VALIDATION OF SLUDGE APPLICATION RATE ADVISOR (SARA) MODEL ASSUMPTIONS AND SIMULATION RESULTS THROUGH FIELD STUDIES ACROSS AGROECOLOGICAL ZONES

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Report to the
WATER RESEARCH COMMISSION

by

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EXECUTIVE SUMMARY

Two separate but complementary studies (field mineralization and lysimeter studies) were conducted at different areas. The field sludge nitrogen mineralisation studies were conducted at selected representative sites across five of the six South African agro-ecological zones. These study sites include: Arid (Upington), semi-arid (Rustenburg), sub-humid (Johannesburg and Bethlehem), humid (Durban), and super-humid (Nelspruit). The lysimeter study was conducted at the Experimental farm of the University of Pretoria to quantify maize grain yield and nitrate leaching from sludge applied according to the SARA model recommendations.

This study aimed to validate the SARA model through field mineralisation studies and controlled field lysimeter studies. The study objectives were as follows: 1) to validated the nitrogen mineralization rates used by SARA model to predict sludge recommendations using field studies from across South African agro-ecological zones, and 2) to quantify maize grain yield and nitrate leaching from sludge applied, according to the SARA model recommendations, using commercial inorganic fertiliser as benchmark under contrasting rainfall regimes (moderate and high).

To achieve the above objectives, field mineralisation studies were conducted in five of the six South African agroecological zones with lysimeter studies conducted at University of Pretoria's experimental farm. The field incubation study was conducted from October 2017 to June 2018 (first sampling season) and 2017 to 2020 (second sampling) on six sites representing five South African agroecological zones. Porous ceramic cups filled with a soil-sludge mix (treatment) and soil alone (control) were used for the study. The cups were buried 15 cm deep below the soil surface. Net nitrogen mineralisation from the cups was estimated as the difference between initial and final organic N applied. The lysimeter study was conducted from November 2018 to April 2020.

The net N mineralization rate during the first year of field incubation study across agro-ecological zones followed similar trends as the model predictions but the values from the field studies were generally higher than those from the model predictions, except for the arid zone. The net N-mineralization model simulation results at the end of the third year of study also followed similar trends but slightly lower than the field mineralization studies. This was mainly due to the abnormally high rainfall experienced especially in the sub-humid areas of Johannesburg. Net N mineralization rate varied significantly among agro-ecological zones. Net N mineralization rate for the first year of field incubation was 10% (arid), 31% (semi-arid), 34% (sub-humid – Bethlehem), 46% (sub-humid-Johannesburg), 48% (humid), and 57% (super-humid). At the end of the third year of the field incubation study across agroecological zones, three-year cumulative net N mineralization was 42±7.8% (arid), 73±4.5% (sub-humid), 73±2.5% (humid), and 69±9.1% (super humid).

Results from lysimeter studies indicate that maize grain yield and cumulative nitrate leaching varied across treatments. Maize grain yield from sludge treated land was similar with that of inorganic fertiliser both under the higher rainfall (simulated sub-humid) and lower rainfall

(simulated humid) zones. The only exception was the second year under the subhumid zone, during which the rainfall was lower, yield was lower from the sludge treated land. Nitrate leaching was higher under the humid compared with the subhumid zone during both growing seasons. The difference was, however, only significant ($P<0.05$) during the second growing season. It was also apparent that nitrate leaching was higher from inorganic fertilizer treatments than sludge treatments in the sub-humid zone and from sludge treatments than inorganic fertilizer in the humid zone for both years. The difference was, however, significant ($P<0.05$) only during the second season.

Sludge applied according to crop N demand, estimated by the SARA model, produced comparable maize grain yield with commercial inorganic fertiliser without a considerable influence on nitrate leaching. Therefore, SARA model could be used as Decision support system for sludge application across South African agro-ecological zones.

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CHAPTER 1: INTRODUCTION

Food insecurity is a fundamental challenge of the century in sub-Saharan African countries. These countries remain on the top globally with people living in hunger and starvation (Clover, 2003). Thirty-three per cent of people living in Africa are malnourished (Kidane et al., 2006). The foremost food insecurity causes are the degradation of agricultural lands, a rapid rise in human population, and the prevailing climate variability (Hendrix and Glaser, 2007; Vlek et al., 2010). Land degradation is predominantly caused by continuous cultivation with low remediation, using low organic matter soil amendments and land-use change practices (Mwangi, 1997; Sheffield et al., 2014). A need exists to restore the degraded lands of the sub-Saharan African countries, including South Africa, supporting food security.

Sludge-use in agricultural land improves soil organic matter, supply crop nutrients and, therefore, rehabilitate degraded agricultural lands (Bravo-Martín-Consuegra et al., 2016; Antonelli et al., 2018; Hamdi et al., 2019). Rehabilitating degraded agricultural lands could be significant in improving land productivity, subsequently crucial in poverty alleviation (Mohamed et al., 2018). Sludge recycling in agricultural lands aligns to the United Nations Sustainable Development Goals (UNSDGs), specifically to Sustainable Developmental Goals (SDG) 12: "Responsible consumption and production, Target 12.5, promote the substantial reduction of waste generation through prevention, reduction, recycling and reuse" (UN, 2015). Other SDGs aligned to sludge-use in agricultural lands include:

- SDG 1 (poverty alleviation)
- SDG 2 (zero hunger)
- SDG 6 (clean water and sanitation)
- SDG 13 (climate action) and SDG 15 (life on land)

Sludge, with acceptable quality for agricultural use, is applied according to crop N requirements (Barbarick et al., 2017). A large N fraction in sewage sludge is realised in organic form. Crops, however, use N through NH_4 and $N0_3$. It is, therefore, essential that the organic N is mineralised before plants can benefit. The mineralisation of organic N is influenced to a substantial extent by the availability of water and soil temperature (Ogbazghi et al., 2016; Tesfamariam et al., 2015). Direct extrapolation of studies conducted under a specific climatic condition, sourcing a specific sludge type to other climatic zones and sludge types can compromise both the environment and crop yield. To discuss this challenge, the

WATER RESEARCH COMMISSION of South Africa funded the following three consecutive projects:

- K5/1724/3 (2007/08 to 2010/11 financial year)
- K5/2131 (2011/12 2014/15 financial year)
- K5/2477//3 (2015/16 to 2017/18 financial year)

This study followed on these three consecutive projects. Ensuing these projects, an addendum to the South African sludge guidelines – Volume 2, was developed. To help implement the addendum to the guidelines, a database computer model, scanning, analysis, response, and assessment (SARA) was developed. Both the addendum and the database model were developed based on computer model scenario simulations. The simulations were conducted using the SWB-Sci model, parametrised through controlled laboratory incubation studies. These were calibrated and validated exercising field experiments from one agroecological zone – Johannesburg area (sub-humid). The addendum to the sludge guidelines (Tesfamariam *et al*., 2018) emphasises the urgency for field validation of the recommendations, based on model scenario simulations, conducting field mineralisation studies across South African agroecological zones.

Excess nutrients are detrimental to plant growth (Brady, 2013) and polluted water bodies. As water moves over the soil surface, it transfers decomposing organic matter, fertilisers, pesticides, and sediments. The nutrient loads of such runoff waters cause nutrient enrichment of water bodies, such as lakes and dams. This nutrient enrichment causes the undesirable proliferation of aquatic plants (eutrophication). Nitrogen in water bodies is oxidised into nitrite and nitrate, depleting levels of dissolved oxygen. This causes the death of fish and other aquatic creatures (Cameron & Haynes, 1986). Water percolating beyond the root zone may also transmit nutrients in a dissolved form, contaminating groundwater.

Contaminated water presents a hazard for humans, animals, and plants. A consequence of groundwater nitrate pollution is methemoglobinemia or blue baby syndrome, which can cause death in infants (Croll & Hayes, 1988). It is of extreme importance for South Africa to protect its scarce groundwater resources from such contamination caused by agricultural activities. Sludge application influence, according to crop nutrient requirement on nitrate leaching below the crop root zone, should, therefore, be investigated. The information from such studies will provide the sustainability of sludge-use in agricultural lands concerning its effect on groundwater contamination.

The study comprises sustainable development solutions as the desired outcome. This indicates validating the addendum to the South African sludge guidelines through field experiments across South African agroecological zones. This facilitates sustainable sludge-use in agricultural lands by enhancing crop production while minimising environmental effects through nitrate leaching to groundwater and surface water bodies.

This study aimed to validate the SARA model through field mineralisation studies and controlled field lysimeter studies. The study objectives were as follows:

1) to validated the nitrogen mineralization rates used by SARA model to predict sludge recommendations using field studies from across South African agro-ecological zones

2) to quantify maize grain yield and nitrate leaching from sludge applied, according to the SARA model recommendations, using commercial inorganic fertiliser as benchmark under contrasting rainfall regimes (moderate and high).

Field mineralisation studies across agroecological zones and lysimeter studies were conducted to achieve the objectives. To achieve the above objectives, field mineralisation studies were conducted in five of the six South African agroecological zones with lysimeter studies conducted at University of Pretoria's experimental farm. A field incubation study was conducted from October 2017 to June 2018 (first sampling season) and 2017 to 2020 (second sampling) on six sites, representing five South African agroecological zones.

CHAPTER 2: LITERATURE REVIEW

2.1 General introduction

Countries strive to fulfil their mandates to achieve the UN-adopted SDGs. This ambitious agenda hopes to discuss 17 core areas of public concern. Fifteen of these (goals four and 17 excluded) relate directly or indirectly to waste recycling, with predominant conceptual emphasis on food security, health, and the environment. Wastewater sludge recycling in agricultural lands serves as a source of water (liquid sludge), crop nutrients (both solid and liquid sludge), improving soil's physical and chemical properties. It is, therefore, crucial in reducing the pressure on freshwater bodies (liquid sludge) and lowering greenhouse gas emissions from fertiliser production.

As a member of the UN, South Africa funded local research projects related to wastewater sludge recycling in agricultural lands. Sludge guidelines were developed and updated, including decision support systems for applying sustainable sludge in agricultural lands (SARA) model. To advance sustainability, validation of the SARA model was warranted. This was conducted by employing field studies from across the South African agroecological zones, which was the mandate of the current research project.

2.2 Wastewater sludge as a nitrogen fertiliser source

2.2.1 Background information

Beneficial agricultural sludge-use as a source of nutrients and soil ameliorant is a well-known practice (Singh & Agrawal, 2008; Bettiol & Ghini, 2011; Roig et al., 2012; Krol et al., 2015). Sewage sludge-use in agricultural lands improves the following:

- Soil organic matter (Navas et al., 1998; Munn et al., 2000; Marx et al., 2004)
- Soil structure (Objeda et al., 2007; Hamidpour et al., 2012)
- Soil physical properties, such as soil porosity (Tsadilas et al., 2005; Angin & Yaganoglu, 2011)
- Bulk density (Ramulu, 2002; Tsadilas et al., 2005)
- Aggregate stability (Angin & Yaganoglu, 2011)

Sewage sludge can also be crucial in improving soil hydraulic properties, such as saturated hydraulic conductivity (Aggelides & Londra, 2000), infiltration rate, and water holding capacity (Epstein, 1975; Gupta et al., 1977; Aggelides & Londra, 2000; Objeda et al. 2003; Glab et al., 2008; Kengne et al., 2014; Krol et al., 2015).

Sewage sludge application in agricultural soils improves soil chemical properties by:

- Decreasing and increasing the soil pH (depending on the source) (Achiba et al., 2009; Angin & Yaganoglu, 2011; Hamidpour et al., 2012)

- Increasing cation exchange capacity (Epstein et al., 1976; Kladivko & Nelson, 1979; Soon, 1981)
- Serving as a source of crop nutrients, such as nitrogen and phosphorus (Aggelides & Londra, 2000; Objeda et al., 2003; Angin & Yaganoglu, 2011; Bouajila & Sanaa, 2011)

Sewage sludge also improves the following, attributable to its higher organic matter and nutrient content:

- Soil microbial activity
- Soil biochemical activities
- Soil respiration
- Soil enzymes activities and biomass (Ros et al., 2003; Bhattacharyya et al., 2003; Singh & Agrawal, 2008; Bettiol & Ghini, 2011).

Improving the core soil quality parameters (soil physical, chemical, and biological properties) is essential to improved crop production (Morera et al., 2002; Cogger et al., 2001; Lavado et al., 2006; Tesfamariam, 2009; Kengnea et al., 2014, Krol et al., 2015). The improvement level in soil's physical, chemical, and biological properties from sewage sludge applied to agricultural soils depends on the sludge quality and composition, influenced by the sludge source, treatment process, and sludge drying techniques.

2.2.2 The fertiliser value of wastewater sludge

According to Finck (1995), soil fertility is a complex term, encompassing a combination of components, such as:

- soil texture
- soil reaction
- soil depth
- nutrient content
- soil microbial activity
- organic matter content and composition
- content or absence of potentially toxic substances

Sewage sludge contributes towards improving soil fertility attributable to its macro-and micronutrients, and carbon. Sewage sludge holds significant amounts of N and P, significant as a supplement or substitute for inorganic fertiliser (Hall, 1986). Sewage sludge also holds essential plant micronutrients, such as Cu, Zn, Mn, and B, besides the well-known macronutrients (N and P), (Ekama, 1993; Snyman & Van Der Waals, 2004). Table 2.1 presents the mean macronutrient content of South African sewage sludge. According to these data, South African sludge has appreciable organic matter, which could be vital in restoring South African soils, highly degraded attributable to several reasons. These include warm temperature, accelerating the mineralisation of soil organic matter, leaving the soil with low organic matter content (Kirschbaum, 1995).

Table 2.1: Typical nutrient content of dry South African sludge

Nutrients	Range (%)
Total N	3.2-4.5
Total P	1.5-1.7
Total K	0.2-0.3
Organic content	40-70

Source: Snyman and Herselman (2006).

2.3 Nutrient and organic matter composition of sludge as affected by wastewater treatment and post-treatment techniques

Sewage sludge comprises organic matter, plant nutrients, organic, and inorganic pollutants, and micro-and mesofauna (Roig et al., 2012; Krol et al., 2015). The organic component of sludge composite encompasses soluble compounds, hemicelluloses, cellulose, lignin, lipids, and fats (Mottet et al., 2010). A larger fraction of the organic matter comprises soluble compounds (Parnaudeau et al., 2004; Zhao et al., 2011; Malobane, 2014). The wastewater origin influences the sludge composition (Banegas et al., 2007; Malobane, 2014). The employed wastewater and sludge treatment methods could further influence the sludge biochemical composition (Cogger et al., 2004). Table 2.2 presents the typical constituents of primary and secondary sewage sludge (Nilsson & Dahlstrom, 2005).

Anaerobic and aerobic sludge digestion is the most applied wastewater treatment technique, producing a stable sludge, whereas the activated sludge treatment process produces unstable sludge (Chan et al., 2009). The most exploited sludge dewatering/drying techniques include cake filtration, centrifugation, bed drying, thermal drying, and belt pressing (Dirkzwager & Hermite, 1988; Matsuoka et al., 2006). In South Africa, drying beds is the dominant sludge drying technique (Snyman & Van Der Waals). Drying beds are cost-effective compared with mechanical and thermal drying techniques (WEF, 2009).

Post wastewater treatment sludge drying techniques are vital for agricultural use as this significantly influences the transport costs of sludge from wastewater treatment plants to receiving soils (Kouloumbos et al., 2008). The drying techniques employed, however, could influence the quantity and plant availability of N in the sludge. N is the essential element based on the sludge recommendation. The effect of the most commonly used drying technique in South Africa (drying beds) on the N and C contents and compositions of sludge is, therefore, detailed in the following sections.

Table 2.2: Typical composition of primary and secondary sewage sludge

	Units	Primary sludge	Secondary sludge
Dry solids (DS)	g/L	12.0	9.0
Volatile solids (VS)	%DS	65.0	67.0
pH		6.0	7.0
Carbon	%VS	51.5	52.5
Hydrogen	%VS	7.0	6.0
Oxygen	%VS	35.5	33.0
Nitrogen	%VS	4.5	7.5
Sulphur	%VS	1.5	1.0
C:N ratio		11.4	7.0
Phosphorus	%DS	2.0	2.0
Chlorine	%DS	0.8	0.8
Potassium	%DS	0.3	0.3
Aluminium	%DS	0.2	0.2
Calcium	%DS	10.0	10.0
Iron	%DS	2.0	2.0
Magnesium	%DS	0.6	0.6
Fat	%DS	18.0	8.0
Protein	%DS	24.0	36.0
Fibres	%DS	16.0	7.0
Calorific value	kWh/t DS	4200.0	4100.0

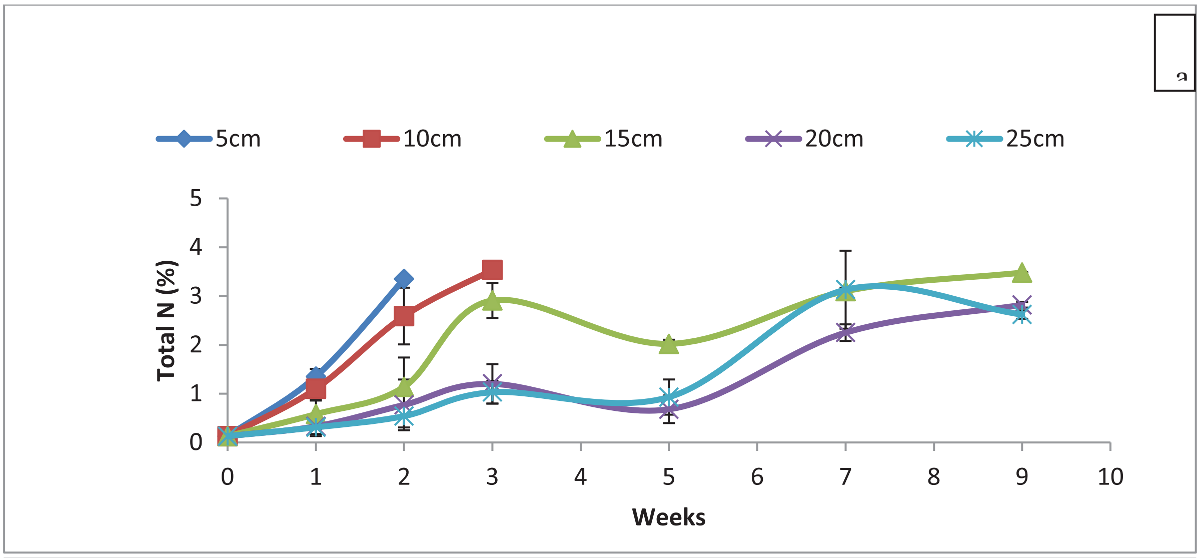
DS: dry solid

VS: volatile solids

Source: Nilsson and Dahlstrom (2005).

2.4 Wastewater sludge N content and composition as affected by sludge drying depth on beds

Tesfamariam et al. (2018) reveals that the total N content of wastewater sludge signified an increasing trend during the drying process. This excludes autumn between weeks three and five (drying depths of 15, 20, and 25 cm) and between weeks seven and nine (drying depths of 25 cm) (Figure 2.1). This contrasts with the total C pattern, decreasing as the time of the drying process progressed.



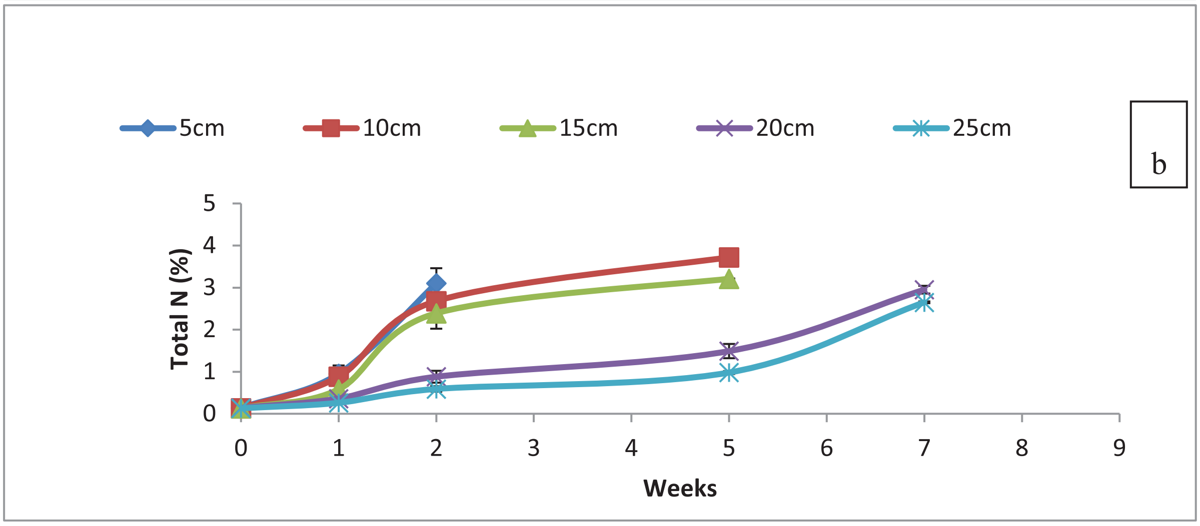


Figure 2.1: Total N changes at 5, 10, 15, 20, and 25 cm sludge drying depths, a) autumn and b) winter seasons (I = SDT)

Source: Tesfamariam et al. (2018).

The N increase could be attributed to the C loss from the substrate during the decomposing organic matter, leading to an increase in the concentration of other elements, including N in the substrate. At the end of the drying period, the highest N was recorded for the 10 cm drying depth (3.53% for autumn; 3.72% for winter) and lowest for the 25 cm drying depth (2.62% for autumn; 2.65% for winter) (Figure 2.2).

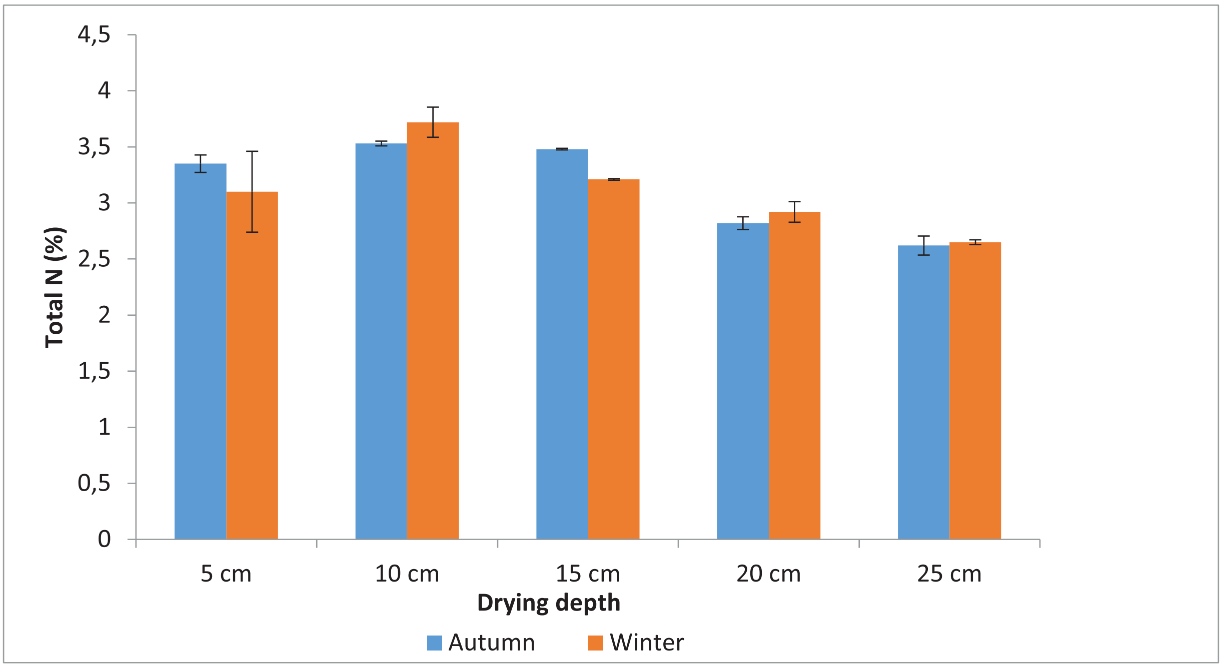


Figure 2.2: Total N content (%) (oven-dry mass basis) of anaerobically digested sludge, dried at five drying depths (I = SDT)

Source: Tesfamariam et al. (2018).

The lower total N content recorded at 20 and 25 cm depths is attributable to two reasons:

- Enhanced NH_3 volatilisation
- Denitrification losses from the middle to lower sections of the drying beds, remaining wet for a longer time.

The wet and warm environment in the middle of the lower sections of the drying beds accelerates organic matter decomposition, releasing NH_4^+. An increase in the concentration of NH_4^+, was observed between weeks three and seven in autumn and weeks two and five in winter for the 20 and 25 cm drying depths. This led to an increase in the concentration gradient between the atmospheric NH_3 and the NH_3 in the sludge. This resulted in ammonia diffusion from the sludge strata to the atmosphere, therefore, negatively affecting the total N sludge content (Al-Malack, 2014). A rapid NH_4^+ increment was observed in the 20 and 25 cm drying depths between weeks three and seven in autumn and weeks two and five in winter. This was followed by a rapid decline in NH_4^+ concentration in the following two weeks. During those two weeks of observed NH_4^+ decline, the NO_3^- content of the strata did not increase. It continued decreasing, indicating that the NH_4^+ content decline was not attributable to nitrification processes but to ammonia volatilisation. The continuous decline in the nitrate concentration despite the increase in the NH_4^+ content is attributed to two reasons:

- The inhibition of nitrification is attributable to the high NH_4^+:NO_3^- ratio because a high NH_4^+:NO_3^- ratio inhibits the nitrification process (Tesfamariam, 2009)
- Denitrification attributable to high soluble organic carbon and higher moisture content, accelerating the denitrification process with the aid of denitrifying bacteria (Bernal et al., 1998)

Tesfamariam et al. (2018) reported a strong correlation (r=0.96) between sludge drying depth and total N (%) (Figure 2.3b). According to the correlation function, The N sludge content increased as the sludge drying depth augmented from 5 cm to 10 cm; the N content gradually decreased as the drying depth increased above 10 cm. Wastewater treatment plants using drying beds to dewater their sludge for agricultural purposes may, therefore, consider developing such correlations, as the drying process is climate dependent.

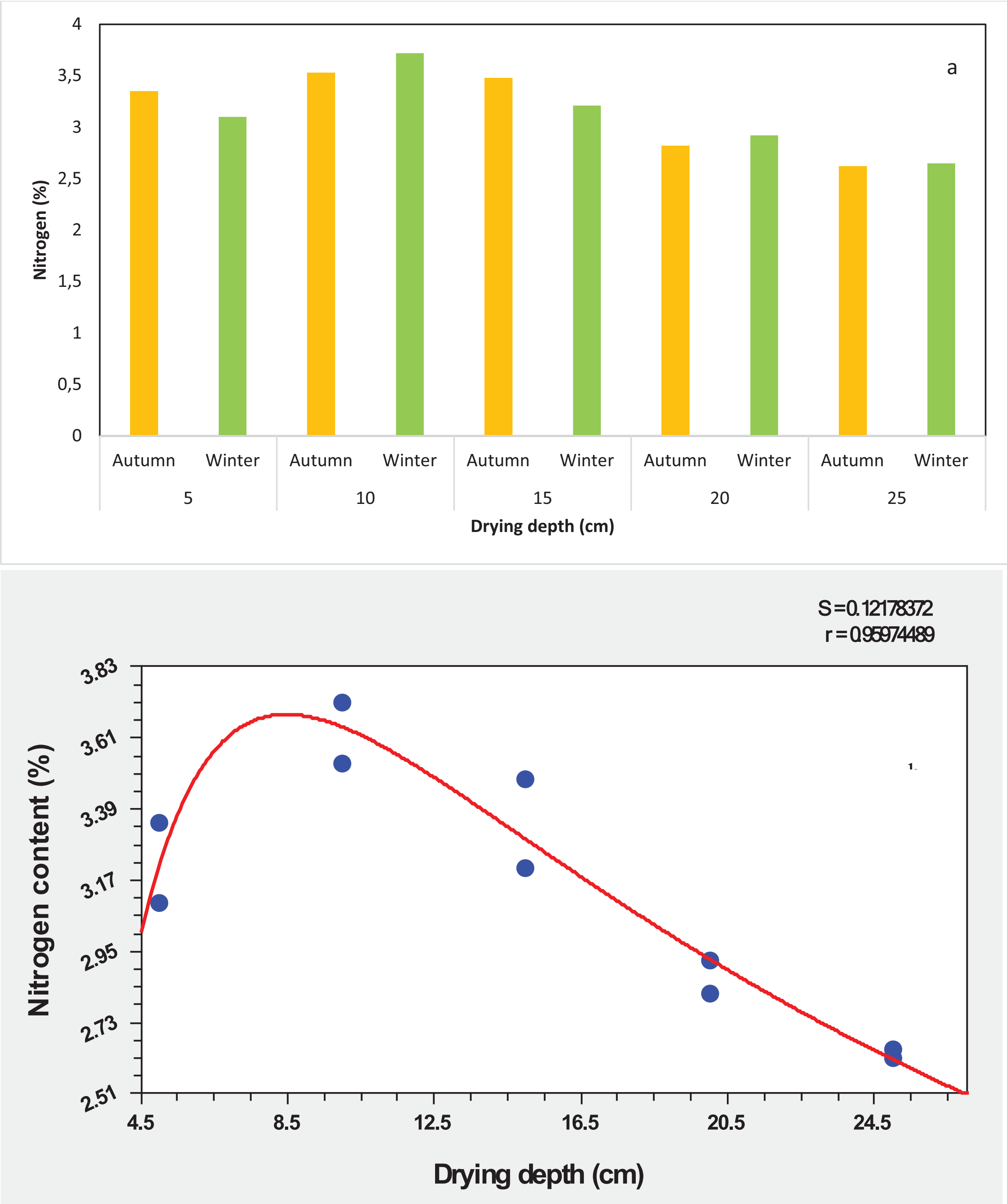


Figure 2.3: (a) Sludge N content as a function of drying depth; (b) correlation between drying depth and sludge N content (combined data for autumn and winter)

Source: Tesfamariam et al. (2018).

2.5 Wastewater sludge organic matter composition, affected by sludge drying depth on beds

Tesfamariam et al. (2018) indicated that the carbon content of wastewater sludge dried on beds increased as the sludge drying depth elevated from 5 cm to 10 cm. A further increase resulted in a gradual carbon decline (Figure 2.4). The organic matter composition of the dried sludge, however, did not show noticeable differences among drying depths (Figure 2.5). The soluble compounds, accounting for over 70% of the organic matter, were 75.9%; 77.8%; 78.2%; 75.7%; and 78.8% for sludge dried at 5; 10; 15; 20; and 25 cm drying depths, respectively. Other compounds, such as lipids, were low for the 15 cm drying depth. The reported higher proportion of soluble compounds agrees with findings by Parnaudeau et al. (2004); Mottet et al. (2010); Zhao et al. (2011) and Malobane (2014).

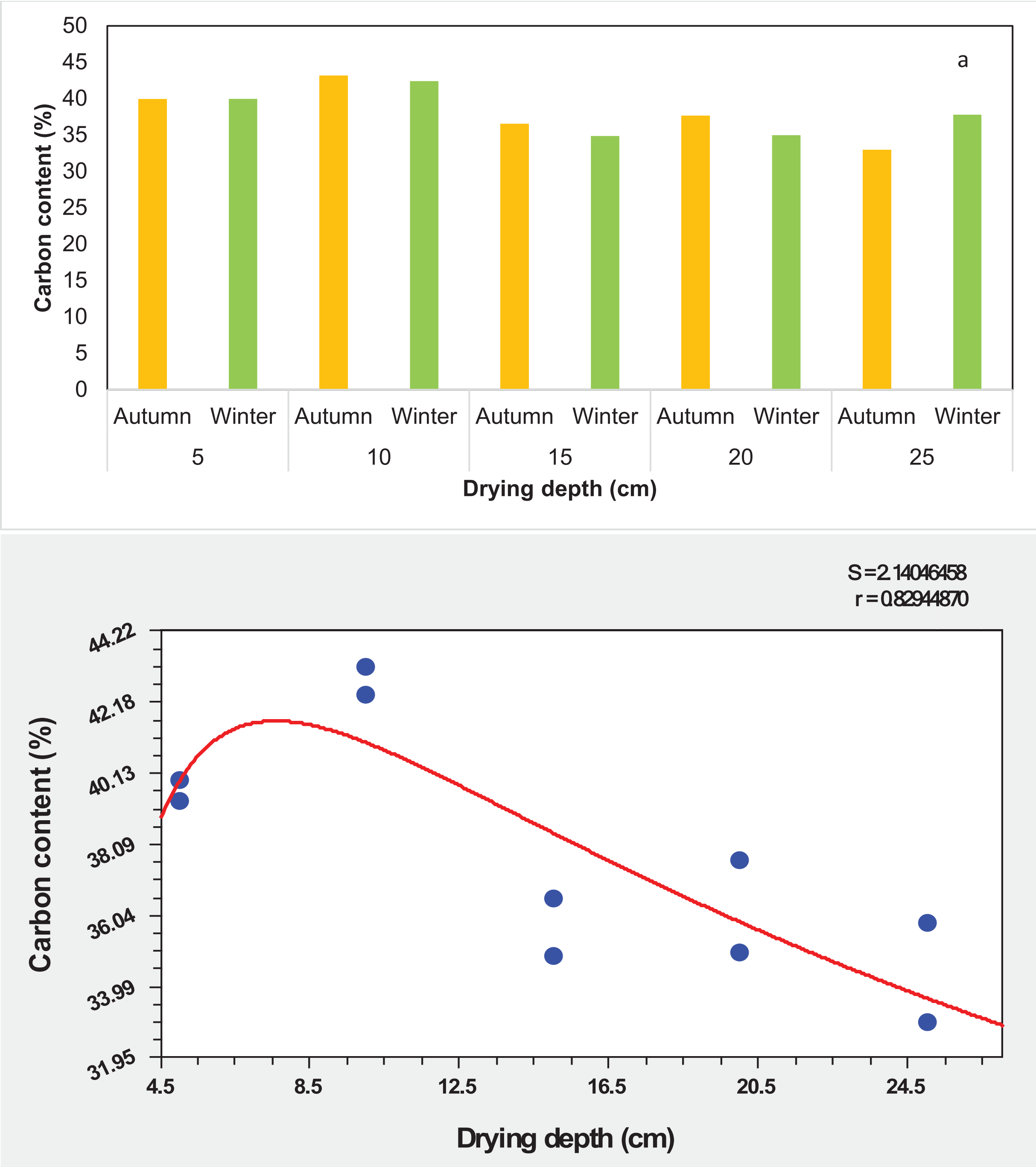


Figure 2.4: (a) Sludge C content as a function of drying depth; (b) A correlation between drying depth and sludge carbon content (combined data for autumn and winter)

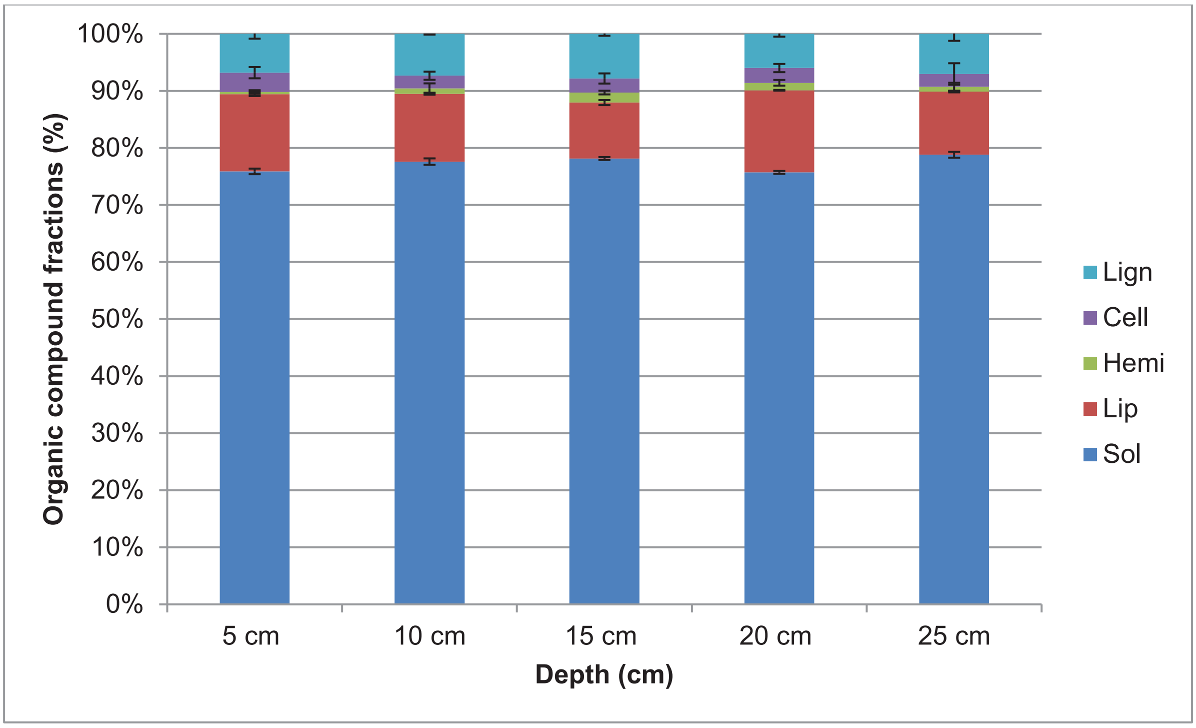


Figure 2.5: Organic compound fractions of anaerobically digested sludge at five drying depths (5, 10, 15, 20, and 25 cm)

NB. Sol= Soluble organic compounds, Lip= Lipids, Hemi= Hemicellulose, Cell= Cellulose and Lign= Lignified fraction (error bars indicated standard deviation around the mean value).

2.6 Organic matter decomposition and nitrogen mineralisation from sludge-amended soils

Adding organic materials to the soil results in various compounds decomposing to release simpler organic and inorganic compounds (Brady & Weil, 2008). The biological breakdown of organic compounds is termed 'decomposition' (Juma, 1999; Brady & Weil, 2008). Organic compounds decompose at fluctuating rates (Hattori & Mukai, 1986; Lerch et al., 1992; Parnaudeau et al., 2004). Organic compounds are usually categorised into:

- Rapidly decomposing (sugars, starches, proteins)
- Slowly decomposing (hemicellulose, cellulose, fats, waxes, resins, and lipids)
- Lignified or recalcitrant compounds, depending on their chemical structure (Alexandra & José, 2005; Brady & Weil, 2008)

C is released through carbon dioxide during organic matter decomposition, while nitrogen is released in an ammonia form (Nayak et al., 2013). Soil organisms use organic matter from various organic materials. These are added to the soil as a source of food and energy. Nutrients (N, S, and P) are also released into the soil (Alexandra & José, 2005; Benbi & Richter, 2002; Brady & Weil, 2008).

A substantial proportion of the N in wastewater sludge is organic and should be transformed into inorganic forms before plants can benefit from it (Tesfamariam et al., 2015; Lasa et al., 1997; Vieira et al., 2005; Cogger et al., 2001). Chemical, physical, and microbial-mediated processes engage in transforming organic N into inorganic N forms (mineralisation). Nitrogen mineralisation (ammonification) is the process of decomposition of a fraction of organic N into ammonium (NH_4^+) by soil microbes (Benbi & Richter, 2002; Alexandra & José, 2005; Brady & Weil, 2008; Sekoma, 2008). Bacteria and fungi are the microorganisms involved in the ammonification process, desolating the nitrogenous waste while releasing NH_4^+, expressed in Equation 1 (Bernhard, 2010).

$$R\text{-}NH_2 + 2H_2O \rightarrow OH^- + R\text{-}OH^- + NH_4^+ \text{ (Brady \& Weil, 2008)} \qquad \text{Equation 1}$$

Several factors influence the mineralisation rate. These include the organic composition of the material, soil type, and other abiotic factors, such as the ambient soil temperature, moisture, oxygen availability, pH, supply of nutrients, and salinity (Foth & Ellis, 1997). The C:N ratio of the material influences the mineralisation of organic substances and residues, affecting the net mineralisation or immobilisation rate (Vinten & Smith, 1993). Carbon to nitrogen ratio (C:N ratio) approximates the important parameter energy:nitrogen (E:N) ratio. Low C:N ratio organic residues have faster mineralisation rates, causing net mineralisation. According to Cameron & Haynes (1986), the lowest C:N ratio is 8:1, established in microbial biomass. The C:N ratio of clover, beans, and lucerne is in the range between 13:1 and 23:1. Cereal straw and other mature plant stalks hold 60:1 to 80:1 C:N ratios. The C:N ratios of some plant materials with N free lignin and residual substances from several peat soils with high C:N ratios are inadequate sources of energy for most microorganisms (Jansson & Persson, 1982).

Nitrogen mineralisation increases as the soil-water content and temperature increase to a certain degree. When the soil temperature and moisture levels exceed the threshold level, the decomposition rate decreases for every increment in the corresponding temperature or moisture level. For instance, when the soil moisture level exceeds the field capacity, the mineralisation rate decelerates attributable to low oxygen availability for the microbes to digest the organic matter (Constantine, 2008). When the temperature increases from cold to warm, the mineralisation rate upsurges; however, the mineralisation rate decelerates when the temperature exceeds the threshold level (Tesfamariam, 2009).

2.7 Organic matter decomposition from sludge-amended soils

The type and chemical composition of organic material added, biological, and environmental properties of the soil influence the decomposing rate of the various organic materials

(Brussaard, 1994; Brady & Weil, 2008). Similarly, the biochemical composition of wastewater sludge dictates the rate of C decomposition (Parnaudeau et al., 2004; Smith & Tibbett, 2004). The proportion of the various organic compounds in organic material is essential in determining the organic matter decomposition rate (Parker & Sommers, 1983; Parnaudeau et al., 2004).

The initial N content and C:N ratio is the first litter chemistry, emphasising the potential decomposition rate of biological organic material (Tian et al., 1993; Valenzuela-Solano & Crohn, 2006; Lashermes et al., 2010). Plant litter studies indicated that low C:N ratio materials are characterised by rapid decomposition and less immobilisation than materials with higher C:N ratios (Muhammad et al., 2011). Low decomposition rates characterise organic materials rich in lignin, cellulose, and hemicellulose (Trofymow et al., 2002; Santiago, 2007; Baddi et al., 2004; Parnaudeau et al., 2004).

The soluble organic matter fraction predominates the organic matter fraction of all sludge. These include sugar, proteins, starch, fatty acids, amino acids, nucleic acids, and other monosaccharides (Alexandra & José, 2005). This group of compounds is the first to decompose, crucial during organic matter decomposition. The carbon from hemicellulose, cellulose, fats, waxes, resins, and lipids decomposes at a slower rate compared with the soluble fractions (Brady & Weil, 2008).

Malobane (2014) indicates that cumulative C decomposition per kg organic C applied was the highest for activated sludge (587 g kg^{-1} organic C applied) followed by thermally hydrolysed sludge (362 g kg^{-1} C applied); it was the lowest for anaerobically digested (191 g kg^{-1} C applied) (Figure 2.6).

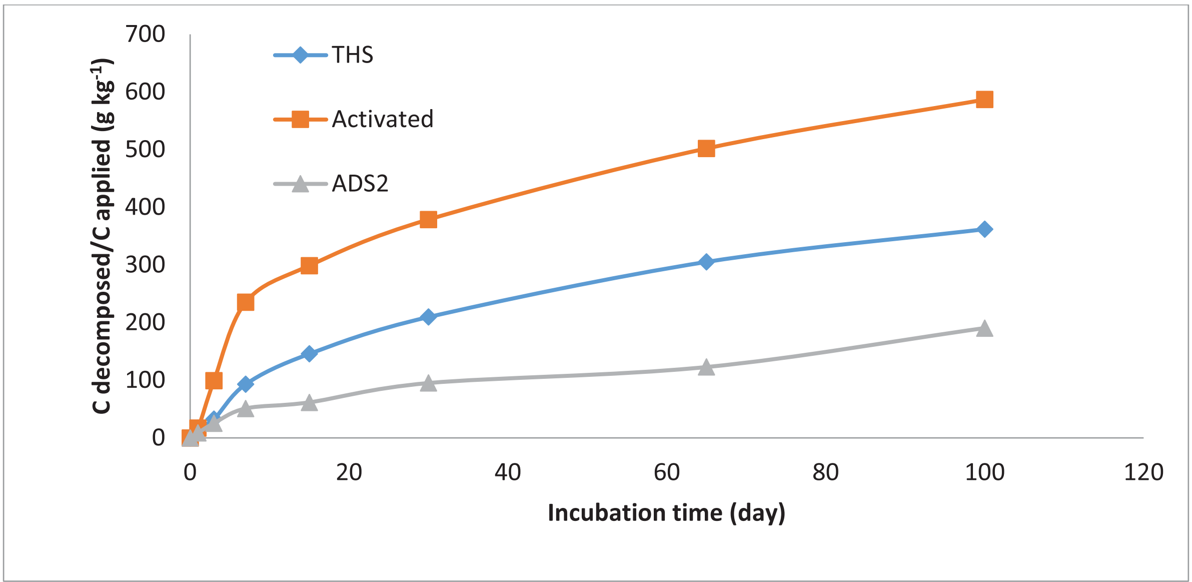


Figure 2.6: Carbon decomposition of anaerobically digested sludge (ADS2), activated sludge (Activated), and thermal hydrolysis sludge (THS) during a 100-day incubation study

Source: Malobane (2014).

2.8 Nitrogen mineralisation from sludge-amended soils

Sludge types have fluctuating N-mineralisation rates attributed to the differences in their composition (Parker & Sommers, 1983; Serna & Pomares, 1992; Parnaudeau et al., 2004; Smith & Tibbett, 2004). The differences in sludge N-mineralisation rates among wastewater sludge could be attributed to the differences in sludge sources and treatment (Epstein et al., 1976; Parker & Sommers, 1983; Serna & Pomares, 1992; Mattana et al., 2014), including post-treatment drying techniques (Tesfamariam et al., 2021).

According to Parker and Sommers (1983), activated sludge had the highest mineralisable N (40%), followed by raw and primary sludge (25%), anaerobically digested sludge (15%), and composted sludge (8%) of the total N applied during a 16-week incubation study. Similarly, Malobane (2014) reported the highest net N-mineralisation rates from activated sludge (45%) followed by THS (34%) and anaerobically digested (18%) in a 14-week incubation study (Figure 2.7).

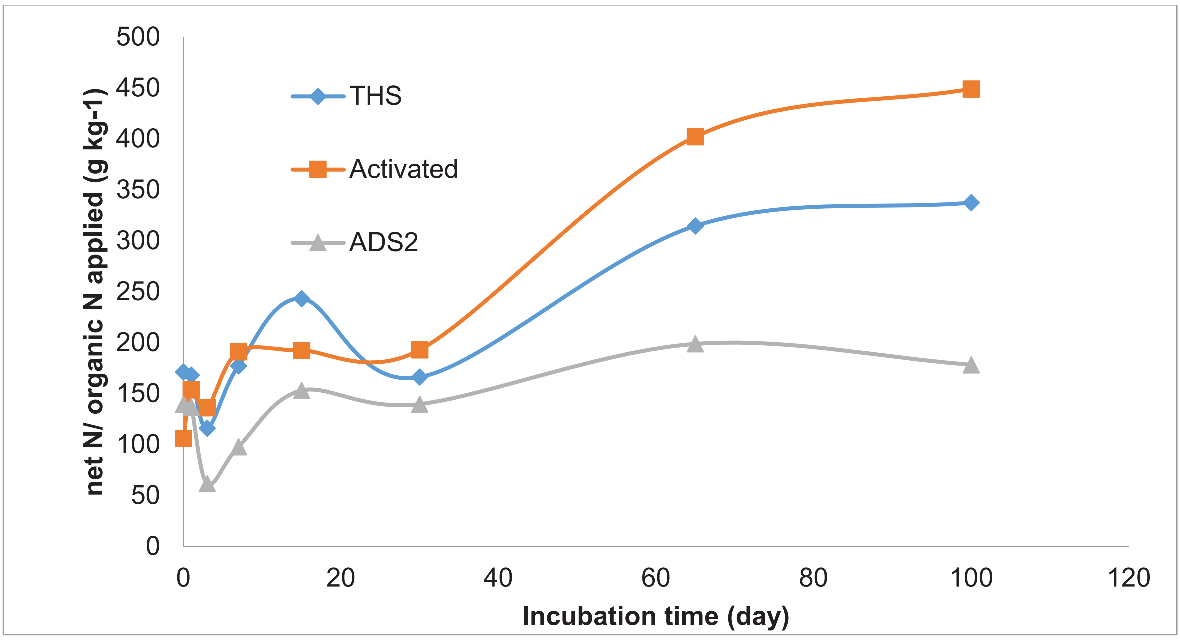


Figure 2.7: Net N mineralised per unit kg organic N applied from anaerobically digested sludge (ADS2), activated sludge (Activated), and thermally hydrolysed sludge (THS)

Source: Malobane (2014).

Serna and Pomares (1992) and Hernández et al. (2002) confirm that aerobically digested sludge provided higher N-mineralisation than anaerobically digested sludge. In contrast, activated sludge was reported to provide significantly higher mineralisation than digested sludge attributable to its high active organic carbon and less stable compounds (Hsieh et al., 1981).

According to Malobane (2014), the highest N-mineralisation rates from activated sludge were attributed to the highest initial N content (4.95%) compared with the other three, ranging between 2.81% and 3.13%. He further explained that the initial N concentration influences the N amount that can be mineralised (Valenzuela-Solano & Crohn, 2006; Lashermes et al., 2010). Activated sludge had the lowest lignin fraction (3.21%) compared with the other three, ranging between 8.7% and 15.96% by mass of the total organic matter. Studies indicated that lignin harms N-mineralisation (Baddi et al., 2004).

Malobane (2014) also reported thermal hydrolysing benefits in improving the N availability. According to Malobane, despite a 9.6% difference in the initial total N content between THS (3.13%) and ADS2 (2.83%), N-mineralisation from THS (57.11 g N kg^{-1} C applied) was 61% higher than ADS2 (22.27% g N kg^{-1} C applied) at the end of the incubation study. Such difference is attributed to the higher lignin fraction (15.96%), lignin:N ratio (1.40), and C:N ratio (8.78) of ADS2 compared with THS (lignin=8.7%, lignin:N ratio=0.50, C:N ratio=5.77). Higher lignin:N ratio causes low net N-mineralisation attributable to N immobilisation (Baddi et al., 2004; Parnaudeau et al., 2004).

Recent methods of wastewater treatment, such as thermal hydrolysis at temperatures ranging from 40 to 180ºC, break the cell walls of biological organic materials in the sludge, improving degradability (Neyens & Baeyens, 2003). Rigby et al. (2016) indicate that thermally dried mesophilic anaerobically digested sludge had a larger mineralisable pool of N than dewatered mesophilic anaerobically digested sludge, despite its lower total and mineral N content. Similar findings were reported by Matsuoka et al. (2006) and Fernández et al. (2007). According to these authors, thermal drying increased the easily mineralisable fraction of N and C in the sludge.

2.9 Other nitrogen transformation processes and dynamics in sludge-amended soils

Once the organic N from sludge is mineralised into inorganic form through mineralisation, the by-product inorganic N (ammonium) is exposed to further transformation and mobility. These transformation processes include immobilisation, volatilisation, nitrification, denitrification, and sometimes leaching.

2.9.1 Immobilisation

Immobilisation is the net incorporation of mineral nitrogen, mainly NH_4^+, into organic forms (microbial tissue) during the decomposition process (Vinten & Smith, 1993). It is a process functioning in the opposite direction of mineralisation. The residue decomposition with a high C:N ratio causes a negative net residue nitrogen mineralisation or immobilisation. This, however, changes with the growth and stabilisation of the microbial population resulting in a decline of the C:N ratio and the release of mineralisable nitrogen.

Nitrogen immobilisation occurs through biological and abiotic processes (Brady & Weil, 2008). Biological immobilisation ensues when microorganisms, decomposing organic N compounds, transform inorganic N into organic form in the absence of enough inorganic N to build the microbial biomass (Brady & Weil, 2008; Sekoma, 2008). The chemical transformation process during immobilisation is presented below in Equation 2:

$$Energy + NO_3^- - 1/2O_2 \rightarrow NO_2^- \rightarrow 4H^+ + energy + NO_2 - O_2 \rightarrow OH^- + R\text{-}OH + NH_4^+ - 2H_2O \rightarrow R\text{-}NH_2,$$

(Brady & Weil, 2008). Equation 2

The influence of immobilisation in agricultural lands is observed through a periodic decline in plant-available nitrogen after the incorporation and decomposition of cereal straw and other mature plant stalks (Powlson et al., 1987). Immobilisation is a temporal process, reversed as time progressed.

The opposite of mineralisation can also be termed 'net immobilisation' (Benbi & Richter, 2002; Sekoma, 2008), and the difference between the two is named net mineralisation. Net mineralisation indicates the plant-available nutrients (Benbi & Richter, 2002).

2.9.2 Nitrification

Nitrification is a process of oxidising NH_4 (the by-product of mineralisation) into nitrite (NO_2) and then to nitrate (NO_3^-) by chemoautotrophic bacteria (Bernhard, 2010). Nitrate is the most stable form of plant-available inorganic N in the soil environment. Howard-Williams and Downes (1993) classified the following two processes responsible for nitrification:

- Autotrophic nitrification
- Heterotrophic nitrification

Autotrophic nitrification is the process of transforming ammonium or ammonia to nitrite by a group of bacteria called *Nitrosomonas* sp. and *Nitrosolobus* sp. followed by transforming nitrite to nitrate by a separate group of bacteria called *Nitrobacter* sp.

Heterotrophic nitrification is the oxidation of reduced organic nitrogen compounds to oxidised nitrogen species. The two-stage nitrification process was expressed by Brady and Weil (2008) as follows:

$$NH_4^+ + O_2 \rightarrow 4H^+ + energy + NO_2^-$$ (Brady & Weil, 2008) Equation 3

$$NO_2^- + 1/2O_2 \rightarrow energy + NO_3^-$$ (Brady & Weil, 2008) Equation 4

Similar to the mineralisation process, the ambient soil temperature, soil moisture, pH, and soil oxygen supply influence the nitrification process (Foth & Ellis, 1997). Nitrification occurs mostly under aerobic environmental conditions.

2.9.3 Denitrification

Denitrification is the gaseous loss of nitrogen through dinitrogen gas (N_2), nitric oxide (NO), and nitrous oxide (N_2O) (Foth & Ellis, 1997; Bernhard, 2010) mediated through anaerobic bacteria (St Luce et al., 2011) (Equation 5).

$$NO_3^- \rightarrow N0_2^- \rightarrow NO \rightarrow N_2O \rightarrow N_2 \quad \text{Equation 5}$$

The following critical conditions need to be accomplished for denitrification to transpire: the presence of nitrate, an anaerobic environment, and a C source (Constantine, 2008; Stenger et al., 2013). Factors influencing the nitrification process, such as soil-water content, soil temperature, and soil pH, also affect the denitrification rate (Foth & Ellis, 1997; Constantine, 2008; Tesfamariam, 2009). The denitrification rate increases as the soil moisture content exceed the field capacity. It is higher in high clay content soils than sand-dominated soils attributable to the higher proportion of micropores in clay-dominated soils. The soil pH affects the gas type produced (Stenger et al., 2013).

The N loss through denitrification has negative environmental effects attributable to its high greenhouse validity besides reducing the plant-available N in the soil. This is despite its favoured use by wastewater treatment plants to lower the nitrate concentration to acceptable concentrations before releasing it to the environment.

2.9.4 Ammonia volatilisation

Ammonia volatilisation is the gaseous loss of NH_3 from the soil surface to the atmosphere (Haynes & Sherlock, 1986). Volatilisation occurs because of the deviation in vapour pressure gradient between ammonia in solution and the ambient air (Freney et al., 1983) and can be huge. Ammonia volatilisation is high from initially wet soils (below saturation), allowed to dry slowly (Fenn & Escarzaga, 1977). This is because the evaporative water loss promotes ammonia volatilisation by increasing or maintaining the concentration of ammonia in the soil solution over time (Haynes & Sherlock, 1986). Conversely, ammonia volatilisation lacks from exceptionally low soil-water content soils (Nelson, 1982). Ammonia volatilisation is high from the surface NH_4^+ application compound fertilisers compared to incorporated fertilisers (Fenn & Kissel, 1976; Quemada et al., 1998).

NH_3 sources for volatilisation include inorganic fertilisers applied to the soil through NH_4^+ compounds and decomposing organic nitrogenous sources, such as sludge. The following factors affect the ammonia volatilisation rate:

- Soil pH
- Soil temperature
- Soil cation exchange capacity (CEC)
- Soil-water content
- Wind speed
- Fertiliser application method (surface, incorporation)
- Fertiliser pH (Nelson, 1982; Freney et al., 1983)

Ammonia volatilisation increases with an intensified soil and fertiliser pH, wind speed, and soil temperature (until 45ºC) (Nelson, 1982). It is suppressed at low pH, cool temperatures, and low wind speeds (Freney et al., 1983). Fine et al. (1989) reported a loss of 87% of the mineralised nitrogen through ammonia volatilisation from activated sewage sludge. Beauchamp et al. (1978) indicate that 20% of total nitrogen applied was lost as ammonia during the first week of decomposition from anaerobically digested liquid sludge. Ammonia volatilisation highly depends on the sludge treatment process, and whether the sludge is left on the surface or incorporated (Table 2.3).

Table 2.3: Ammonia volatilisation rates from Northwest Biosolids applied in western Washington (maritime climate) adapted from Henry et al., 1999)

Treatment method				Volatilisation rate (%)
	Liquid (incorporated)			20 to 40
			Agronomic rate	14 to 50
			Double agronomic rate	25 to 49
			Lime amended	45 to 134
Anaerobically		Incorporated	Reduced pH	12 to 28
digested	Dewatered	Surface applied		51-127
Aerobically digested (incorporated)				6 to 12
Lagooned (incorporated)				4 to 20
Lime-stabilised (incorporated)				14 to 22
Drying bed (incorporated)				2 to 5
Oxidation ditch (incorporated)				9-23

Source: Adapted from Henry et al. (1999).

2.9.5 Crop nitrogen uptake

Nitrogen is mostly absorbed by plants through NO_3 during a convective flow of soil water to plants in response to transpiration from the canopy (Olson & Kurtz, 1982). This is because of the low attraction of NO_3^- to the soil colloid compared with NH_4^+, which has a higher force of attraction to the soil colloids. This low force of attraction between the NO_3^- and soil colloids facilitates the NO_3^- to be carried by mass flow to the plant roots. The rapid NH_4^+ nitrification in most soil conditions is significant in minimising its availability for crop uptake (Haynes & Sherlock, 1986). NH_4^+ could be the major source of N to crops under soil conditions unfavourable for nitrification, such as anaerobic and acidic soil conditions (Haynes & Sherlock, 1986).

2.9.6 Nitrogen leaching

Nitrate salts are highly soluble; therefore, nitrate moves with water and can be easily leached through soil (Wang et al., 2003; Esteller et al., 2009). The leached NO_3^- can be lost to groundwater or drainage system because of downward movement through the soil in percolating water (Prasad & Power, 1997). A global concern exists over increasing NO_3 groundwater contamination attributable to improper agricultural practices (Brye et al., 2001). A primary problem is a potential threat to human and animal health attributable to NO_3 surface and groundwater pollution (Ross, 1989). Some physical soil properties, such as texture and structure, can influence NO_3^- leaching; it influences the water movement through the soil (Esteller et al., 2009; Di & Cameron, 2010).

The challenges with excessive nitrate leaching from agricultural lands are associated with ill-timed fertiliser applications and inadequate fertilisation strategies. In rural communities, groundwater is a major drinking water source. Increasing nitrate levels in groundwater led to limited groundwater use for human consumption in countries, such as India, the USA, and Africa (Shrimali & Singh, 2001). High nitrate content in drinking water is of great concern to human health. In infants, ingestion of nitrate water causes methemoglobinemia, a condition known as a baby-blue syndrome, which may lead to death. Eutrophication is another environmental problem associated with excess nitrogenous and phosphorus fertilisers in agricultural lands. Nitrates and phosphates from nutrient-rich soils carried through runoff and soil erosion into surface water bodies, such as rivers or lakes, promote algal bloom deplete oxygen in water bodies, and limit sunlight penetration into the water, leading to the death of living organisms (Schindler, 2006).

Proper fertiliser application strategies are, therefore, vital for sustainable agricultural production regardless of the source of fertiliser (commercial inorganic fertiliser, manure, or sludge).

CHAPTER 3: VALIDATION OF NITROGEN MINERALISATION ASSUMPTIONS IN THE SARA MODEL USING FIELD STUDIES IN DIFFERENT AGRO-ECOLOGICAL ZONES OF SOUTH AFRICA

3.1 Abstract

Understanding N dynamics following sludge application is critical for N management in agroecosystems. Sludge used in agricultural lands as a source of plant nutrients gained global momentum. A field incubation study was conducted from October 2017 to June 2018 (first sampling season) and 2017 to 2020 (second sampling) on six sites, representing five South African agroecological zones. Porous ceramic tubes filled with soil-sludge mix (treatment) and soil alone (control), were employed in the study. The tubes were buried at 15 cm depth below the soil surface. Net nitrogen mineralisation from the tubes was estimated as the variance between initial and final organic N applied. The net N-mineralisation rate varied significantly among agroecological zones.

During the first incubation year, the lowest mineralisation rate was recorded in an arid zone (10%) and the highest in the super-humid zone of Nelspruit (57%). The net N-mineralisation rate the first field incubation year was 10% (arid); 31% (semi-arid); 34% (subhumid – Bethlehem); 46% (subhumid – Johannesburg); 48% (humid); and 57% (super-humid). The net N-mineralisation rate during the first year of field incubation study across agroecological zones followed similar trends as the model predictions. The values from the field studies were higher than those from the model predictions, except for the arid zone. At the third-year end of the field incubation study across agroecological zones, the net N-mineralisation was 42% (arid); 73-76% (subhumid); 73% (humid); and 69% (super-humid).

The net N-mineralisation model simulation results for three to six years followed similar trends; 35% to 40% (arid); 44% to 52% (Subhumid); 52% to 57% (humid); and 58% to 65% (super-humid) but slightly lower than the field mineralisation studies attributable to the abnormally high rainfall experienced especially in the subhumid areas of Johannesburg. The model simulation results were reasonable, considering the values used by the model as the average for 20 years, including high, moderate, and low rainfall. The sludge recommendation rates generated by the SARA model are generally acceptable and could be employed across South African agroecological zones.

3.2 Introduction

Recycling municipal wastewater sludge through land application for agricultural benefits gained global attention and momentum (Masunga et al., 2016, Rigby et al., 2016). This was a viable and sustainable sludge utilisation option relative to landfilling, incineration, and ocean dumping (Latare et al., 2014). Sludge contains a wide range of macro-and micronutrients highly required for plant growth (Christie et al., 2001, Uggetti et al., 2012). The presence of major nutrients, such as N and P, provides sludge with an ameliorative effect (Latare et al.,

2014) on non-fertile soils. Sludge also contains a substantial amount of organic matter, rendering it a reliable soil conditioning material (Singh & Agrawal, 2008). Sludge can be used as a soil amendment option to improve degraded and low soil fertility (Mattana et al., 2014) and subsequently enhance agricultural productivity (Singh & Agrawal, 2008, Singh & Agrawal, 2010, Barry et al., 2019).

Several benefits are associated with sludge-use in agricultural lands. Such benefits include cost saving for farmers and wastewater treatment plants (WWTPs) (Smith & Tibbett, 2004) and reduced environmental pollution as sludge would be taken as a resource rather than as waste. Synthetic fertilisers are out of reach for several smallholder farmers, especially in the developing world. Some authors suggest that organic amendments or sludge be substituted for chemical fertilisers (Boudjabi et al., 2017, Diacono & Montemurro, 2010, Mehrotra et al., 2016). Sludge as an alternative plant nutrient source would reduce expenses associated with purchasing chemical fertilisers by farmers (Price et al., 2015).

Among the essential nutrients required for plant growth, nitrogen is one of the major plant nutrient requirements and most limiting element in crop production (Rigby et al., 2016, Graham & Vance, 2000). Plants need N as a precursor for chlorophyll synthesis (Henry et al., 1999) improving crop yields. Whenever soils are infertile, such required nutrients must be met through some external sources, and these could be through synthetic fertilisers or organic amendments, such as sludge, compost, or manure application.

Sludge N content is widely variable. Largely, N, other nutrients, and the general chemical composition of sludge is influenced by certain factors. These include quality of wastewater sources (domestic or industrial origin) and the treatment processes involved in the wastewater treatment plant (Sharma et al., 2017). The N content range of sludge was reported as 3% to 6% (Eldridge et al., 2008; Rigby et al., 2016); of which 50% to 90% of sludge N is in organic form (Sommers, 1977). The organic N must be mineralised into inorganic forms (nitrate ($NO_3^- - N$) and ammonium ($NH_4^+ - N$)) to be available for plant uptake (Mbakwe et al., 2013; Ogbazghi et al., 2016). Understanding the soil organic matter transformation and N dynamics in the cropping systems is a prerequisite for predicting plant-available nutrients (Rigby et al., 2016), proper accountability of nutrient budgets, and making informed decisions without compromising environmental quality (Cogger et al., 2011). Such accountability is important in reducing or limiting nutrients application above crop demands. An excess nutrient application could lead to a significant accumulation of nutrients, such as N ($NO_3^- - N$) and P in the soil profile (Diacono & Montemurro, 2010). It may be leached or eroded and transported into surface or underground water bodies. Such nutrients loaded into the ecosystem are detrimental to human health and would cause environmental pollution that could cause eutrophication (US EPA, 2002), endangering aquatic life.

Considering sustainable sludge-use and minimising nutrient accumulation in the soil profile, researchers (Torri et al., 2017; Tesfamariam et al., 2013; Lu et al., 2012) observed that several directives and guidelines (National Research Council, 2002, Snyman & Herselman, 2006; Walker et al., 1994) advocate for sludge application based on crop N requirements. Since sludge is high in organic compounds and organic N fractions that must be mineralised, an application should consider certain factors. These include mineral N pool size, mineralisable N and N-mineralisation rate, and its organic matter content and quality.

This would reduce the chances of over or under application of crop N (Rigby et al., 2016) at any provided time. Nitrogen mineralisation is the primary process ensuring organic N transformation into mineral N that can be absorbed by plants. It is a biological process influenced by an array of factors. Such factors must be considered when planning for sustainable sludge management options in agricultural lands. Governing factors affecting N-mineralisation in sludge-use in agricultural lands including the initial quality and quantity of organic matter of the material; the C:N ratio of the material; soil type; soil pH; rainfall; and temperature (Mohanty et al., 2011; Rigby et al., 2016; Diacono & Montemurro, 2010).

Intensive research documented a vast literature collection on N-mineralisation from sludge-amended agricultural soils globally; however, most of such studies were either laboratory incubation work (Masunga et al., 2016; Azeez & Van Averbeke, 2010; Mohanty et al., 2011; López-Tercero et al., 2005) or greenhouse experiments (Abdelhafez et al., 2018), including modelling studies (Ogbazghi, 2016; Gil et al., 2011). These were conducted under controlled environmental conditions. From these study findings, extrapolations were made in attempting to estimate real field N-mineralisation. This is attributable to a few studies directly conducted in the field on N-mineralisation in sludge and other organic material amended soils (Atallah et al., 2011; He et al., 2000; Henry et al., 2000; Gilmour et al., 2003).

The major drawback of conducting field studies, especially across various climatic zones, is cost-effectiveness. Extrapolations of findings from computer models and laboratory studies in estimating N-mineralisation in the field are still considered strongly valid and useful. Since these studies are mostly conducted under controlled conditions, they may lack strong and proper representativeness of the reality of in-field response attributable to several factors existing in the field. An understanding of N transformation under real field conditions remains critical for adequate prediction of net N-mineralisation and influences decision-making in N management programmes with limited risk to the environment.

This section of the report, therefore, focuses on quantifying the N-mineralisation of sludge-amended soils in six sites of the five agroecological zones of South Africa, characterised by varying climatic and edaphic conditions. The main objective of this study was to conduct a field experiment and estimate the potential field N-mineralisation of sludge-amended soils in five South African agroecological zones.

3.3 Materials and methods

3.3.1 Sites

The study was conducted at six sites in South Africa. Figure 3.1 indicates the locations of the sites, including Upington in arid (A); Rustenburg in semi-arid (B); Bethlehem (C); and

Johannesburg (D) both in subhumid; Durban in humid (E); and Nelspruit (F) in the super-humid zones.

The sites were selected to represent five of the six South African agroecological zones. Two sites (C and D) of the five sites were within the same agroecological zone. These two sites exhibit contrasting temperatures.

Figure 3.1: Experimental sites (A to F) as shown on Google Earth (© 2019 AfriGIS (Pty) Ltd. US Dept of State Geographer Image LandSat/ Copernicus, Data SIO, NOAA, U.S. Navy, NGA, GEECO)

3.3.2 Weather and soil conditions during the study

Weather data and soil temperatures at each study site were measured during the experimental period. The measured parameters included daily rainfall and daily maximum and minimum temperatures. For Bethlehem, Rustenburg, Johannesburg, and Nelspruit sites, the parameters' data were measured and collected from the Soil, Climate and Water Department of Agricultural Research Council (ARC) weather stations, Upington International Airport weather services provided Upington (Northern Cape Rural-TVET College) site data while South African Sugarcane Research Institute (SASRI) provided the Durban site data.

The soil temperature data readings were measured with Thermochron Hi-Res (-40ºC to +85ºC) Acc 0.5ºC soil temperature sensors (*i-buttons*), Fairbridgetech, South Africa. The sensors were calibrated using cold chain Thermodynamics – a multi-profile software package supplied with the *i-buttons* before installation. They were calibrated to record temperature data for the study duration at four hour intervals. During installation, the *i-buttons* were positioned in the supplied silicone waterproof enclosures and sealed with corks and buried at the depths of 15 cm below the surface along with the porous ceramic tubes (Figure 3.2).

3.3.3 Sludge used for the field experiment

In this study, anaerobically digested sludge dried to 90% solid content was used. The sludge was in the drying beds for 45 days to reach a 90% solid content. Drying of the sludge was conducted at 25 cm thickness (drying depth) on sand drying beds during spring, spanning from August to September 2017.

3.3.4 Field experiment

The field N-mineralisation study was conducted at each of the sites, using porous ceramic tubes, following the method proposed by Weaver et al. (1994). The porous tubes were 200 mm long, 20 mm, and 23 mm for inner and outer diameter, respectively (Figure 3.2), with pore sizes of 2.5 μm. The pores of the tubes allowed the movement of water, air, and dissolved solutes; they were small enough to restrict the movement of solid materials to and from the tubes.

Figure 3.2: The porous ceramic tube used for the field incubation

Representative soil samples were collected from the top 15 cm of the study site. The soils were homogenised before filling the tubes. The tubes filled with soil alone and a soil-sludge mix treatment, each replicated three times, were buried horizontally at 15 cm soil depth, as indicated in Figure 3.3. The ceramic tubes were filled with 65 g of soil (maximum capacity of the tubes) or soil-sludge mix. Sludge was applied to the soil-sludge mix at a maximum application rate of 10 tonnes ha^{-1}, as stipulated in the South African sludge guidelines (Snyman & Herselman, 2006). The sludge was mixed thoroughly with the soil before filling and burying the tubes.

As emphasised by Tesfamariam (2009), the 10 tonnes ha^{-1} application rate was institutionalised into the guidelines as the maximum rate (unless supported through research), following some laboratory studies and short-term (one to two years) field studies in South Africa. Previously, the maximum sludge application rate was pegged at 8 tonnes ha^{-1} year^{-1}. This rate was based on some studies conducted in other countries, therefore, could not be strongly qualified without local studies. Each study site was cleared of vegetation before the burial of ceramic tubes during the entire period to avoid plant root interference.

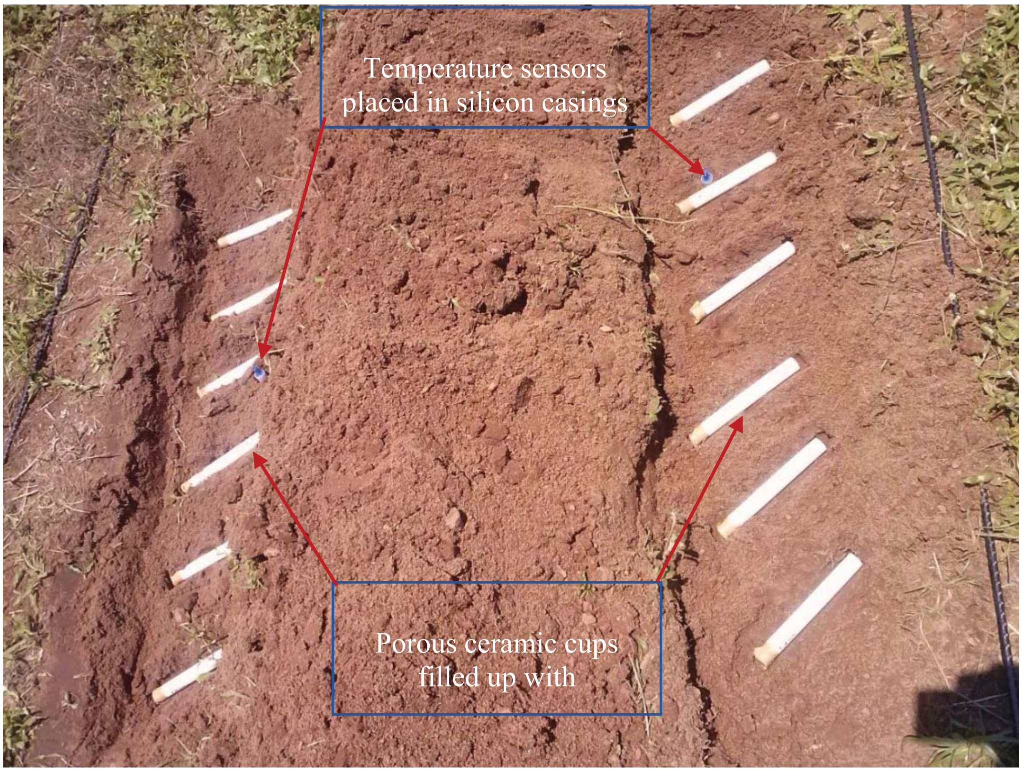


Figure 3.3: Field layout of the porous ceramic tube during incubation

3.3.5 Soil sampling

Portions (30 g) of each homogenised soil sample prepared for the incubation, from both the soil alone and soil-sludge mix samples, were collected for analyses of selected chemical and physical properties. These were named initial samples. Samples were transported to the University of Pretoria in a portable cooler box at 4^{o}C. The samples were then transferred to a cold room and were kept at -20^{o}C until further analyses. To prepare for analyses, air-dried samples were pulverised to pass through a 2 mm sieve before analyses.

The incubation study was conducted for nine months in the first season, and 26 months in the second season. After the incubation period lapsed, the ceramic tubes were carefully dug out from the corresponding sites at each agroecological zone with maximum care to avoid damaging the tubes. These were the seasonal last samples. Similarly, the soil temperature sensors were dug out and taken along. For the second season, the soil temperature sensors were malfunctioning; therefore, no data could be retrieved from the sensors for the second season. The samples were handled, transported, and prepared for analyses in the same manner described above.

3.3.6 Physico-chemical property analyses

Soil salinity (EC) and pH were measured in a soil-water suspension, extracted from a saturated paste (1:2.5 soil to water ratio). Soil EC was measured using a Consort C861 EC meter and soil pH using a Consort C830 pH meter, multi-parameter analyser, Sep Sci, Belgium. Soil samples ground to pass 2 mm sieve were also extracted using 1 *M* KCl (1:10 that is soil (g) to KCl ml ratio) for ammonium and nitrate analyses. The extracts were analysed using the colourimetric method with Lachat Auto-analyser (Lacht Quick Chem Systems, Milwaukee, MI) USA. The NO_3^- - N, and NO_2^- - N were analysed using Ion Chromatography. Total carbon (TC) and total nitrogen (TN) were analysed by the total combustion method using a Carlo Erba Na1500 C/N/S analyser (Carlo Erba Strumentazione, Milan, Italy).

Inductively Coupled Plasma-Optical Emission Spectrometry (ICP-OES) was used for total elemental analyses of Ca; Na; Mg; Mn; S; Fe; Al; K; Cu; Zn & P; and heavy metals (Hg; Cd; Cr; V; Pb; Ni and As for sludge only), after microwave-assisted nitric acid – perchloric acid mixture digestion. Plant-available phosphorus was determined using the Mehlich III (M3) test for soil samples, collected from study sites and by P Bray1 for sludge. Soils and sludge organic carbon and organic matter (OM) were determined after the wet oxidation method by Walkley and Black (1934). Soil textural analysis was conducted using the hydrometer method following OM removal by hydrogen peroxide oxidation.

3.3.7 Computation of the net N-mineralisation from amended soils

The sludge N-mineralisation rate was estimated using a residual method, as described by Griffin et al. (2005) and Azeez and Van Averbeke (2010). At any sampling time, Mineral N / inorganic N was calculated as the difference in mineral N released between soil-sludge mix treatment (amended soil) and control (unamended soil) (Hanselman et al., 2004); that is:

$$\text{Mineral N}_{(t = 0;\ 1)} = \text{Mineral N}_{(\text{amended } (t=0;\ 1))} - \text{Mineral N}_{(\text{control } (t=0;\ 1))}$$

$$\text{N-mineralisation rate (NMR) (\%)} = \frac{\text{Organic N}_{\text{applied } (t=0)} - \text{Organic N}_{\text{recovered } (t=1)}}{\text{Organic N}_{\text{applied } (t=0)}}$$

Where; Organic N = Total N less mineral N (NH_4^+–N + NO_3^-–N) at *t=0* (initial) or *t=1* (final) sampling time.

3.3.8 Statistical analyses

The SAS 9.4 version statistical package was employed to assess the treatment effects on the N-mineralisation rate and other parameters analysed. The data were subjected to a one-way analysis of variance (ANOVA) at a threshold *P*-value of 0.05, with the site as a factor of six levels. Where treatment effects were significantly dissimilar, the Duncan Multiple Range test ($\alpha = 0.05$) was used to separate the means.

3.4 Results and discussion

3.4.1 Sites and soils' physico-chemical properties

The study was conducted at six sites across five agroecological zones of South Africa (Figure 3.1). The sites exhibited a wide variation in their soil types and soil properties. Table 3.1 and Figure 3.1 present the soil types and other chemical properties of the sites. In the Upington and Nelspruit sites, soils are characterised by a sandy texture, whereas Bethlehem and Durban's sites are loamy sand soils. These sites' soils were characterised by over 70% sand and little clay content whereas Johannesburg and Rustenburg sites had loam and sandy loam soils respectively with 50% sand and 50% of clay plus silt (Table 3.1).

Table 3.1: Sites and the respective soil pH, EC, and texture analyses

Site	Agro-ecological zone	pH	EC	Textural analysis			
				Clay	Silt	Sand	Textural class
			*m*S m^{-1}	%	%	%	
Upington	Arid	6.83	44	2.5	8.8	88.7	Sandy
Rustenburg	Semi-arid	5.47	9.7	13.2	33	53.8	Sandy loam
Bethlehem	Subhumid	5.52	12	10.2	13.4	76.3	Loamy sand
Johannesburg	Subhumid	6.28	8.6	17.8	33	49.2	Loam
Durban	Humid	7.25	9.5	10.3	17.9	71.8	Loamy sand
Nelspruit	Super-humid	5.66	119	3.8	7.5	88.7	Sandy

The soils' pH ranged between 5.5 and 7. pH levels in such range ideal to support soil microbial activities in agricultural ecosystems, favourable for biological processes, such as OM decomposition and N transformation, especially when coupled with favourable climatic conditions.

Figure 3.1G, H, and I present nitrogen content before sludge amendment. The total N content in the top 15 cm layer of soils in the study sites, varied between 0,07% in the humid zone of Durban to 0.2% in the subhumid zone of Johannesburg. Most of the soils had similar total N concentrations (0.07% to 0.1%), whereas the subhumid zone of Johannesburg was twice as high as the other sites.

The inorganic N was dominated with NO_3^-–N and was as high as 160 mg kg^{-1} in the super-humid zone against its NH_4^+–N of 16 mg kg^{-1} (Figure 3.1H and I). The land offered at Nelspruit station for the experiment never received fertiliser before. It was a fallow land, where grass would be allowed to grow, mowed, and left to decompose on the land over years. Attributable to this reason coupled with samples collected well in the dry season, there was little microbial activity and movement of NO_3^-–N, therefore, elevated levels of this N fraction were observed relative to other sites. The arid, semi-arid, and subhumid had around 60, 12 and 23 mg kg^{-1} of NO_3^- N respectively and these were ten, three, and four times higher in NO_3^-–N than their respective NH_4^+–N content.

For subhumid_J and humid soils, NO_3^-–N content was the same between these two zones; however, the subhumid_J had a four times higher NH_4^+–N (14.5 mg kg^{-1}) than its NO_3^-–N (3.9 mg kg^{-1}) whereas the humid zone indicated no difference between the two N fractions (Figure 3.4H and I). Other selected soil chemical parameters (Figure 3.4 and Figure 3.5) were also strongly variable among sites with subhumid of the Johannesburg site, indicating high concentration for several of the essential elements compared to other sites.

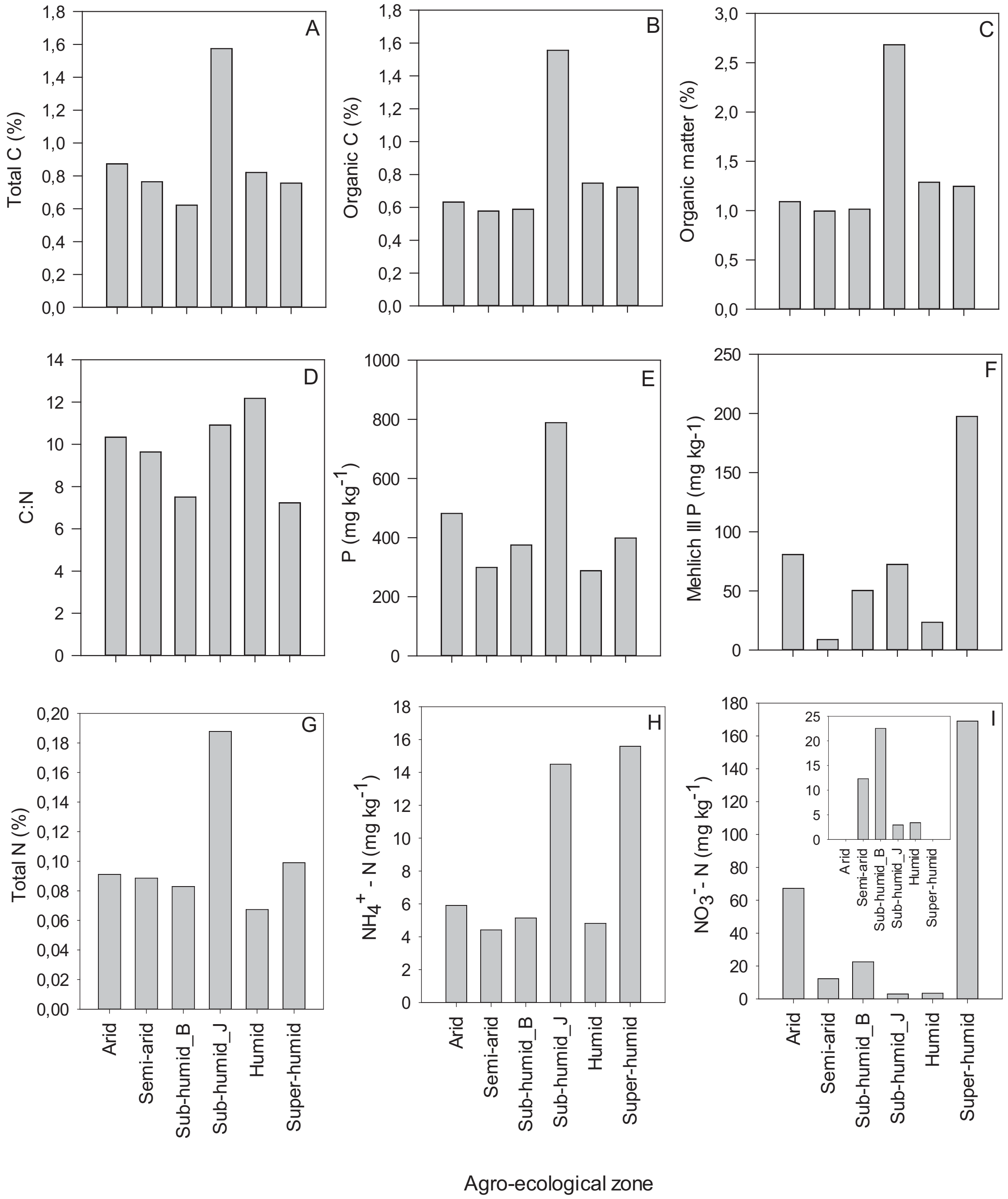


Figure 3.4: Carbon (TC & Organic C), organic matter, phosphorus (total P & MIII P) content C:N ratio and nitrogen (total N, ammonium N (NH4+ - N) and nitrate-N (NO3- - N)) of the soils per site measured before sludge amendment.

The insert on Graph I is included to zoom out the NO3- -N scale for values below 25 mg kg^{-1}.

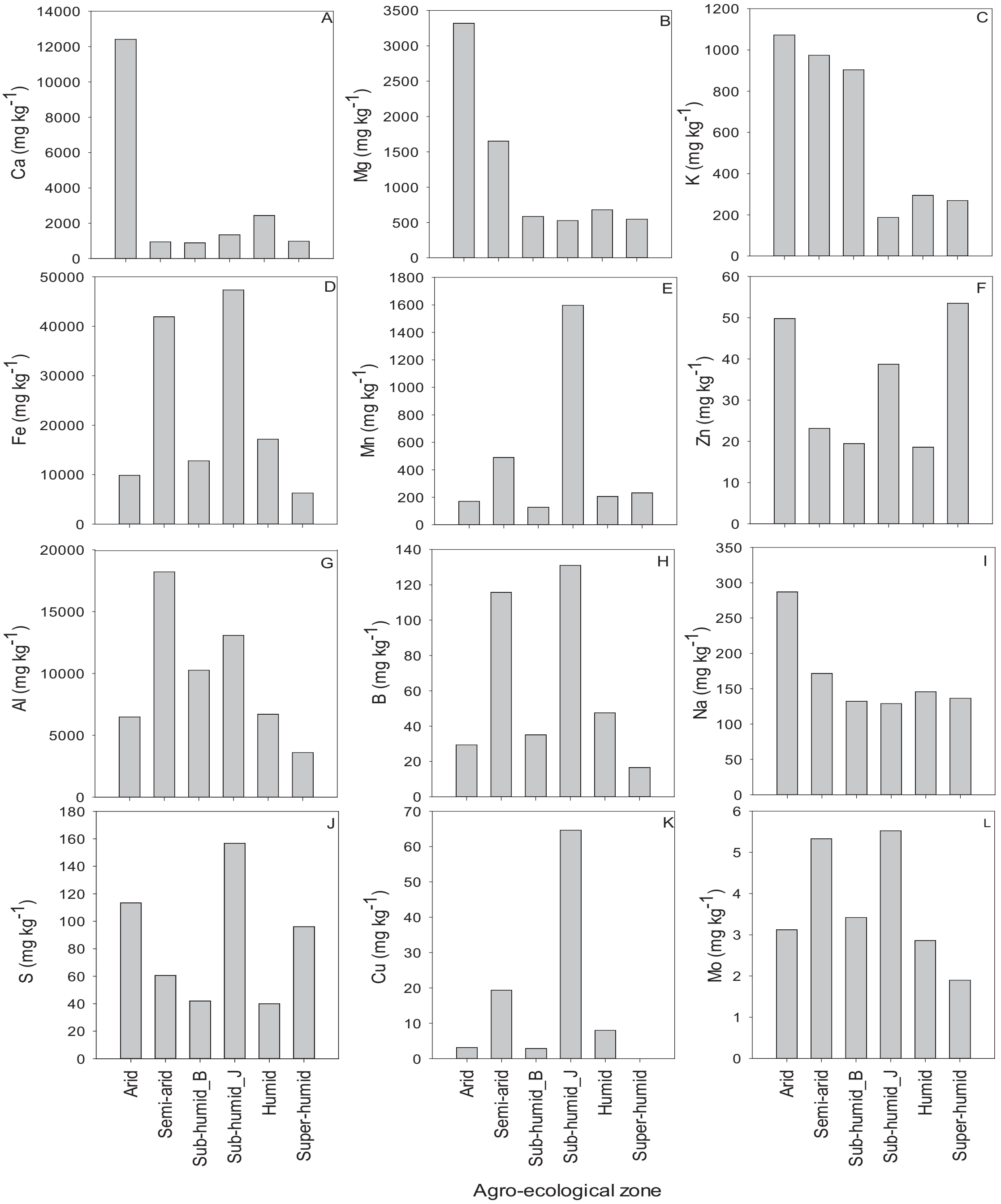


Figure 3.5: Macro-and micronutrients concentration (mg kg^{-1}) of the soils per site measured before sludge amendment

3.4.2 Sludge's chemical properties

Selected chemical properties of the sludge used in this study (Table 3.2). The sludge pH was within the recommended level that would not harm the receiving soils. A pH of 6.4 was recorded for the sludge material, suitable for promoting solubility and availability of the required nutrients for crop growth while reducing the dissolutions and uptake of heavy metal elements.

Table 3.2: Chemical properties of sludge material used in the field incubation study

Parameter	Unit	Concentration	Parameter	Unit	Concentration
$pH_{(H2O)}$		6.4			
EC	mS m^{-1}	937			
Total elemental composition					
Total N	%	4.2	Na	%	0.2
Total C	%	34	S	%	1.1
C:N		8	Al	%	1.2
OM	%	42	Fe	%	1.7
Organic C	%	25	Cu	mg kg^{-1}	242
NH_4^+ - N	%	0.7	Zn	mg kg^{-1}	1700
NO_3^- - N	mg kg^{-1}	4.7	Mo	mg kg^{-1}	13
P	%	1.9	Co	mg kg^{-1}	13
K	%	0.2	Cr	mg kg^{-1}	218
P Bray	mg kg^{-1}	23	Cd	mg kg^{-1}	3.1
Mg	%	0.5	Pb	mg kg^{-1}	180
Ca	%	2.5	V	mg kg^{-1}	31
Mn	mg kg^{-1}	763	Ni	mg kg^{-1}	112
B	mg kg^{-1}	70	Se	mg kg^{-1}	5.7

pH is a crucial factor in the agroecosystems, as it controls the availability of plant nutrients and soil microbial activities. The EC of the sludge's material was above the expected threshold level. It recorded 937 *m*S m^{-1}, therefore, 337 *m*S m^{-1} above the critical level of 600 *m*S m^{-1}. This indicates slightly higher salt content in the sludge material. The applied sludge satisfied the requirements of the guidelines governing threshold levels of heavy metals that would discredit its suitability for agricultural land application as stated in the American (Walker et al., 1994) and South African (Snyman & Herselman, 2006) guidelines. All heavy metals listed in these guidelines were below minimum concentrations that would limit using the sludge. Based on the South African sludge classification system by Snyman and Herselman (2006), the sludge material used in the current study was within pollutant Class A and qualified to be applied in agricultural lands.

The total N content of 4.2% was recorded for the sludge material. The greater portion of inorganic N was NH_4^+-N with 0.72% while NO_3^--N was less than 0.001%. Inorganic N constituted 17.1% of the total N, implying that a greater fraction (82.9%) of the total N was organic N. This agrees with previous literature, stating that a larger portion of sludge N is the organic fraction (Rigby et al., 2016; Zare & Ronaghi, 2019). An average C:N ratio of eight was observed. The application of organic materials with a high C:N ratio would delay the availability of mineral N. The sludge-use in agriculture would not affect N-mineralisation and availability of mineral N for plant uptake attributable to its low C:N ratio. The sludge used was well stabilised and would influence OM transformation and N-mineralisation.

3.4.3 Weather data

Figure 3.6 presents soil temperature data recorded during the study in the 2018 season. Temperature is one of the influential and driving factors of microbial activities within the soil environment. Nitrogen mineralisation, as a biological process, is influenced by temperature (Dessureault-Rompré et al., 2010; Ogbazghi et al., 2016). It is evident in the literature that the optimum temperature range within which high N-mineralisation could be achieved is between 25°C to 35°C (Sierra et al., 2001; Guntiñas et al., 2012; Hu et al., 2017). The soil temperatures recorded during the study were highly variable, from one agroecological zone to another. In the early days of the study, in October 2017, the temperatures were in the lower range with subhumid_B and super-humid zones, recording as low as 19°C, followed by semi-arid, subhumid_J, and humid zones with their initial temperatures being around 24°C.

The highest temperature of 33°C at the beginning of the study was observed in an arid zone (Figure 3.6). As the season progressed, the soil temperatures gradually increased in all agroecological zones to a maximum of around ±25°C for the subhumid and humid zones, ±30°C for semi-arid and super-humid, and 37°C in mid-January in the arid region. At the end of January, the temperatures dropped until the end of the study. In the final three months of the study, the soil temperatures of the subhumid and humid zones were below the optimum range for microbial activities. The temperatures in these three zones dropped faster than the arid, semi-arid, and super-humid zone (Figure 3.3).

This implies that the latter three agroecological zones had a relative advantage of an extra month of warmer soil temperatures (≥20°C) that could be favourable for soil microbial activities than the former zones. In all instances during the study period, the subhumid and humid zones had the lowest temperatures, followed by semi-arid and super-humid. The arid zone site recorded the highest temperatures for the better part of the season, though it dropped to below the super-humid zone in the final two months of the study.

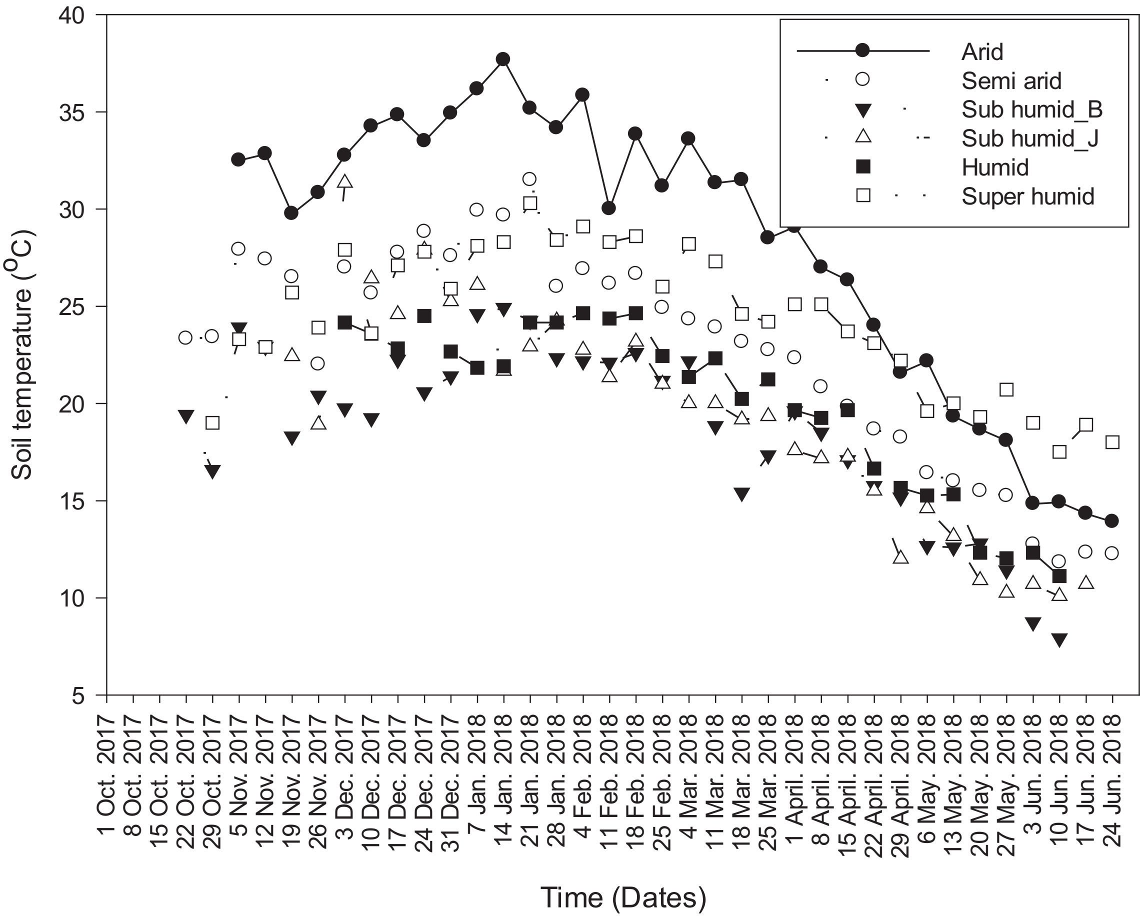


Figure 3.6: Mean daily soil temperatures (^oC) (Daily temperatures plotted on weekly interval) for the 2018 season, recorded at 15 cm depth during the field incubation period

The soil temperatures of the subhumid sites were lower than in other agroecological zones. This is attributable to the general climatic conditions of these areas, experiencing more cold and low temperatures, especially in subhumid_B, than the other zones. This site exists in comparatively higher altitudes (Table 3.3) than other sites, therefore, low temperatures were recorded.

Soil temperatures trends indicated some relatedness to the agroecological zones' mean daily air temperatures experienced during the study (Table 3.3). The two subhumid zone sites had low air temperatures, observing their mean daily temperatures hardly reaching 20^oC (Table 3.3) for the greater part of the study period except for a period between January and March 2018; however, with 1-2 ^oC colder in the second season for subhumid_B.

Table 3.4 and Figure 3.7 respectively, present the cumulative and monthly rainfall received per site during the study period. Rainfall was highly variable across sites. Concerning the general South Africa ago-ecological zones' rainfall data presented in the work by Ogbazghi et al. (2016), it was observed that the two study seasons had below normal rainfall in the arid and super-humid zones, above normal at subhumid-J, and near normal at the other sites in 2018. Except for the humid site, the second season had low rainfall received across sites, although the rainfall totals were of two rain seasons combined (Table 3.4).

Table 3.3: Mean air temperatures and altitude at each site during the study period

Site	Agroecological zone	Altitude[1] (m.a.s.l)	Mean Air Temperature (^{o}C)										
			Oct-Dec	Jan-Mar	Apr-Jun	Aug-Oct		Nov-Jan		Feb-Apr		May-Jul	
			2018			2019	2020	2019	2020	2019	2020	2019	2020
Upington	Arid	809	24.1	27.3	17	17	20	25	26	26	25	17	15
Rustenburg	Semi-Arid	1130	23.9	24.4	16.9								
Bethlehem	Subhumid-B	1721	17.7	19.7	12	14	15	21	20	19	18	11	9
Johannesburg	Subhumid-J	1583	18.8	19.7	13.5								
Durban	Humid	96	21.7	24.4	21.1	19	18	24	25	25	21	20	17
Nelspruit	Super-humid	376	22.4	25.1	20.1	21	22	25	25	25	23	19	17

[1]m.a.s.l represent metres above sea level.

Table 3.4: Cumulative precipitation during the study period at each site, compared with normal precipitation

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Site	Agroecological zone	Seasonal precipitation (mm)		
		During study period		Normal[2]
		2017-2018	2018-2020[1]	
Upington	Arid	123	74	200-400
Rustenburg	Semi-Arid	476	-	401-600
Bethlehem	Subhumid-B	789	1154	601-800
Johannesburg	Subhumid-J	1033	-	601-800
Durban	Humid	944	2139	801-1000
Nelspruit	Super-humid	748	1492	>1000

[1] Rainfall totals recorded are the combined totals of two rainfall seasons, 2019 and 2020.

[2]Observed from Schoeman and Van Der Walt (2004).

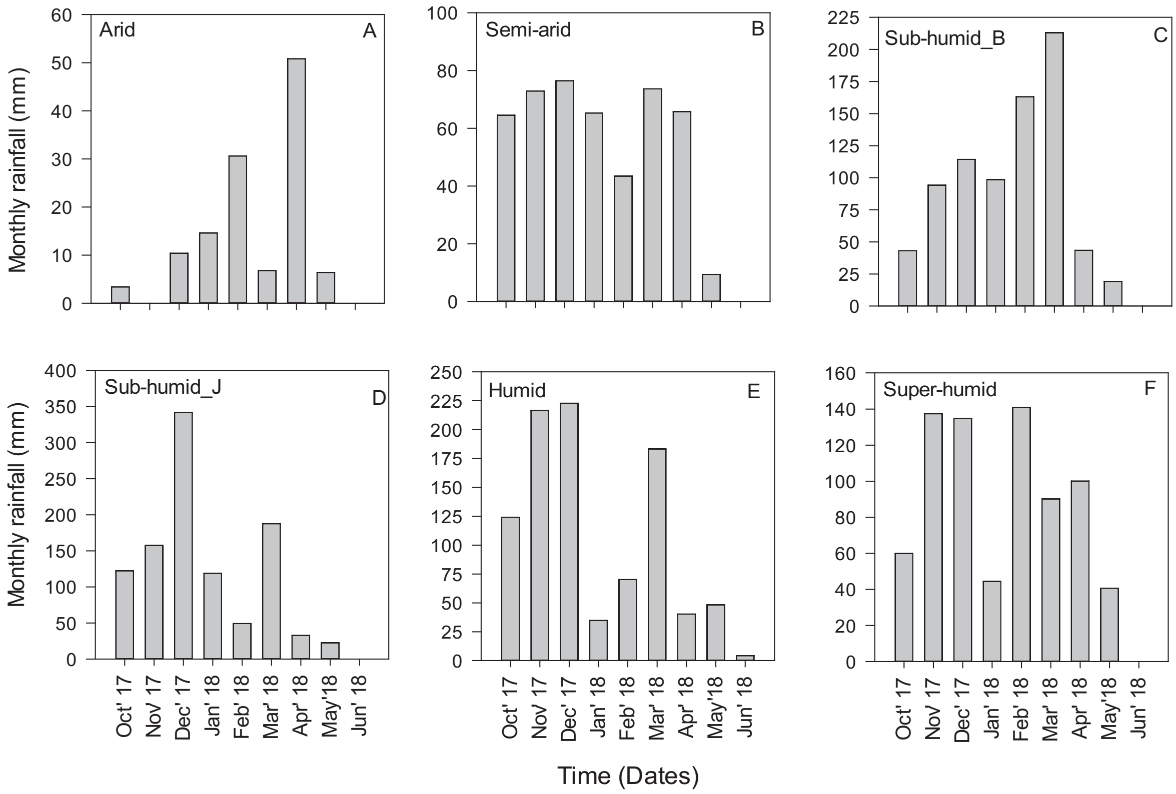

Figure 3.7: First season monthly total rainfall (mm) data recorded per site (A to F) during the incubation period

3.4.4 Total nitrogen, nitrate, and ammonium nitrogen dynamics in soil

3.4.4.1 Total nitrogen

The study site soils indicated a varied inherent total N content. The initial total N content of the control treatments (unamended) soil ranged between 676 mg kg^{-1} to 1877 mg kg^{-1} (Table 3.5a). Following sludge application at a rate of 10 tonnes ha^{-1}, the initial soil-sludge mix indicated an increase of total N content at all sites. The total N content increased with variable magnitudes, based on agroecological zones after sludge application (Table 3.5a). The expected increase in N content following sludge application was 291 mg N kg^{-1} soil based on the sludge total N concentration of 42000 mg kg^{-1} (4.2%) and a sludge application rate of 10 Mg ha^{-1}. The observed increases in total N ranged from 170 mg kg^{-1} in the Durban site of humid to 955 mg kg^{-1} in the Johannesburg site (subhumid).

In Rustenburg (semi-arid), Bethlehem (subhumid), Durban (humid), and Nelspruit (super-humid), the changes in total N content were similar in magnitude across these sites (Table 3.5a) and close to the expected increase. The humid zone had the lowest change in total N content post sludge application with 25% of the soil-sludge mix initial total N content being contributed by the sludge. This was followed by the semi-arid and super-humid zones, both recording 27%

while the subhumid_B had 30% as the total N change realised from sludge application. The highest total N change was observed in the arid and subhumid_J zones where 40% and 51% of their total N in soil-sludge mix initial samples was contributed by sludge respectively relative to their control initial samples (Table 3.5a).

At the end of the first season of the incubation in 2018, the total N content of the soil-sludge mix in the ceramic tubes decreased variably among sites. The magnitude of decrease was low in the 2020 season, relative to 2018. The highest total N decrease was observed at the Nelspruit site in the super-humid zone with a magnitude of 51%, followed by a 37% reduction in Durban. Upington, Johannesburg, Rustenburg, and Bethlehem sites indicated a reduction in total N by 12%, 21%, 23%, and 27% respectively; the opposite was observed by the end of the 2020 season (Table 3.5a).

Table 3.5: The initial (measured at beginning of the study) and the end of sampling season total N, nitrate – N and ammonium – N concentrations for the unamended soil (control) and soil – sludge mix (treatment) across agroecological zones

Site	Agroecological zone	Unamended soil			Soil-Sludge Mix		
		Sampling time					
		Initial	Year 2018	Year 2020	Initial	Year 2018	Year 2020
a. Total N (mg kg^{-1})							
Upington	Arid	920(34)[a]	872(10)	683(75)	1291(83)	1134(124)	793(15)
Rustenburg	Semi-Arid	897(92)	740(107)	nd	1135(82)	874(4)	nd
Bethlehem	Subhumid-B	829(60)	642(13)	613(67)	1077(104)	781(31)	663(75)
Johannesburg	Subhumid-J	1877(104)	1747(78)	1670(45)	2832(91)	2247(65)	1860(165)
Durban	Humid	676(17)	455(65)	440(66)	846(37)	536(90)	513(86)
Nelspruit	Super-humid	990(57)	542(135)	530(92)	1259(32)	623(105)	603(90)
b. Nitrate – N (mg kg^{-1})							
Upington	Arid	67(20.3)	82(6.6)	40.26(5.46)	129(7.56)	90(6.61)	40.01(3.2)
Rustenburg	Semi-Arid	12(0.45)	2(0.47)	nd	16(7.23)	3(0.23)	nd

Bethlehem	Subhumid-B	23(9.41)	2(0.48)	0.33(0.29)	33(3.21)	3(0.58)	1(0.3)
Johannesburg	Subhumid-J	3(0.26)	4(0.97)	6.07(0.82)	6(0.43)	4(0.34)	6.33(1.8)
Durban	Humid	3(0.15)	2(0.27)	4.54(1.31)	3(0.28)	3(0.27)	5.85(0.63)
Nelspruit	Super-humid	172(13.58)	3(0.71)	4.1(0.84)	229(5.45)	3(0.52)	6.36(1.63)

c. Ammonium – N (mg kg^{-1})

Upington	Arid	6(0.59)	1(0.28)	7.54(1.96)	35(2.09)	1(0.31)	9.5(1.76)
Rustenburg	Semi-Arid	4(0.8)	1(0.16)	nd	54(2.3)	2(0.76)	nd
Bethlehem	Subhumid-B	5(1.35)	0.4(0.06)	4.34(0.42)	33(2.98)	0.8(0.43)	4.97(1.75)
Johannesburg	Subhumid-J	15(1.82)	2(0.39)	16.08(2.7)	42(4.88)	2(0.91)	18.98(3.03)
Durban	Humid	5(0.88)	2(0.31)	12.06(0.4)	24(8.39)	1(0.14)	11.47(0.37)
Nelspruit	Super-humid	16(0.85)	2(0.57)	5.63(0.57)	49(3.41)	2(0.55)	6.25(0.78)

[a] Number in parenthesis are standard deviations of means (n = 3).

nd = no data

The declining total N from the initial concentrations confirms various N transformation processes occurring during the incubation period. Such processes were indicated to have decelerated in the second season. During incubation, several N transformation processes occur, such as mineralisation, immobilisation, ammonification, and nitrification. These processes would change the total N into distinct forms that could either be NH_4^+–N or NO_3^-–N fractions, therefore, exposing N to diverse pathways causing it to leave the soil systems. NO_3^-–N can be leached out of the system as mineral N or denitrified and would be lost as ammonia gas through volatilisation. Volatilisation was unlikely under the incubation conditions where the tubes were buried underground. As a cation, the NH_4^+ – N fractions could either be retained at the soil exchange sites or transformed through the act of nitrifying bacteria (nitrification), converted into nitrate and lost through leaching (Azeez & Van Averbeke, 2010).

3.4.4.2 Inorganic nitrogen

The study sites' soils had dissimilar magnitudes of native NH_4^+–N (Figure 3.4H and Table 3.5c) and NO_3^-–N concentration (Figure 3.4I and Table 3.5b). The unamended soils of Upington and Nelspruit sites had the highest NO_3^-–N content with 67 and 172 mg kg^{-1}, respectively, while the other sites recorded less than 25 mg kg^{-1} (Table 3.5b). The high native NO_3^-–N concentrations observed at these sites could be attributed to Upington being in an arid zone associated with erratic rains and limited leaching, therefore, NO_3^-–N accumulation. Nelspruit site is on previous grass-covered land that would be cut and left to decompose, resulting in NO_3^-–N builds up. Sampling on sites was conducted in spring (dry period) with no rains, while the NO_3^-–N leached off the soil system before sampling.

The observed sludge NO_3^-–N at application was extremely low (4.7 mg kg^{-1}). The initial soil–sludge mix indicated higher NO_3^-–N than expected; 129 mg kg^{-1} NO_3^-–N was recorded after sludge application in the arid zone. The Johannesburg site in the subhumid had exceptionally low native nitrate concentration (2.94 mg kg^{-1}); it only indicated a two-time increase of NO_3^-–N from sludge application (Table 3.5b) while in Durban (humid) there was no increase in this form of N; the site recorded a decrease in NO_3^-–N. At Rustenburg (semi-arid), Bethlehem (subhumid) and Nelspruit (super-humid) sites 4, 10 mg kg^{-1}, and fifty-seven were realised after sludge application respectively (Table 3.5b). Some N transformation such as nitrification is possible, occurring between the time of sludge addition, sampling, and the time of analysis, therefore, higher observed NO_3^-–N relative to the expected sludge contribution in the soil-sludge mix, particularly for Upington and Nelspruit.

By the end of the incubation period of the 2018 season, most of the sites, especially those in medium to high rainfall receiving areas, had small concentrations of NO_3^-–N. Most of the sites recorded below 5 mg kg^{-1} for their final NO_3^-–N content by the end of the first season, except for the Upington site, retaining 90 mg kg^{-1}. This was an opposite scenario by the end of the 2020 season, observing a little higher NO_3–N content across sites relative to 2018. High rainfall in the semi-arid, subhumid (Bethlehem), and super-humid was possibly the crucial factor, promoting the loss of NO_3^-–N in these agroecological zones, compared to the Upington site in the arid zone for the 2018 season.

Figure 3.4H and Table 3.5c present the concentrations of observed NH_4^+–N across study sites. Johannesburg and Nelspruit had the highest native NH_4^+–N of 15 and 16 mg kg^{-1}, respectively (Table 3.5c). All other sites recorded NH_4^+–N between 4 and 6 mg kg^{-1}. Sludge applications increased NH_4^+–N by an average of 31 mg kg^{-1} across sites, ranging from 19 mg kg^{-1} at Durban to 50 mg kg^{-1} at Rustenburg. The expected increase in NH_4^+–N concentration was 50 mg kg^{-1}, based on the sludge NH_4^+–N concentration of 7200 mg kg^{-1} (0.72%), and a sludge application rate of 0.45 g per 65 g soil (10 Mg ha^{-1}).

Contrary to the trend observed on NO_3^-–N losses highly varying from site to site, NH_4^+–N losses were similar in magnitude across all sites, ranging between 94% and 97% by the end of the first season study period. By the end of the 2018 study period, both unamended soils (control) and soil-sludge mix treatments from the buried ceramic tubes, recorded exceptionally low concentrations of NH_4^+–N and NO_3^-–N compared to the concentrations measured at the beginning of the study. It is, therefore, evident that larger fractions of inorganic N left the soil system during the incubation period, especially in the 2018 than the 2020 season. The strongest viable way where NH_4^+–N could have been lost is through nitrification, resulting in NO_3^-–N leaching of the system or, to a lesser extent, through fixing by soil microbes.

3.4.4.3 Organic nitrogen

Figure 3.8 presents the measured organic N (Initial) just after sludge application and its concentration at the end of two sampling seasons study period. Sludge application led to varying proportions of organic N, differing with the site and agroecological zone attributable to their inherent dissimilarities in soil OM. Initially, sludge application observed 280 mg kg^{-1} of organic N being added in an arid region, 185 mg kg^{-1} and 210 mg kg^{-1} added to the semi-arid and Bethlehem (subhumid), respectively; while 151 mg kg^{-1} was recorded in Durban (humid), and 178 mg kg^{-1} was observed at Nelspruit (super-humid). The highest organic N was recorded at Johannesburg (subhumid) with 923 mg kg^{-1}. Over the incubation period, organic N decreased significantly among the agroecological zones with larger magnitudes of decrease recorded in 2018, relative to the 2020 season. The decrease in organic N between the initial and the ultimate values recorded in Figure 3.8 implies the N transformation.

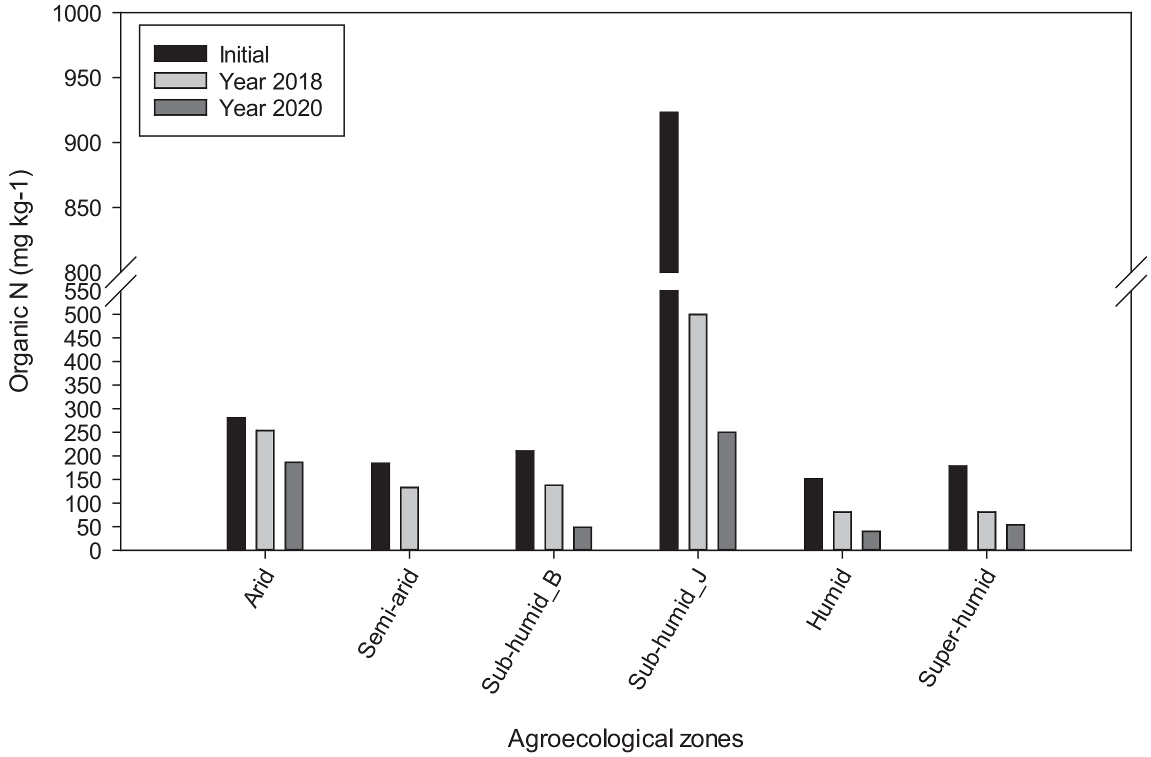

Figure 3.8: The initial (applied plus soil inherent organic N measured at the beginning of the study) and the final (measured at the end of 2018 and 2020 seasons) organic nitrogen in mg kg^{1} for soil-sludge mix treatments across agroecological zones

Transforming organic N into inorganic N during field incubation would cause mineral N to leave the soil system through pathways, such as leaching or denitrification, or be retained in the soil colloid exchange sites, which could be observed through laboratory analyses. The decrease in organic N occurred at dissimilar magnitudes, based on agroecological zones, ranging from 10% to 57% in 2018 and between 12% and 42% in 2020 (Figure 3.9). A smaller percentage of organic N transformation was recorded in the arid region. This site, therefore, retained 90% of its initially applied organic N proportion by the end of the 2018 season period. The same scenario of exceedingly small organic N decrease was also observed in the super-humid zone in the second season than all other sites.

These scenarios are possible to have been driven by below normal rains they received during the incubation period (Table 3.4), therefore, presenting limited soil moisture required to influence soil microbial activity and action on the degradation of organic N. The slowed organic N decrease across sites in the second season implies that most of the fast-degradable organic N and OM were transformed during the first season. As a result, the residual N and OM carryover in the second season could slowly degrade; therefore, the limited change of organic N decrease in the 2020 season.

3.4.5 Sludge mineralisation under field conditions

3.4.5.1 Net N-mineralisation of applied sludge

Estimating N-mineralisation under field conditions is essential for N management in the agricultural cropping systems. Figure 3.9 presents the net N-mineralisation rate of sludge-amended soils at varying sites in fluctuating climatic conditions. The seasonal N-mineralisation was estimated as the difference between the initial and final organic N content as a percentage of the initially applied organic N. Field incubation annual N-mineralisation ranged from 10% to 57% in 2018 and 12% to 42% at the end of the 2020 incubation period. The highest N-mineralisation of 57%, was recorded in the super-humid Nelspruit region.

In the second season of 2020, the same site recorded a 12% annual N-mineralisation only. The second highest annual percentage N-mineralisation was recorded in the humid region of Durban (48%), followed by the subhumid region of Johannesburg with 46%, and Bethlehem with 34%. Johannesburg recorded an annual percentage N-mineralisation of 46%, 12% higher than Bethlehem (34%), despite both sites being under the same agroecological zone in the first season. While the lowest mineralisation occurred in the arid region of Upington (10%), the semi-arid area of Rustenburg had a three times higher net annual percentage N-mineralisation (31%) than Upington. The net annual percentage N-mineralisation varied significantly (p = 0.001) among agroecological zones in the 2018 season, while no significant difference was observed in the 2020 season (Figure 3.9). Relative to the annual percentage N-mineralisation observed in the 2018 season, there is an agreement with previous model simulation studies by Ogbazghi et al. (2016) for some similar agroecological zones.

Net annual percentage N-mineralisation was minimal in the second season of 2020. Considering that N-mineralisation after the first season was from the residual organic N of the initially applied N, the percentage became low attributable to a reduced amount of organic N to be mineralised and limited OM to be degraded. The second season annual percentage net N-mineralisation from this study are, however, related to previously reported rates from the second-year net N-mineralisation of residual organic N (Cogger et al., 2004).

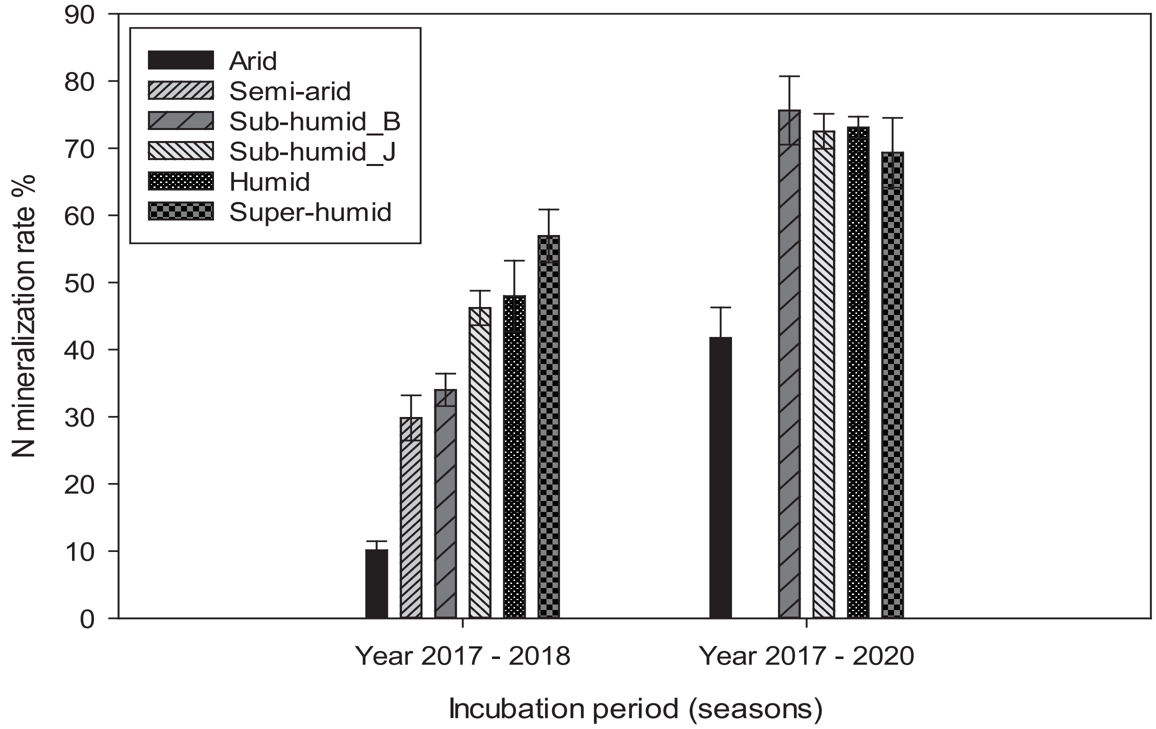

Figure 3.9: Net N-mineralisation (%) during the field incubation study per agroecological zone. Error bars = standard deviation of replications around the mean (n = 3)

The annual percentage net N-mineralisation did not differ significantly between the semi-arid and subhumid in Bethlehem. No statistical difference was observed in the annual percentage net N-mineralisation among subhumid in Johannesburg and humid in Durban in 2018. In the second season, a statistical difference was observed between super-humid and all other agroecological zones with the former, recording the smallest annual percentage net N-mineralisation of 12% relative to 25% and above, observed in all other zones.

A magnitude difference existed among these agroecological zones in 2018, minimal in the 2020 season. In other field studies, Eldridge et al. (2008) reported 54%, 48%, and 45% mineralisation of applied organic N over 12 months period of incubation from 12, 24, and 48 tonnes ha^{-1} application of granulated sludge, respectively. Hanselman et al. (2004) observed 18% to 20% mineralisation from organic N added through sludge within three days of application in various *in situ* incubation methods, with the highest percentage (47%) recorded at day forty-five after sludge application in loamy soils. Their associated laboratory study recorded the highest mineralisation of 66% by the end of 180 days of study.

The N-mineralisation was influenced by climatic factors where rainfall was observed as the primary driver of N transformation. This saw the magnitude of the annual net N-mineralisation correlated with the amount of rainfall received in an area. The drier regions exhibited a low annual N-mineralisation; however, the percentage increased with the increase in rainfall, observed from the Rustenburg site (semi-arid) to the super-humid zone of Nelspruit. The high annual percentage N-mineralisation at the Johannesburg site that did not vary significantly with humid zone could be attributable to its high rainfall received over the season, high initial OM, and C content of the site (Figure 3.4A to C), and possible excess sludge applied. High C content would have influenced the proliferation of soil microbial population, enhancing microbial activities because of high energy and food supply (Azeez & Van Averbeke, 2010; Rigby et al., 2016), therefore, high organic N transformation and subsequently high OM decomposition.

This site received a high and well-distributed rainfall over the study period (Table 3.4 and Fig 3-4D). High water availability could have created an extended period of favourable conditions for N-mineralisation than the other agroecological zones. The findings of the current study concur with some previous studies (Paul et al., 2003) from which a direct positive relationship between water content and N-mineralisation was noticed. The N-mineralisation of the current study presents a similar pattern to the results observed in a modelling study by Ogbazghi et al. (2016). This study observed an increase in the percentage net N-mineralisation with an increase in wetness.

3.4.5.2 Mineralised N, implications, and nitrogen management

The decline in soil inherent and sludge applied organic N during the incubation period indicates that decomposition occurred and, subsequently, N-mineralisation occurred. Based on the amount of organic N applied per site, 27, 41, 78, 423, 71, and 103 mg kg^{-1} of the applied organic N were mineralised in the 2018 season (Figure 3.10) and these translate to 10%, 31%, 34%, 46%, 48% and 57% N-mineralisation rate for arid, semi-arid, subhumid_B, subhumid_J, humid and super-humid agroecological regions respectively against 27 mg kg^{-1} to 240 mg kg^{-1} in 2020 season (Figure 3.10). Smaller magnitudes of mineralised N were recorded in the second season in subhumid_J, humid, and super-humid with 240, 41, 27 mg kg^{-1} respectively and higher than 2018 season for arid (111 mg kg^{-1}) and subhumid zone (89 mg kg^{-1}) (Figure 3.10).

Of the various magnitudes of mineralised N observed in the study, only small fractions of the inorganic (NH_4 + NO_3) N were recovered from the soil and soil-sludge mix in the ceramic tubes. The inorganic N proportions retained were only 17.8%, 0.98%, 1.27%, 0.21%, 0.19% and 0.33% of the total mineralised N (Table 3.5b/c) for the arid, semi-arid, subhumid_B, subhumid_J, humid and super-humid agroecological zones respectively in 2018 season. Relative to other agroecological zones, the proportion of mineralised N retained in the ceramic tubes was highest in the arid zone of Upington and low at all the other sites. The Upington site of the arid region retained high levels of mineralised N attributable to lack of water to leach off the mineral N from the soil system since it received low rainfall (Table 3.4) throughout the 2018 season compared to other regions. An opposite trend was observed in the 2020 season, where higher values of inorganic N were recorded relative to the 2018 season. The observed higher inorganic N values in the second season relate to low total rainfall received across sites compared to the rains received in the 2018 season.

The experimental sites were kept weed-free during the study period. Since the contents were enclosed in the tubes, therefore, losing inorganic N was not attributable to uptake by any plants. The ceramic tubes are porous and allow water and air movement between the soil inside and outside the tubes. This, therefore, mimics the natural scenario in the fields, where nitrate mobility within soil layers and ammonia volatilisation and denitrification losses from the soil to the atmosphere are common phenomena. The most probable cause of losses was attributable to leaching or denitrification pathways.

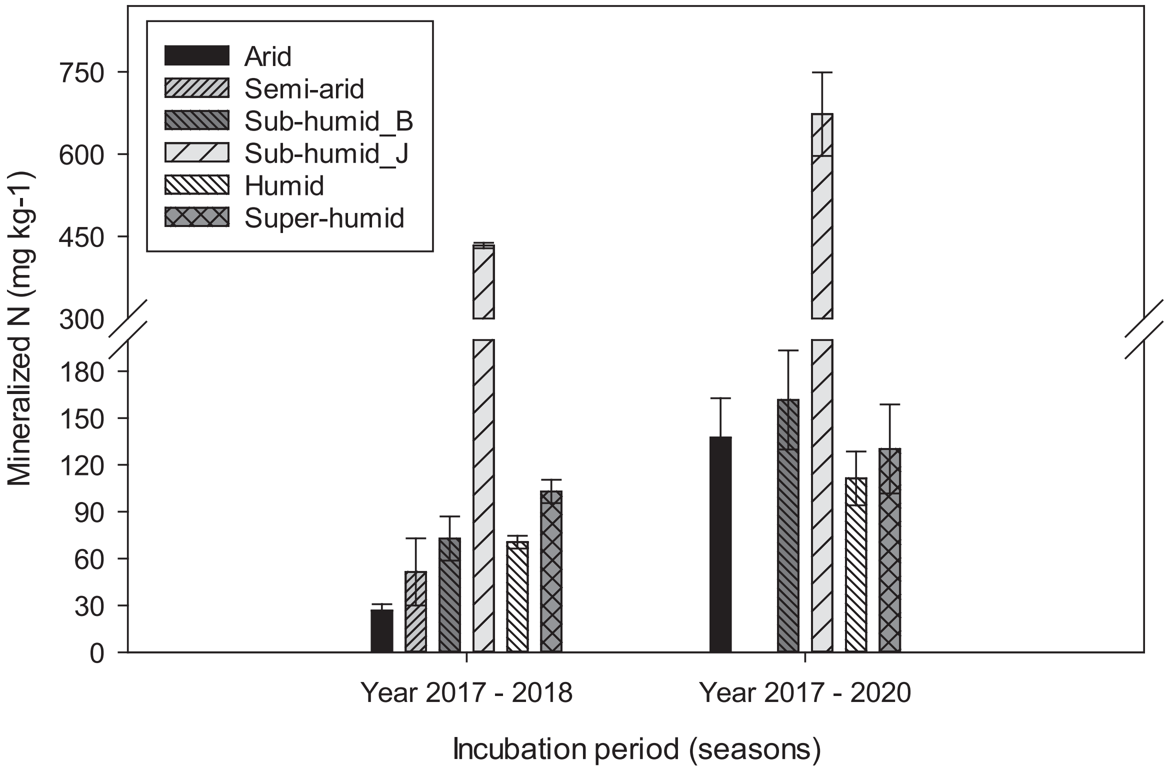

Figure 3.10: Mineralised N (mg kg^{-1}) for the field incubation study per agroecological zone. Error bars = standard deviation of replication means (n = 3)

Well-planned N management strategies are essential when using organic materials, such as sludge, as N sources in cropping systems. Fertilisation programmes need to be strongly scrutinised and ascertained to minimise the challenges of N losses, especially through leaching. If the application is conducted randomly, this would cause contamination of environments and pollution of underground and surface waters. Aspects, such as crop N requirements, soil types, and texture of soils, to which the sludge is targeted, and N-mineralisation rate should be important in N fertilisation programmes.

3.4.6 Validation of SARA model nitrogen mineralisation assumptions using field studies from across South African agro-ecological zones

A good agreement exists between the trends of the model predicting the N-mineralization produced by Ogbazghi et al. (2016), used in the SARA model, and the field estimates in this study (Table 3.6). There were some differences between model predicted values and those obtained from the field studies. These differences are attributed to the variation in rainfall and temperature experienced in the field study compared with the weather data used for model simulation by Ogbazghi et al. (2016). Field N-mineralization (%) measurements during the first year of the study were similar to those used by the SARA model for semi-arid region (Rustenburg), subhumid region (Bethlehem), and humid region (Durban). While the N-mineralization values of the SARA model were lower than field measurements in super humid (Nelspruit), subhumid (Johannesburg), and arid (Upington) regions.

Table 3.6: Net N-mineralisation at each site determined by field incubations and model predictions

Site	Agroecological zone	Net N mineralization[1]				
		Model Prediction[2] (%)			Field Incubation (%)	
		First year	Three years	Six years	2017-2018	2017-2020
Upington	Arid	24 (1.3)	35(2)	40(2)	10 (2.4)	42 (7.8)
Rustenburg	Semi-Arid	28 (1.9)	40(2)	45(3)	29 (5.8)	-
Bethlehem	Subhumid-B	29 (1.5)	-	-	34 (4.2)	76 (8.8)
Johannesburg	Subhumid-J	29 (1.5)	44(5)	52(10)	46 (4.4)	73 (4.5)
Durban	Humid	37 (2.7)	52(6)	57(7)	48 (9.2)	73 (2.5)
Nelspruit	Super-humid	42 (1.1)	58(9)	65(9)	57 (6.8)	69 (9.1)

[1] Figures in parentheses represent standard deviation from the mean, n = 3.

[2] Mean values as observed by Ogbazghi et al. (2016).

Rigby et al. (2016) detail potential factors influencing N-mineralization, such as rainfall, temperature, soil texture, and pH. The observed variation in mineralisation rates, especially in the subhumid (Johannesburg) region, between model simulations used by SARA model and the current field study, could be driven by the differences in rainfall besides the possible differences in the N content of sludge used in the study. Rainfall performs an important function in promoting N-mineralisation. In the current field study, the super-humid zone received low rainfall (748 mm) than the subhumid in Bethlehem (789 mm) and Johannesburg (1033 mm) and humid (944 mm) agroecological zones (Table 3.4) in the first season. This could have been the reason behind the non-significant differences in field N-mineralisation measurements among the subhumid, humid, and super-humid zones (Figure 3.7).

The current study also had two sites representing the subhumid zone (Bethlehem and Johannesburg) attributable to the differences in temperatures of these sites. The N-mineralisation rate in Johannesburg (46%) was 12% higher than that of Bethlehem (34%) during the first incubation year. This variation, however, disappeared towards the end of the three-year study, with both sites scoring similar cumulative N-mineralisation percentage (Bethlehem 76±8.8; Johannesburg 73±4.5). The initial variation observed between these two sites in the same agroecological zone could be attributed to the differences in rainfall, temperature, and edaphic properties. The three months mean temperatures recorded during the study (Table 3.3) indicated minimal difference (<1.5^{0}C) between the two sites.

The mean monthly temperatures strongly indicate that the Bethlehem site in the cold climatic zone and the month-to-month temperature differences ranged between 0.2^{0}C to 2^{0}C lower than the Johannesburg site. Temperature variation was also reported in the work of Ogbazghi et al. (2016), observing that Bethlehem temperatures were lower than Johannesburg, with a range of 2^{0}C to 7^{0}C per year. This is supported by the elevation of the two sites where Bethlehem is at an altitude of 138 m higher than Johannesburg (Table 3.3). Rainfall variation, with these temperature differences, could significantly explain the 12% N-mineralisation variances between these two sites. Similar trends were reported by Ogbazghi et al. (2016) where this was attributed to relatively higher rainfall and temperature. Johannesburg site received the highest rainfall among all sites and had favourable air temperature during the first study year (Table 3.3 and Table 3.4), while Bethlehem had 250 mm lower rainfall than the former, characterised with comparatively low temperatures (Table 3.3).

Although Nelspruit had an inferior rainfall compared to subhumid and humid sites over the first season, the site recorded 57% N-mineralisation during this study. This was the highest compared to other sites and is 15% higher than the modelled results. This site was in the grass before the experiment and then converted to bare soil. Its grass would be left to decompose on the site, stimulating microbial activity and mineralisation, leading to the high initial NO_3^-–N values. A closer observation of the initial and final total N values for the unamended soil at Nelspruit (Table 3.5a) indicated a large decrease in comparison with the other sites, suggesting greater mineralisation of N from the soil. This is consistent with the conversion from grass, suggesting increased microbial activity. The increased microbial activity might also have led to increased mineralisation of the sludge, resulting in the high mineralisation rate observed.

Temperature and soil moisture were reported as major drivers of N-mineralisation in previous studies (Wang et al., 2006). Ogbazghi (2016) had a scenario indicating an increase in rainfall levels, while the air temperatures remained low for the arid and semi-arid regions. This indicated an insignificant influence on N-mineralisation in response to the increased rainfall with low air temperatures. A similar scenario was observed during the current field study. Bethlehem site received high rainfall; however, it was associated with low temperatures experienced over the study period, resulting in lower N-mineralisation than the Nelspruit site, which had a comparatively low rainfall (40 mm less than Bethlehem) but with favourable temperatures, promoting N-mineralisation.

Guntiñas et al. (2012) reported a direct influence of temperature on N-mineralisation, while Song et al. (2018) noted a rise in N-mineralisation, nitrification, and ammonification when the temperatures were elevated. Tejada et al. (2002) revealed that N-mineralisation was more profound at 25°C than at 15°C. Dridi and Gueddari (2019) attributed low inorganic N from N-mineralisation to low temperatures from January to February in their study in Tunisia.

The N-mineralisation rate variation among the agroecological zones strengthens that a "*one-size-fits-all*" idea in sludge application rates could not be the best to implement in N management strategies. Although crop N demand could be low in arid regions relative to areas receiving moderate to high rainfall, to provide adequate crop N requirement in drier regions from sludge application. The application could be supplemented with commercial mineral fertilisers if needed during dry years, or the application should be based on crop N requirements. These strategies would supplement the nitrogen divergence that could be created by the low N-mineralisation experienced in such regions, especially in the first year of application.

3.5 Conclusions

The results of this field incubation study indicated that the N-mineralisation rate varies with agroecological zones and climatic conditions. The results suggest that higher N-mineralisation rates should be expected in high rainfall receiving areas and low in arid regions. There was a good agreement in the trends and some agroecological zones the rates of the field estimation of N-mineralisation from the current study across South African agroecological zones. The findings indicate that sludge recommendations generated by the SARA model are reasonable and robust and could be a decision support device to implement the South African sludge guidelines regarding sludge application in agricultural lands. The study findings indicate that rainfall and temperatures perform a major function in N-mineralisation. The study findings strongly support previous model simulations by SWB-Sci that a single sludge rate is not applicable across sites of fluctuating climatic characteristics in meeting crop N requirements. The study also indicated that the residual method of determining N-mineralisation, based on the differences between initial and final organic N, is reliable.

CHAPTER 4: VALIDATION OF SARA MODEL SLUDGE RECOMMENDATION USING INORGANIC FERTILIZER AS BENCH MARK: LYSIMETER STUDY

4.1 Abstract

Sludge land application based on crop N requirement is widely adopted to safeguard the environment. N availability to crops depends on the mineralisation rate determined by climatic factors. The SARA model was developed to estimate site-specific sludge application rates. A two-year lysimeter study was conducted to quantify maize yield and nitrate leaching from sludge applied according to recommendations by the SARA model using commercial inorganic fertiliser as benchmark under two rainfall regimes simulating (humid and subhumid agroecological zones). Four treatments (inorganic fertiliser + humid rainfall, inorganic fertiliser + subhumid rainfall, sludge + humid rainfall, sludge + subhumid rainfall), each treatment replicated three times, were randomly allocated to 12 lysimeters. Results indicated that maize grain yield and cumulative nitrate leaching varied across treatments.

Maize grain yield did not differ significantly between sludge treatments and inorganic fertiliser treatments for both seasons and rainfall regimes, except during the second season under the simulated subhumid zone, which received lower rainfall compared to the first year. Nitrate leaching was higher under the humid compared with the subhumid zone for both growing seasons. The difference was, however, only significant ($P<0.05$) during the second growing season. It was also apparent that nitrate leaching was higher from inorganic fertiliser treatments than sludge treatments in the subhumid zone and sludge treatments than inorganic fertiliser in the humid zone for both years.

The difference was significant ($P<0.05$) during the second season. To conclude, sludge applied according to crop N demand as estimated by the SARA model produced comparable maize grain yield with commercial inorganic fertiliser without significant negative effects on the environment through nitrate leaching below the root zone under the subhumid zone. Further study is recommended to ascertain the assumption that because of higher temperature in the humid agroecological zone, maize grain yield is expected to be higher from sludge application than inorganic fertiliser accompanied by less nitrate leaching.

4.2 Introduction

Waste recycling and protecting water bodies are essential to environmental sustainability. With an end goal of achieving this, the wastewater treatment plants (WWTP) are committed to developing strategies to protect the environment from the ever-increasing wastes emanating from the industry. Sludge solid waste from WWTP comprises high OM content between 45% to 65% (Yang et al., 2018; Kominko et al., 2017; Haynes et al., 2009; Snyman & Herselman 2006; Rowell et al., 2001), and varying components of essential plant nutrients with nitrogen constituting the greatest percentage of 1.7% to 5.8% as compared to 0.4% to 4.1% phosphorus and 0.1% to 1.1% potassium (Snyman &Van Der Waals, 2004; Smith & Vasiloudis, 1991).

Nitrogen is identified as the most limiting nutrient in crop production because it is a major constituent in amino acids and nucleic acids with essential functions of chlorophyll production for photosynthesis; however, soil nitrogen occurs in quantities less of crop demand (Brady & Weil 2013) and, therefore, must be added as inorganic fertiliser or organic manure. Absorption of nitrogen by plants occurs in forms of nitrate (NO_3^-) and ammonium (NH_4^+) ions, and nitrate being highly mobile poses a threat to groundwaters, especially when nitrogen is applied excessively (Paramashivam et al., 2016; Esteller et al., 2009; Daniels et al., 2003) and/or under high rainfall/irrigation events following fertiliser applications (Tesfamariam et al., 2015; Elfving 1982).

To safeguard groundwaters from nitrate pollution, sludge is land applied for agricultural production based on the nitrogen demand of crops. With best management practices, restrictions on N inputs from sludge are placed by several international communities, such as the United Kingdom (UK), United States of America (USA), and Australia, as reviewed by Rigby et al. (2016). Sludge application rates in these countries are estimated using plant-available N (PAN) and must not exceed crop N requirements. The UK takes considers soil type and seasonal rainfall, and nitrate vulnerable zones where the maximum application rate is 250 kg ha^{-1} total N.

These guidelines and restrictions are hinged on that the total N in sludge contains 70% organic N (Rowell et al., 2001). Inorganic N must be released through microbial-mediated decomposition/mineralisation for crop uptake. The released inorganic N is absorbed by plants, leached below the root zone in the presence of excess water, or lost in gaseous forms through volatilisation or denitrification processes. These processes are complex in regulating the nitrogen balance in such agricultural systems and are associated with soil pH, aeration, moisture, and temperature directly or indirectly influenced by climatic variations, such as rainfall and atmospheric temperature (Ogbazghi et al., 2016; Er et al., 2005; Wang et al., 2004).

Rainfall varies enormously across ecological zones within a geographical region. For instance, in South Africa, the mean annual rainfall varies between <200 mm to >1000 mm across the six agroecological zones: Desert, arid, semi-arid, subhumid, humid, and super-humid (Schoeman & Van Der Walt, 2004). In a model simulation study, a consistent trend was established between cumulative annual N-mineralisation from sludge and rainfall across the agroecological zones (Ogbazghi et al., 2016), indicating the significance of rainfall on N leaching in these zones as further reported by Ogbazghi et al. (2019). Following the findings from previous studies in South Africa suggesting site-specific sludge application rates (Tesfamariam et al., 2015), successive studies were launched to accommodate the wide range of climatic conditions.

Using the SWB-Sci computer model, scenario simulations of N-mineralisation, mean annual maize yield, and nitrate leaching were run based on an upper limit of 10 t ha^{-1} for all South

African agroecological zones (Ogbazghi et al., 2016). The results from the scenario simulations were used to develop a database computer model (SARA model), estimating the sludge application rate considering the agroecological zone, crop type, crop target yield, method of sludge application, cropping system, sludge, and initial soil physiochemical properties, and N-mineralisation rates of sludge used. It was recommended to validate the SARA model recommendations and potential negative environmental effects through nitrate leaching through field studies across agroecological zones and controlled lysimeter studies that simulate varying rainfall regimes using commercial fertiliser as a benchmark. This study aimed to quantify maize yield and nitrate leaching from sludge applied according to recommendations by the SARA model, using commercial inorganic fertiliser as a benchmark.

4.3 Materials and methods

4.3.1 Experimental site

To quantify maize yield and nitrate leaching from sludge applied according to recommendations by the SARA model using commercial inorganic fertiliser as a benchmark, a two-year field study was conducted on a lysimeter facility at the Hatfield experimental farm, the University of Pretoria at latitude 25°45′ S and longitude 28°16′ E, and 1370 m above sea level. Pretoria is within the subhumid agroecological zone of South Africa with an average annual rainfall of between 600 mm to 800 mm (Gbetibouo & Hassan, 2005). The average annual rainfall of Pretoria is 692 mm (1981-2010) (South African Weather Service, 2019).

4.3.2 Field trial

4.3.2.1 Experimental structure

There were 14 drainage lysimeters arranged in two rows (Figure 4.1): each row comprising seven lysimeters on either side of underground housing, with a spacing of 0.8 m in-between lysimeters. Only 12 of the lysimeters were used for the study (six on each side of the housing). Each lysimeter is a metal-built cylinder with a dimension of 2.4 m diameter, 1.3 m depth, and a volume of 6.1 m^3. The lysimeters were packed with uniform soil layers for over three decades and have not been disturbed since then; therefore, the soil monoliths were assumed to be in a natural state.

Figure 4.1: Experimental site during land preparation indicating lysimeters at both sides of the underground housing

Located inside the underground housing are large plastic drums (volume ≥120 litres) beneath each lysimeter column where leachates were collected through drainage pipes (Figure 4.2). Each plastic drum has a valve at the base for the collection of leachates, and a lid to protect leachate from any contamination, such as dust or debris. There was also an installed automatic pump that evacuates excess water from the floor of the housing in the event of flooding. A lighting facility was available for analysing leachate on-site at night when necessary.

Figure 4.2: Inside the underground housing, indicating plastic drums used for leachate collection

4.3.2.2 Treatments and experimental design

Four treatments comprising sludge + humid rainfall, sludge + subhumid rainfall, inorganic fertiliser + humid rainfall, and inorganic fertiliser + subhumid rainfall, was randomly allocated to the lysimeters and replicated three times. Sludge (anaerobically digested) and commercial inorganic fertiliser were applied based on recommendations by the SARA model and Fertiliser Society of South Africa (FSSA, 2007) as presented in Table 4.1:

Table 4.1: Sludge and commercial inorganic fertiliser application rates as used in the study

		Subhumid rainfall	Humid rainfall
First-year trial	Sludge	16.5 Mg ha^{-1} + 104 kg KCl ha^{-1}	31.7 Mg ha^{-1} + 160 kg KCl ha^{-1}
	Inorganic fertiliser	155:63:104 kg NPK ha^{-1}	258:69:160 kg NPK ha^{-1}
Second-year trial	Sludge	1.6 Mg ha^{-1} + 33.4 kg KCl ha^{-1}	12.7 Mg ha^{-1} + 40.7 kg KCl ha^{-1}
	Inorganic fertiliser	145:22:26 kg NPK ha^{-1}	220:30:53 kg NPK ha^{-1}

4.3.2.3 Land preparation, treatment application, and planting

Land tilling was conducted manually using hoe while sludge and inorganic fertiliser were applied by broadcasting and immediately incorporated into the top 0-10 cm of soil, and then soil levelled using a rake. Maize seeds (PAN 6439for the first year and IMP 52-11R for the second year) were sown a day later at the rates of 40,000 (subhumid) and 60,000 (humid) seeds ha^{-1} at 3 cm depth.

While P was applied once at planting, N and K inorganic fertilisers were applied in two splits: 30% at planting and 70% seven weeks after planting. Attributable to the low K content of sludge (Tesfamariam et al., 2015), K was added to all sludge treatments through inorganic KCl (Table 4.1) as estimated by the SARA model.

4.3.2.4 Sludge incubation for mineralisation studies

A soil-sludge mix was incubated in the porous ceramic tube in the sludge-treated lysimeters at 10 cm depth to quantify N-mineralisation, using the method as explained by Henry et al. (2000).

Organic N (ON) mineralisation was calculated as:

ON mineralisation = $[(Mo * ONo) - (M_1 * ON_1)] / (Mo * ONo)$

Where M_0 = Initial mass of sludge

M_1 = Final mass of sludge

ON_0 = Initial concentration of organic N

ON_1 = Final concentration of organic N

4.3.2.5 Irrigation and simulation of humid rainfall

During the establishment stage, all plants received 10 mm uniform irrigation every three or four days in the absence of rainfall until tasselling, in the first year, and for four weeks after emergence, in the second year. In both years, once-of uniform irrigation of 5 mm was also applied to all treatments immediately after the second split inorganic fertiliser application.

Since the experiment was conducted in the subhumid region, humid treatments intermittently received supplemental rainfall through pressure compensated drippers operating at a pressure of 100 kPa. The humid region has an annual rainfall of 801-1000 mm (Mpofu et al., 2020, Schoeman and Van Der Walt 2004), and irrigation applied to mimic the actual rainfall distribution of the humid zone by topping onto the natural rainfall of the study site. To achieve this, ten years of daily rainfall data from SASRI, Durban (humid zone), and experimental site (subhumid zone) were compared (Table 4.2). Data indicated that during the ten years the humid region had a consistently higher rainfall frequency range, therefore, 10 mm or 20 mm of irrigation was chosen as the top-up for the wetter months (January to March) and 5 mm for the drier months (April and May). Additional water application to the humid zone was terminated at the end of the crop's physiological maturity.

Table 4.2: Descriptive statistics summary of ≤10 years (2008 to 2018) rainfall data for maize planting season in the humid and subhumid agroecological zones in South Africa

	Humid (Durban)							Subhumid (Pretoria)						
	Dec	Jan	Feb	Mar	Apr	May	Jun	Dec	Jan	Feb	Mar	Apr	May	Jun
n =	10	10	10	10	10	10	10	9	10	10	9	10	9	10
Mean total (mm)	115	108	86	110	42	55	16	141	137	123	132	45	17	5
Mean monthly (mm)	6.6	8.0	6.9	8.5	4.8	7.7	3.8	11.0	12.7	13.3	15.8	44.5	7.3	2.5
Max. daily (mm)	71.4	77.9	77.6	104.2	42.4	106.9	61.3	85.5	62.0	73.5	113.8	55.0	30.8	19.2
Frequency range (days)	10-26	9-18	9-16	9-18	7-12	3-9	1-8	8-20	4-6	4- 4	1-15	1 10	1-5	1-2

n is the number of years of rainfall data

Sources: SASRI weatherweb station Mt Edgecombe – SASRI [MET-29] [29º42'15"S] [31º2'45"E] [96 m] (Durban)

Pretoria University Proefplaas weather station 0513435A4 (Pretoria)

The rainfall simulation was targeted during dry periods and did not exceed the maximum monthly frequency of rainfall for the humid agroecological region. During the trial, the actual rainfall amount in the humid agroecological region (Durban) was 689.1 mm in the first year and 398 mm in the second year. These were similar to the total rainfall applied under humid rainfall treatments, as seen in Table 4.3.

Table 4.3: Total amount of irrigation and rainfall (natural and simulated) in mm received by maize plants during the trials

		First-year	(28 December 2018 to 6 June 2019)			Second-year	(6 February-31 May 2020)		
		Natural rainfall	Uniform irrigation	Simulated rainfall	Total rainfall received	Natural rainfall	Uniform irrigation	Simulated rainfall	Total rainfall received
Humid	In-H	398.8	75	135	608.8	228.9	35	120	383.9
	Bio-H	398.8	75	135	608.8	228.9	35	120	383.9
Subhumid	In-S	398.8	75		473.8	228.9	35		263.9
	Bio-S	398.8	75		473.8	228.9	35		263.9

In-H: Inorganic fertiliser-Humid, Bio-H: Sludge-Humid, In-S: Inorganic fertiliser-Subhumid, Bio-S: Sludge-Subhumid

Figure 4.3: First-year maize plants at six weeks after planting

4.3.3 Weather data of subhumid and humid agroecological zones during the field trials

Weather data were collected from Hatfield experimental farm automated weather station less than 500 m from the experimental site representing the subhumid region and from SASRI weather station, Durban representing the humid region. Table 4.4 indicates the mean maximum, minimum, and average temperatures, and total rainfall for both regions for the study period.

Table 4.4: Mean minimum, maximum, and average temperatures, and total rainfall of subhumid and humid agroecological zones during field

* No data found $^{+}$Incomplete data

		First year								Second year						
		Subhumid (Pretoria)				Humid (Durban)				Subhumid (Pretoria)				Humid (Durban)		
Month	Week	Temperature °C			Rainfall mm	Temperature °C			Rainfall mm	Temperature °C			Rainfall mm	Temperature °C		
		max.	min.	ave.	total	max.	min.	ave.	total	max.	min.	ave.	total	max.	min.	ave.
Dec.	3-4	*	*	*	10.9^{+}	29.2	20.9	25.1	32.1	-	-	-	-	-	-	-
Jan.	1-2	28.9	17.2	23.0	114.5	29.1	19.6	23.7	23.4	-	-	-	-	-	-	-
	3-4	30.8	16.4	23.6	54.8	27.9	19.6	23.7	91.7	-	-	-	-	-	-	-
Feb.	1-2	27.7	17.6	22.6	106.0	30.4	21.2	25.8	74.1	28.7	15.7	22.2	14.7	30.0	21.5	25.8
	3-4	29.7	15.5	22.6	41.5	30.2	21.2	25.7	65.8	28.2	16.0	22.1	12	28.1	20.0	24.0
Mar.	1-2	30.8	17.4	24.1	0.6	30.2	22.2	26.2	104.6	26.6	13.4	20.0	28.4	29.0	19.6	24.3
	3-4	31.0	15.9	23.4	7.5	30.9	22.8	26.8	68.1	25.3	16.2	20.8	37.9	27.9	19.6	23.8
April	1-2	24.8	13.5	19.2	47.0	29.3	20.5	24.9	47.0	22.3	15.2	18.7	95.8	26.3	17.1	21.7
	3-4	25.2	12.3	18.8	17.7	29.3	20.8	25.0	191.8	20.3	12.51	16.4	40.1	25.9	14.5	21.5
May	1-2	26.2	10.7	18.4	0	30.2	19.8	25.0	1.1	21.3	9.9	15.6	0	26.6	12.4	20.5
	3-4	24.1	7.8	15.9	0	28.2	17.7	22.9	5.4	19.2	7.8	13.5	0	25.5	10.8	18.9
June	1-2	22.0	5.0	13.5	0	27.4	14.8	21.1	8.4	-	-	-	-	-	-	-

4.3.4 Sampling and maize measurements

4.3.4.1 Leachate

Leachate was collected from drums in the underground house following each heavy rainfall event during the first year and after every drainage during the second year. The leachate samples were analysed for nitrate concentration on-site using a multi-parameter colourimeter (Move 100 Spectroquant®, model 173632, Merck).

4.3.4.2 Soil

Soil samples for diagnostic physical and chemical property determination were collected from the centre of each lysimeter at 0-30, 30-60, 60-100 cm depth intervals before sludge and inorganic fertiliser applications using a soil auger. The samples were prepared for laboratory analysis by air-drying and passing through a 2 mm sieve. Additional undisturbed soil samples were collected using a core sampler for bulk density determination from one of the extra lysimeters not used for the study.

4.3.4.3 Soil-water content measurement

The soil-water content measurement was conducted using a site calibrate neutron water metre. The neutron water meter was calibrated using one of the lysimeters not used for the study. Weekly or bi-weekly readings, depending on the rainfall, were taken throughout the maize growing period.

4.3.4.4 Plant sampling

During the growing season, canopy cover was measured using Ceptometer every 10 to 15 days as of 35 days after planting (DAP) until 100 DAP. Whole plant samples (five plants (subhumid) and seven plants (humid)) were collected from each lysimeter at physiological maturity for above-ground biomass determination. The samples were partitioned into leaves, stems, and grain. The partitioned samples were oven-dried at 65^{O}C to constant mass and weighed.

4.3.5 Physico-chemical analysis of soil, plant, sludge, and leachate

Soil and sludge OM content was determined using the Walkley and Black (1934) method. Exchangeable cations (Na, K, Ca, Mg) were analysed using ICP-OES after extraction using 1 M ammonium acetate. Soil and sludge electrical conductivity and pH were analysed with glass electrodes in a 1:2.5 solid: distilled water slurry. Plant-available P was estimated using the Bray-1 extraction method. Nitrate and ammonium were extracted using 1 M KCl and measured colorimetrically with a UV/visible spectrophotometer (Pharmacia LKB – Ultraspec III). Soil effective cation exchange capacity (ECEC) was calculated as the sum of 1M ammonium acetate extractable bases and 1M KCl extractable acidity.

Sludge, soil, and plant samples were analysed for TC and nitrogen using a Carlo Erba NA 1500 C/N analyser (Carlo Erba Strumentazione, Milan, Italy). Total P in sludge and soil was extracted by microwave-assisted acid digestion while trace metals were digested with nitric and perchloric acid then analysed with ICP-OES. Nitrate concentrations in leachate were analysed on-site using a multi-parameter colourimeter (Move 100 Spectroquant® Merck) with a nitrate test kit (Merck cat. no. 1147730001) of measuring range 0.5-15.0 (± 0.31 accuracy) mg l^{-1} NO_3^- – N. This employed the USEPA approved nitrospectral method. Nitrate leached was calculated as concentration multiplied by the volume of leachate. Soil textural class was determined after separating the particles into sand, clay, and silt fractions using the hydrometer method.

4.3.6 Statistical analysis

Data were analysed statistically using the General Linear Model (GLM) of SAS software 9.4 (SAS Institute, Cary, NC). The Least Significant Difference (LSD) at a 5% probability level was used to compare means.

4.4 Results and discussion

4.4.1 Physico-chemical properties of soil

Table 4.5a displays the physical and chemical properties of the soil before the treatment application at the first-year trial. The lysimeters employed in the study contained soils of similar textural classes. This offered a fair comparison of nitrate leaching among treatments. The clay fractions in all strata of the soil profile were moderate to high, ranging from 30% to 41%. The topsoils (0-30 cm) were sandy clay loam while the subsoils (30-100 cm) – either clay loam or clay. A consistent decrease was established in the sand fraction, with an increase in the clay fraction with depth. Clay may have translocated from the top layer to the lower layers through eluviation. This may have been attained over years of non-disturbance; therefore, it could be assumed that soil monoliths were close to the natural state before the study.

Soil texture is crucial in affecting nitrate leaching (Lai et al., 2020; Padilla et al., 2018; Simmelsgaard, 1998). The ratio of sand to clay ranging from 1.5 to 1.8 at 0-30 cm depth and 1.1 to 1.3 at 0-60 cm depth indicates that the soils are fairly drained; however, nitrate may accumulate in the lower layers attributable to lower sand to clay ratio of ≤ 0.9 (Tahir & Marschner, 2017). Most South African soils are highly weathered and naturally deficient in essential nutrients (Maqubela et al., 2009; Barnard & du Preez, 2004). The chemical properties of soil used for the study indicated similar characteristics to uncultivated soils in most parts of South Africa (Du Preez et al., 2011; Mandiringana et al., 2005).

The topsoils were moderately acidic, with a pH of 5.2±0.1. The soil pH controls microbial activities, and, therefore, N-mineralisation. The soil pH was within the optimal range (5 to 7) in most microbial group activities (Pietri & Brookes, 2008). The total and organic carbon contents were exceptionally low (<1%). The available P and exchangeable K ranged from 7.79 to 13.51 and 30.20 to 32.71 mg kg^{-1}, respectively, below the critical limits required for optimum maize growth (FSSA, 2007). Exchangeable Mg and Ca were also low (FSSA, 2007) ranging from 20.14 to 22.69 and 40.26 to 46.09 mg kg^{-1}, respectively.

Table 4.5: Physico-chemical properties of soils in the lysimeters used for the study

Lysimeter ID*	Depth	Sand	Clay	Silt	Texture	pH	Bray-1 P	K	NO_3^--N	NH_4^+-N	Total N	Total C	Organic C	ECEC
	cm		%					mg kg^{-1}				%		meq/ 100 g
A: Soil properties before treatment application during the first-year trial														
In - H	0-30	53 ± 0.8	30 ± 1.8	17 ± 1.3	SCL	5.2 ± 0.11	13.51 ± 2.6	30.40 ± 1.6	4.47 ± 0.5	12.35 ± 5.0	0.057 ± 0.005	0.69 ± 0.06	0.63 ± 0.10	1.49 ± 0.03
	30-60	40 ± 0.6	32 ± 1.3	28 ± 0.7	CL	5.4 ± 0.04	4.96 ± 0.5	32.97 ± 3.1	3.18 ± 0.2		0.043 ± 0.002	0.45 ± 0.03	0.31 ± 0.02	
	60-100	32 ± 0.3	41 ± 1.2	27 ± 1.3	C	5.5 ± 0.07	5.74 ± 1.4	29.39 ± 2.1	3.20 ± 0.4		0.029 ± 0.001	0.25 ± 0.002	0.17 ± 0.04	
Bio - H	0-30	50 ± 1.8	33 ± 2.1	17 ± 0.3	SCL	5.3 ± 0.06	7.79 ± 1.1	30.20 ± 2.3	5.44 ± 0.4	11.04 ± 1.9	0.069 ± 0.004	0.89 ± 0.09	0.67 ± 0.06	1.34 ± 0.02
	30-60	41 ± 2.6	39 ± 4.0	20 ± 1.5	CL	5.3 ± 0.07	6.60 ± 1.3	31.64 ± 3.2	2.96 ± 0.6		0.052 ± 0.006	0.61 ± 0.10	0.55 ± 0.15	
	60-100	35 ± 1.0	39 ± 4.1	26 ± 3.5	CL	5.4 ± 0.10	7.61 ± 1.4	32.70 ± 4.5	3.75 ± 0.4		0.043 ± 0.009	0.45 ± 0.13	0.32 ± 0.12	
In - S	0-30	52 ± 1.4	32 ± 1.4	16 ± 0.3	SCL	5.2 ± 0.11	12.55 ± 2.6	32.71 ± 1.7	3.56 ± 0.8	6.73 ± 0.3	0.061 ± 0.003	0.76 ± 0.05	0.65 ± 0.03	1.47 ± 0.06
	30-60	40 ± 0.4	37 ± 2.7	23 ± 2.5	CL	5.5 ± 0.12	5.45 ± 0.3	32.11 ± 3.0	2.46 ± 0.8		0.048 ± 0.002	0.53 ± 0.02	0.39 ± 0.10	
	60-100	34 ± 1.3	40 ± 2.7	26 ± 1.4	C	5.4 ± 0.04	5.24 ± 0.7	30.03 ± 2.2	1.81 ± 0.1		0.040 ± 0.003	0.39 ± 0.05	0.33 ± 0.06	
Bio - S	0-30	50 ± 0.4	34 ± 1.8	16 ± 1.8	SCL	5.1 ± 0.13	11.12 ± 1.2	32.17 ± 2.7	6.48 ± 3.7	9.45 ± 2.1	0.065 ± 0.002	0.78 ± 0.04	0.62 ± 0.04	1.56 ± 0.06
	30-60	42 ± 0.8	40 ± 1.5	18 ± 2.2	C	5.4 ± 0.14	5.41 ± 1.8	29.07 ± 3.8	3.50 ± 0.6		0.055 ± 0.009	0.62 ± 0.14	0.42 ± 0.09	
	60-100	37 ± 1.7	40 ± 1.3	23 ± 1.1	C	5.4 ± 0.06	4.38 ± 0.4	25.55 ± 0.5	2.87 ± 0.9		0.038 ± 0.005	0.37 ± 0.07	0.23 ± 0.04	
B: Soil properties after first year trial crop harvest and before second-year trial treatment application														
In - H	0 -10						27.64 ± 1.9	112.5 ± 9.0	10.49 ± 0.7	4.76 ± 0.9	0.107 ± 0.012			
Bio - H	0 -10						32.49 ± 2.6	120.7 ± 16.0	19.91 ± 5.4	3. 86 ± 1.2	0.133 ± 0.012			
In - S	0 -10						29.04 ± 2.2	135.8 ± 6.4	12.87 ± 5.3	3.49 ± 0.6	0.097 ± 0.007			
Bio - S	0 -10						22.14 ± 2.8	96.0 ± 4.2	23.52 ± 7.0	4.00 ± 1.0	0.103 ± 0.009			

In-H = Inorganic fertiliser*Humid rainfall, In-S = Inorganic fertiliser*Subhumid rainfall, Bio-H = Sludge*Humid rainfall, Bio-S = Sludge*Subhumid rainfall.

SCL = Sandy clay loam, CL = Clay loam, C = Clay

± standard error of mean

Soil CEC, as a function of type or amount of clay, exhibits a significant relationship in the leaching of anionic nutrients (Gaines & Gaines, 1994), performing as an important indicator of nitrate leaching in simulation studies (De Filippis et al., 2021; Li et al., 2006; Vachaud & Chen, 2002). Although the topsoils held a moderate clay content, the CEC measured as ECEC, was below two meq/100 g, attributable to a high sand fraction (50 to 53%). Soils were also deficient in nitrogen with mean values < 0.1%. Along with most crops, maize requires high N inputs (Nasielski et al., 2019; Dai, 1998); uptake mostly occurs through nitrate ions. Soil nitrate-N was below the lower limit (20 mg kg^{-1}), considered as optimum for maize crops (Blackmer et al., 1989).

Before the second-year trial, soil nutrient composition indicated increases in total N and nitrate (NO_3^-) but decreases in ammonium (NH_4^+) concentrations (Table 4.5b) relative to the initial soil before treatment application. NH_4^+ could be lost or transformed into the environment through volatilisation, nitrification, immobilisation, and fixation. NH_4^+ loss through volatilisation ensues from its transformation to ammonia gas. Ammonia production in the soil system is temperature, moisture, and pH-dependent (Dari et al., 2019). It mostly occurs at maximum fertiliser exposure to the atmosphere. Since the pH of sludge and the initial topsoil were low, and with the immediate incorporation of sludge and inorganic fertiliser into the soil at application, it is assumed that nitrogen losses through volatilisation would be insignificant.

Decreases in NH_4^+ may be attributed to the nitrification process converting NH_4^+ to NO_3^- by the increased microbial transformation activities resulting from the freshly supplied nutrients (Nugroho et al., 2006; Phillips et al., 2000). Several studies agree with this finding; for example, in an incubation study by He et al. (2000), NO_3^- was identified as the consistently dominant mineral N after periodic mineralisation evaluations, despite NH_4^+ being the higher N mineral before the experiment.

Conversely, a previous study on sludge N-mineralisation in the subhumid and humid agroecological regions in South Africa indicated decreases in both NO_3^- and NH_4^+ after one year incubation (Badza, 2020). The decreases may be attributable to the extremely low clay fraction (10% to 18%) of the soils used for the study, implying that nitrification may have occurred but NO_3^- was lost to the environment. Nitrification is of great relevance in N cycling and plant nutrition in agricultural soils. This process can also lead to nitrate leaching.

4.4.2 Physico-chemical properties of sludge

The concrete bed-dried anaerobically digested sludge in the trials was collected from the East Rand Waterworks, Johannesburg, South Africa. Some properties of the sludge are indicated in Table 4.6.

Table 4.6: Some Physico-chemical properties of sludge used in the study (n=3)

Parameter	Unit	First-year Value	Second-year Value
Moisture	%	25	27
pH	-	6.5	6.9
EC	ms m^{-1}	998	1127
Total Nitrogen	%	2.71	3.4
NO_3^- - N	mg kg^{-1}	21.98	22.08
NH_4^+ - N	mg kg^{-1}	24.13	26.91
Organic Carbon	%	15.69	19.6
Total Phosphorus	%	3.07	3.0
Potassium	mg kg^{-1}	180.86	3145.5
Magnesium	mg kg^{-1}	494.0	1122.3
Sodium	mg kg^{-1}	155.89	2650.6
Calcium	mg kg^{-1}	3703.67	6940.8
As	mg kg^{-1}	6.7	4.5
Cd	mg kg^{-1}	4.5	6.6
Cr	mg kg^{-1}	165.0	218.0
Cu	mg kg^{-1}	283.7	364.0
Hg	mg kg^{-1}	1.7	0.6
Mn	mg kg^{-1}	767.7	1091
Ni	mg kg^{-1}	124.0	156.0
Pb	mg kg^{-1}	63.4	81.9
Zn	mg kg^{-1}	1717.7	1695.0

The sludge held a pH of 6.5 and 6.9 for the first and second seasons, respectively. Depending on the treatment process, the pH of most sludge ranges from slightly acidic to highly alkaline (Kominko et al., 2017; Tesfamariam et al., 2013; Corrêa et al., 2005). In low pH soil, it is expected to increase pH with its application, consequently boosting microbial activities. Under heavy rainfall or irrigation, soil acidity may increase with an increase in nitrate leaching as more hydrogen ions associated with nitrate remain in the soil, unless counteracted by the buffering capacity of clay (Jansen Van Rensburg et al., 2009). The electrical conductivity of both sludges was high (9.98 and 11.3 *m*s cm^{-1} for the first and second year, respectively) (Table 4.6).

The OM content of sludge was high; 27 and 34% for first- and second-years' sludge, respectively, but lower than the range (45% to 65%) reported in most sludge (Yang et al., 2018; Kominko et al., 2017; Haynes et al., 2009; Snyman & Herselman, 2006). Elemental concentrations of heavy metals (Pb, Zn, and Cu) of concern, except for Cd, were above the range of acceptable sludge for land application in South Africa (Snyman & Van Der Waals, 2004). The toxicity of these heavy metals is soil-dependent concerning absorption, fixation, and leaching.

The electrical conductivity of both sludges was 9.98 and 11.3 *m*s cm^{-1}. OM contents were high at 27 and 34% for first- and second-years' sludge, respectively, but lower than the range

(45% to 65%) reported in most sludge (Yang et al., 2018; Kominko et al., 2017; Haynes et al., 2009; Snyman & Herselman, 2006). Elemental concentrations of heavy metals of concern (As, Cd, Cr, Cu, Hg, Mn, Ni, Pb, & Zn), were all within an acceptable range for agricultural use and could be categorised as “class a”.

Inorganic nitrogen concentration of sludge for both years was less than 1% of the total nitrogen, indicating that 99% of the TN was in organic form. Nitrate leaching from the application of such sludge is, therefore, highly dependent on the rate of N-mineralisation. The C:N ratio of the sludge for both years was six. Sludge C:N ratio is one predictor of the N-mineralisation rate (Er et al., 2005; Gilmour & Skinner, 1999). The higher the C:N ratio the lower the mineralisation rates (Lu et al., 2012; Knowles et al., 2011). For any organic material, a C:N ratio of between one and 15 is adequate for optimum N-mineralisation (Brust, 2019).

4.4.3 Maize nitrogen uptake, growth, and yields

The grain and leaf nitrogen uptake were higher from the inorganic fertiliser compared to sludge-treated soils under both rainfall scenarios for both years (Table 4.7). In the first year, leaf nitrogen uptake by maize planted to the inorganic fertiliser treatment was 14% (humid) and 21% (subhumid) higher than that planted to sludge-amended soils.

Table 4.7: Maize nitrogen uptake (kg ha^{-1}): Inorganic fertiliser versus sludge under humid and subhumid rainfall regimes

		First year		Second year	
		Leaf	Grain	Leaf	Grain
Humid rainfall	Inorganic fertiliser	65.18 ± 5.9	215.33 ± 14.0	80.45 ± 7.2	135.34 ± 24.7
	Sludge	55.83 ± 6.0	149.45 ± 9.7	66.22 ± 10.5	118.47 ± 34.5
Subhumid rainfall	Inorganic fertiliser	51.72 ± 5.1	123.03 ± 7.9	42.31 ± 3.3	100.52 ± 14.1
	Sludge	40.66 ± 2.2	112.03 ± 22.5	21.23 ± 2.2	23.00 ± 1.3

Grain N uptake was 31% (humid) and 9% (subhumid) higher from the inorganic fertiliser treated soils than those treated with sludge. During the second year, leaf N uptake was 18% (humid) and 50% (subhumid) higher from the inorganic fertiliser treated soil; grain N uptake was also 12% (humid) and 77% (subhumid) higher than the inorganic fertiliser treated soil compared with the sludge-treated soil.

The maize grain yield was higher in the humid compared to the subhumid treatments for both years (Figure 4.4). Generally maize grain yield was lower during the second year compare with the first year. The drastic drop in maize grain yield during the second year was mainly attributed to the low rainfall experienced during the season (Table 4.3 and Table 4.4). The decline in yield was much more severe in the sludge-treated lysimeters than in the inorganic fertiliser attributable to the decline in nitrogen mineralisation (Figure 4.5). According to the field incubation study, nitrogen mineralisation during the second year was 8% (humid) and 10% (subhumid) lower than in the first year. The effect of low rainfall on sludge-treated lysimeters was double-fold, directly through water deficiency, harming the photosynthesis process and dry matter production, and indirectly by adversely affecting the nutrient release rate through OM decomposition.

The aforementioned findings contrast the previously simulated results for maize yield in the focus areas (humid and subhumid agroecological zones of South Africa), indicating significantly (P<0.05) higher maize yield in sludge than in inorganic fertiliser on even lower sludge application rates (Ogbazghi et al., 2019). Studies confirm similar or higher yields from sludge application as compared to inorganic fertiliser (Barbarick et al., 2017; Cogger et al., 2013; Koenig et al., 2011; Jaber et al., 2005).

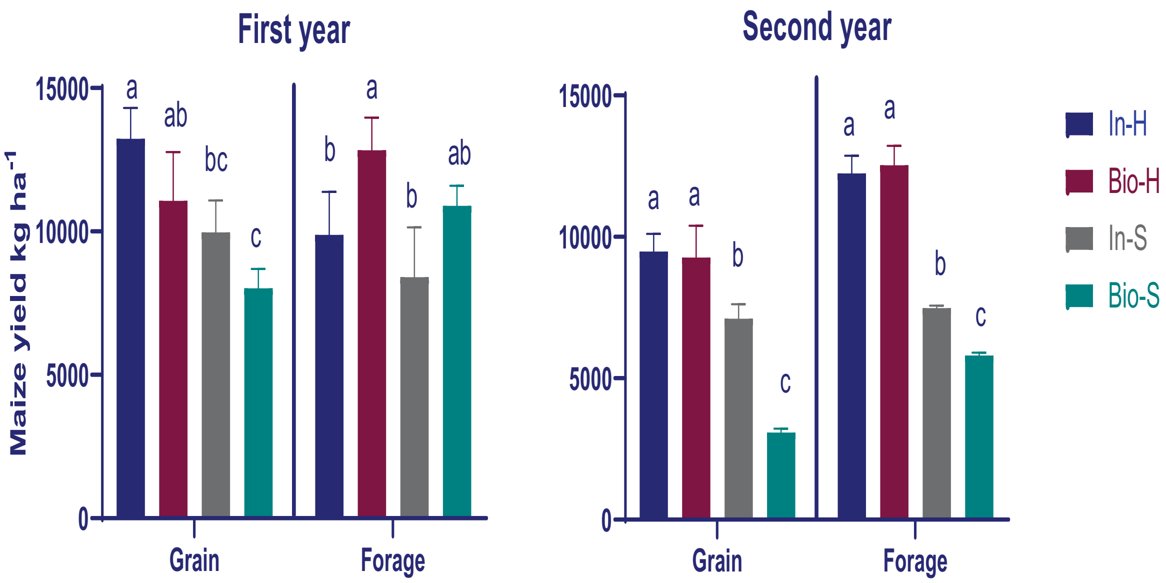

Figure 4.4: Maize grain and forage yields as affected by sludge versus inorganic fertiliser under humid and subhumid rainfall distribution

Error bars represent the standard error of the means. Alphabets indicate the significance (P<0.05) of a yield type across treatments.

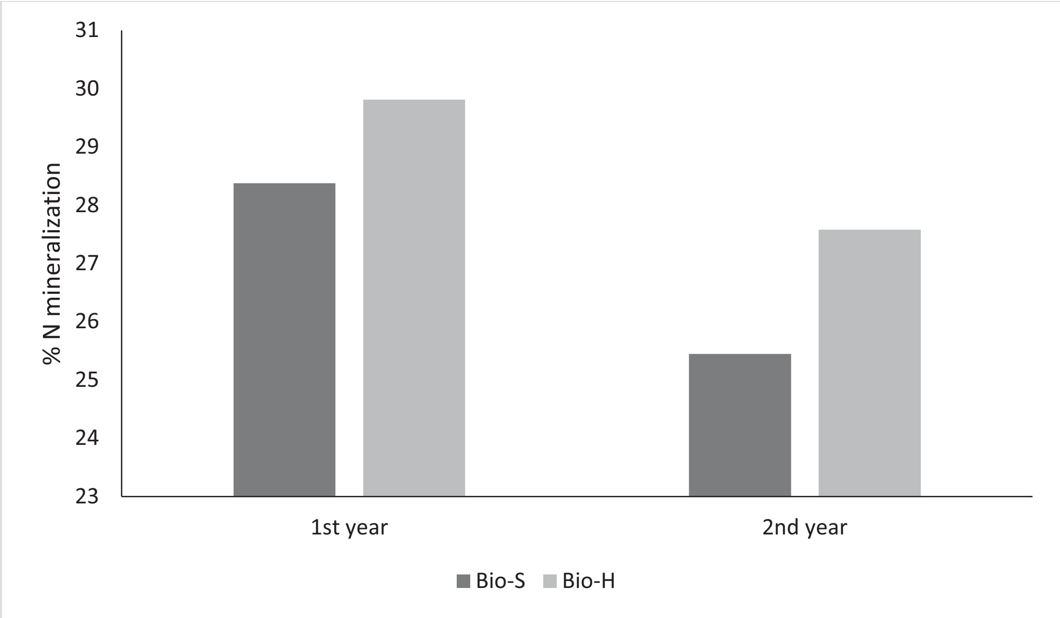

Figure 4.5: N-mineralisation for incubated sludge under sludge treatments alone

It was also apparent that forage yield was higher than grain yield for all sludge-treated lysimeters in both years, indicating a lower harvest index. Conversely, grain yield was higher than forage yield for all inorganic fertiliser treated lysimeters during the first growing season, indicating a higher harvest index. The maize leaf area index for the current study was measured only during the second-year trial (Figure 4.6). Leaf area index (LAI) performs an essential function in characterising the physiological development of crops. LAI correlates to photosynthesis, respiration, dry matter production, and final grain yield (Roth et al., 2018 Liu & Pattey, 2010; Bréda, 2003) and can access crop growth progression on-site.

Results indicated LAI increased gradually and peaked at 65 days after sowing (DAS), then slowly declined and tapered towards 100 DAS. At this stage, the leaves started drying out. Sixty-five DAS corresponded with the onset of the reproductive stage (R1) of the maize variety planted. At this stage, N uptake reached its maximum for leaves blade and sheaths, husky leaves, stem, cob, and tassels, after which N begins remobilisation to the grains (Ross et al., 2013). Attributable to high N uptake at this stage, NO_3^- leaching is expected to reduce unless in case of heavy rainfall (Srivastava et al., 2020). The LAI of maize plants followed an order: Bio-H>In-H>In-S>Bio-S. This trend was similar for forage and grain yields. The higher LAI of Bio-H compared to the In-H supports the reported higher forage yield for Bio-H (Figure 4.4) as the leaves are the photosynthetic factory contributing to biomass production. The reason for the lower grain yield of Bio-H is the low assimilate partitioning to the grain.

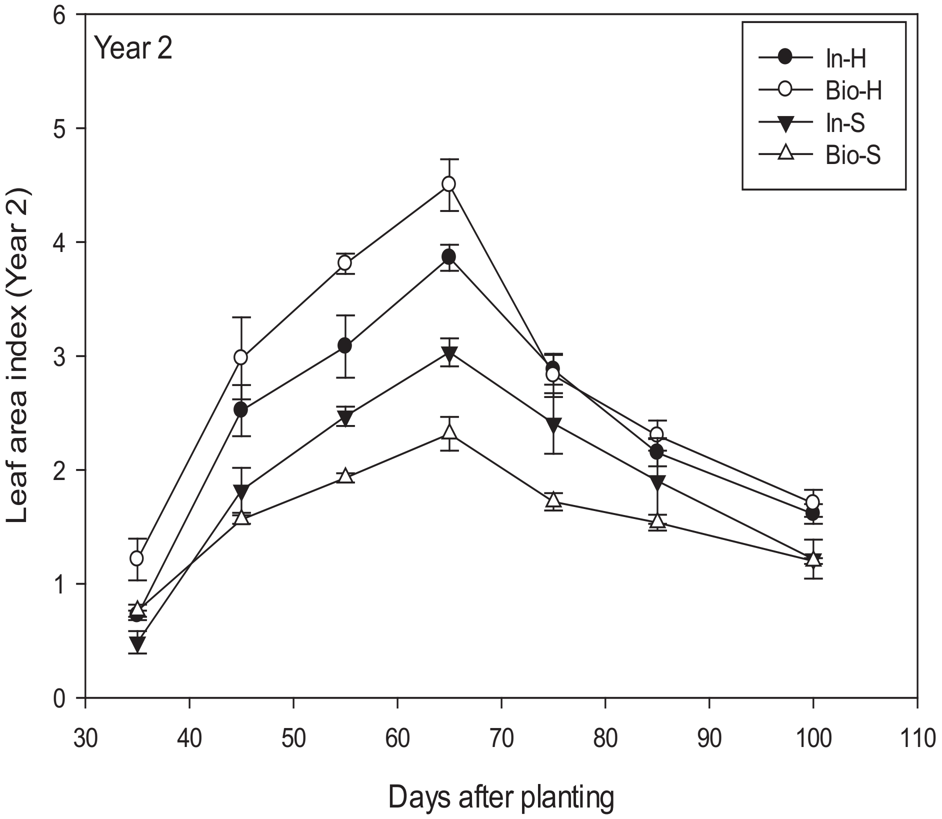

Figure 4.6: Second-year maize leaf area index in inorganic fertiliser and sludge-amended soils under humid and subhumid rainfall

4.4.4 Nitrate leaching

Generally, cumulative nitrate leaching was relatively higher under the simulated humid zone than subhumid, attributed to the higher rainfall leading to potential leaching of nitrate below the root zone (Figure 4.7). The relatively higher nitrate leaching from the humid zone during the second season compared with similar treatment during the first season, is most probably attributed to the low rainfall (Table 4.2), causing low crop biomass production, leading to lower nitrate uptake from the soil. It, therefore, leads to higher nitrate accumulation in the soil profile, exposing it to potentially high leaching in the event of high rainfall. Both the rainfall amount and frequency influenced the nitrate leaching (Figure 4.8 and Figure 4.9).

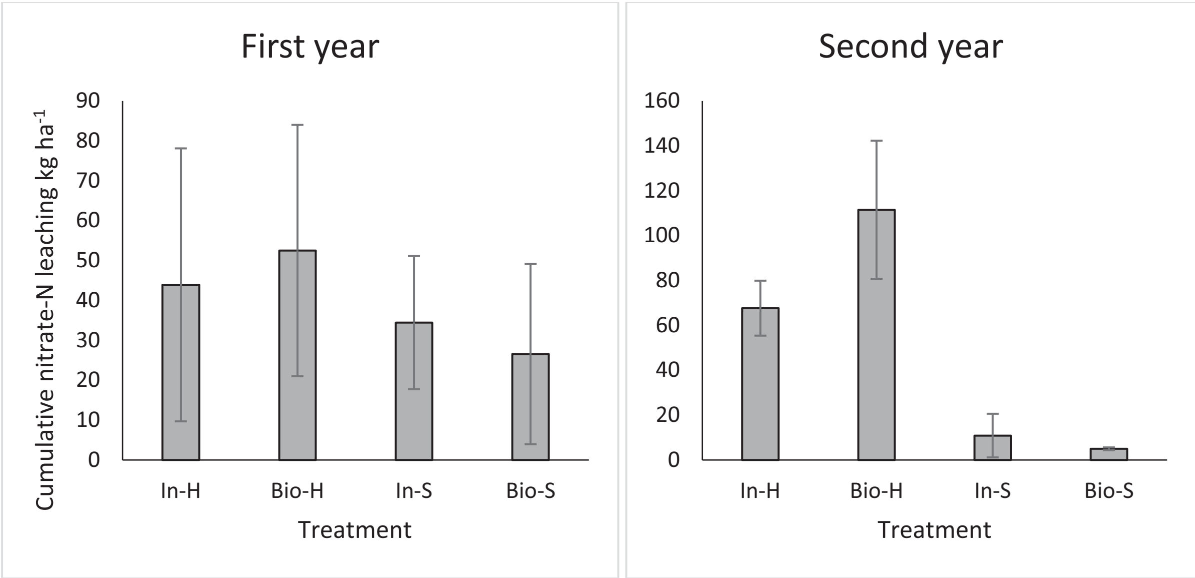

Figure 4.7: Cumulative nitrate-N (kg ha^{-1}) leaching from sludge versus inorganic fertiliser under humid and subhumid rainfall distribution

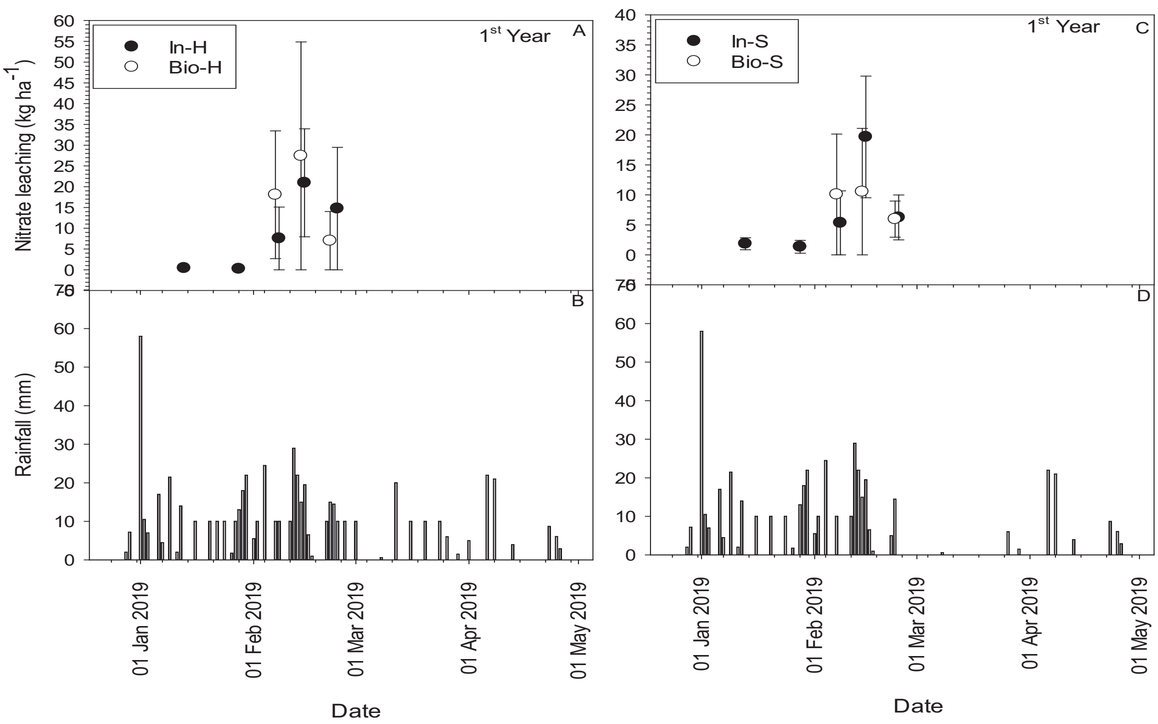

Figure 4.8: Nitrate leaching responses to inorganic fertiliser and sludge application under humid and subhumid rainfall during the first-year trial

In the first-year trial, 160 litres of leachate (not stated in the table) were collected from all the treatments at the first sampling event; however, the nitrate concentration was below the lower detection limit (0.2 mg l^{-1}) quantifiable by the instrument. Similarly, nitrate concentration in leachate from sludge treatments on the second and third sampling events was below the instrument's measuring range. High organic N (99%) in sludge may explain the delayed nitrate leaching from sludge treatments. Dilution of nitrate concentration in the leachate, though, could be the essential factor, leading to no detection or low nitrate concentration during the first three sampling events before the higher concentrations recorded on 8 February 2019 (Figure 4.8).

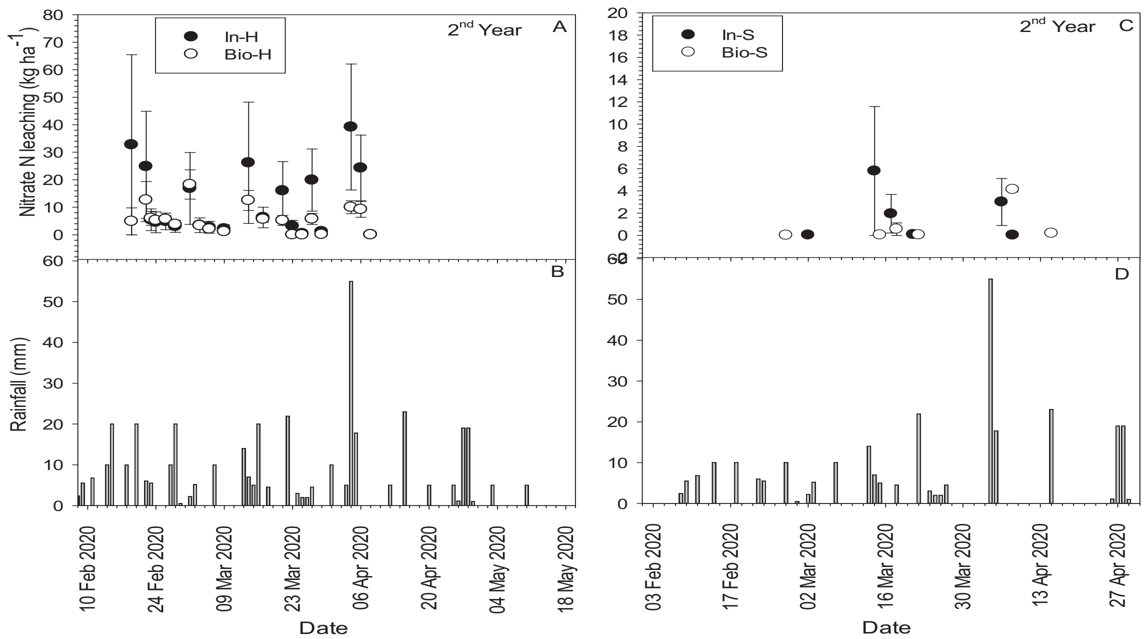

Figure 4.9: Nitrate leaching responses to inorganic fertiliser and sludge application under humid and subhumid rainfall during second-year trial

N uptake increases as a plant grow, concurrently reducing N leaching. More nitrate leaching was expected as the crop reached physiological maturity (growth stage V5-V6). Uptake is reduced, but no drainage occurred at this stage (April to May) as rainfall dwindled. On 15 February 2019, in the first year, the highest nitrate leaching occurred at the V12-V14 stage of growth. At this stage, N uptake was low coupled with five days of consistent rainfall (on the 11^{th}, 12^{th},13^{th}, 14^{th} and 15^{th}) totalling 95.5 mm, leading to high nitrate leaching. This nitrate concentration was higher under humid but lower under subhumid for sludge compared to inorganic fertiliser.

The second-year nitrate leaching recorded two peaks under humid rainfall and one peak under subhumid rainfall, all from inorganic fertiliser. The humid rainfall treatments had these peaks at V2 (two-leaf stages) and R1 (onset of the reproductive stage). At V2, the N uptake was extremely low with relatively low rainfall; however, at R1, N uptake was high with high rainfall. This indicates the importance of the crop growth stage on nitrate leaching. Under the subhumid rainfall, the highest nitrate leaching occurred at V10-V12.

4.4.5 Soil residual total nitrogen

Inorganic fertiliser and sludge applications increased soil N after harvest in both trials. As expected, sludge treatments had higher residual soil N after the growing periods compared to inorganic fertiliser, attributable to high organic N (not readily available) versus readily available N in inorganic fertiliser. Soil residual N followed the order: Bio-S > Bio-H > In-H > In-S in the first year (Figure 4.10A) and Bio-H > Bio-S > In-S = In-H in the second year (Figure 4.10B). Comparison between inorganic fertiliser and sludge treatments indicated that in the first year, soil residual N from sludge was 21% and 35% higher than inorganic fertiliser under humid and subhumid rainfalls, respectively. In the second year, however, residual N was 40% higher in sludge than in inorganic fertiliser under humid rainfall, while sludge and inorganic fertiliser under subhumid rainfall had similar values of residual N.

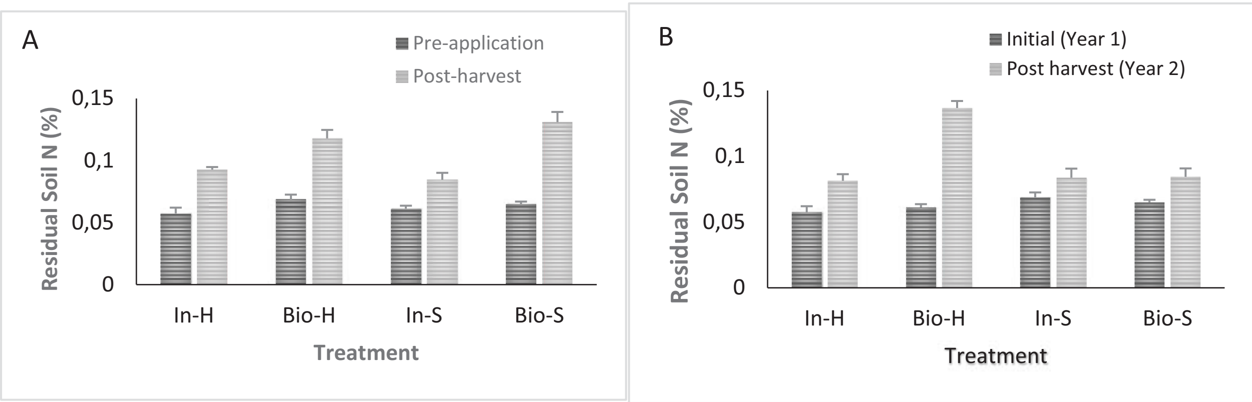

Figure 4.10: Residual soil N; (A) after the first-year trial; (B) after the second-year trial

Error bars represent the standard error of means.

4.4.6 Soil residual inorganic N

Before the study, nitrate concentrations were below the lower limit for optimum maize growth (Blackmer et al., 1989). The concentrations were higher, 4 to 6 mg kg^{-1} NO_3^--N, at the topsoil (0-30 cm) compared to the subsoils (30-60 and 60-100 cm), ranging from 1.8 to 3.8 mg kg^{-1} NO_3 – N (Figure 4.11). The higher nitrate concentration at the topsoil could be associated with higher organic carbon (Fuss et al., 2019) since the plots were not cultivated or applied fertiliser for at least three decades before the trial. Residual nitrate in the soil was determined after the second-year trial and was lower than the initial concentrations.

Results confirm a higher nitrate concentration at 60-100 cm stratum than the upper soil strata. This may be attributed to one or more of these three scenarios: heavy rainfall (Nair et al., 2020; Tesfamariam et al., 2015, Hagmann, 1994); lower sand to clay ratio at 60-100 cm stratum and higher sand fraction at the topsoil (Tahir & Marschner 2017; Yang et al., 2007); and maize luxury N consumption at the root zone (Nasielski et al., 2019; Blackmer & Schepers, 1994) resulting in low residual NO_3^--N after harvesting.

At the topsoil (0-30 cm), there was a decrease in residual NH_4^+ under sludge application but increases in the inorganic fertiliser treatments. The above results indicated that the high soil residual total N observed under sludge-humid rainfall treatment is mostly in organic form. This may cause excessive nitrate leaching at the onset of rain before the subsequent planting season.

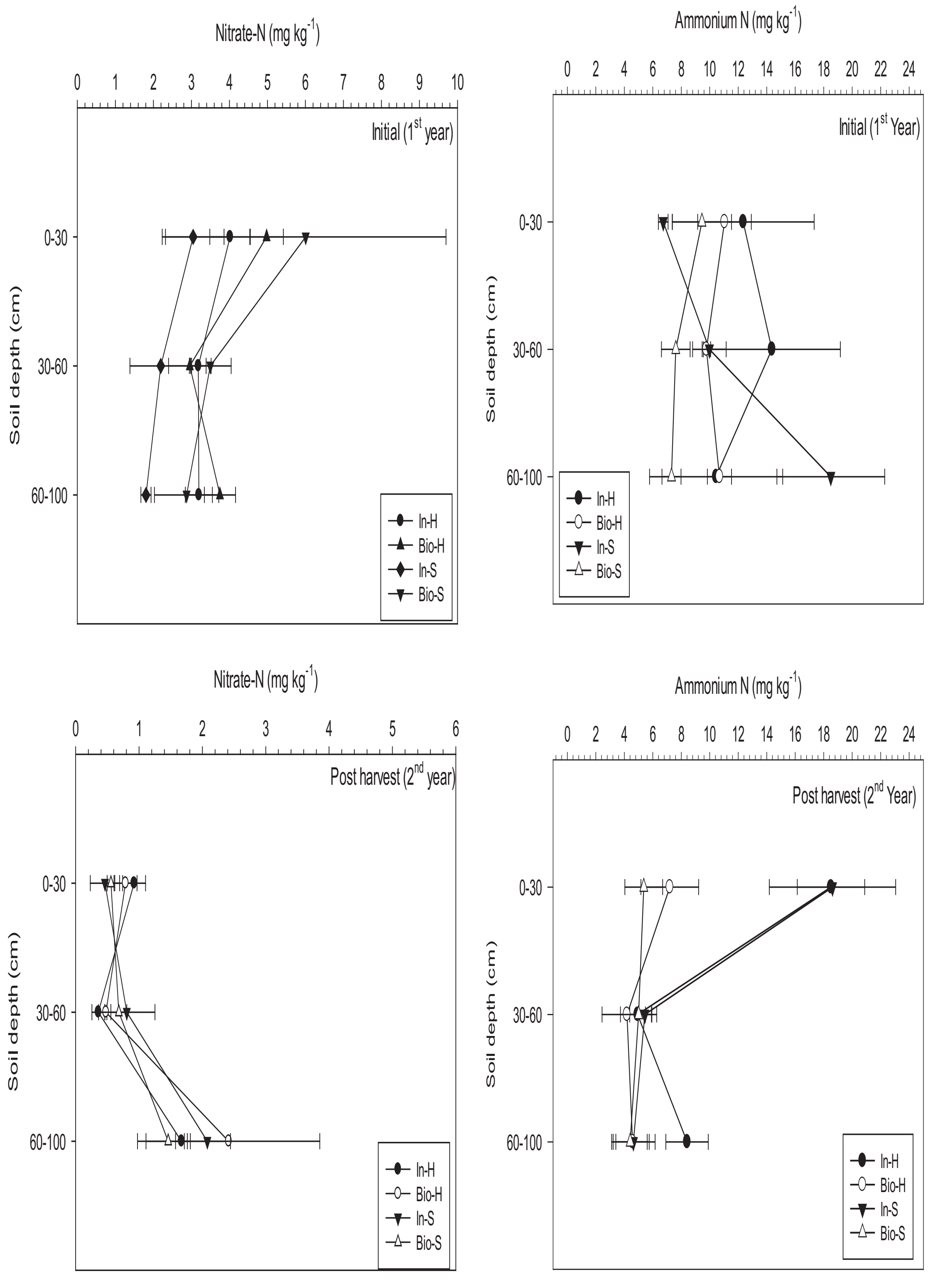

Figure 4.11: Residual soil inorganic nitrogen with depth

4.4.7 Validation of SARA model sludge recommendation rate

SARA's sludge recommendation rate was validated through lysimeter studies using commercial inorganic fertilizer as a standard under two simulated rainfall regimes (subhumid and humid). For the given rainfall regimes, both inorganic fertilizer and SARA recommended sludge rates were applied according to the Fertiliser Society of South Africa handbook. To validate the reliability of the sludge recommendation rates based on SARA model, we used the maize grain yield and nitrate leaching beneath the root zone from inorganic fertilizers.

There was no statistically significant ($P<0.05$) maize grain yield difference between inorganic fertiliser and sludge-treated plots according to SARA model recommendations of the similar agroecological zone except during the second year for the subhumid zone (Figure 4.4). During the second year, the rainfall under the subhumid zone was low, therefore, the grain yield from the sludge-treated plots according to SARA model recommendations was lower than those treated with commercial inorganic fertiliser.

Maize grain yield was above or within the target range (8 t ha^{-1} for the subhumid zone and 11 t ha^{-1} for the humid zone (FSSA 2007)) in the first year, but lower in the second year for both the inorganic fertilizer and sludge treated according to recommendation by SARA model. In the first year, the highest mean grain yield was 13,230 kg ha^{-1} (inorganic fertiliser) and 11,067 kg ha^{-1} (sludge) under the humid rainfall and 9,973 kg ha^{-1} (inorganic fertiliser) and 8,017 kg ha^{-1} (sludge) in the subhumid. In the second year, maize grain yield from the inorganic fertiliser and sludge-treated lysimeters was 9,487 kg ha^{-1} and 9,272 kg ha^{-1}, under humid and 7,103 kg ha^{-1} and 3,078 kg ha^{-1}, respectively under the subhumid regimes. Concerning the target yields, the percentage decreases in yield during the second year were 14%, 16%, 11%, and 62% for In-H, Bio-H, In-S, and Bio-S, respectively.

Cumulative nitrate leaching varied across treatments in both years (Figure 4.7). In the first year, there was no statistically significant ($P<0.05$) nitrate leaching difference between treatments that received inorganic fertilizer and those treated with sludge according to SARA model recommendations. Nitrate leaching was slightly higher from the sludge-treated lysimeter in the humid region and the inorganic fertiliser treated lysimeter in the subhumid environment. In the second season, however, nitrate leaching was significantly ($P<0.05$) higher from the sludge-treated soil under the humid environment and from the inorganic fertiliser treated soil in the subhumid zone, though not statistically significant (Figure 4.7).

In the second year, 112 kg NO_3-N ha^{-1} was lost through leaching from sludge, while 68 kg NO_3-N ha^{-1} was leached from inorganic fertiliser treatments under humid rainfall. Under subhumid rainfall, inorganic fertiliser treatment lost 11 kg NO_3-N ha^{-1}, whereas sludge lost only 5 kg NO_3-N ha^{-1} from the soil through leaching. Percentage differences revealed that under humid rainfall, sludge had a 54% higher cumulative nitrate leaching than the inorganic fertiliser. Conversely, inorganic fertiliser had 39% higher cumulative nitrate leaching than the sludge under subhumid rainfall. It is evident in the second-year trial that inorganic fertiliser had less frequent but higher cumulative nitrate leaching. Sludge, conversely, had lower concentration but more frequent nitrate leaching, cumulating to higher concentration. This may be attributed to the slow release of inorganic N from sludge. In conclusion, the sludge recommendation provided by the SARA model provides a reasonable maize grain yield comparable to inorganic fertiliser under subhumid rainfall without compromising the groundwater through nitrate leaching.

4.5 Conclusion

Sludge applications, mostly based on N crop demand, must be considered in the diverse environmental conditions to estimate application rates, paramount to environmental protection. Results from the current study indicate that maize grain yield from sludge-amended soils was similar to inorganic fertiliser. The only exception was during the second year under the subhumid zone when the rain was below the normal average. The effect of low rainfall in sludge-treated maize crops is double-fold; it lowers crop nutrient availability, especially nitrogen, attributable to the decrease in mineralisation rate and low photosynthesis due to water deficiency.

Nitrate leaching somewhat increased with the availability of rainfall, therefore, it was higher in humid compared to subhumid agroecological zones. Nitrate leaching did not vary between sludge and inorganic fertiliser treated soils within the same agroecological zone. In conclusion, the sludge recommendation provided by the SARA model provides a reasonable maize grain yield comparable to inorganic fertiliser under subhumid rainfall without compromising the groundwater through nitrate leaching. It is recommended to replicate this study under field conditions of the humid agroecological zone to ascertain these results attributable to the disparities in temperatures of humid and subhumid agroecological zones.

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