment recording the lowest whilst the highest pH level was observed from the control (Fig 3.5). Generally, there was no significant change in soil pH throughout the incubation period. pH levels did not differ significantly amongst the treatments (effluent, effluent + SSP, effluent + struvite, and the control).

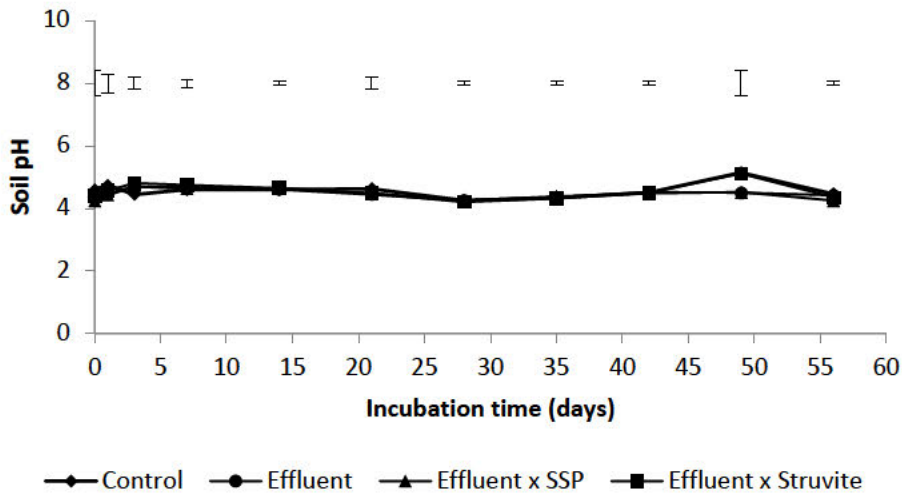


Figure 3.5. Changes in soil pH during incubation time of 56 days from co-applied treatments with effluent relative to single applications. The error bars represent the standard error of means.

3.4.2.2 Ammonium-N and Nitrate-N

The effluent + SSP and effluent + struvite had higher initial ammonium N concentrations relative to effluent alone and control treatments (Fig 3.6). Ammonium-N concentrations decreased gradually to 10 mg/kg between days 0 and 28, and beyond this period, the concentrations started to constantly level up until day 56 of the incubation. Overall, the observed concentrations were not significantly different ($P > 0.05$) between treatments throughout the incubation.

Nitrate-N concentrations in the soil treated with effluent were also investigated over a period of 56 days (Figure 3.7). There were significant differences observed ($P < 0.05$). Nitrate-N content in all treatments was initially low and increased steadily with time from day 7 up to day 56. The effluent + SSP and effluent + struvite treatments released significantly more nitrates than the control treatment, especially after 49 and 56 days of incubation.

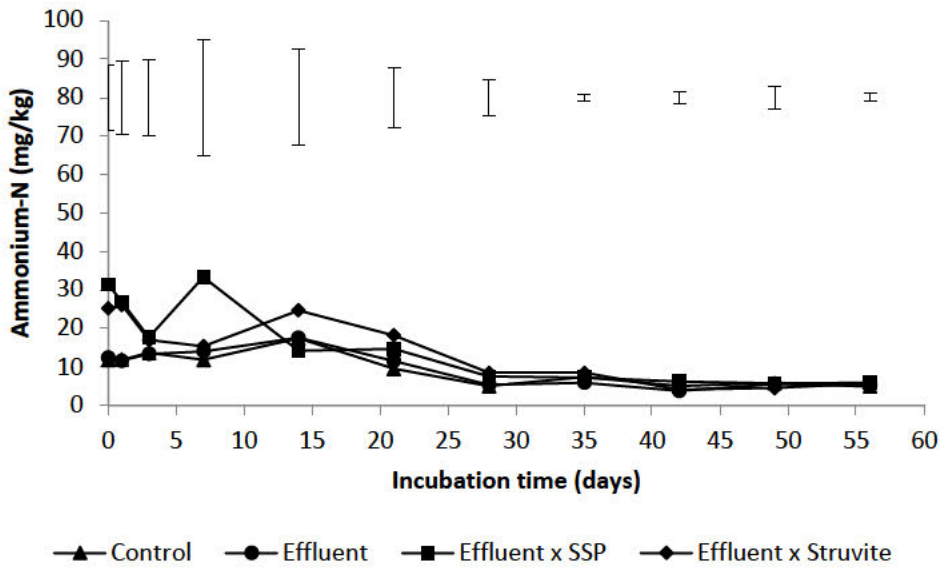


Figure 3.6. Changes in $\text{NH}_4\text{-N}$ concentration during incubation time of 56 days from co-applied treatments with effluent relative to single applications. The error bars represent the standard error of means.

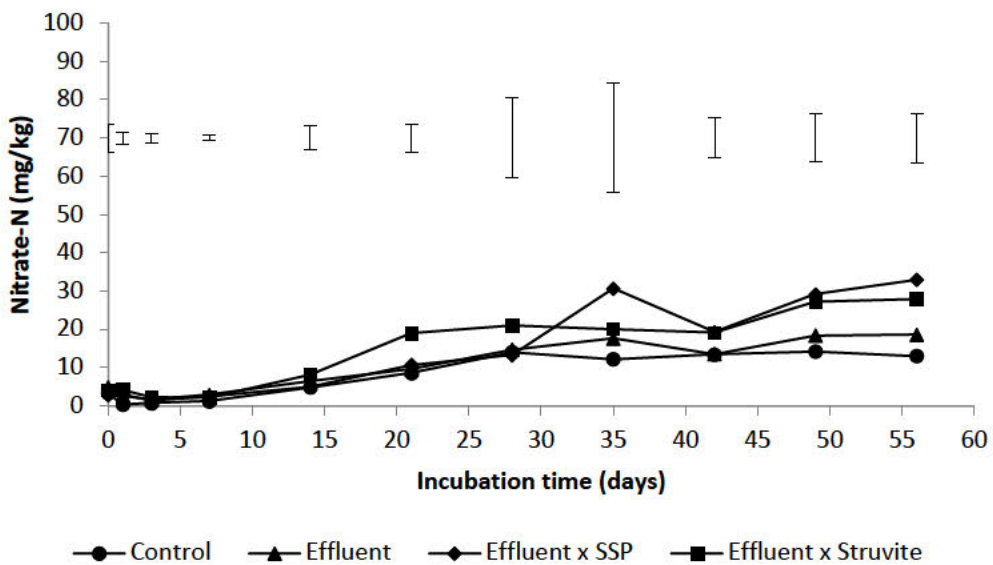


Figure 3.7. Changes in $\text{NO}_3\text{-N}$ concentration during incubation time of 56 days from co-applied treatments with effluent relative to single applications. The error bars represent the standard error of means.

3.4.2.3 Extractable-P

The extractable-P trend observed for the Cartref soil treated with effluent, effluent + struvite, effluent + SSP, and the control ranged between 10–15 mg/kg and was constant in all treatments (Fig 3.8) throughout the incubation period. Treatments did not differ significantly ($P>0.05$) in extractable- P concentrations throughout the incubation period. However, the combination of effluent + struvite released higher (12.85 mgP/kg) at day 3 than 10.75 mg/kg P released from the effluent + SSP treatment.

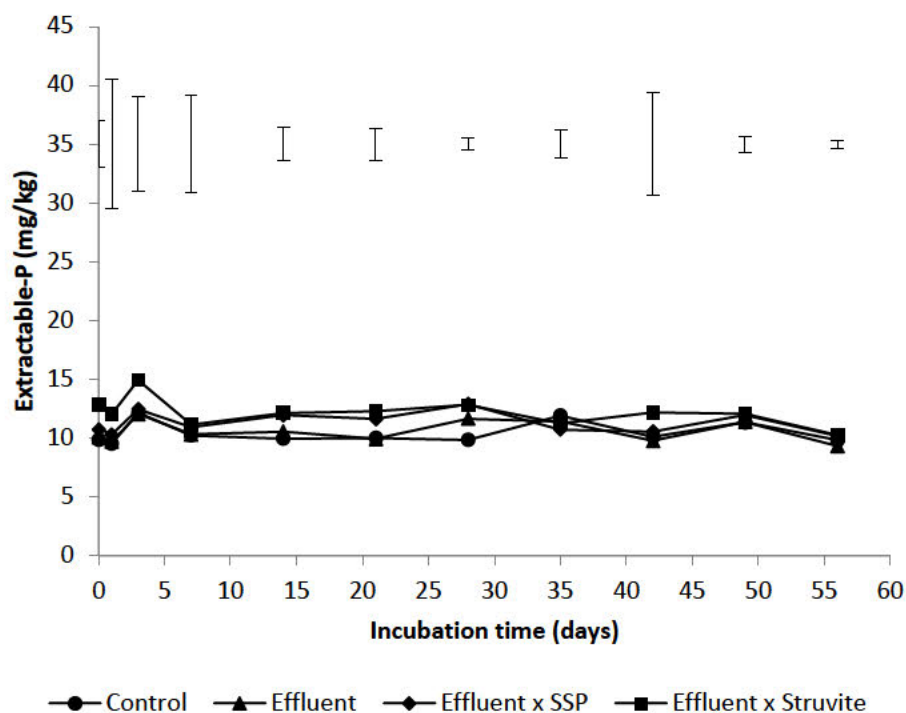


Figure 3.8. Changes in Extractable-P concentration during incubation time of 56 days from co-applied treatments with effluent relative to single applications. The error bars represent the standard error of means.

3.5 Discussion

The progressive decline of ammonium- N over the incubation period corresponded with an increase in the nitrate- N levels. This could be explained by nitrification because during the mineralization process ammonium- N is first released into the soil, and the nitrifying bacteria oxidize ammonium- N into nitrate- N, resulting in decreasing in ammonium- N content and an

increase in the nitrate- N (Murugan and Swarnam, 2013). However, ammonium- N tends to accumulate in soils if the nitrification process is inhibited. This means that not all the quantities of ammonium- N converted to nitrate- N during nitrification. The process of immobilisation can also prevent ammonium from undergoing nitrification (Bengtsson et al., 2003). This process occurs when ammonium gets used up by microorganisms for their N requirements.

The ammonium- N and nitrate- N release pattern from the commercial chemical fertilizers (SSP and urea) was rapid at the initial stages. The two commercial fertilizers solubilise faster and tend to be readily available when applied to the soil. Most of the ammonium and nitrates released from struvite alone were lower compared to urea. Struvite formation uses low quantities of N and high amounts of P from the urine (Udert and Wachter, 2011), this explains the lower N content in the struvite than urea. The lower initial N content in struvite resulted in lower ammonium N and nitrate N. When struvite was combined with urea, mineral- N (ammonium and nitrate) was released early in the incubation, and so the nitrification process happened fast. The fast nitrification resulted from the acidity of the Cartref soil, as also reported by Bame *et al.* (2013). This scenario means that struvite can provide mineral nutrients slowly to plants during the growing season, and some external sources of N (urea used in this study) may need to be incorporated to meet the crop N needs in the early days of growth. Unlike the readily available commercial chemical nutrient sources with high solubility, struvite has low solubility of 0.2 g/L in water (Barak and Stafford, 2006). According to Nicolardot *et al.* (1994), when struvite is applied to the soil, nutrient release is largely a result of microbial nitrification of ammonium constituent rather than simple dissolution. Lind et al. (2000) mentioned that struvite has its nutrients released in a biologically based slow-release mechanism. The combination of HEDM (struvite) and commercial (urea) performed better on ammonium- N release than when SSP + urea was applied together (Fig 3.3). The main reason behind this difference in pattern is the level of initial N concentration from these combinations where 200

+ 27 kg N/ha was applied from struvite + urea compared to SSP + struvite in which struvite alone was the source of N (27 kg N/ha) for the treatment.

During the second incubation study, the nutrient release patterns were investigated from the application of the DEWATS effluent alone or in combination with struvite and or SSP into the Cartref sandy soil. Similar to the first experiment observations, higher ammonium- N and nitrate- N were released rapidly in the initial stages where effluent was in combination with SSP or with struvite than when it was applied alone. The effluent is high in organic matter and organic N content that need to undergo decomposition and mineralization processes respectively (Yu, 2012). These are microorganisms-mediated processes that require enough N for microbial use for energy supply before it is released for plant uptake. A negative N period is therefore created especially in the early days of application as most of the available N would be taken up by microbes acting on the organic matter (Kuzyakov *et al.*, 2000; Ngole, 2010). The liquid nature of the effluent used could also have caused a significant N- loss. According to Argo and Biernbaum, (1995), ammonium- N in liquid fertilisers or materials could be lost quickly to the air as the liquid material evaporates. A combination of effluent and commercial SSP performed better than when effluent + struvite on P release. This could strongly be attributed to the greater solubility of SSP than struvite making P more available in the former than the later treatment. Although SSP has more soluble P than struvite, this did not result in any significant differences ($P>0.05$) in extractable-P amongst the treatments throughout the incubation. The low P content in the sandy soil (Cf) was probably due to the high drainage of the soil. According to findings by Bame *et al.* (2013), Cf loses more P due to its coarse texture where there is no retention of Al/Mn/Fe minerals.

Mineralisation and immobilisation, are pathways most relevant to this study which might be responsible for the trends observed in the release of P in the sandy soil. Mineralisation increases plant-available P while immobilisation hinders P availability (Hyland *et al.*, 2005). The limited

variability in extractable-P over time in all the treatments could be explained by the acidity state of the soil as shown by the low pH levels which also did not change with incubation time. In acidic conditions, concentrations of most nutrients, including P, are low and some of these nutrients are also taken up by microorganisms for their metabolism (Hue *et al.*, 1986). This is also supported by Tully *et al.*, (2013) who reported that the concentration of phosphorus is generally lower at a pH of <5.5 than at higher pH. Phosphorus is most available for plant use when the pH ranges between 6 and 7 units (Nelson *et al.*, 1953). Liming is a common practice that is used to increase the soil pH in acidic soils making them -non-acidic. In addition, liming improves P concentration and beneficial microbe populations in the soil (Fageria and Moreira, 2011).

3.6 Conclusion

The study concludes that the HEDM performed better with respect to available nitrogen release, but not phosphorus, when combined with other nutrient sources to optimise N and P than when used solely in an acidic sandy soil. Struvite as a source of P may be comparable with SSP which means that struvite could be used to supply the nutrient P instead of SSP. The concentration of extractable-P from the different sources can be suppressed under acidic conditions and therefore the addition of lime could be required to increase soil pH, and ultimately enhance the availability of P. Optimising N and P supply through combining AF effluent and struvite or with inorganic fertilisers could potentially be a better option for balanced crop nutrients supply than when applied individually.

CHAPTER FOUR: THE EFFECT OF STRUVITE AND DEWATS EFFLUENT ON THE GROWTH, NUTRIENT UPTAKE, AND BIOMASS PRODUCTION OF MAIZE.

4.1 Abstract

The use of human excreta-derived matter (HEDM) like struvite and treated effluent could be alternative sources of N and P for smallholder agriculture in sub-Saharan Africa (SSA). The combined application of HEDM with inorganic commercial fertilisers could provide much-needed nutrients for crop growth. The study was conducted in a tunnel to determine the effects of the application of struvite and treated effluent from the anaerobic filters (AF) on the growth, nutrient uptake, and biomass production of maize. The fertilizers were applied at half-recommended (0.5) and recommended (R) rates. The study was set up as an 9 x 2 factorial experiment in a completely randomised design (CRD) with the following factors: fertilizer combinations (8 levels- (i) struvite + urea (R); (ii) struvite + urea (0.5), (iii) struvite + effluent (R); (iv) struvite + effluent (0.5); (v) SSP + effluent (R); (vi) SSP + effluent (0.5); (vii) SSP + urea (R) and (viii) SSP + urea (0.5) and the control. The second factor was maize variety with two (2) levels that is 'Colorado' and 'IMAS'. All treatments were replicated 3 times. The findings showed that the average maize plant height, leaf number, and chlorophyll content were significantly higher for plants applied with SSP and effluent at both half and recommended application rates. There were significant differences ($P < 0.05$) in final maize dry mass and cob + grain size parameters between the different treatments at both application rates. The findings of the study suggest that a combined nutrient management system could be an efficient fertiliser application method for growing maize, supplementing, and reducing the quantities of chemical commercial fertilisers that can be used.

Keywords: Fertilizer combination, Application rate, Variety, Human-excreta derived materials

4.2 Introduction

Maize (*Zea mays* L.) is one of the most important cereal crops in sub-Saharan Africa (SSA) (Edmonds *et al.*, 2009). Major yield losses of maize have been reported in sub-Saharan Africa, mainly due to poor soil fertility (United Nations, 2019). While the use of fertilizers has been shown to result in subsequent increases in crop yields, the high costs associated with these fertilizers pose a major challenge to most small-scale farmers (CSO, 2012). Furthermore, phosphate reserves are limited, and based on global predictions, there are concerns that the reserves can run out and thus pose a threat for future fertilizer production (Bonvin, 2013). Also, the production of nitrogen fertilizers using atmospheric N is very expensive. The continuous losses of the nutrients due to harvesting, soil erosion, and leaching, with limited replenishment, exacerbate the challenge. Under these circumstances, the utilization of varieties tolerant to low soil N is an attractive option (Hirel *et al.*, 2001; Davis, 2013) among other alternatives.

Varieties that are adapted to low input (nutrient-limited) conditions have developed efficient mechanisms through which they can take up important mineral elements (nitrogen and phosphorus) from the soil and utilize them efficiently for dry matter production (Regier *et al.*, 2012). For example, the Improved Maize for African Soils ('IMAS') genotypes have been developed by the International Maize and Wheat Improvement Centre (CYMMIT) to be better at capturing the small amount of fertilizer that African farmers can afford. These varieties use the nitrogen they take up more efficiently to produce grain and are regarded as low nitrogen tolerant varieties (James, 2015). The 'IMAS' has been developed to produce higher yields when compared to other varieties on poor, infertile soils and under drought conditions. These varieties are developed using conventional breeding that offers a 20% yield advantage over varieties that have been commonly used (James, 2015). However, efficiency in nitrogen uptake and nitrogen utilization of these varieties when applied to organic nutrient sources (crop residues and human excreta) are still not well understood. Also, information is still limited on

how efficiently do 'IMAS' varieties respond to the use of HEDMs as nutrient sources compared to an ordinary commercial maize variety such as 'Colorado'.

The use of HEDMs including urine products and treated wastewater could be potential solutions to poverty alleviation in sub-Saharan Africa (Andersson, 2015; Andersson et al., 2016; Nansubuga et al., 2016). This could immensely contribute to the reduction of hunger in low-income communities, and move towards achieving the UN Sustainable Development Goal number two (WWAP, 2017). The use of HEDMs may be essential in addressing the adverse environmental effects of using commercial chemical fertilizers, at the same time providing innovative ways of dealing with the problems of lack of sustainable disposal of waste and wastewater. There is limited information on the use of HEDM such as struvite and the Decentralised wastewater treatment system (DEWATS) effluent for maize production as sources of nitrogen and phosphorus in combination with commercial chemical fertilizers. The results in Chapter 3 showed that a greater amount of mineral N was released from DEWATS effluent treated soil when co-applied with either struvite or SSP than when the effluent was applied alone. This suggested that the co-application of the two HEDM could be of significance as a nutrient source. However, whether the observed N and P release would translate into favourable crop growth and dry matter yield need to be explored.

Therefore, this study aimed to determine the implication of co-application of DEWATS effluent and struvite as sources of nitrogen and phosphorus respectively, on maize growth and yield. The specific objectives of the study were to: (i) Assess the potential of the two excreta products (struvite and DEWATS effluent) as fertiliser sources in combination with other commercial fertilisers (urea and SSP); and to (ii) investigate the effects of co-applying struvite and DEWATS effluent on the growth, nutrient uptake, and biomass production of maize. The

following research question was raised; do waste-based fertilizers perform better when combined together or when combined with commercial chemical fertilizers on the growth, nutrient uptake, and biomass production of maize?

4.3 Materials and method

4.3.1 Experimental set-up

The study was conducted in the Controlled Environment Facilities (CEF), greenhouse (tunnel O) at the Agriculture Campus of the University of Kwa-Zulu Natal (KZN), Pietermaritzburg, South Africa (29° 37'S, 30° 24'E). The sandy soil was collected within 0–0.3 m depth at KwaDinabakubo near Durban, KZN under natural grassland. The collected soil was air-dried and sieved using a 2 mm mesh.

Respective fertiliser (struvite, urea, and single superphosphate (SSP)) were applied to the soil according to rates described in Table 4.1 and mixed thoroughly to allow uniform distribution within the soil. The inorganic fertilisers that were applied were urea (46% N), single superphosphate (10.5% P), and potassium chloride (52% K) based on soil analysis. The potassium (K) was added to all treatments to make sure that it would not be limiting to the plant growth. The recommended application rates were 200 kg N/ha and 60 kg P/ha targeted for 12 t/ha maize yield. These recommended rates translated to the application rates of 2.54 g of struvite per 20 kg soil. Single superphosphate (SSP) and urea were used as the inorganic components of the fertilizer combinations and were applied at rates of 3.05 g and 2.34 g per 20 kg soil respectively.

Table 4.1: Amount of each fertiliser source added (g/pot).

Fertiliser combination	Struvite	Urea	SSP
Control	-	-	-
Effluent + struvite	2.54	-	-
Effluent + SSP	-	-	3.05
Urea + struvite	0.145	2.195	-
Urea + SSP	-	2.195	3.05

The effluent was supplied to meet the N and requirement of maize at the recommended application rate after soil analysis. Where effluent surpassed 100% FC, the discrepancy was added the preceding day

4.3.2. Experimental design and management practices

A pot tunnel experiment was set up as a 9 x 2 factorial experiment in a completely randomised design (CRD) with the following treatments: fertilizer combinations (8 levels- (i) struvite + urea (recommended rates); (ii)) struvite + urea (half recommended), (iii) struvite + effluent (recommended rates); (iv) struvite + effluent (half recommended); (v) SSP + effluent (recommended rates); (vi) SSP + effluent (half recommended); (vii) SSP + urea (recommended rates); (viii) SSP + urea (half recommended) and the control. The second treatment was maize variety with 2 levels –‘ Colorado’ and ‘IMAS’. Each treatment was replicated 3 times to give a total of 54 experimental units of 20 litre pots filled with soil. Three maize seeds were planted per pot and were thinned 3 weeks after planting to one plant per pot. The amount of water applied as irrigation was based on Cartref soil water requirements. An irrigation schedule to satisfy 70 % plant available water (PAW) was calculated to a rate of 1.7 litres per 20 kg soil to be applied throughout the experiment (Equation 1). For the treatments receiving effluent. the effluent was applied as a source of nutrients through irrigation and was based on Cartref soil

water requirements. The total volume of effluent that was added during the experiment was 13.6 litres per pot. Pesticides and herbicides were not used in this experiment, and weeds removal was done by hand.

$$\text{Readily available water content} = \text{PAW} \times \text{SAWDL} \times \text{Soil Mass} \dots \text{Equation 1}$$

Whereby: PAW= plant available water (%), SAWDL = soil available water depletion level (%), and soil mass = soil contained per pot (taken to be 20 kg).



Figure 4.1. The layout of the experimental trial in the tunnel.

4.3.3 Crop growth and nutrient uptake

Crop growth variables collected on a weekly basis were plant height and leaf number. The effects of each treatment on maize plant height and the number of leaves were investigated over a period of 56 Days After Planting (DAP). Plant height was measured from the soil surface to the tip of the uppermost leaf and the number of leaves was also counted on the same plant height measurement. Data on chlorophyll content was collected once and it was at 28 DAP. Chlorophyll content was measured using a CCM200 chlorophyll meter (Opti sciences Inc., USA). To determine tissue nutrient concentration, maize leaf samples were collected at the

tasselling stage by cutting the ear leaf. The samples were oven-dried at 70°C until a constant mass was achieved (that is after 72 hours) and ground using the Wiley mill equipped with 20-, 40- and 60-mesh screens according to methods described by Kalra (1997). Samples were then digested and analysed for macro and micronutrients following methods described by Riekert and Bainbridge (1998) at the Soil Fertility and Analytical Services Division, of the KwaZulu-Natal Department of Agriculture and Rural Development (CEDARA). The plant tissue N concentrations were analysed using the LECO® TruSpec Micro CNS analyser while the other nutrients were determined using inductively coupled plasma optical emission spectroscopy (ICP-OES) Vista MPX after the acid digestion (Riekert and Bainbridge, 1998). At the harvesting stage, cob + grain mass(g), fresh and dry mass(g) were collected. The plants were cut from 1cm above the soil surface and fresh mass was measured directly after harvesting using a 5 kg balance with an accuracy of ± 0.01 g. The plants were then dried at 70°C for 72 hours to determine dry mass.

Plant nutrient uptake calculation:

$N\% \times \text{Dry matter} = N \text{ uptake (mg/pot)}$

$P\% \times \text{Dry matter} = P \text{ uptake (mg/pot)}$

4.3.4 Data analysis

The data was analysed using GenStat® 18th edition (VSN International, 2017). The data were subjected to analysis of variance (ANOVA) and means were separated at a 5% level of significance.

4.4 Results

4.4.1 Crop growth and biomass

4.4.1.1 Plant height

There were significant differences ($P < 0.05$) in plant height among treatments on the two maize varieties. Plant height accumulation increased significantly with time during the period of 56

DAP (Fig 4.2). The fertilizer combination PE 0.5, PU 0.5, SE 0.5, and SU 0.5 differed significantly in plant height among with the recorded means of 0.99 m, 0.91 m, 0.84 m, and 0.83 m respectively (Fig 4.3). Significant differences in height were also observed between the two maize varieties with ‘IMAS’ recording 0.84 m and 0.79 m for ‘Colorado’ (Fig 4.4).

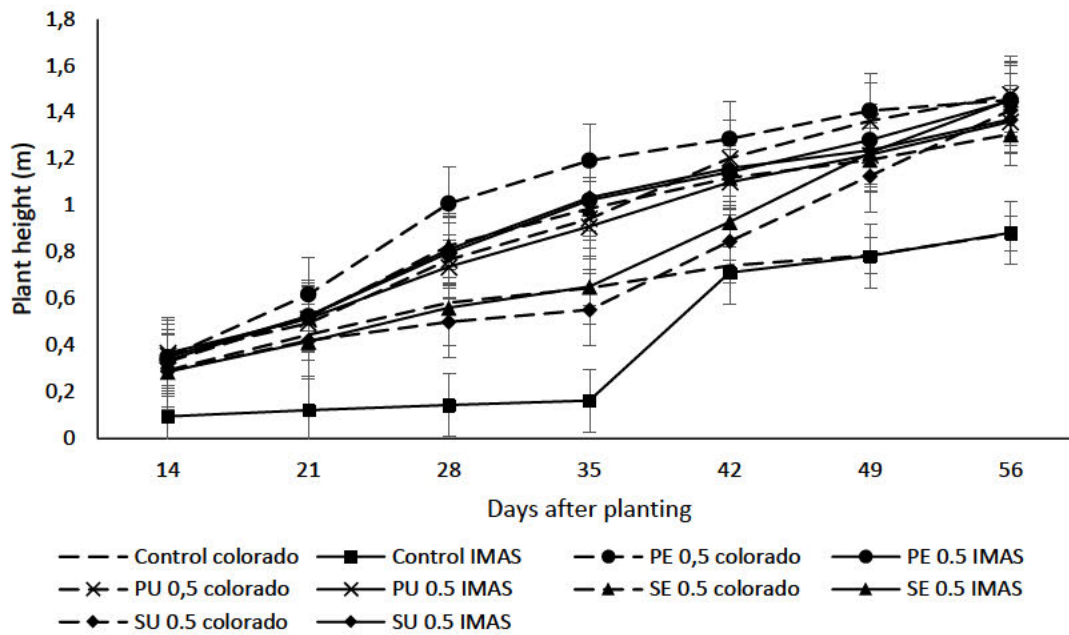


Figure 4.2. Maize cumulative average plant height (m) in response to the different fertilizer combinations applied at half-recommended rates over a period of 56 days after planting. Error bars= standard error of means.

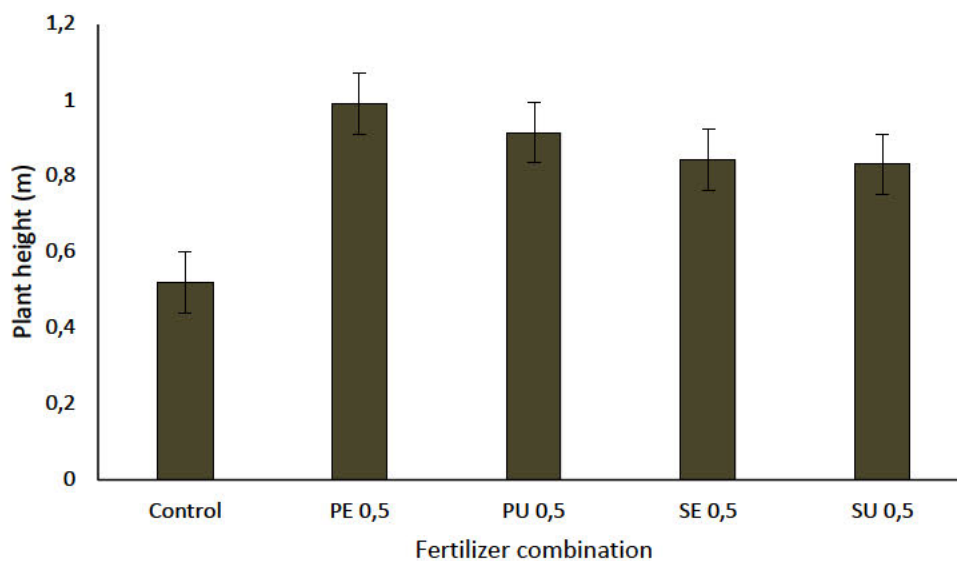


Figure 4.3. Average plant height (m) in response to the different fertilizer combinations applied at half recommended rate 56 days after planting. Error bars= standard error of means.

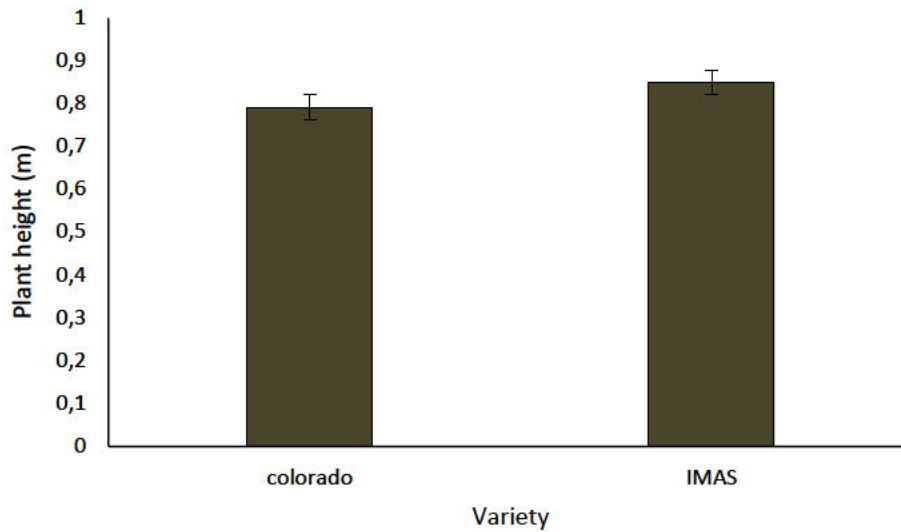


Figure 4.4. Average maize varietal plant height (m) at half recommended rates 56 days after planting. Error bars represent standard error of means.

There were no significant differences ($P>0.05$) in plant height when observing the interaction of different treatments (fertilizer combinations and the two maize varieties) during the period 56 DAP (Figure 4.5). The treatment PE, PU, SE, and SU differed significantly ($p < 0.05$) from each other in plant height with recorded means of 1.04 m, 0.94 m, 0.84 m, and 0.80 m respectively (Fig 4.6). Plant height also differed significantly between the two maize varieties with 'IMAS' recording higher height (0.86 m) compared to 'Colorado' (0.80 m) (Fig 4.7).

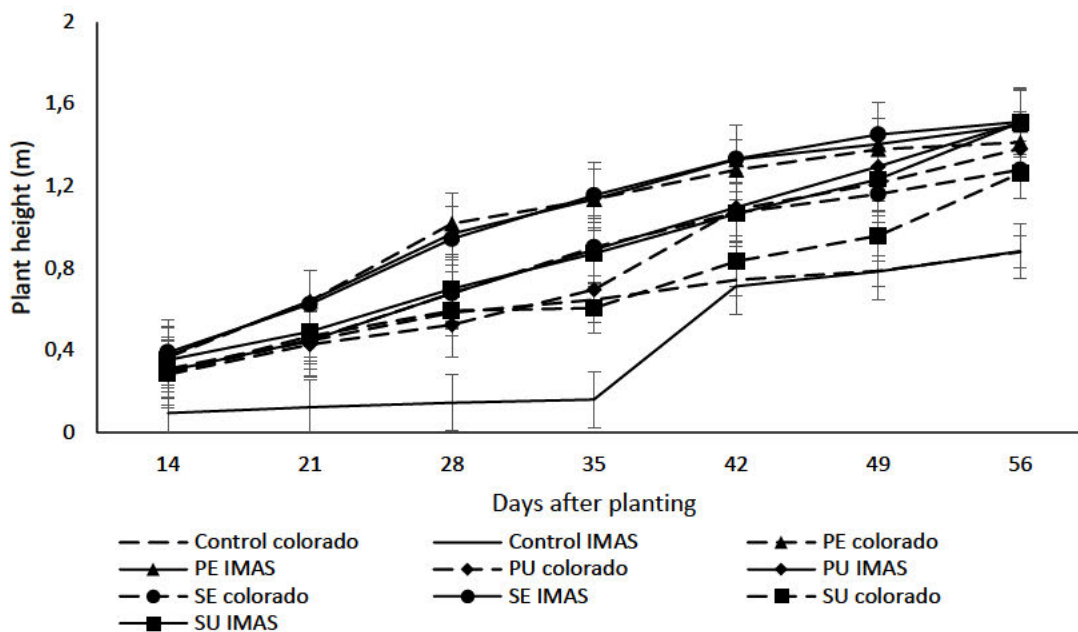


Figure 4.5. Maize cumulative average plant height (m) in response to the different fertilizer combinations applied at recommended rates over a period of 56 days after planting. Error bars represent standard error of means.

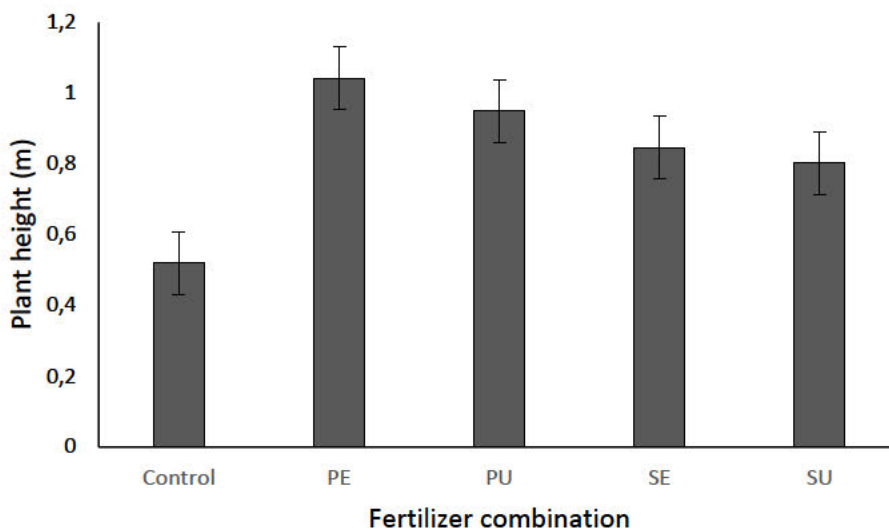


Figure 4.6. The average plant height (m) in response to the different fertilizer combinations applied at recommended rates 56 days after planting. Error bars represent standard error of means.

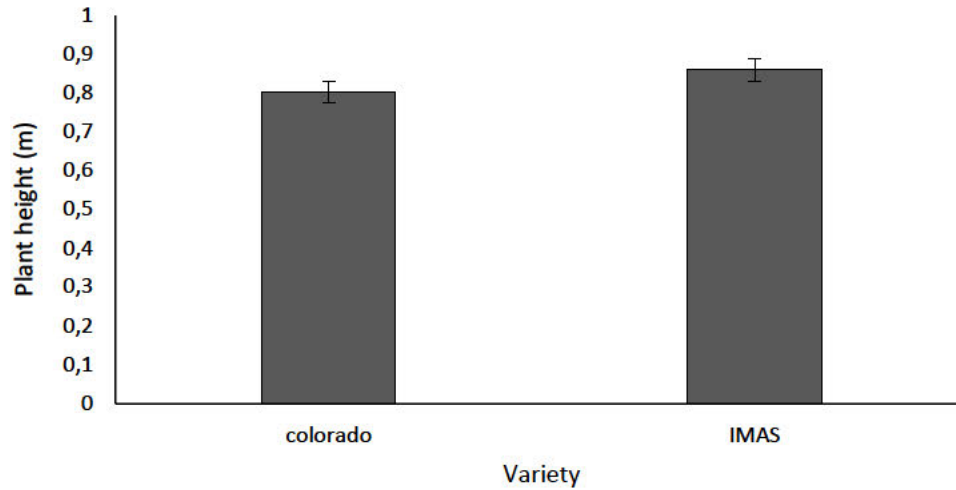


Figure 4.7 Average maize varietal plant height (m) at recommended rates 56 days after planting. Error bars represent standard error of means.

4.4.1.2 Leaf number

Leaf number differed significantly ($P < 0.05$) amongst the treatments 56 DAP. As expected, the number of leaves increased over time for all treatments (Fig 4.8). The fertilizer combinations PE 0.5, PU 0.5, SE 0.5, and SU 0.5 showed significantly different leaf number with the means of 9.66, 9.09, 9.14, and 8.94 leaves respectively (Fig 4.9). The final number of leaves was also significantly different between the two maize varieties ‘IMAS’ (9.28) and ‘Colorado’ (8.75) (Figure 4.10).

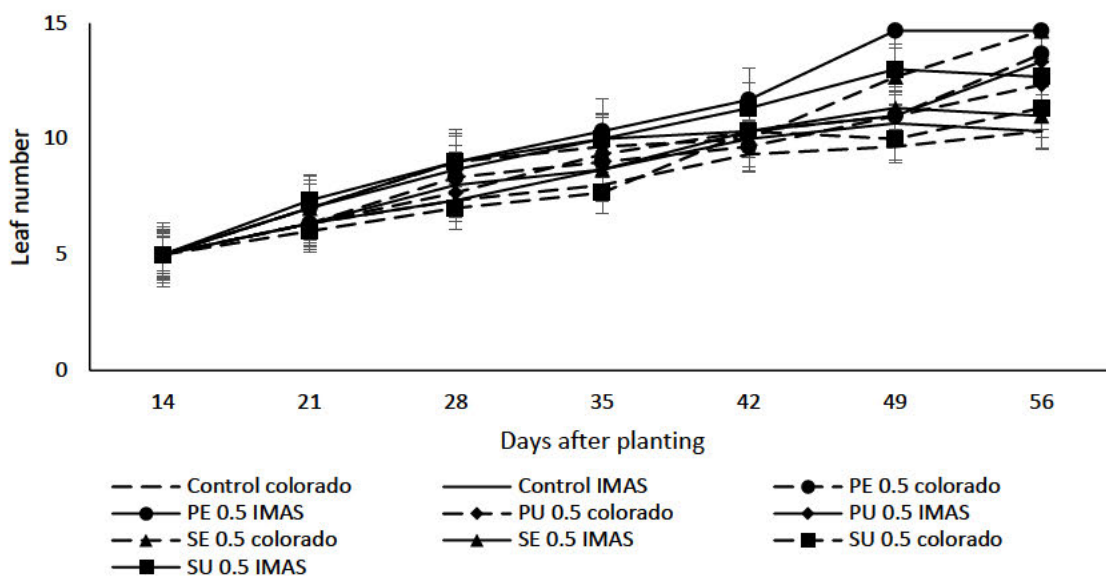


Figure 4.8. Average leaf number of maize varieties in response to the different fertilizer combinations when applied at half recommended application rates over a period of 56 days. Error bars represent standard error of means.

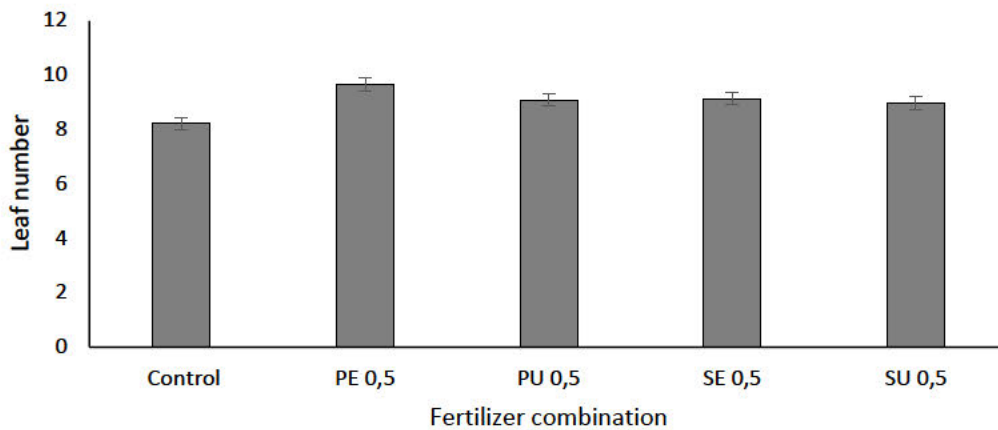


Figure 4.9. Average leaf number in response to the different fertilizer combinations when applied at half recommended application rates 56 days after planting. Error bars represent standard error of means.

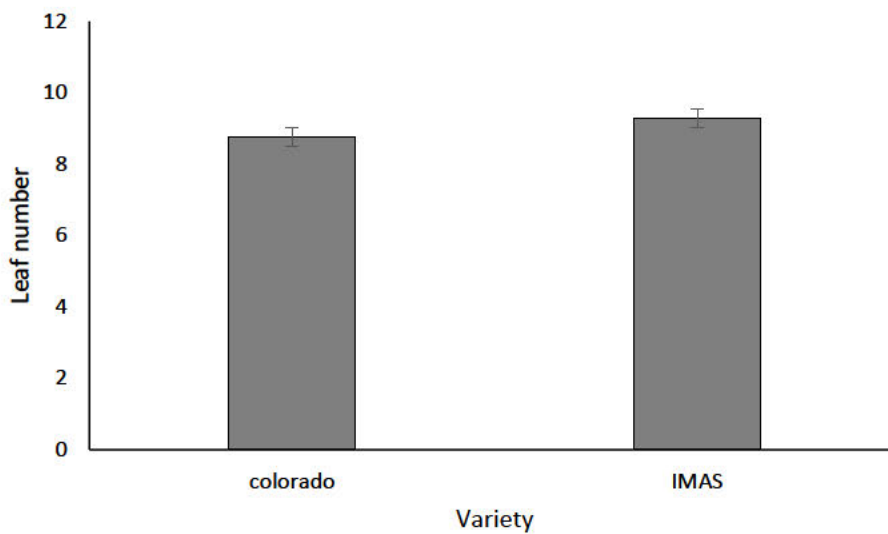


Figure 4.10. Average varietal maize leaf number at half recommended rates 56 days after planting. Error bars represent standard error of means.

The interaction between fertiliser combinations and maize variety did not show significantly different ($P > 0.05$) leaf number accumulation over the study period of 56 DAP (Fig 4.11). However, there were significant differences observed among the fertilizer combinations with

PE, PU, SE, and SU recording average leaf number of 10.19, 8.92, 9.88, and 8.97 respectively (Fig 4.12). The two maize varieties ‘Colorado’ and ‘IMAS’ also showed highly significant different leaf numbers with means of 8.87 and 9.62 respectively (Fig 13).

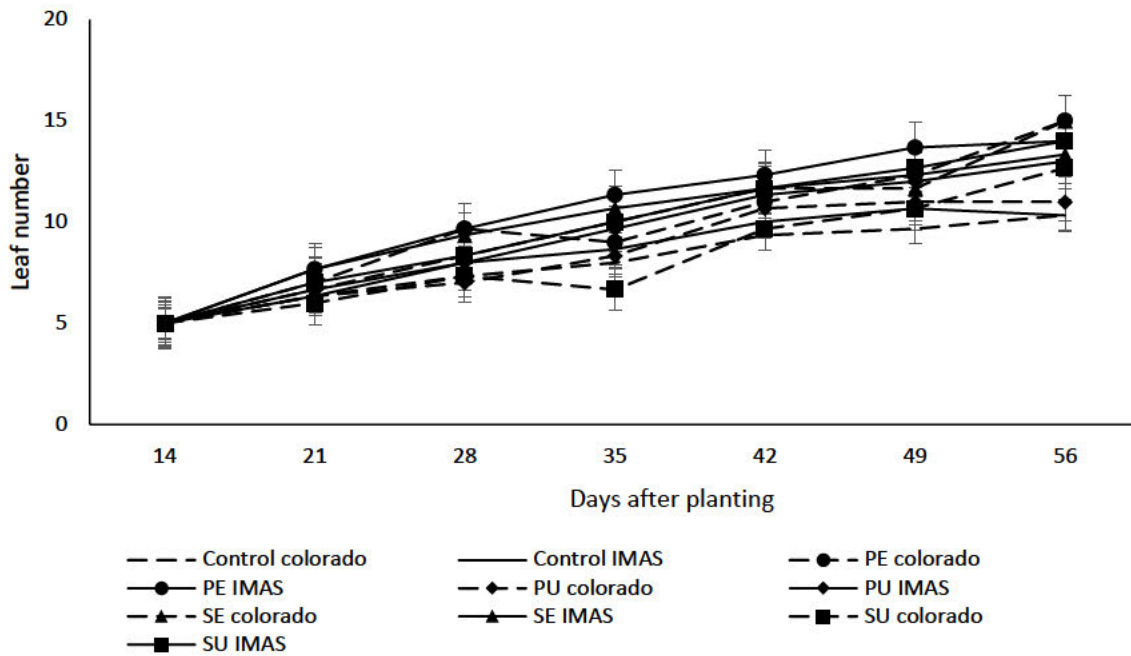


Figure 4.11. Average leaf number of maize varieties in response to the different fertilizer combinations when applied at recommended application rates over a period of 56 days. Error bars represent standard error of means.

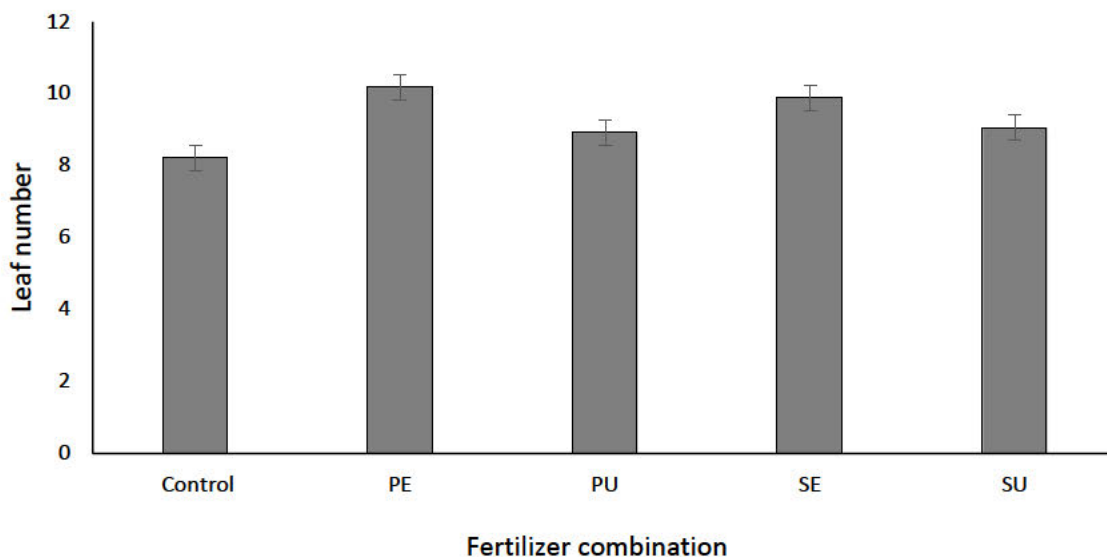


Figure 4.12. Average leaf number in response to the different fertilizer combinations when applied at half recommended application rates 56 days after planting. Error bars represent standard error of means).

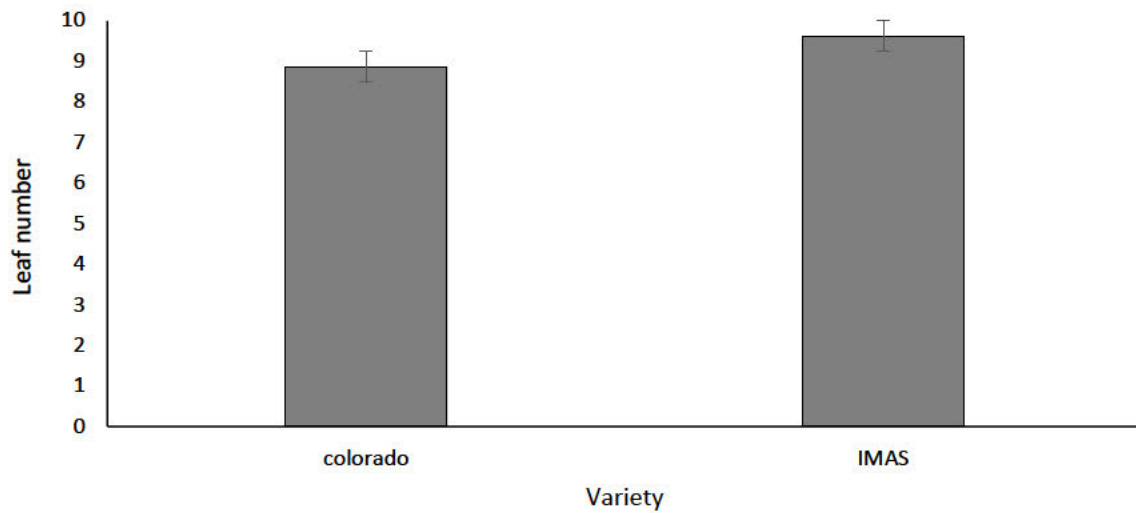


Figure 4.13. Maize average leaf number at half recommended rates, error bars represent standard error of means.

4.4.1.3 Chlorophyll content

The interaction between fertilizer combination and maize variety did not differ significantly ($P > 0.05$) in chlorophyll content. The highest chlorophyll content was recorded in treatment PE with 43.67 followed by SE (41.33), PE 0.5 (41) all in 'IMAS' variety and with the lowest recorded for 'IMAS' observed from the control treatment (35.67) (Fig 4.14). The recorded chlorophyll content for the half-recommended fertilizer combinations ranged from highest to lowest in the order of; PE 0.5 (36.25), SU 0.5 (35.73), PU 0.5 (35.26), and SE 0.5 (34.7), whilst under the recommended fertilizer combinations the chlorophyll content values were PE (39.03), SE (36.15), PU (35.2), SU (34.6) and lowest recorded from the control with 33.27 (Fig 4.15). The two maize varieties also differed significantly ($P < 0.05$) in chlorophyll content. The 'IMAS' variety had on average the high chlorophyll content with 39.78 relative to the 'Colorado' which had 31.38 (Fig 4.16).

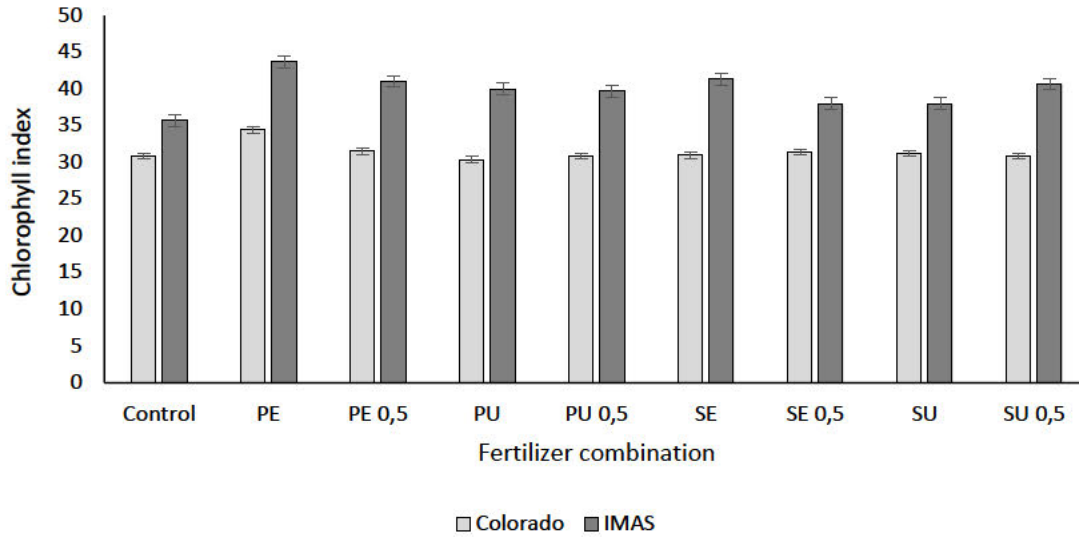


Figure 4.14. Average maize chlorophyll content for different fertilizer combinations at recommended and half recommended rates. error bars represent standard error of means.

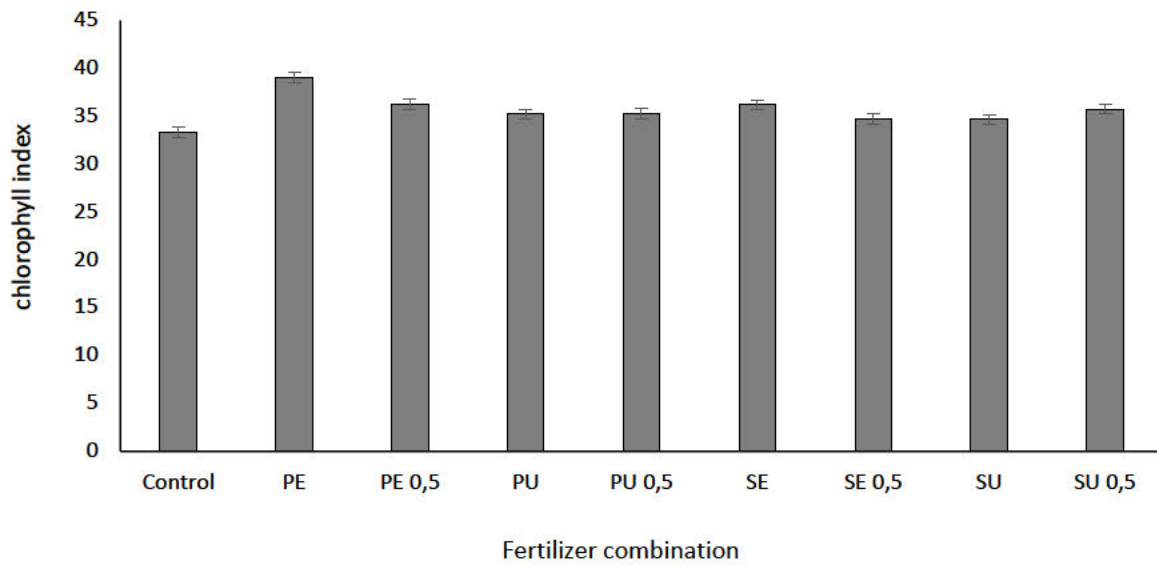


Figure 4.15. Average chlorophyll content for all fertilizer combinations at recommended and half recommended rates 56 days after planting. Error bars represent the standard error of means.

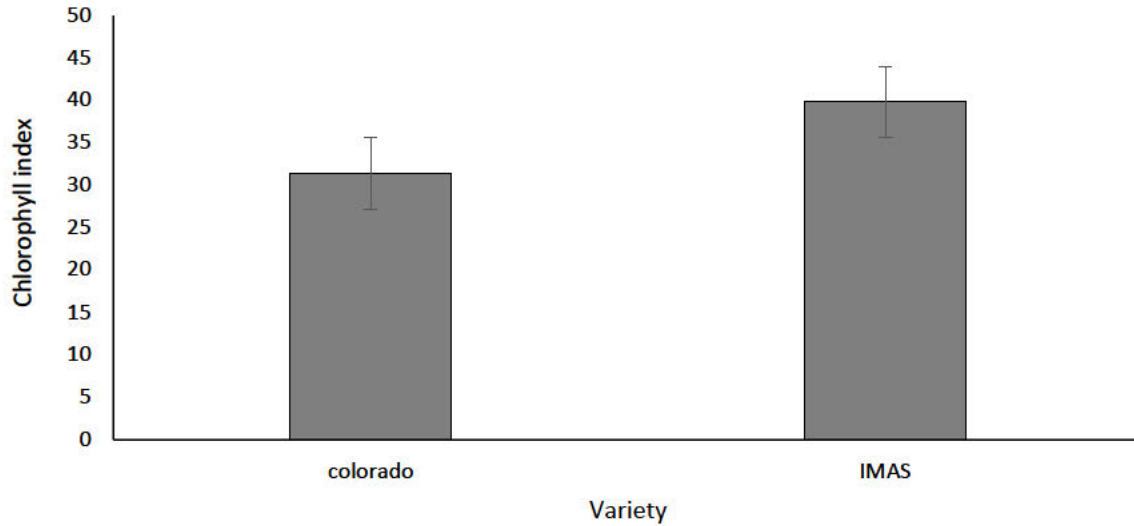


Figure 4.16. Average chlorophyll content for the two maize varieties 56 days after planting. Error bars represent the standard error of means.

4.4.1.4 Final dry matter

The interaction of fertilizer combination and maize variety did not show a significant effect ($P > 0.05$) in final dry matter. However, the different fertilizer combinations had a significant effect on dry matter production. Half recommended (0.5) treatments, SU 0.5 produced greater dry mass with 34.6 g, SE 0.5 (34.5 g), PU 0.5 (23.7 g) and PE 0.5 (18.5 g). The plants that received recommended application rates had a greater dry matter with SE recording 36.7 g which was followed by PU (35.7 g), PE (29.6 g), and SU (25 g). The control treatment accumulated much lower dry matter than all the other treatments and recorded an average of 12.1 g (Fig 4.17). The two maize varieties 'Colorado' and 'IMAS' did not differ significantly ($P > 0.005$) in dry matter production and had means of 27.0 and 28.6 g respectively.

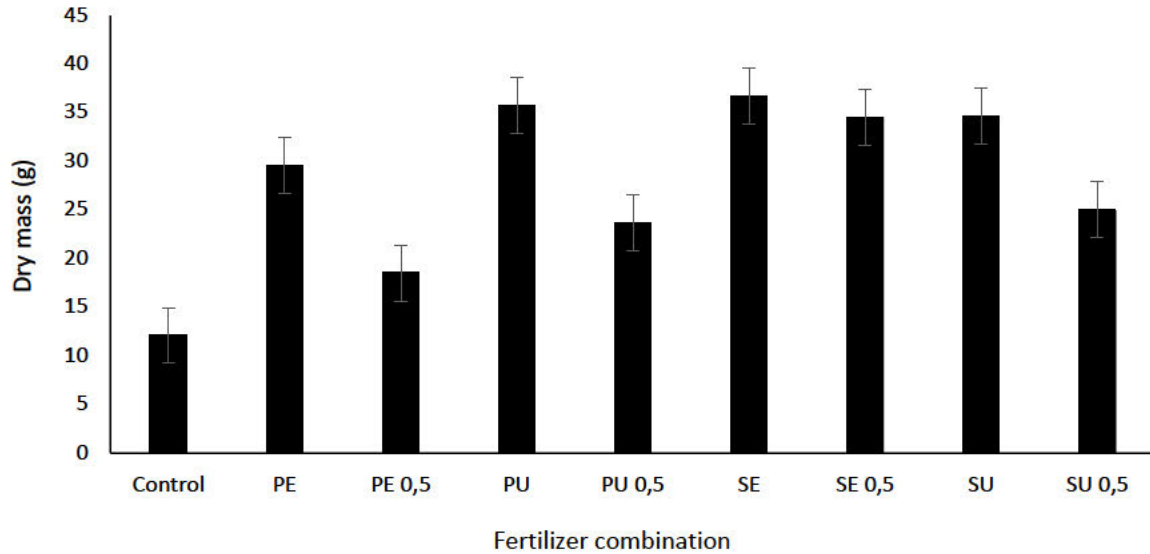


Figure 4.17. Accumulated dry matter production under different fertilizer combinations applied at recommended and half recommended rates. (n= 28; mean \pm Error bars represent standard error of means).

4.4.1.5 Cob mass

The interaction between fertilizer combination and maize variety did not significantly show an effect ($P > 0.05$) in cob mass. However, there were significant differences ($P < 0.05$) among the fertilizer combinations. From the half-recommended treatments, greater average cob size was recorded in the PE 0.5 (56.7 g) followed by SE (42.8 g), SU 0.5 (41 g), and PU (39.5 g). Whilst in the recommended treatments higher average cob sizes were in the SE (71.2 g) and then SU (53.8 g), PE (58.5 g), and PU (62.5 g) (Fig 4.18). All the fertilizer combinations produced significantly larger cobs than the control. The two maize varieties were significantly different ($P < 0.05$) in cob + grain mass. ‘IMAS’ produced cobs of higher mass averaging 61.7 g, while ‘Colorado’ had an average of 36.1 g (Fig 4.19).

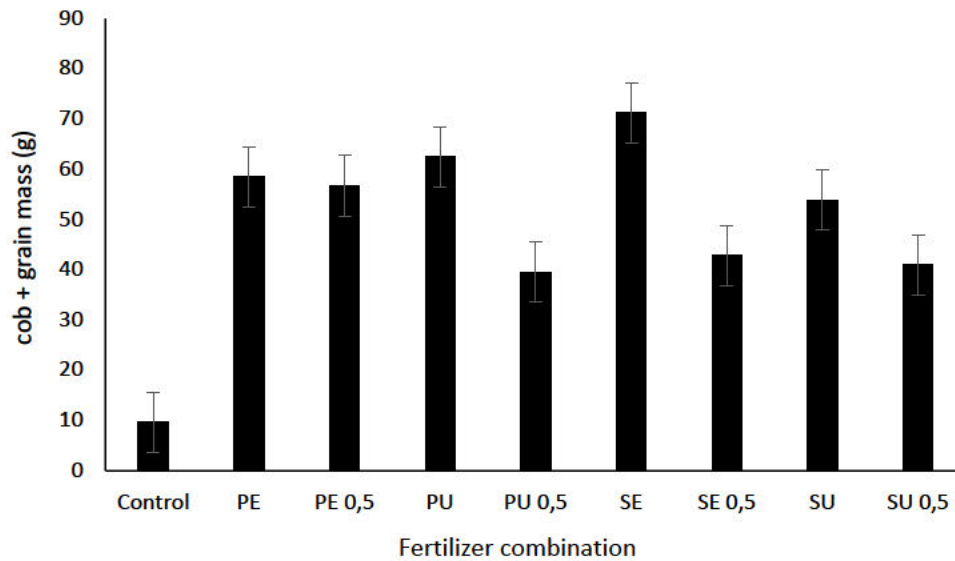


Figure 4.18. The average cob + grain mass per plant from the different fertilizer combinations applied at recommended and half recommended application rates. Error bars represent the standard error of means.

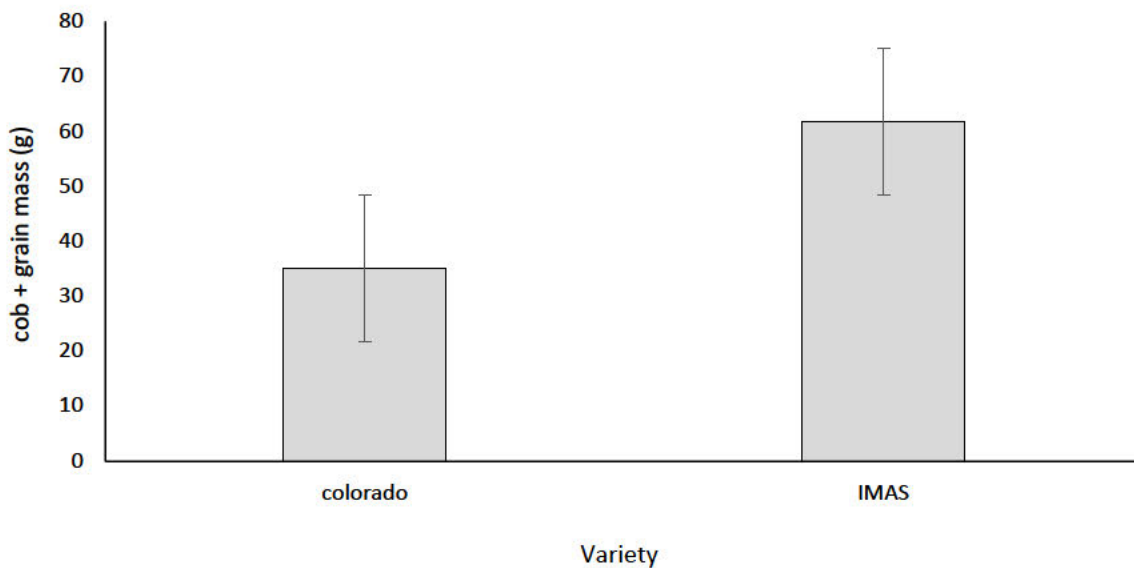


Figure 4.19. The average cob + grain mass of the two maize varieties. Error bars represent the standard error of means.

4.4.1.6 Tissue nutrient concentration of macronutrient N and P in maize.

The concentrations of N and P nutrient uptake were significantly different ($p < 0.05$) among fertilizer combinations of PE, PU, SE, and SU (Table 4.2 and Table 4.3).

Table 4.2. Tissue concentration of macronutrient N and plant nutrient uptake of the different fertilizer combinations of struvite, effluent, and commercial fertilisers urea and SSP on maize.

Treatment combination	N %	N uptake (mg/pot)
control	1,15	13,9
SSP x effluent (PE)	1,69	50,1
SSP x effluent (PE) 0,5	1,98	36,63
SSP x urea (PU)	1,09	38,91
SSP x urea (PU) 0,5	1,33	31,56
struvite x effluent (SE)	2,29	84,05
struvite x effluent (SE) 0,5	1,51	52,14
struvite x urea (SU)	1,94	48,57
struvite x urea (SU) 0,5	1,57	54,37

$p < 0.001$; SED=0.167; cv%=17.9

Table 4.3. Tissue concentration of macronutrient P and plant nutrient uptake of the different fertilizer combinations of struvite, effluent, and commercial fertilisers urea and SSP on maize.

Treatment combination	P %	P uptake (mg/pot)
control	0,1	1,3
SSP x effluent	0,15	6,99
SSP x effluent 0,5	0,15	2,97
SSP x urea	0,11	4,53
SSP x urea 0,5	0,075	0,52
struvite x effluent	0,19	7,02
struvite x effluent 0,5	0,125	2,18
struvite x urea	0,13	3,71
struvite x urea 0,5	0,065	2,22

$p < 0.001$; SED = 0.016; cv%= 29.9

4.5 Discussion

The pot trial assessed the combination of HEDM sources (struvite and effluent) and inorganic fertilisers (urea and SSP) on selected maize parameters. Treatment combinations were applied at recommended and half-recommended (0.5) application rates. This was to compare whether the application of fertilizers at the half-recommended rates can supply essential nutrients for plant growth and how would they compare with treatments that are applied at the recommended rates in sustainable meeting the crop nutrient needs in a more economical way. Nutrient management practices encourage the use of low inputs for maximum crop production as the

prices of conventional fertilizers keep on rising putting a financial burden on small-scale farmers who have already been facing a challenge on fertilizer access (Handmer *et al.*, 2012).

The fertilizer combination PE (0.5) and PU (0.5) produced much taller plants than SE (0.5) and SU (0.5) (Figure 4.3). Similar results were obtained when recommended application rates were applied (Figure 4.6). This could be attributed to the addition of SSP which provided P to the plants in readily available forms for early crop utilization. The high performance of maize plants treated with PE could also be attributed to the fact that there was a constant supply of nutrients in both early and late stages of maize growth, which agreed with several studies using the same type of effluent (Bame *et al.* 2014; Musazura *et al.*, 2019). The tissue concentration of P was much higher for PE and the uptake as well (Table 4.4). The effluent (E) was being applied as irrigation allowing retention in soil rather than once-off which resulted in a continuous supply of ammonium- N for conversion into nitrate- N (Adler *et al.*, 2005). In sandy soil, NH_4^+ - N has a low ability to form electrostatic bonds due to low clay and organic colloids to impair losses of fertilizer N (Medina, 2006). This means that nutrients can be available for plant uptake depending on the root structure of the crop.

Fertilizer combination containing struvite (S) produced shorter plants than the other treatments. Under this treatment, the maize plants were grown however, they did not reach physiological maturity. This could be associated with the fact that struvite is a slow-release fertilizer source for P which potentially resulted in the plant growth delay. The control treatments accumulated shorter growth throughout the growing period.

The fertilizer combination PE (0.5) and SE (0.5) produced plants that had a higher number of leaves than PU (0.5) and SU (0.5) (Figure 4.9). Similar results were obtained when recommended application rates were used (Figure 4.12). PE and SE combinations contained the DEWATS AF effluent which is rich a source of nitrogen and high organic matter. The

nature of the N in the effluent is in the form of Ammonium-N which mineralizes faster as it is in liquid form to be taken up by the plants (Gutterer *et al.*, 2009). The constant supply of the effluent then allowed for increased leaf growth of the plants. The results from plant height and number of leaves best explain the higher chlorophyll content of the plants treated with effluent which is a result of the amount of N and P that was taken up by the maize plants (Table 4.2; Table 4.3). Bame *et al.* 2014 reported similar results, that the above-ground nutrient uptake of maize was significantly higher ($p < 0.05$) in the effluent-irrigated pots than in the equivalent treatment for water-irrigated pots.

Final maize dry mass was highest in the fertilizer combination struvite + effluent (SE), SSP + urea (PU), struvite + urea (SU) than SSP + effluent (SE) at both recommended and half recommended application rates (Figure 4.17). Magwaza *et al.* 2020 observed similar results for plant fresh and dry biomass in tomatoes which was significantly higher in plants grown in wastewater supplemented with chemical fertilizer than when the ABR effluent was solely applied. In terms of nutrient uptake, these treatments (SU and SE) made more N and P available to the plants resulting in greater biomass accumulation. These findings agree with González-Ponce *et al.* (2009) who reported that the response of lettuce head fresh weight and P uptake exhibited statistically significant quadratic relationships for both single superphosphate and struvite. The study found that struvite was more effective than single superphosphate in increasing lettuce yield and P uptake. Uysal and Kuru (2013), Prabhu and Mutnuri (2014), and Uysal *et al.* (2014) also reported that struvite fertiliser is a better than commercial fertilizers.

Phosphorus in plant cells is important during the process of photosynthesis, converting solar energy to increased crop growth and higher grain production. Struvite contains 12.6 % P while SSP contains 14.5 % P which may have contributed to the greater accumulated biomass at struvite-treated plants. Uysal *et al.* (2010) have suggested that struvite's effectiveness as a P fertilizer is favoured in soils with acidic pH. The soil used for the study was a sandy soil with

pH 4.21 and the slow release of nutrients by struvite may have been more available at the later stages of plant growth. This agrees with results obtained from cob + grain size where larger cobs were from plants treated with SE and SU (Figure 4.18). The high P uptake was observed in these treatments (Table 4.3). The high dry matter among PE, PU, SE, and SU treatments compared to zero fertiliser treatment was attributed to the nutrient characterisation of the sources, there are more nutrients in fertilisers sources than in pure water. The high average final dry matter and cob + grain size for all treatments was when the application was at recommended rates for optimum maize production.

The overall nutrient uptake of N and P observations from plant tissue analysis from the different treatments show that the maize plants did not take many nutrients from the soil, this is when compared to the initial input. This agrees with Fonseca *et al.* (2005a) who reported that the use of secondary-treated sewage effluent on adequately fertilized maize plants did not increase plant N uptake. These may be a result of nutrient losses through processes such as leaching as well as volatilisation of N and non-availability of P (Bame *et al.*, 2013, 2014). When comparing the two varieties of maize under PE, PU, SE, and SU treatments, both varieties performed well in terms of overall growth and biomass accumulation. However, the ‘IMAS’ variety performed better than the ‘Colorado’. This means that both varieties can be used by farmers and would give greater yields, especially under unfavourable soil conditions.

4.6 Conclusion

The study aimed at assessing the efficiency of HEMD (struvite and DEWATS effluent) on maize growth and biomass production. Plant height, leaf number, chlorophyll content, dry matter, N and P uptake, and grain + cob yields were higher when DEWATS effluent was applied, particularly when combined with struvite and SSP. The major finding of the study is that the HEDM plant nutrient sources (struvite and effluent) can be used as alternative nutrient sources for growing maize, supplementing, and reducing the quantities of chemical commercial

fertiliser used. Agriculture may not have to rely solely on struvite as a source of P and effluent as a source of N for crop production. Therefore, a combined nutrient management system could be an efficient fertiliser application method for better maize yield. However, to reliably make informed decisions, these findings need to be tested under field conditions where climatic conditions always fluctuate during the season.

CHAPTER FIVE: GENERAL DISCUSSION, CONCLUSION, AND RECOMMENDATION

5.1 General discussion

The increase in population has resulted in a greater need to increase food production which has resulted in greater use of fertilizer materials in agriculture. This has also caused the prices of fertilizers to rise and becoming more difficult to access especially for smallholder farmers. The need to find alternative sources of nutrients has resulted in research in source separation, and in some cases, processing of human urine and faeces. The main objective of this study was to determine the availability of N and P in soils treated with struvite, effluent, and in combination with commercial fertilizers (urea and SSP) and their effects on the growth and biomass production of maize.

The higher accumulation in plant height, leaf number, and chlorophyll content in the soils treated with combinations SSP x effluent, struvite x effluent, struvite x urea, and SSP x urea, than the control, suggested that these HEDMs (struvite and effluent) can be used as fertilizer materials to supply nitrogen and phosphorus. This was supported by the results of the incubation studies which showed that combining SSP x effluent and struvite x urea resulted in higher ammonium N and nitrate N. The higher ammonium N and nitrate N of the effluent during the early and late stages of incubation suggested that this treatment would make more N available for plant growth. (Bame et al., 2013) conducted studies on the behaviour of DEWATS effluent in different soils of Kwazulu-Natal and the findings from their pot's experiments showed that the addition of DEWATS effluent increased vegetative growth (plant height, dry mass, and leaf area) of maize and banana plants.

Urea releases more N as it quickly breaks down into ammonia which is converted into nitrates for plant uptake, which supports results from the incubation study where urea alone and struvite x urea released high ammonium N and nitrates N. Struvite contains about 5.7% N which is in

an inorganic form that is not readily available to plants but needs to solubilise first and then released slowly to the plant. This could have also contributed to the higher nutrient released by soil and taken up plants as observed on the plant tissue concentration. This is in agreement with Richard *et al.* (2001) who highlighted that the ammonium N contained in struvite could be used as available to plants like that in any ammonium N fertilizer.

The combination of struvite x effluent and SSP x urea resulted in higher extractable P than the other treatments and the control during the incubation. This agrees with the amount of P that was taken up by the plants which resulted in greater uptake, cob + grain mass, and dry matter production of maize in those treatments. The results of grain +cob weight followed the same trend as those of dry matter yield, where treatments with effluent, especially when combined with struvite, had higher than the other fertilizer combinations. This suggests that greater dry matter resulted in higher grain + cob, all as a result of higher N and P uptake, particularly from the HEDM products. These findings agree with Tanski *et al.* (2005) who reported that cucumber fertilised with urine collected from households had better yields than those in rows fertilized with commercial fertilizer. Furthermore, Guzha *et al.* (2005) also reported that growing maize with the help of toilet compost and urine on poor sandy soils was beneficial in low-income areas.

The benefit of alternative waste management of urine and DEWATS effluent is by reducing pollution/eutrophication of surface water while getting better yields than when chemical fertilisers were used, especially in combination for a better balance of nutrients. The application of fertilisers at recommended and half recommended rates produced similar results in terms of plant height, leaf number, and chlorophyll content. This implies that combining fertilizer sources even at half the recommendation rate proved to be a potential option and this could reduce fertilizer costs with favourable crop growth. However, the recommended application rates were better performing in terms of cob + grain size and final dry matter.

5.2 Conclusion and Recommendations

The current work has shown that optimising N and P supply through a combination of AF effluent and struvite or with inorganic fertilisers could potentially be considered as a better option for providing a more balanced supply of nutrients than when applied separately. There was better performance observed in plant height, leaf number, chlorophyll content, dry matter, N and P uptake and grain + cob yields of the maize plants the fertiliser combinations containing the effluent the at both recommended and half recommended application rates. The soil incubation experiment showed that there were significant ($P < 0.05$) differences observed among the different fertilizer combinations and zero fertilizer. The combination of HEDMs and commercial nutrient sources released more ammonium N and nitrate N than sole applications and when commercial SSP + urea was applied together. Ammonium N declined over time and nitrate N increased rapidly over time. The findings suggested that the fertiliser combination of HEDMs and commercial fertiliser increased nutrient N availability to the soil. Extractable P release did not change over time in all treatments and also the uptake was low in the plant tissue. Hence further incubation studies need to be done on the effect of these nutrient combinations on phosphorus sorption and nutrient release in varying soil types. Also, the choice of which fertilizers can be combined together for optimum nutrient availability in the soil and plant growth needs to be investigated. For farmers who may use this method, it would be recommended they apply the fertilizers at the recommended rates for optimum growth for that specific crop. Moreover, the basis for determining the accurate amount of supplementary fertilizer to be added to the effluent still needs to be established. It is also recommended that future studies must be conducted to understand fully the effect of the fertilizer combinations through field experiments with a variety of crops. The effect of these combinations on soil physical properties and nutrient dynamics needs further investigation to understand fully the potential, limitations, and any possible drawbacks from using such methods.

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Appendices

Incubation study

Analysis of variance

Variate: Ammonium_mg_kg

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	48.40	24.20	1.06	
Rep.*Units* stratum					
Treatment	8	12568.95	1571.12	69.02	<.001
Day	10	32031.72	3203.17	140.71	<.001
Treatment.Day	80	12058.68	150.73	6.62	<.001
Residual	196	4461.87	22.76		
Total	296	61169.62			

Analysis of variance

Variate: Nitrate_mg_kg

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	6.91	3.46	0.25	
Rep.*Units* stratum					
Treatment	8	8999.63	1124.95	80.21	<.001
Day	10	42187.30	4218.73	300.79	<.001
Treatment.Day	80	10453.92	130.67	9.32	<.001
Residual	196	2749.04	14.03		
Total	296	64396.80			

Analysis of variance

Variate: P_mg_kg

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	7.496	3.748	1.33	
Rep.*Units* stratum					
Treatment	8	243.465	30.433	10.79	<.001
Day	10	147.363	14.736	5.23	<.001
Treatment.Day	80	354.556	4.432	1.57	0.006
Residual	196	552.645	2.820		
Total	296	1305.524			

Analysis of variance

Variate: pH

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Rep stratum	2	0.06767	0.03384	2.16	
Rep.*Units* stratum					
Treatment	8	1.89154	0.23644	15.10	<.001
Day	10	10.43202	1.04320	66.63	<.001
Treatment.Day	80	6.36332	0.07954	5.08	<.001
Residual	196	3.06873	0.01566		
Total	296	21.82328			

Tunnel study

Plant height

Half recommended

Analysis of variance

Variate: Plant_height_m

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Combination	4	5.41125	1.35281	122.95	<.001
Variety	1	0.18231	0.18231	16.57	<.001
Time_DAP	6	24.26587	4.04431	367.58	<.001
Combination.Variety	4	0.95268	0.23817	21.65	<.001
Combination.Time_DAP	24	0.87325	0.03639	3.31	<.001
Variety.Time_DAP	6	0.09831	0.01638	1.49	0.186
Combination.Variety.Time_DAP	24	0.93785	0.03908	3.55	<.001
Residual	140	1.54035	0.01100		
Total	209	34.26188			

Recommended
Analysis of variance

Variate: Plant_height_m

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Combination	4	6.52574	1.63143	97.40	<.001
Variety	1	0.17157	0.17157	10.24	0.002
Time_DAP	6	24.58661	4.09777	244.64	<.001
Combination.Variety	4	1.20821	0.30205	18.03	<.001
Combination.Time_DAP	24	1.28384	0.05349	3.19	<.001
Variety.Time_DAP	6	0.20768	0.03461	2.07	0.061
Combination.Variety.Time_DAP	24	0.39725	0.01655	0.99	0.486
Residual	140	2.34502	0.01675		
Total	209	36.72591			

Leaf number
Half recommended

Analysis of variance

Variate: Leaf_number

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Combination	4	45.7810	11.4452	17.05	<.001
Variety	1	14.9333	14.9333	22.24	<.001
Time_DAP	6	1270.2571	211.7095	315.31	<.001
Combination.Variety	4	47.6857	11.9214	17.76	<.001
Combination.Time_DAP	24	42.5524	1.7730	2.64	<.001
Variety.Time_DAP	6	10.0667	1.6778	2.50	0.025
Combination.Variety.Time_DAP	24	30.6476	1.2770	1.90	0.011
Residual	140	94.0000	0.6714		
Total	209	1555.9238			

Recommended
Analysis of variance

Variate: Leaf_number

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Combination	4	104.9810	26.2452	35.11	<.001
Variety	1	29.7190	29.7190	39.75	<.001
Time_DAP	6	1406.5905	234.4317	313.57	<.001
Combination.Variety	4	10.4476	2.6119	3.49	0.009
Combination.Time_DAP	24	54.8857	2.2869	3.06	<.001
Variety.Time_DAP	6	15.3143	2.5524	3.41	0.004
Combination.Variety.Time_DAP	24	19.0190	0.7925	1.06	0.397
Residual	140	104.6667	0.7476		
Total	209	1745.6238			

Chlorophyll index

Analysis of variance

Variate: Chlorophyll

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Combination	8	119.979	14.997	3.43	0.005
Variety	1	952.308	952.308	217.94	<.001
Combination.Variety	8	42.766	5.346	1.22	0.314
Residual	36	157.305	4.370		
Total	53	1272.357			

Final dry matter

Analysis of variance

Variate: Final_drymatter_g

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Combination	8	3574.5	446.8	3.66	0.003
Variety	1	35.2	35.2	0.29	0.594
Combination.Variety	8	877.7	109.7	0.90	0.527
Residual	36	4390.4	122.0		
Total	53	8877.8			

Cob + grain mass

Analysis of variance

Variate: cob_grain_mass_g

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Combination	8	15493.4	1936.7	5.14	<.001
Variety	1	9546.7	9546.7	25.32	<.001
Combination.Variety	8	4936.9	617.1	1.64	0.149
Residual	36	13576.0	377.1		
Total	53	43553.0			

N uptake

Analysis of variance

Variate: N_uptake_mg_pot

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Combination	8	18553.7	2319.2	6.47	<.001
Variety	1	0.5	0.5	0.00	0.972
Combination.Variety	8	666.7	83.3	0.23	0.982
Residual	36	12896.5	358.2		
Total	53	32117.3			

P uptake

Analysis of variance

Variate: P_uptake_mg_pot

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Combination	8	126.274	15.784	6.86	<.001
Variety	1	6.182	6.182	2.69	0.110
Combination.Variety	8	3.616	0.452	0.20	0.990
Residual	36	82.875	2.302		
Total	53	218.946			

ANOVA table for treatment analysis

Sources of variance	Degree of freedom (n-1)
Factor A- fertilizer combination	8
Factor B- variety	1
Fertilizer combination \times variety	8
Residual	36
Total	54