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Closing the loop: crop yield response and soil health implications of human excreta-derived fertilizer application in dryland agriculture

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Introduction: Human excreta-derived fertilizers, such as sewage sludge-based co-compost and urine, offer a potential pathway to improve soil fertility and crop productivity in dryland agriculture while promoting circular bioeconomy practices. However, their agronomic performance and environmental safety remain underexplored.

Methods: A six-month field experiment was conducted at the Bishopstowe Agricultural Living Lab using a randomized complete block design with five treatments (urine, urine + co-compost, chicken manure, no fertilizer, and conventional fertilizer) and four replications. Chili (*Capsicum annuum*) yield, soil nitrogen dynamics (mineralization, ammonium, nitrate), enzyme activities (urease, β -glucosidase, phosphatases), microbial communities, soil carbon fractions, and groundwater contamination risks (*Escherichia coli*, nitrate) were assessed.

Results: Chili yield, soil N mineralization, ammonium and nitrate concentrations, enzyme activities, microbial community composition, and active carbon did not differ significantly across treatments ($p > 0.05$). In contrast, organic carbon and extractable phosphorus were significantly higher in the urine + co-compost and chicken manure treatments. Although co-compost contained high organic carbon, associated microbial activity was lower than that observed with chicken manure. Groundwater contamination by *E. coli* and nitrate was not significantly affected by excreta-derived amendments during the trial period.

Discussion and Conclusion: Excreta-derived amendments enhanced soil organic carbon and phosphorus without compromising groundwater quality, highlighting their potential role in resilient and sustainable food systems. Nevertheless, their safe use requires management practices such as salinity control and adherence to WHO sanitation guidelines. The study was limited by its short duration and single-site scope; therefore, long-term (>2 years) and multi-location trials are recommended to fully assess agronomic, soil health, and environmental outcomes of excreta-derived fertilizers in dryland agriculture.

KEYWORDS

biostimulants, circular bioeconomy, climate change, human excreta fertilizers, regenerative agriculture, soil microbiology

1 Introduction

Sub-Saharan countries are burdened with poor waste management, pollution, food insecurity, and biodiversity loss. Management of human excreta from onsite sanitation systems is of great concern in Sub-Saharan Africa. About 80% of the 2.8 billion people globally served by pit latrines are from Sub-Saharan Africa (Strande et al., 2014). Studies from 39 global South cities show that 58% of human excreta is not safely managed, 14% of fecal sludge is contained but not emptied, 13% is emptied but not delivered to the treatment plants, and 2% that reaches the treatment plant is not treated (Peal et al., 2020). This exposes communities to public health risks, and studies have linked outbreaks of major diarrheal diseases to poor faecal sludge management (Mamera et al., 2021; Ngasala et al., 2019; Back et al., 2018). This concerns South Africa, a water-scarce country, considering rolling out boreholes as part of the National Groundwater Strategy (NGS) for potable water supply in rural areas (WRC, 2017). Therefore, improving fecal sludge collection, transport, and treatment is essential to protect public health and groundwater resources.

Apart from sanitation, food insecurity is another challenge faced in South Africa. About 21.7% of Black Africans, especially in rural areas, are severely food insecure (STATSSA, 2025). Apart from poverty, unemployment, and reliance on social grants, poor crop yields are drivers of food insecurity in South Africa. South African soils are not productive, some being acidic and nutrient-deficient. About 19.5% of soils in the Western Cape have a pH (KCl) < 5 (Liebenberg et al., 2020), natural acidification has been reported to affect about 16 million ha of cultivated lands in South Africa (van Huyssteen and du Preez, 2023) and this has been problematic, especially for smallholder farmers, who are mainly producing under rainfed conditions (van Huyssteen and du Preez, 2023). This is exacerbated by nutrient mining and the minimal use of organic fertilizers (Anyega et al., 2021), and persistent droughts (Orimoloye, 2022). Elagib et al. (2024) reported that by 2050, intensifying droughts will likely affect food security in 4.30 and 28.8% of areas under dryland agriculture in wet and dry regions, respectively. About 65% of Sub-Saharan soils are degraded due to minimal use of organic matter and nutrient mining (Zingore et al., 2015). This also threatens soil health as well as the capacity to produce food for future generations optimally (Louw et al., 2014; Montgomery and Bikié, 2021). Addressing these challenges requires improving soil health and climate-resilient farming practices to safeguard food security.

The closed-loop circular bioeconomy is an innovative approach to solving the intertwining food security and sanitation challenges in South Africa (Sekabira et al., 2023). Human excreta can be valorized into soil organic amendments, such as co-compost (Nikiema et al., 2020). This aligns with the South African National Environmental Management Waste Act 59 of 2008 (DFFE, 2019), which emphasizes the transition towards zero landfilling of organic waste by 2050. While at a global scale, the co-compost production and agricultural use are important for eliminating hunger (Sustainable Development Goal or SDG 2), ensuring good health and well-being of people from exposure to hazardous waste (SDG 3), mitigate climate change from landfills emissions by sequestering carbon in soils (SDG 13) and prevent soil biodiversity loss while enhancing life on land (SDG 15) (UN, 2025).

The agricultural use of co-compost has several agronomic benefits, including increasing soil organic matter, water and nutrient use efficiency, soil health, and crop yields (Suddick and Six, 2013; Yaseen et al., 2020; Fuhrmann et al., 2022). However, this has human health concerns regarding heavy metals, pathogens, and micropollutants. These challenges are addressed in the World Health Organization's (WHO) Sanitation Safety Planning guideline through the application of a multibarrier approach. Part of the multibarrier approaches in treating human excreta to eliminate pathogens is done by thermophilic temperatures attained during co-composting. When it comes to heavy metals, the South African regulations require the co-compost to meet certain minimum limits (RSA, 1947), which are possible by mixing a certain portion of sewage sludge with green waste, which is generally low in heavy metals. The only challenge comes with micropollutants and antibiotic resistance genes, which have been detected in sewage sludge, but their impacts on the environment and health risks are still grey and subject to ongoing research.

The use of biofertilizers as a climate change adaptation to maintain soil biodiversity for sustainable agriculture in uncertain rainfall conditions of dryland agriculture is a well-articulated topic, but with some grey areas. The long-term ability of organic fertilizers to increase soil health in terms of microbial activity and water retention capacity for sustainable dryland agriculture was evidenced using sewage sludge alone (Desjardins et al., 2025). The same was observed by Lucchetta et al. (2023) from 3 years of applying animal manure-derived co-compost in vineyards. The use of human urine alters soil ecology and microbial diversity, but there are concerns regarding soil microbial activity (Yu et al., 2025; Orwin et al., 2010; Abdul Rahman et al., 2021; Bertram, 2009). Conversely, in controlled greenhouse experiments, Rumeau et al. (2024) observed that the high salinity of one-year-stored urine had little effect on soil bacterial communities (only ~3 % of taxa shifted); however, given the greenhouse setting and short-term scope, these findings may not extend to open-field conditions. A study done using a combination of human urine and faecal sludge-derived co-compost was done in controlled experiments, focusing on cabbage physiological performance, not soil health (Häfner et al., 2023).

There is a dearth of studies on the use of sewage sludge and greenwaste co-compost in combination with human urine to complement soil conditioning and nutrient supply for crops, especially in dryland agriculture. This baseline case study sought to bridge the gap by assessing the potential of using human excreta-based organic amendment (co-compost) and biobased fertilizer (urine) to promote crop production under dryland agriculture at Bishopstowe Agroecological Living Lab (BALL) in South Africa.

1.1 Hypothesis

The use of sewage sludge and green waste-derived co-compost together with human urine significantly increases chili (*Capsicum annuum*) yield, improves microbial activity, abundance, and evenness in a single season of dryland agriculture, without contaminating groundwater with *E. coli*.

2 Materials and methods

2.1 Study contextualization

This study was informed by sanitation management practices, challenges, and opportunities in agricultural production systems of small-scale communal farmers from uMsunduzi Municipality, South Africa. Pit latrines serve the rural farmers, while urban farmers use flushing toilets connected to the wastewater treatment plant. The fecal and sewage sludge that is not managed can be valorized into sewage sludge. In addition to that, farmers are using organic materials such as cow dung and chicken manure in their agricultural fields (Sekabira et al., 2023). The farmers receive some extension services from the Department of Agriculture, Land Reform and Rural Development (DALRRD), where they learn about applying inorganic fertilizers in their fields (Zondi, 2023). Due to financial constraints, they rarely have irrigation and depend mostly on dryland farming. They grow various crops, including Yellow maize (*Zea mays* L.), Swiss chard (*Beta vulgaris* L.), green and dry beans (*Phaseolus vulgaris* L.), and Chilies (*Capsicum annum* L.) (Adey et al., 2004). Linking resource recovery and reuse with existing indigenous knowledge systems is important to spearhead the concept to the next level, bringing impact to communities while transforming existing food systems for future generations.

2.2 Study site characteristics

The study was done at the Bishopstowe Agroecological Living Lab (BALL) located in uMshwati Local Municipality within uMgungundlovu District Municipality (29°35'09.9" S 30°28'45.4"E). The BALL is a newly established agroecological hub where green technologies are tested, demonstrated, and transferred to potential beneficiaries. The soils are generally red clay soils with >50% clay content down to 1 m depth (Table 1). The slope is approximately 7.4°, and the altitude ranges from 451–50 m above sea level.

2.3 Experimental materials

The co-compost used during the study was made by mixing one part of sewage sludge from the Ixopo Wastewater Treatment Plant with three parts of green organic waste from various garden service providers in Pietermaritzburg, South Africa. The co-composting was done in windrows for 5 to 6 months to ensure that the organic matter is fully digested and free from pathogens due to thermophilic temperatures. The compost attained temperatures between 32°C and

55°C. The 55°C was maintained for at least three weeks, which was enough to sterilize the pile.

The urine used to supplement crop nutrients was collected from the Durban Fresh market public urinal, courtesy of a Non-Governmental Organization called Asiye eTafuleni (AET), following standard methods for urine collection, handling, and transportation (Tilley et al., 2014). Chicken manure was obtained from a small-scale chicken farmer at the BALL. The physicochemical properties of materials used during the study are shown in Table 2.

2.4 Experimental methodology

Twenty plots of 12 m² (3 m × 4 m) were laid out in a randomized complete block design consisting of five treatments to four replicates. The treatments were co-compost + urine (T1), poultry manure (T2), urine (T3), no fertilizer (T4) and conventional fertilizer (T5).

2.4.1 Trial establishment and management

Studies were done in dryland farming to represent agricultural practices commonly done by small-scale farmers in South Africa, who cannot afford to establish irrigation systems. The experiments were set on a land of 17 × 15.5 m. The land was ploughed and disked a month earlier. Plots 3 m wide and 4 m long, giving an area of 12 m², were made using hand hoes. Organic fertilizers (co-compost and chicken manure) were applied based on crop N requirements recommended by the Fertility and Advisory Services (FAS) of the KwaZulu-Natal Department of Agriculture and Rural Development (DARD) soil analysis results. Organic fertilizers have a slow nutrient release rate, so the application rate was calculated following Equation 1 according to the methods recommended by Tesfamariam et al. (2020).

$$\text{Co-compost application} = \frac{\text{Crop N requirements (kg ha}^{-1}\text{)}}{\text{Mineralization rate (\%)} \times \text{compost N content (\%)}} \quad (1)$$

In Equation 1, the mineralization rate for the BALL site (29%) was obtained from Ogbazghi et al. (2016) with an assumption that Pietermaritzburg is within the sub-humid region. The compost N content was obtained from the FAS analytical results. The organic fertilizers were applied on each planting station at a rate of 20 tons per hectare a month prior to planting.

Chilies (*Capsicum annum*) of the variety Star 6,604 was selected as a test crop due to its potential for processing into chili paste and

TABLE 1 General soil physical properties of the experimental field at BALL at three different soil depths.

						Moisture retention	
Soil depth	Clay	Fine silt	Sand	Texture class	Bulk density	FC	PWP
m	%				kg m ⁻³	m ³ m ⁻³	
0.3	51.3	16.3	32.4	Clay	1.1	0.36	0.21
0.6	55.7	16.7	27.6	Clay	1.1	0.36	0.21
0.9	59	17	24	Clay	1.1	0.36	0.21

Clay has particles <0.002 mm, fine silt has 0.02–0.002 mm, and sand has 0.02–2 mm.
FC, Field capacity; PWP, Permanent wilting point.

TABLE 2 Physicochemical characteristics of materials used during the study in comparison with the South African fertilizer, farm feeds, Agricultural Remedies and Stock Remedies Act of 1947 (RSA, 1947).

Characteristic	Co-compost	Urine	Chicken manure	Standard limits
pH	6.3	8.8	6.1	6–9 ^a
EC ($\mu\text{S cm}^{-1}$)	1996.7			
C: N ratio	13.28			
Moisture (%)	31.2	95	39.8	<40%
Total C (%)	20	–	21	>17.4% ^a
Total N (%)	1.2	0.2	5.5	0–6% ^a
Phosphorus (%)	0.5	0.01	2.2	0.25% ^a
Potassium (%)	0.3	0.08	2.3	0.2% ^a
Calcium (%)	1.3	0.0014	10.6	3.0% ^a
Magnesium (%)	0.2	0.003	0.9	0.3 ^a
Sodium (%)	0.07	0.01	0.47	<3 ^a
Manganese (mg kg^{-1})	–	–	400	–
Iron (mg kg^{-1})	–	–	1,000	–
Aluminium (mg kg^{-1})	2,052	–	600	–
Cadmium (mg kg^{-1})	1	–	–	<40
Chromium (mg kg^{-1})	300	–	–	<1,200
Cobalt (mg kg^{-1})	5	–	–	–
Copper (mg kg^{-1})	101.5	–	–	<1,500
Lithium (mg kg^{-1})	10	–	–	–
Nickel (mg kg^{-1})	20	–	–	<420
Lead (mg kg^{-1})	25	–	–	<300
Zinc (mg kg^{-1})	330	–	500	<2,800
<i>E. coli</i> (CFU/100 g)	<3	–	–	<1,000
Ascaris (Viable eggs/kg)	25	–	–	62.5

^aDenote ranges that are expected for an ideal co-compost without legal implications.

Source: CCME (2005).

powder by the farmers at BALL, and according to the WHO (2006) multibarrier approach specifications have lower microbial contamination risks. The seedlings obtained from a local registered nursery were planted on 17 November 2022 at a spacing of 0.5 m × 0.5 m, and the test crop lasted for 6 months. The inorganic fertilizers were applied at recommended rates (Urea: 326 kg ha⁻¹, Single Super Phosphate (SSP): 1,960 kg ha⁻¹, Potassium Chloride (KCl): 558 kg ha⁻¹), which directly translates to 150:196:290 (N: P: K). The urea was split-applied; 50% was applied a week after planting, the other 50% was applied a month after planting (25%) and 3 months after planting (25%). The SSP and KCl were applied once off at planting. The fertilizers were banded and covered up by the soil soon after application. The urine was stored for 1 month at room temperature, as recommended by WHO (2006). The stored urine N content value of 3.9 g L⁻¹, which was obtained from previous studies (Odindo et al., 2022) was used to calculate crop N requirements following Equation 2.

$$\text{Urine} \left(\text{L m}^{-2} \right) = \frac{\text{Crop N requirement} \left(\text{g m}^{-2} \right)}{\text{N content in urine} \left(\text{g L}^{-1} \right)} \quad (2)$$

The urine was diluted with water at a ratio of 3:1 and applied at the same time as inorganic fertilizers (a week, a month, and 3 months after planting), directly to the soil surface of each plant using the watering container to avoid foliar contact (Richert et al., 2010). An integrated weed, pest, and disease management program was implemented. The selected chili variety has a general tolerance to leaf diseases; however, agrochemicals were not used to control pests and diseases because they were not identified.

The data obtained from the National Aeronautics and Space Administration (NASA) access viewer database (NASA, 2022) was used to monitor climatic parameters for optimum crop growth. Comparisons were made between the amounts of rainfall received on the site during the crop growing period with historic monthly mean rainfalls averaged for >30 years. Water balances were done by calculating the monthly average crop evapotranspiration following Equation 3.

$$\text{ET}_{\text{crop}} = \text{ET}_{\text{pan}} \times K_c. \quad (3)$$

Whereby the ET_{crop} is the historic average crop evapotranspiration, ET_{pan} is the historic average reference evapotranspiration measured

from the class A evaporation pan, and K_c is the crop factor value for the chili crop. A simple water balance assessed the rainfall sufficiency for crop production by subtracting average monthly ET_{crop} from actual monthly rainfall.

Soil chemical and biological properties were measured before planting and after harvesting. Samples for chemical analyses were collected within the 0.3 m soil depth from five different spots within each plot. These were bulked to form composite samples for each experimental unit. The samples were submitted to the Fertility and Advisory Services (FAS) of the Department of Agriculture Land Reform and Rural Development laboratory for the analysis of organic carbon, cation exchange capacity, nutrients (organic N, mineral N, extractable P, extractable K, Ca, Mg, Cu, Al and Fe), soil pH (KCl), acid and base saturation according to the standard methods (Manson et al., 2020).

2.4.2 Crop yield

The chili harvesting started 12 weeks after transplanting. Only red chilies were harvested, leaving green ones and flowers. The fresh mass from the harvested chilies was measured immediately using a balance with ± 0.02 g accuracy. The yield per harvest was determined according to Equation 4.

$$\text{Yield (kg ha}^{-1}\text{)} = MA \times \frac{10,000 (\text{m}^2)}{1,000 (\text{g})} \quad (4)$$

Whereby the “M” is the mass of chilies in grams per plant. “A” is the area per plant, referring to plant spacing of 0.5×0.5 m (0.025 m^2). The cumulative yield was then determined as the sum of mean yield values from all the treatments and three harvests obtained from the tables of means calculated by the GenStat software.

2.4.3 Soil microbiology

About 200 g of soil was sampled from the 0.3 m topsoil level, within a 0.1 m radius of the plant, before planting and after the final crop harvest. Each sample was placed in zip-lock plastic bags at room temperature for not more than 5 days and submitted to Sporotec Soil Microbiology Laboratories within the Department of Microbiology at Stellenbosch University to analyze microbial communities and soil enzymatic activity. Microbial communities are important in nutrient recycling. The more even the taxa, the more stable it is to continually carry ecosystem services (Drosos et al., 2023). Therefore, microbial communities were analyzed using the molecular fingerprinting technique, which generated a profile for each composite soil sample (Singh et al., 2006).

Soil biochemistry is an important indicator of soil health that can help understand the influence of certain soil amendments on soil ecology, which drives optimal microbial processes. When organic matter decomposes in the soil, microorganisms release various enzymes that break down complex molecules such as urea, cellulose, and phosphates, producing carbon dioxide and inorganic nutrients (Yu et al., 2025). Therefore, N mineralization, urease activity, β -glucosidase, PO_4^{3-} -P, alkaline and acid phosphatase, microbial activity, and organic and active carbon were measured according to standard methods (Karlen et al., 2021). The aerobic incubation method was used to determine soil mineralization after incubating soil samples at 25°C for 14 to 28 days. Initial and final

concentrations of inorganic nitrogen (NO_3^- -N and NH_4^+ -N) were extracted with 2 M KCl and measured colorimetrically using the Thermo Scientific™ Gallery™ Discrete Analyzer, to calculate net N mineralization (Maynard et al., 2007). The β -glucosidase was determined by incubating a soil sample with a substrate containing p-nitrophenyl- β -D-glucopyranoside in a buffer solution at pH 6, 37°C and for 1 h. The reaction was stopped by adding calcium chloride, and the absorbance was measured at 400 nm using a spectrophotometer. The Bray II method was used to test the PO_4^{3-} -P, after shaking a soil sample with a solution of 0.03 M NH_4F and 0.1 M HCl and then filtering. The Murphy-Riley method was used to colorimetrically measure PO_4^{3-} -P at 882 nm absorbance. Alkaline and acid phosphatases activity was determined after incubation in a buffer solution, which was acidic (acid phosphatase) or alkaline (alkaline phosphatase), and p-nitrophenyl phosphate as the substrate. The samples were measured spectrophotometrically at 405 nm to quantify the enzyme activity. The fluorescein diacetate (FDA) hydrolysis assay was used to estimate total microbial activity in soil by measuring the enzymatic conversion of FDA into fluorescein. The amount of fluorescein produced, quantified spectrophotometrically, reflects the collective activity of microbial enzymes such as esterases, lipases, and proteases (Adam and Duncan, 2001). The Walkley-Black method was used to measure organic C, and the potassium permanganate (KMnO_4) oxidation was used to measure active and labile C forms.

2.4.4 Groundwater sampling

Groundwater samples were analyzed for suitability as irrigation water and human health safety by the drinking water quality standards. The borehole water samples were collected before planting and after 6 months. These were submitted to the salinity laboratory of the FAS for the analysis of salinity, Na, Ca, Mg, K, Cu, Mn, Fe, Zn, Total alkalinity, SAR, salinity class, and Cl^- according to standard methods (APHA AWWA & WEF, 2017). Using human excreta products is likely to contaminate groundwater with pathogens. Water samples were monitored at a six-month interval and analyzed for nutrients (N and P) and pathogens (*E. coli*) according to Velkushanova et al. (2021).

2.5 Data analysis

The GenStat 21st Edition statistical package software was used for data analysis (VSN International, 2022). The quantitative data was subjected to the analysis of variance (ANOVA) at a 5% significance level. The soil microbial community and biochemical data were subjected to multivariate analysis of variance (MANOVA) at a 5% significance level since there were 19 variables to be considered. Where $p < 0.05$, the Bonferroni multiple comparison test was done to compare treatment differences.

3 Results and discussion

3.1 Climatic information

The climatic information of the BALL for a six-month period (November 2022 to June 2023) is shown in Figures 1A–C. The

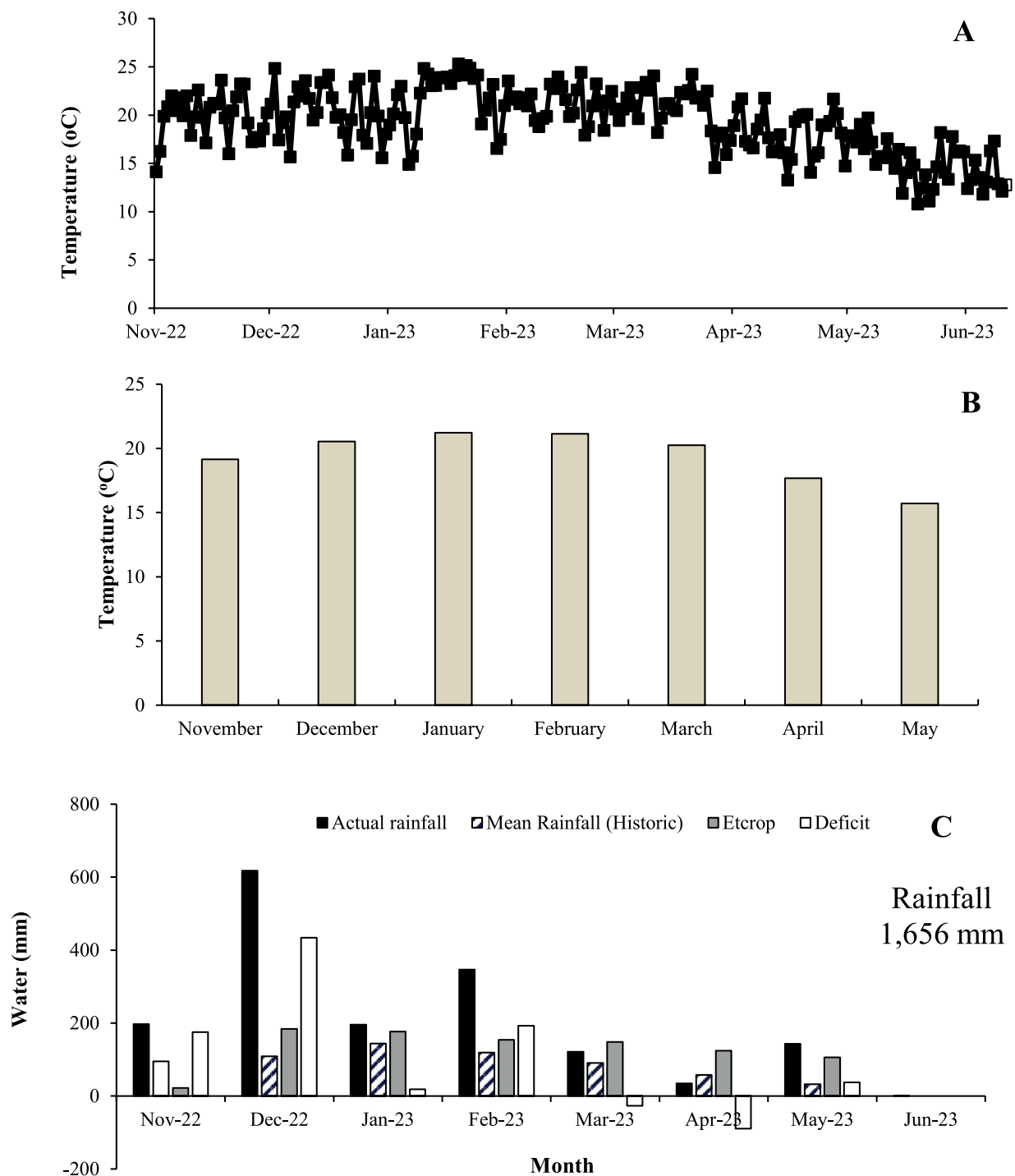


FIGURE 1

The climatic information for Bishopstowe Agroecological Living Lab for a 6-month period showing mean temperatures during the growing period (A), historic monthly mean temperatures (B) and total rainfall (C).

mean daily temperature ranged from 10.8°C to 25.1°C. Chili peppers require a daily maximum temperature of between 20°C and 27°C, and a minimum 15°C for optimum growth (Welbaum, 2015). The mean daily temperatures recorded on the site during the growing period (> 15°C) were ideal to support the optimum growth of chili peppers.

Figure 1C shows that the BALL site received more rainfall during the growing season than the historical average calculated over 25 years. This is a positive indicator from a climate change perspective, especially when droughts threaten food production systems, as Elagib et al. (2024) reported. The irrigation deficit was high in March and April and lower during other months (Figure 1C). During those

TABLE 3 Analysis of variance showing repeated measures for chili yield in five different treatments.

Source of variation	Degrees of freedom	Sum of squares	Mean of squares	Variance ratio	F probability
Replication stratum	3	0.1	0.01	0.09	
Replication × subject stratum					
Treatment	4	1.9	0.48	2.45	0.1
Residual	12	2.3	0.19	2.98	
Replication × Subject × Date stratum					
Degrees of freedom correction factor 0.6833					
Date	2	1.4	0.69	10.53	0.001
Date x treatment	8	2.6	0.33	5.08	0.002
Residual	22	1.4	0.07		
Total	51	8.2			

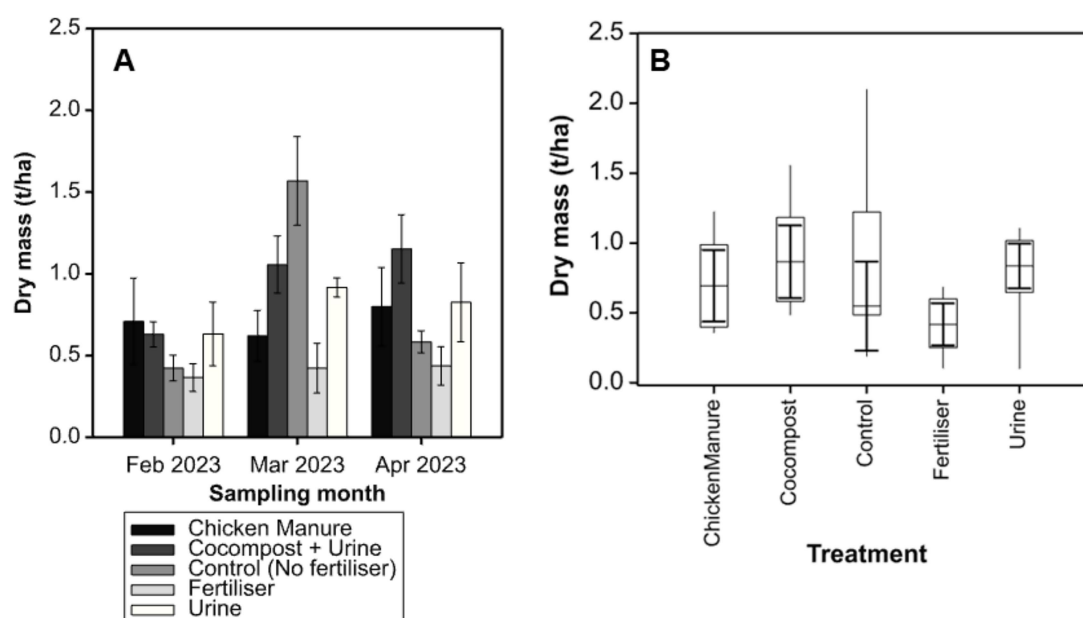


FIGURE 2

Five treatments showing treatment differences ($n = 4$, $p < 0.05$, mean \pm standard error of difference) over time (A) and boxplots (showing median values, 25–75% quartiles and non-parametric standard error of median, $n = 4$) for chilies yield over the three harvesting months (1; February, 2; March and 3; April) (B).

periods, the farmer should have irrigated the crop for optimum yields. This is when dryland becomes challenging because rainfall is not evenly distributed across the season.

3.2 Crop yield

The analysis of variance for the yields of a chili crop is shown in Table 3. The repeated measures on yield significantly differed among treatments over the sampling time.

Figure 2A reports mean values for crop yields amongst the five treatments and across three sampling months. Crops amended with co-compost + urine yielded more red chilies than the no fertilizer and fertilizer treatments during the first month. This indicates that the co-compost + urine treatment grew faster, as exhibited by shorter

days to flowering and subsequent ripening. The increase in yields exhibited by chilies from the no-fertilizer treatment and the abrupt decline in the third harvest were caused by the ripening of fruits and flowers not previously harvested.

The chili pepper yield median values did not significantly differ across all treatments, including the no fertilizer treatment (Figure 2B), indicating the site's inherent fertility. Co-compost is an organic amendment that should undergo mineralization, depending on the soil biotic and abiotic factors, to release bioavailable nutrients (Perez et al., 2023). Studies done using sewage sludge in South Africa found that the mineralization rate depends on the agroecological region's climatic conditions, with super-humid areas exhibiting higher mineralization rates (Ogbazghi et al., 2019). This implies that to cater to the yield gap, the co-compost should be applied earlier (Tesfamariam et al., 2020). Even a survey done on farmers who were

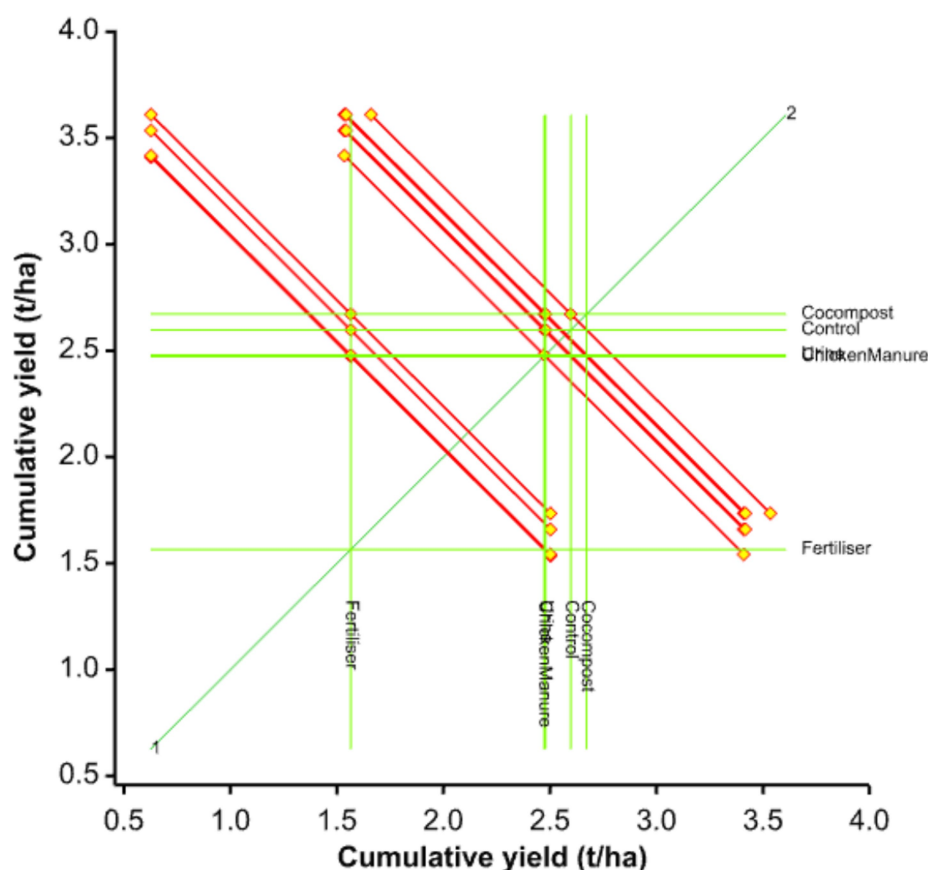


FIGURE 3
Cumulative chili yield shown by mean–mean scatter plot ($n = 4$) for three monthly harvests.

using fecal matter-derived co-compost in India found that crop yields increased by 90% for an average of 2 years (Fendel et al., 2022). Increased lettuce yield after applying co-compost was also reported under dryland agriculture in Malawi (Kamanga et al., 2024). During this study, the co-compost was applied at 20 tons per hectare, double the conventional application rate, to synchronize nutrient bioavailability with crop requirements during critical growth stages. This means that N was not limiting in co-compost amended crops, further explaining the insignificant differences even with the inorganic fertilizer treatment. Similar results were reported with regard to the use of livestock manure on chili, in which the double application rate increased the yields of chili in the clay soil type (Khaitov et al., 2019).

A cumulative yield analysis was also done, and the mean–mean scatter plot is shown in Figure 3. The cumulative mean values for the chili yield were as low as 1.6 tons/ha (fertilizer treatment) and as high as 2.7 tons/ha (co-compost), with a range of between 0.6 tons/ha (1) and 3.6 tons/ha (2). The rainfall was sufficient to promote chili crop growth (Figure 1). Chili peppers are very sensitive to drought due to their broad range of transpiring leaves, shallow root system, and high stomatal conductance (Girmay and Wale, 2019). Given adequate irrigation, chili crops can attain high yields of between 5.4 and 9 tons/ha (Girmay and Wale, 2019). However, the 3.6 tons/ha maximum cumulative yield was enough and not affected by periods of water deficits during March and April (Figure 1). This is confirmed in a crop modeling study using

the SWB Sci model, whereby the simulated harvestable dry mass for the same chili variety ranged from 4 to 5.8 tons/ha, while the results validated using empirical findings reported between 2.11 and 3.68 tons/ha of dry yield (Abebe, 2010). Dry chili yields of between 1.5 tons/ha (conservative conditions) and 4 tons/ha (target) are expected in South Africa (KZNDARD, 2019). Higher yields could have been attained if both red and green chilies had been harvested. Chilies attain higher yields if harvested frequently at a 10–14-day interval because removing both red and green chilies stimulates flowering (KZNDARD, 2019).

3.3 Soil microbiology

Mean squares for soil health indicators in different fertilizer amendments are shown in Table 4. The application of urine and co-compost did not significantly change most of the measured soil health indicators, except for organic C, extractable P, and microbial activity, which significantly differed at the 5% significance level.

3.3.1 Organic carbon

The mean differences in organic C are reported in Table 5. Comparisons amongst the treatments showed significant differences ($p < 0.05$) in organic C between no fertilizer and co-compost + urine (Table 5). The soil organic C significantly increased with the

TABLE 4 Mean squares for soil biochemical analyses and microbial communities in different fertilizer amendments ($n = 4$).

Source of variation	DF	Missing value	Soil moisture	Organic C	Electrical conductivity	pH (water)	NO ₃	NH ₄	Nmin–NH ₄
Replication	3		0.001	0.03	700	0.6	8.4	17	0.2
Treatment	4		0.0004	0.11*	2,117	0.02	21	12	1
Residual	11	–1	0.001	0.03	6,015	0.1	47	40	1
Total	18								

Source of variation	DF	Missing value	Bacterial Shannon index	Bacterial Simpson's index	Bacterial species richness	Fungal Shannon index	Fungal Simpson's index	Fungal species richness	P (Bray II)
Replication	3		0.13	0.005	17	0.09	0.001	37	3,500
Treatment	4		0.02	0.002	2	0.06	0.002	27	9,800*
Residual	11	–1	0.74	0.01	190	0.2	0.004	57	2,700
Total	18								

Source of variation	DF	Missing value	Active carbon	Acid phosphatase	Alkaline phosphatase	Urease	β -glucosidase	Microbial activity
Replication	3		3,939	75,505	473,571	30	4,500	3,067
Treatment	4		14,059	97,282	74,081	21.4	7,740	17,359*
Residual	11	–1	4,308	161,853	198,323	8.2	6,134	3,246
Total	18							

*Significant differences at the 5% level.

application of co-compost + urine compared to no fertilizer, urine, or fertilizer treatments. This is because co-compost contains high carbon content (Table 2). Depending on the forms of organic C, the compost plays a role in sequestering C when non-digestible forms are left to accumulate in the soil (Zhao et al., 2020). In this case, the active organic C fractions did not significantly differ across all treatments (Table 5). The stable organic C forms increase water and nutrient retention capacity, which is important for climate change adaptation.

3.3.2 Microbial activity

The microbial activity was significantly lower in co-compost + urine treatment compared to chicken manure amendment, despite the two differing insignificantly in the observed soil organic C value (Table 5). This observation is frequently reported, as microbial activity is regulated by a combination of factors, including the physical, chemical, and biological properties of the soil (edaphic factors) and the quality of the substrate (Wang et al., 2024; Furtak and Gałazka, 2019; Muneer et al., 2022; Cao et al., 2016). The study was done on one soil type, ruling out the effect of soil properties as an explanation. Soil moisture content also plays an important role in microbial activity. Amending soils with organic matter increases soil moisture retention capacity, further stimulating microbial activity in a water-limited soil (Martín-Lammerding et al., 2021; Hueso et al., 2012). All the treatments were subjected to dryland conditions, but the soil moisture content did not significantly differ ($p > 0.05$) across all treatments (Table 5). Even the microbial activity in the no-fertilizer treatment did not significantly differ from the co-compost + urine (Table A1). These findings explain that, while it is recognized that organic amendments increase organic matter, which boosts microbial activity, their impact within one season was not effective. Even most studies that looked at the effects of organic amendments on microbial activity were done

over long term of at least 2 years (Desjardins et al., 2025; Sayre et al., 2023; Wang and Jiao, 2022). Further, calling for long-term monitoring of the relationship between organic fertilizer amendment and improved soil moisture retention in dryland agriculture.

Variations in organic fertilizer quality, such as C: N ratio, pH, nutrient content, organic matter decomposability, type of organic matter, and maturity, contribute to microbial activity. Characteristics of organic amendments used are shown in Table 2. Chicken manure has high N content (5%) compared to co-compost (1.2%). The C: N ratio plays a role in microbial activity; soil amendments with high C: N ratios have low decomposition rates because microbes have to source N from somewhere, while low C: N ratios provide a balance between required C and N (Li et al., 2018), and initiate protein synthesis required for enzymes and reproduction (Wang and Jiao, 2022), leading to higher microbial biomass. The higher NPK content in chicken manure (Table 2) explains higher microbial activities in the soil compared to the use of co-compost, regardless of the time frame. When it comes to the time frame Jin et al. (2022), found that a single-season application of chicken manure was enough to change the microbial soil processes.

Another possibility could be the stability of the amendment used. Poorly decomposed organic amendments with high moisture content trigger microbial activities because of high concentrations of readily available organic matter, nutrients, microbial loads, and a shift in soil microbial structure (Liu X. et al., 2023). The co-compost used was stabilized by composting for over 5 months, which allowed organic matter to degrade; hence, its application in soil could not trigger microbial activity, just like unstabilized chicken manure. Urra et al. (2019) confirmed that fresh unstabilized chicken and horse manure increase soil microbial activity due to the presence of organic N and P forms. Some studies attributed higher microbial activity in chicken

TABLE 5 Bonferroni test showing treatment differences ($n = 4$; mean \pm standard error of mean deviation) for soil moisture, organic, extractable P, organic carbon, and microbial activity.

Treatment	Soil Moisture (Dry soil/g)	P (Bray II) ($\mu\text{g g}^{-1}$ dry soil)	Organic carbon %	Microbial activity (mg fluorescein- kg^{-1} soil- h^{-1})
Chicken manure	0.937 ^a \pm 0.015	103 ^a \pm 16	0.81 ^{ab} \pm 0.08	62 ^a \pm 8
Co-compost + Urine	0.945 ^a \pm 0.013	145 ^a \pm 47	1.10 ^b \pm 0.12	14 ^{bc} \pm 7
No Fertilizer	0.940 ^a \pm 0.011	30 ^b \pm 12	0.66 ^a \pm 0.11	31 ^b \pm 17
Fertilizer	0.945 ^a \pm 0.012	38 ^b \pm 18	0.73 ^{ab} \pm 0.04	9 ^c \pm 2
Urine	0.965 ^a \pm 0.010	43 ^b \pm 23	0.75 ^{ab} \pm 0.02	13 ^{bc} \pm 5

Superscript letters (a, b, c) indicate statistically significant differences among treatments within each column. Values sharing the same letter are not significantly different ($p > 0.05$), while values with different letters are significantly different ($p < 0.05$) according to the Bonferroni test.

manure-amended soil to changes in soil microbial community structure (Chen et al., 2023; Jin et al., 2022). The studies that reported similar results of chicken manure, although it was not clearly mentioned to be composted, were done for 2 years in soils with a pH of 8.8 (Chen et al., 2023).

The presence of xenobiotics such as heavy metals and micropollutants sewage sludge-derived co-compost is raising significant concerns about their ecotoxicity. High concentrations of heavy metals affect soil microbial activities by disrupting cationic exchange processes while altering soil pH (Abd-Elhalim et al., 2025). However, the sewage sludge-derived co-compost was low in heavy metals, which was below the South African fertilizer legislation standard limits (Table 2). Even the co-compost pH was within the optimum ranges for agricultural use, and the reported soil pH did not significantly differ (Table 5). Meaning that the co-compost was environmentally safe for agricultural use. However, Panneerselvam et al. (2022) discussed the importance of micropollutants such as drugs and pesticides, which might find their way into the wastewater treatment system, contaminating the resulting sludge. Micropollutants are deemed emerging contaminants because there are no consistent regulations to guide their use against ecotoxicity (Yu et al., 2025; Häfner et al., 2023; Gul et al., 2022). This is one of the grey areas in the use of human excreta-derived fertilizers, requiring further studies.

Higher microbial activity is not always good for agriculture because increased microbial biomass immobilizes inorganic N, making it unavailable for crop uptake. Meaning that co-compost is a good soil amendment in dryland agriculture because of its C: N ratio from mixing organic garden waste and sewage sludge.

Closing nutrient loops and enhancing soil health using biobased fertilizers such as urine and co-compost is important. The findings showed that even the microbial activity in co-compost + urine, urine, and fertilizer treatments was significantly ($p < 0.05$) lower than no-fertilizer amendment (Table 5). This was not expected because organic carbon from co-compost and mineral nutrients provided by urine were supposed to boost microbial activity. There are mixed findings on this matter in the literature. Yu et al. (2025) found that the application of human urine in soil can increase microbial activity due to the presence of urea, which stimulates urease activity, as well as the presence of dissolved organic carbon and a shift in soil pH due to an increase in electrical conductivity. This was reported in similar studies, whereby the addition of bovine urine increased microbial activity after stimulating ammonium-nitrifying bacteria (Orwin et al., 2010). Despite high soil salinity due to the use of urine, the microbial activity still increases in a similar way to applying inorganic fertilizers (Rumeau et al., 2024), which is consistent with the lack of significant

differences between urine and fertilizer treatments. However, higher microbial activity in non-amended soil shows that the application of amendments altered natural processes in the short run. A meta-analysis by Liu W. et al. (2023) found that there are various factors that can stimulate microbial activity, an initial change in soil pH being one of them. However, this cannot be a cause in this case because the soil pH did not significantly differ across all treatments (Table 5). As confirmed in literature, sometimes the application of urine fertilizers elevates the soil salinity, shifting the microbial balance by reducing the microbial balance of sensitive microbes such as *Bacillota* (Yu et al., 2025). The same applies to the use of inorganic fertilizers (Carrascosa et al., 2023; Lian et al., 2022). A $> 25\%$ decline in microbial activities in the soil amended with inorganic fertilizers was also reported (Elbl et al., 2019). A stable soil ecosystem is created when inorganic fertilizers are applied together with manure or organic amendments (Lian et al., 2022). Reduction of microbial activity after the use of co-compost and urine could have been triggered by microbial stress. Microbial stress in this context refers to the ability of microorganisms to adapt to adverse conditions such as pH, temperature, and other extreme conditions (Abdul Rahman et al., 2021). Some microorganisms may succumb due to their inability to synthesize proteins that help them adapt to environmental stresses, as was evidenced in studies done using cow urine (Bertram, 2009). However, in this juncture, the influence of soil salinity in response to the use of urine is not a factor because the electrical conductivity did not significantly differ ($p > 0.05$) across all treatments (Table 5). Underscoring the need for further investigations into this matter with longer-term studies, as proposed by Yu et al. (2025).

3.3.3 Enzymatic activity

The sum of metabolic processes taking place in the soil can be determined by enzymatic activities. This section digs deep into specific enzymes responsible for nutrient cycling. During microbial degradation of organic matter in the soil, enzymes such as β -Glucosidase are released to break down β -1,4-glycosidic bonds in glucosides, oligosaccharides, and various glycoconjugates, including cellulose from organic matter (Adetunji et al., 2017; Bastida et al., 2008; Ansari et al., 2023). When it comes to nitrogen cycling, urease enzymes hydrolyze urea into ammonium-N, which, depending on soil ecological factors such as pH, can further be nitrified by nitrifying bacteria into plant bioavailable forms (Yu et al., 2025). However, both the urease and the β -Glucosidase activities in this study did not significantly differ across all the treatment groups ($p > 0.05$) (Table 5). This further explains that the higher microbial activities in chicken manure were not linked to the amendments

applied. This is further supported by the fact that the soil active carbon (labile carbon) pool, which serves as the readily available food source for microbes (Orwin et al., 2010; Sayre et al., 2023; Li et al., 2024) did not significantly change in response to the amendments applied. This implies that the application time frame and loading rates could have been very short to show significant changes in the soil. Most studies were done for 2 years or beyond (Ansari et al., 2023; Liu W. et al., 2023; Drosos et al., 2023). Therefore, the six-month experimental period, although during the rainy season of the study site, which received 1,656 mm of rainfall (Figure 1C), was not enough to influence soil health.

3.3.4 Nitrogen and phosphorus mineralization

Table 5 reports on N mineralization and extractable P results. The N mineralization rate did not significantly differ amongst all treatments ($p > 0.05$), while extractable P significantly differed ($p < 0.05$). Chicken manure and co-compost had significantly higher median values of extractable P. This is attributed to relatively high extractable P concentrations in organic amendments (Table 2). This cannot be linked to P mineralization, because the acid and alkaline phosphatase activities did not significantly differ in all treatments (Table 5). Several studies have confirmed that the P in untreated urine is generally lower than N and K, and these are lower than in an inorganic fertilizer (Jönsson et al., 2004; Richert et al., 2010; Alemayehu et al., 2020). Low concentrations of extractable P in soils that were applied inorganic fertilizer resulted from high solubility of Single Super Phosphate, uptake by plants, or loss from the topsoil. Organic matter increases the surface area for soil P adsorption capacity, allowing soils to capture and retain inorganic phosphates. This explains why the co-compost and urine combination and chicken manure had higher extractable P median values. The ability of organic amendments such as co-compost to improve nutrient retention by creating adsorption sites that retain cations and anions is well-documented and recommended from environmental and agronomic perspectives (Urrea et al., 2019; Snyman et al., 2006; Fuhrmann et al., 2022). The only challenge is when phosphorus is transported to nearby water bodies through surface runoff, and causes pollution if proper slope management strategies are not put in place (Sharpley, 2016).

3.3.5 Microbial community

Microbial activity may not be a good indicator of soil health in the short run, as it did not show the activity of beneficial microbes around nutrient cycling. Lower microbial activities were observed in co-compost, urine, and even in inorganic fertilizer-amended soils, while relatively higher in the chicken manure and even in unamended

soils. Some other soil biodiversity indicators, such as the Shannon Index, the Simpson Index, and Species richness, can be used as soil health indicators. The Shannon index quantifies the diversity of microbial communities by integrating the number of different species (richness) and their relative abundance (Meng et al., 2016). This has been used in various agroecosystems to assess the effects of various management practices on soil microbial diversity (Meng et al., 2016; Sayre et al., 2023; Drosos et al., 2023). These indicators did not significantly differ across all the treatments (Table 5), indicating that the short-term use of the organic amendments did not significantly change microbial diversity, regardless of notable differences in soil organic carbon compositions and microbial activity. This clearly shows that the microbial responses to organic amendments was not significant over the studied period.

3.4 Groundwater contamination

The sewage sludge is a byproduct of the wastewater treatment processes, whose quality depends on anthropogenic activities from wastewater generation to the final destination. Stormwater intrusion and illegal discharge of industrial waste introduce heavy metals into the wastewater treatment system. Therefore, to safeguard the use of sewage sludge-derived fertilizers, there are regulations to limit the concentrations of heavy metals and pathogens. When it comes to pathogens of concern, *E. coli*, helminths, and Salmonella are used as indicators. However, the heavy metals accumulation was not included in the study because the product used complied with the South African legislation. In this case, *E. coli* was monitored due to potential regrowth after treatment (Sharma and Reynnells, 2016; Kim et al., 2009; Zaleski et al., 2005). Based on findings in Table 6, *E. coli* was not detected in groundwater because of the thermophilic sludge treatment processes that deactivated microorganisms (RUNRES, 2023). The same applies to nitrates, produced from microbial processes, which, due to their negative charge, easily leach in the soil, causing blue baby syndrome to consumers using groundwater. Its absence from groundwater is a good indicator of public health safety. Even though *E. coli* was not detected; potential pathogen risks from biosolids should not be overruled. Therefore, the standard multibarrier approaches recommended by WHO, as well as monitoring the groundwater, should always be implemented. The South African Guideline for utilization and disposal of sewage sludge recommends that nutrients be monitored before and after each planting, while trace elements be monitored at least once in 5 years on each receiving soil (Snyman et al., 2006).

TABLE 6 Groundwater sampling results for *E. coli* and nitrate before planting and 6 months after crop establishment.

Sample number	Sampling time	Nitrate	<i>E. coli</i>
1	Nov	nd	<1
2	Nov	nd	<1
3	Nov	nd	<1
1	May	nd	<1
2	May	nd	<1
3	May	nd	<1

4 Conclusions and recommendations

4.1 Conclusion

This study examined the short-term effects of sewage sludge-derived co-compost and urine on crop yield, soil health, and potential groundwater contamination under dryland farming. Chili yields showed no significant differences among treatments, likely due to the soil's inherent fertility, harvesting practices, and favorable climatic conditions.

In terms of soil health, organic carbon, microbial activity, and extractable phosphorus (P) responded significantly to treatments. Both co-compost + urine and chicken manure increased soil organic carbon, demonstrating their potential for carbon sequestration. However, microbial activity was lower in the co-compost + urine treatment compared to the no-fertilizer and chicken manure treatments. This reduction may have been caused by urine-induced stress on microbial processes, though the short study duration prevents firm conclusions and highlights the need for further research.

Microbial enzymatic activity (urease and β -glucosidase) and microbial diversity (Shannon Index) did not differ significantly among treatments ($p > 0.05$), underscoring the importance of long-term studies. Such studies could better capture the effects of repeated organic matter additions on soil physical and chemical properties, microbial communities, and their functions. Extractable P was significantly higher with co-compost + urine and chicken manure amendments. Yet, acid and alkaline phosphatase enzyme activities showed no significant differences across treatments, suggesting that organic matter degradation was insufficient to release additional nutrients. Thus, these organic amendments appear to have served as direct sources of bioavailable P rather than stimulating soil enzymatic activity. When it comes to safety concerns, the studies showed no significant change in *E. coli* and nitrate content in groundwater. Overall, the findings indicate that sewage sludge-derived co-compost combined with urine has potential for nutrient recycling and carbon sequestration in dryland agriculture.

4.2 Recommendations

Sewage sludge-derived co-compost and urine can be used as an organic fertilizer to ameliorate degraded sandy soils and provide nutrients such as P. However, management practices to control the effects of salinity through urine dilution to minimize potential challenges to the soil microbiota and soil erosion management practices are essential to protect the environment from pollution. Although there were no pathogen and nutrient contamination risks on nearby groundwater resources, adhering to the WHO Sanitation Safety Planning guidelines is essential.

4.3 Limitations of the study

Due to budget constraints, other soil health measurement methods, such as qPCR, were not done to assess actual microbial communities and link them to specific nutrient cycling. The study was done in a single season and at one specific site, which limits its generalizability to other areas. Future studies should extend

monitoring to at least 2 years across ≥ 2 agro-ecological sites, retaining the current treatment structure while adding a co-compost isolate urine effects. A comprehensive agronomic panel will pair yield and profitability metrics with quarterly soil-health diagnostics and microbial community profiling via qPCR/amplicon sequencing to address the present study's methodological gap. Environmental safety assessments will expand from the 6-month snapshot to quarterly groundwater monitoring (nitrate, ammonium, phosphate, EC, *E. coli*, Salmonella, and helminths) and semi-annual soil/heavy-metal screening, embedded within a WHO Sanitation Safety Planning framework. Data will be analyzed with mixed-effects longitudinal models and used to parameterize mineralization and risk models to enable extrapolation to other regions and seasons, directly responding to the limitations identified here.

Data availability statement

The original contributions presented in the study are included in the article/[Supplementary material](#), further inquiries can be directed to the corresponding author.

Author contributions

WM: Project administration, Formal analysis, Visualization, Writing – original draft, Funding acquisition, Supervision, Data curation, Software, Methodology, Conceptualization, Validation, Investigation. NN: Validation, Writing – review & editing, Formal analysis, Data curation, Investigation. BO: Conceptualization, Writing – review & editing. TB: Writing – review & editing, Formal analysis, Investigation, Data curation. AO: Writing – review & editing, Resources, Funding acquisition, Visualization, Supervision, Conceptualization, Project administration. SO: Supervision, Resources, Writing – review & editing, Conceptualization, Funding acquisition, Project administration.

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Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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Supplementary material

The Supplementary material for this article can be found online at: <https://www.frontiersin.org/articles/10.3389/fsufs.2025.1616479/full#supplementary-material>

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Appendices

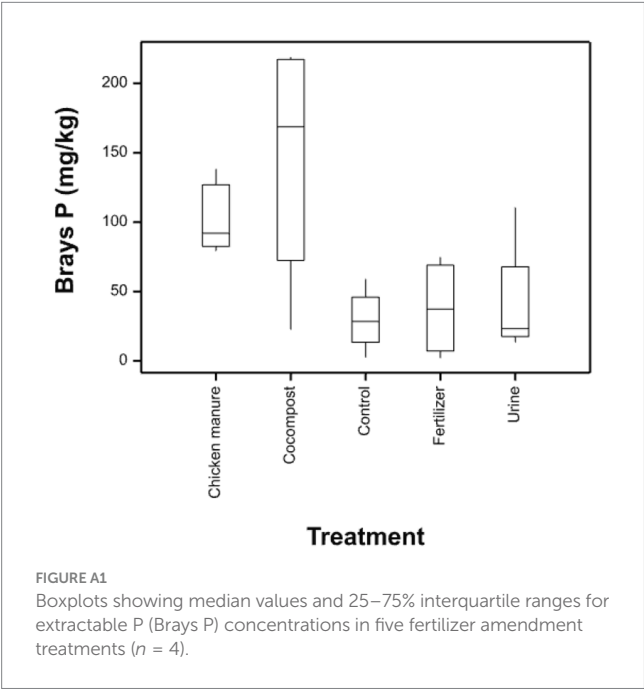


TABLE A1 Comparisons of differences between mean values for organic carbon and microbial activity.

Comparisons	Significant (95% significance level)	
Comparison	Organic C	Microbial activity
No fertilizer vs Fertilizer	No	No
No fertilizer vs Urine	No	No
No fertilizer vs Chicken manure	No	No
No fertilizer vs Co-compost + urine	Yes	No
Fertilizer vs Urine	No	No
Fertilizer vs Chicken Manure	No	Yes
Fertilizer vs Co-compost + urine	No	No
Urine vs Chicken manure	No	Yes
Urine vs Co-compost + urine	No	No
Chicken manure vs Co-compost + urine	No	Yes