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ORIGINAL PAPER

Sewage Sludge Provided Nitrogen To Maize (*Zea Mays* L.) Very Rapidly, Demonstrating a High Short-Term Net Mineralization Rate

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Abstract

Purpose This study aims to evaluate the potential of sewage sludge as a sustainable alternative to traditional farmyard manures, considering the limited availability of such resources in many regions specializing in crop production.

Methods This study compared the application of sewage sludge (SS) and cow manure (CM) at equivalent nitrogen (N) rates in silage maize (*Zea mays* L.) cultivation over two growing seasons. Organic amendments were applied at rates of 50 (SS50, CM50), 100 (SS100, CM100), and 200 (SS200, CM200) kg ha^{-1} of N, alongside an unfertilized control (N0), with three replicates per treatment in a completely randomized design.

Results Dry matter yield (DMY) increased significantly with the applied dose but showed no significant differences between amendments. N availability indices, including the pre-side-dress soil nitrate test, leaf N concentration, and stalk nitrate test, revealed higher N availability from sewage sludge compared to cow manure at equivalent application rates. Apparent N recovery (ANR) by the end of the second growing season was 99.8%, 90.8%, and 74.5% for SS50, SS100, and SS200, respectively, and 76.7%, 75.4%, and 50.5% for CM50, CM100, and CM200, respectively.

Conclusions The rapid mineralization of sewage sludge likely results from its low carbon (C)/N ratio and the absence of lignin and other low-energy organic compounds, which are less metabolically accessible to soil microorganisms. These properties position sewage sludge as an intermediate between conventional organic compost and mineral fertilizers, making it particularly suitable for cropping systems requiring high short-term N availability.

Keywords Organic manure · N mineralisation · Decay series · Apparent N recovery · PSNT · Stalk nitrate test

1 Introduction

The Green Revolution that followed World War II led to a 44% increase in crop yields between 1965 and 2010 (Gollin et al. 2021). The increase in crop yields was facilitated by the adoption of high-yielding varieties, alongside the widespread use of chemical fertilizers, pesticides, and irrigation practices (Weil and Brady 2017). However, intensive farming practices can result in significant global environmental impacts, including reduced biodiversity, soil degradation, and contamination of water bodies and the atmosphere (McDonald et al. 2023; Pisciotta et al. 2015; Poikane et al. 2019; Ribeiro et al. 2019). In the process of agricultural intensification, chemical fertilizers have played a crucial role in boosting crop productivity, but they are also one of the inputs that can cause substantial environmental damage. N fertilizers, in particular, raise significant concerns. The excessive use of N fertilizers results in low N use efficiency, with part of the applied nutrient being lost to water courses

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through leaching (Poikane et al. 2019; Yang et al. 2018) or released into the atmosphere in the form of dinitrogen (N_2) or N oxides (NO_x). Some of these emissions, such as nitrous oxide (N_2O), have a potent greenhouse effect (McDonald et al. 2023; Xie et al. 2024).

Organic amendments, which historically played a significant role in maintaining soil fertility until the first half of the 20th century, are once again being recognized as a means to reduce the reliance on industrial synthetic fertilizers. Numerous studies have demonstrated that the application of organic amendments leads to an increase in soil organic matter, resulting in favourable effects on soil physical properties, biological activity, and nutrient cycling (Afonso et al. 2021; Cardarelli et al. 2023; Dimande et al. 2023; Mondini et al. 2018). A meta-analysis of 769 datasets from 107 research papers underscored that organic amendments increased tomato (*Solanum lycopersicum* L.) yield by 42.2% compared to controls, while also enhancing the content of soluble solids, soluble sugars, lycopene, and vitamin C by 11.9%, 42.2%, 24.0%, and 19.0%, respectively (Gao et al. 2023).

In many regions worldwide, the specialization and mechanization of agriculture have led to a decline in mixed farming systems, resulting in reduced availability of farmyard manure. In response to this challenge, alternative organic materials are being considered for agricultural use. Some of these materials are part of a circular economy strategy, which aims to reuse resources and recover their nutrients in crops (Jastrzębska et al. 2022). The utilization of sewage sludge as an organic amendment in agriculture aligns with the current imperative to promote a circular economy. While sewage sludge typically has a dry matter content ranging from 2 to 8%, dewatering can increase this to values comparable to those of farmyard manure. Additionally, the concentration of nutrients in sewage sludge can be similar to or even higher than those in conventional manures, rendering it a material with high fertilizing value (Buta et al. 2021; Eid et al. 2022; Jastrzębska et al. 2022).

The application of sewage sludge has been shown to enhance soil fertility. Tsadilas et al. (2018) reported an increase in soil organic matter, total N, phosphorus (P), and boron (B) in disturbed lands in China and Greece following long-term application of sewage sludge. In another study involving okra [*Abelmoschus esculentus* (L.) Moench], the use of sewage sludge led to an increase in soil organic C and beneficially decreased soil pH, from 8.38 to 7.34 (Eid et al. 2022). Moreover, Abd Elsalam et al. (2021) reported an increase in soil nitrate and ammonium levels during the maize and fava bean (*Vicia faba* L.) growing seasons after sewage sludge application. Additionally, sewage sludge has been shown to enhance crop productivity in various important crops worldwide (Abd Elsalam et al. 2021; Eid et al. 2022; Jastrzębska et al. 2022; Tsadilas et al. 2018).

Águas do Norte, the company responsible for wastewater treatment plants (WWTP) in the north of Portugal, generates a significant amount of sewage sludge that can be used in agriculture (ADN, 2025). In this region, livestock activity is currently limited, and there is a high demand for organic amendments. However, there is a lack of sufficient studies demonstrating the agronomic value of sewage sludge to both Águas do Norte and local farmers. Therefore, this study aimed to evaluate the potential of sewage sludge from a WWTP for agricultural use. The study involved applying sewage sludge at three different rates in an experimental design that also included equivalent rates of cow manure and an unfertilized control. The rates of organic amendments were selected to achieve uniform N application levels (50, 100, and 200 kg ha^{-1}), as N is a crucial element influencing crop productivity. The test crop chosen was maize grown for silage. The hypothesis proposed was that sewage sludge presents agronomic performance comparable to that of conventional farmyard manure.

2 Materials and Methods

2.1 Site Characterization

This study is based on a two-year field trial conducted at the Poulão farm (41°46'50"N; 6°47'52"W; 701 m above sea level) in Bragança, located in the northeast of Portugal. The experimental plot is part of an eight-year irrigation rotation system, which involves four years of growing forage maize followed by four years of temporary pasture. The study took place during the third and fourth years of the maize phase in the rotation, encompassing two maize growing cycles, with the trials starting in May 2022 and concluding in October 2023.

The climate of the region is classified as Csb under the Köppen system, featuring warm, dry summers and wet winters. The average annual air temperature and precipitation are 12.7 °C and 772.8 mm, respectively (IPMA 2024). The monthly values of air temperature and precipitation observed during the experimental period are presented in Fig. 1.

The soil of the experimental plot is classified as a Eutric Fluvisol (WRB 2022). Its texture is sandy clay loam, with soil separates consisting of 204 g kg^{-1} clay, 239 g kg^{-1} silt, and 557 g kg^{-1} sand. The soil has an organic C content of 17.8 g kg^{-1} (Walkley-Black method), a pH(H2O) of 6.1, and extractable P and potassium (K) levels of 96 g kg^{-1} (P_2O_5) and 106.4 g kg^{-1} (K_2O), respectively, determined using the

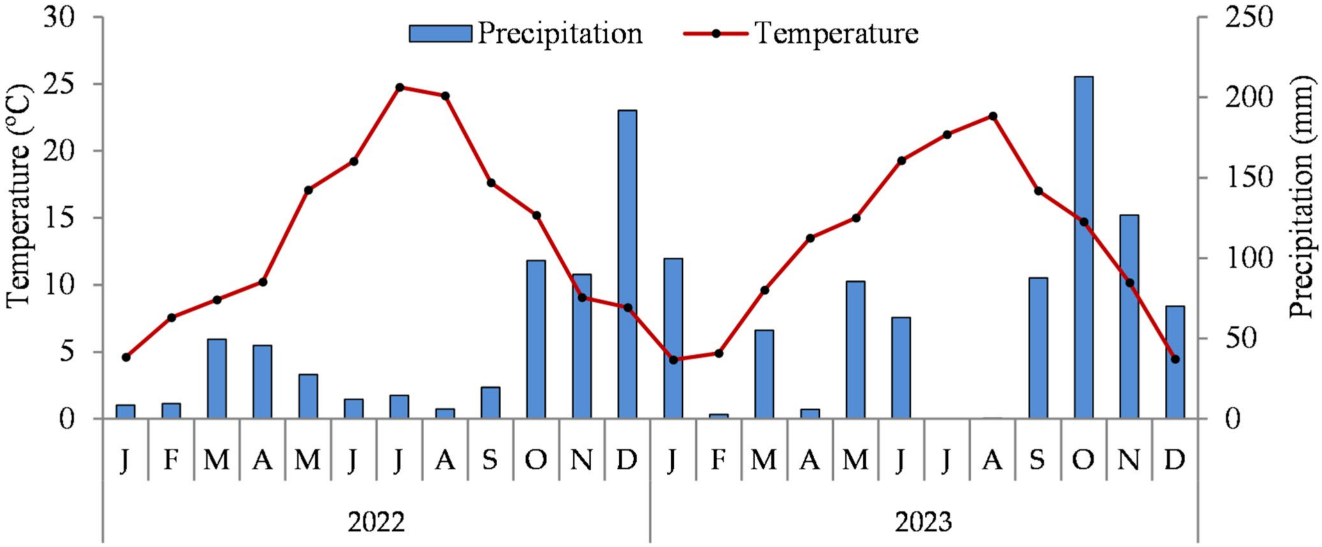

Fig. 1 Average air temperature and monthly precipitation observed during the experimental period at the Sta Apolónia farm weather station, located near the experimental plot

Table 1 Selected properties (average ± standard deviation, $n = 3$) of cow manure and sewage sludge applied as organic amendments in the experiment

Properties	Cow manure		Sewage sludge	
	2022	2023	2022	2023
[1] Moisture (%)	60.6 ± 1.7	63.2 ± 7.4	78.9 ± 4.0	82.0 ± 7.5
[2] Organic carbon (g kg^{-1})	435.2 ± 12.2	415.3 ± 15.9	401.4 ± 2.9	358.1 ± 10.3
[3] pH(H_2O)	9.2 ± 0.1	9.1 ± 0.2	7.7 ± 0.2	8.0 ± 0.2
[4] Nitrogen (g kg^{-1})	22.8 ± 2.4	24.1 ± 0.5	48.5 ± 3.2	49.6 ± 1.2
[5] Phosphorus (g kg^{-1})	6.3 ± 1.3	5.7 ± 0.4	11.2 ± 2.2	16.8 ± 1.3
[5] Boron (mg kg^{-1})	27.8 ± 2.3	25.8 ± 0.6	12.5 ± 0.7	20.2 ± 1.9
[6] Potassium (g kg^{-1})	31.7 ± 4.1	29.0 ± 8.0	1.6 ± 0.3	2.5 ± 0.6
[7] Calcium (g kg^{-1})	8.8 ± 0.6	9.3 ± 1.3	7.2 ± 0.5	20.2 ± 3.0
[7] Magnesium (g kg^{-1})	8.1 ± 0.4	8.8 ± 0.7	2.5 ± 0.2	4.5 ± 1.0
[7] Iron (g kg^{-1})	4.4 ± 0.5	6.3 ± 2.2	5.7 ± 1.1	11.6 ± 3.1
[7] Manganese (mg kg^{-1})	310.6 ± 8.0	343.1 ± 37.9	91.0 ± 7.1	240.5 ± 26.4
[7] Zinc (mg kg^{-1})	112.6 ± 9.2	114.7 ± 6.2	447.5 ± 62.5	790.8 ± 114.9
[7] Copper (mg kg^{-1})	28.0 ± 3.5	32.9 ± 3.4	92.2 ± 5.6	250.6 ± 21.2

[1] Gravimetry; [2] Incineration; [3] Potentiometry; [4] Kjeldahl; [5] Colorimetry; [6] Flame emission spectrometry; [7] Atomic absorption spectrophotometry

Egnér-Riehm method. The cation-exchange capacity (CEC) of the soil is 20.5 cmol$_c$ kg^{-1}.

2.2 Experimental Design and Crop Management

The experiment was arranged in a completely randomized design with a single factor, comprising seven treatments and three replicates. The fertilization treatments included three rates of cow manure (CM), three rates of sewage sludge (SS), and an unfertilized control. The organic amendments were applied at rates corresponding to the application of 50, 100, and 200 kg ha^{-1} of N, with the treatments designated as CM50, CM100, CM200, SS50, SS100, SS200, and Control (N0). Some of the key properties of the organic amendments at the beginning of the trials are presented in Table 1.

Soil preparation began in early April with a pass of a mouldboard plow at a depth of 0.30 m, followed by a pass with a cultivator in the opposite direction. Just before sowing, the plots corresponding to each experimental unit (6 m × 5 m) were marked, and organic amendments were manually applied to the designated plots. The organic amendments were then incorporated into the soil with an additional pass of the cultivator. These operations were carried out in a similar manner in both years of the study. Maize sowing took place on May 17, 2022, and May 10, 2023, respectively. A precision seeder was used, placing maize seeds (hybrid DKC 6181) 0.7 m apart between rows and 0.15 m apart within rows, resulting in a seeding density of 95,238 seeds ha^{-1}.

At growth stage 14 (4 unfolded leaves) (Meier 2018), chemical weed control was carried out using an herbicide containing 44 g L^{-1} of Tembotrione and 22 g L^{-1} of Isoxadifen-ethyl. The commercial product was applied at a concentration of 0.6 L hL^{-1} with a dosage of 2 L ha^{-1}, using a spray volume of 300 L ha^{-1}. The crop was irrigated by a central pivot system, with an estimated annual water consumption of 3,000 m³ ha^{-1}.

2.3 Soil and Plant Sampling

The soil was sampled at the beginning of the study to characterize the experimental plot. Three composite samples (10 cores per sample) were randomly collected across the plot at a depth of 0–0.20 m. Near phenological stage 16 (6 unfolded leaves) (Meier 2018), in both years of the study, a composite soil sample (5 cores per sample) was collected from each experimental plot, also at a depth of 0–0.20 m, to determine the soil's inorganic N content during the plant's active growth phase. At the conclusion of the study in October 2023, the soil was sampled again in each experimental unit (5 cores per composite sample) to assess the effects of the treatments on soil properties, as well as to evaluate the residual inorganic N in the soil resulting from maize fertilization.

During the vegetative growth stage, leaves were collected to assess the nutritional status of the plants. In each experimental unit, six young leaves with fully expanded blades were sampled. The plants were at growth stage 16 (6 unfolded leaves) according to Meier (2018). At the end of the growing season, entire plants were harvested at phenological stage 79 (nearly all kernels have reached final size) (Meier 2018) on September 6, 2022, and August 25, 2023, respectively. The plants were randomly collected from within the plots (10 plants per plot; 5 plants from two adjacent rows). These samples were weighed fresh in the field. A representative subsample was separated from the initial sample, weighed fresh, and then weighed dry after being oven-dried at 70 °C to determine the moisture content. This allowed for the expression of yield results in terms of dry matter per hectare. Additionally, a sample from the lower 15 cm of the stalks was taken to determine nitrate content, which was used as an indicator of N availability in the soil during the growing season.

2.4 Sample Preparation and Laboratory Analysis

Soil samples were first sieved through a 2 mm mesh and then oven-dried at 40 °C. Leaf samples, subsamples of the aboveground biomass, and stalk samples were oven-dried at 70 °C and subsequently ground to pass through a 1 mm mesh.

Soil samples were analyzed for pH (H_2O and KCl) using a soil-to-solution ratio of 1:2.5. The CEC was measured with ammonium acetate at pH 7.0. Organic C was determined via wet digestion using the Walkley-Black method, while extractable P and K were quantified using the Egner-Riehm method with ammonium lactate extraction. Additionally, soil B was extracted using hot water and quantified with the azomethine-H method. These analytical procedures are thoroughly detailed in Van Reeuwijk (2002).

The availability of other micronutrients, including copper (Cu), iron (Fe), zinc (Zn), and manganese (Mn), was assessed by atomic absorption spectrometry following extraction with diethylenetriaminepentaacetic acid (DTPA) buffered at pH 7.3, as per the standard FAO procedure (FAO 2022). Soil inorganic N was determined from soil extracts prepared using 20 g of soil and 40 ml of 2 M KCl. The suspension was shaken for 1 h and subsequently filtered through Whatman No. 42 filter paper. Nitrate and ammonium concentrations in the extracts were then analyzed using UV–Vis spectrophotometry (Baird et al. 2017).

Leaf and plant samples underwent elemental chemical analysis, while stalk samples were specifically analyzed for nitrate concentration. The elemental tissue analyses were conducted using the following methods: Kjeldahl method for N, colorimetry for B and P, flame emission spectrometry for K, and atomic absorption spectrophotometry for calcium (Ca), magnesium (Mg), Cu, Fe, Zn, and Mn following nitric acid digestion of the samples (FAO 2022). Nitrate concentration in the stalk samples was determined using UV-vis spectrophotometry, following the method described by Baird et al. (2017), with a water extract prepared at a dry matter-to-water ratio of 1:50 (m/v).

2.5 Data Analysis

The data were initially tested for normality using the Shapiro-Wilk test and for homogeneity of variances using Bartlett's test. Following these preliminary checks, a one-way analysis of variance (ANOVA) was conducted. For those fertilization treatments that showed significant differences ($\alpha < 0.05$) in the ANOVA, the means were further separated using Tukey's Honestly Significant Difference (HSD) test at a significance level of $\alpha = 0.05$.

3 Results

3.1 Soil Inorganic Nitrogen and Leaf Nitrogen Concentration during the Growing Season

Inorganic N in the soil was measured during the growing season as an index of soil N availability (Table 2). Although this study did not include side-dressing fertilization, the measurement of inorganic N in the soil was conducted at a time corresponding to the widely used pre-side-dress soil nitrate test.

Table 2 Soil nitrate (NO_3^--N), ammonium (NH_4^+-N), and total inorganic nitrogen (Inorg-N) measured at side-dress N application time as a function of sewage sludge (SS) and cow manure (CM) applied at rates of 50 (SS50, CM50), 100 (SS100, CM100), and 200 (SS200, CM200) kg N ha^{-1}, along with a non-amended control (N0)

	2022			2023		
	NO_3^--N	NH_4^+-N mg kg^{-1}	Inorg-N	NO_3^--N	NH_4^+-N mg kg^{-1}	Inorg-N
N0	9.7 a	3.2 b	12.9 b	8.5 b	4.1 bc	12.5 c
SS50	11.9 a	4.3 b	16.1 ab	8.4 b	2.8 c	11.2 c
SS100	13.0 a	4.6 b	17.7 ab	11.2 b	2.9 c	14.1 bc
SS200	13.7 a	7.6 a	21.2 a	17.1 a	4.4 b	21.5 a
CM50	9.7 a	3.7 b	13.5 b	8.1 b	2.3 c	10.3 c
CM100	10.6 a	4.8 b	15.4 ab	9.2 b	2.2 c	11.4 c
CM200	11.0 a	4.4 b	15.4 ab	11.6 b	8.2 a	19.9 ab
Prob.	0.0814	0.0004	0.0099	0.0002	0.0003	0.0002
SE	1.04	0.468	1.32	0.996	0.686	1.364

In columns, means followed by the same letters are not significantly different by the Tukey HSD test (α=0.05)

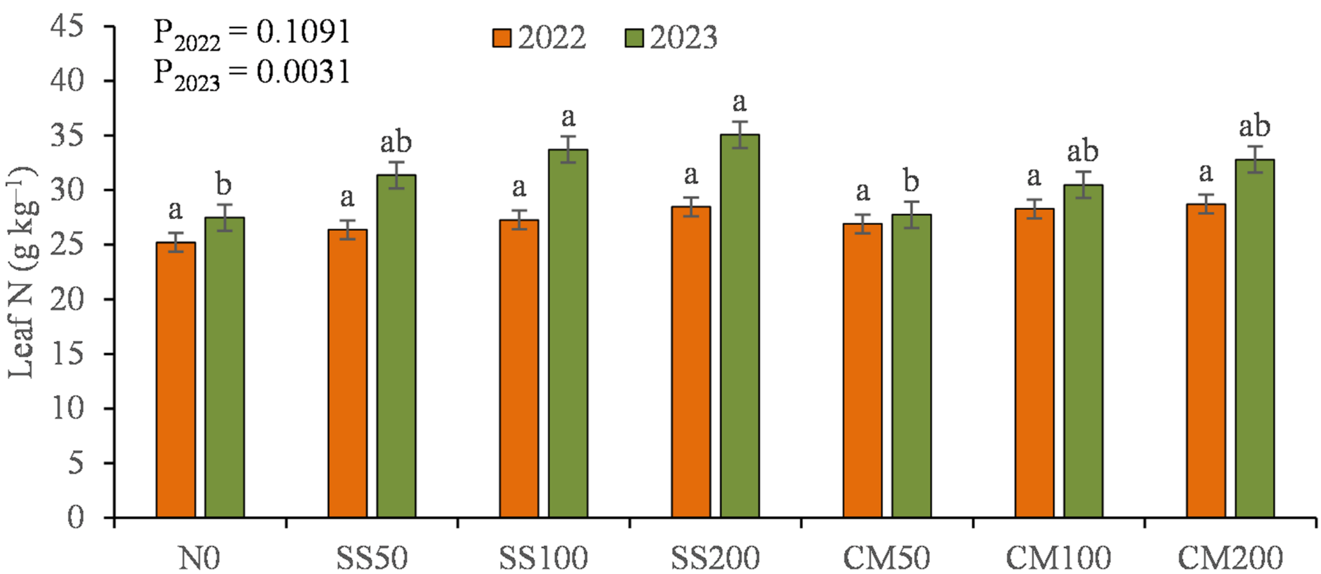

Fig. 2 Leaf nitrogen (N) concentration measured at side-dress N application time as a function of sewage sludge (SS) and cow manure (CM) applied at rates of 50 (SS50, CM50), 100 (SS100, CM100), and 200 (SS200, CM200) kg N ha^{-1}, along with a non-amended control (N0). For each year, means followed by the same letter are not significantly different according to the Tukey HSD test (α=0.05). The error bars represent the standard errors

The nitrate-N concentration in the soil tended to increase with the rate of organic amendment applied, particularly with sewage sludge. In 2022, the mean nitrate-N concentration for the SS200 treatment (13.7 mg kg^{-1}) was higher than that of cow manure treatments (ranging from 9.7 to 11.0 mg kg^{-1}). In 2023, the average concentration for SS200 (17.1 mg kg^{-1}) was significantly higher than that of the other treatments (ranging from 8.1 to 11.6 mg kg^{-1}). Despite significant differences among treatments, ammonium-N levels in the soil were more variable and challenging to interpret. In 2022, the highest ammonium-N concentration was recorded in the SS200 treatment (7.6 mg kg^{-1}), whereas in 2023, the highest value was observed in the CM200 treatment (8.2 mg kg^{-1}). Total inorganic N (Inorg-N) further emphasized the differences between treatments, with significant variations observed in both years of the study. The highest mean values were recorded in the SS200 treatment (21.2 and 21.5 mg kg^{-1} for 2022 and 2023, respectively), although no significant differences were found compared to the CM200 treatment (15.4 and 19.9 mg kg^{-1} for 2022 and 2023, respectively).

In 2022, leaf N concentration did not vary significantly across treatments (Fig. 2). The mean values ranged from 25.2 g kg^{-1} (N0) to 28.7 g kg^{-1} (CM200). However, significant differences between treatments were observed in 2023. A trend of increasing N concentration in the leaves was evident from the control to treatments with higher rates of organic amendments. The mean leaf N concentrations for the SS100 (33.7 g kg^{-1}) and SS200 (35.1 g kg^{-1}) treatments

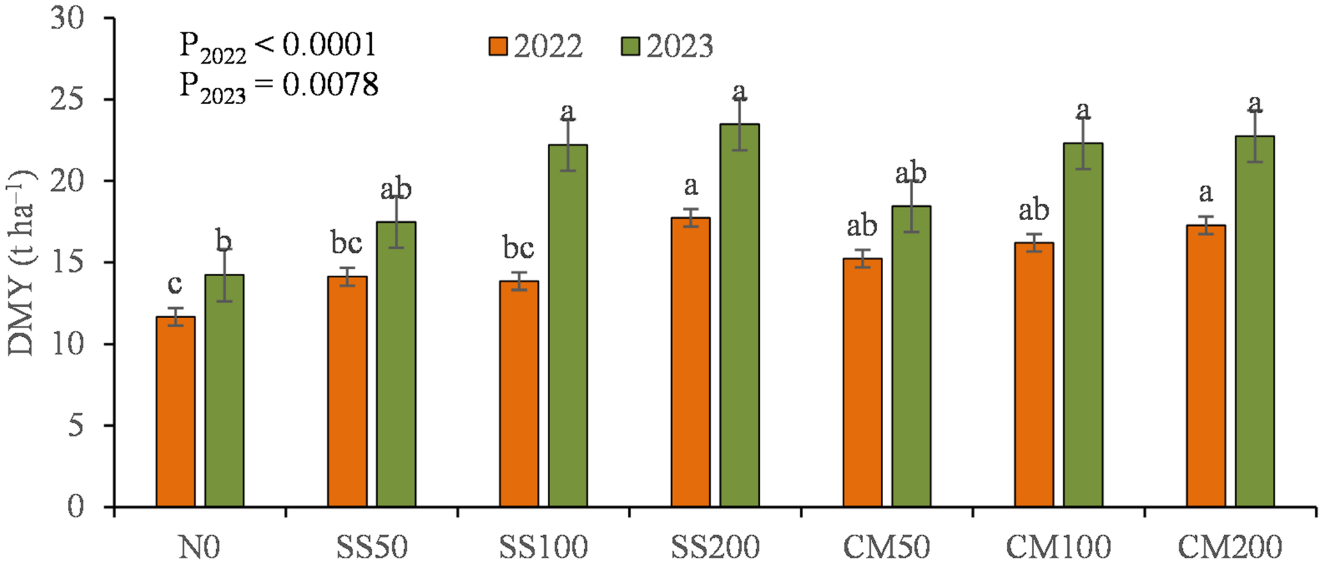

Fig. 3 Dry matter yield (DMY) as a function of sewage sludge (SS) and cow manure (CM) applied at rates of 50 (SS50, CM50), 100 (SS100, CM100), and 200 (SS200, CM200) kg N ha^{-1}, along with a non-amended control (N0). For each year, means followed by the same letter are not significantly different according to the Tukey HSD test (α = 0.05). The error bars represent the standard errors

Table 3 Concentrations of macro and micronutrients in maize tissues in 2022 as a function of sewage sludge (SS) and cow manure (CM) applied at rates of 50 (SS50, CM50), 100 (SS100, CM100), and 200 (SS200, CM200) kg N ha^{-1}, along with a non-amended control (N0)

	Macronutrients (g kg^{-1})					Micronutrients (mg kg^{-1})				
	N	P	K	Ca	Mg	B	Fe	Mn	Zn	Cu
N0	11.2 a	1.7 a	6.2 a	1.8 a	3.5 a	5.4 a	188.0 a	32.5 a	22.9 a	5.8 a
SS50	11.2 a	1.7 a	5.6 a	1.5 a	3.4 a	4.5 a	86.7 b	17.1 b	17.8 a	5.1 a
SS100	12.3 a	1.6 a	6.2 a	1.7 a	3.9 a	5.5 a	67.3 b	27.2 ab	20.4 a	5.3 a
SS200	12.5 a	1.7 a	4.7 a	1.9 a	4.5 a	5.1 a	80.8 b	26.7 ab	27.4 a	5.9 a
CM50	11.0 a	1.7 a	6.5 a	1.8 a	4.1 a	5.3 a	99.9 b	27.6 ab	21.3 a	5.8 a
CM100	11.3 a	1.5 a	6.5 a	1.4 a	3.4 a	5.1 a	76.5 b	26.9 ab	19.5 a	5.4 a
CM200	11.4 a	1.7 a	6.4 a	1.7 a	3.5 a	6.0 a	61.8 b	24.6 ab	18.5 a	5.2 a
Prob.	0.2327	0.9506	0.2044	0.7610	0.0565	0.4477	<0.0001	0.0152	0.1732	0.6193
SE	0.48	0.13	0.52	0.11	0.25	0.44	10.13	2.31	2.43	0.36

In columns, means followed by the same letters are not significantly different by the Tukey HSD test (α = 0.05)

were significantly higher than that of the control (27.5 g kg^{-1}).

3.2 Maize Dry Matter Yield and Nutrient Concentration in Plant Tissue

DMY consistently increased with higher rates of organic amendments (Fig. 3). In 2022, DMY rose significantly from 11.7 t ha^{-1} in the N0 treatment to 17.7 and 17.3 t ha^{-1} in the SS200 and CM200 treatments, respectively. In 2023, similar consistent increases in DMY were observed with organic amendments, ranging from 14.2 t ha^{-1} in the N0 treatment to 23.5 and 22.7 t ha^{-1} in the SS200 and CM200 treatments, respectively.

Applying organic amendments did not significantly increase the concentration of the macronutrients N, P, K, Ca, and Mg in maize tissues in the 2022 trial (Table 3). The average concentrations of these macronutrients in plant tissues ranged as follows: N, 11.0 to 12.5 g kg^{-1}; P, 1.5 to 1.7 g kg^{-1}; K, 4.7 to 6.5 g kg^{-1}; Ca, 1.4 to 1.9 g kg^{-1}; and Mg, 3.4 to 4.5 g kg^{-1}. Regarding micronutrients, the concentrations of B, Zn, and Cu in maize tissues did not vary significantly between treatments. For Fe, there appeared to be a consistent decrease in tissue concentration with increasing rates of organic amendments. In contrast, manganese showed no clear relationship with the type or dose of fertilizer applied.

In 2023, the concentration of macronutrients N, P, K, and Mg in maize tissues varied significantly among treatments (Table 4). N and P levels in plant tissues showed an increasing trend with higher rates of organic amendments, with significantly higher concentrations observed in the SS200 treatment compared to the control. K concentrations also followed a rising trend with increasing rates of organic amendments, although the highest values were recorded

Table 4 Concentrations of macro and micronutrients in maize tissues in 2023 as a function of sewage sludge (SS) and cow manure (CM) applied at rates of 50 (SS50, CM50), 100 (SS100, CM100), and 200 (SS200, CM200) kg N ha^{-1}, along with a non-amended control (N0)

	Macronutrients (g kg^{-1})					Micronutrients (mg kg^{-1})				
	N	P	K	Ca	Mg	B	Fe	Mn	Zn	Cu
N0	9.2 b	1.3 b	4.1 f	1.3 a	2.8 a	4.3 a	157.0 a	19.8 a	27.5 a	7.4 a
SS50	10.4 ab	1.2 b	6.1 de	1.4 a	2.7 ab	4.2 a	88.0 bcd	23.2 a	20.1 ab	7.4 a
SS100	10.0 ab	1.3 b	7.4 cd	1.4 a	2.0 abc	4.9 a	85.5 cd	25.6 a	19.7 ab	8.0 a
SS200	12.0 a	1.7 a	8.3 bc	1.5 a	2.2 abc	4.5 a	68.9 d	21.1 a	20.0 ab	8.0 a
CM50	9.2 b	1.3 b	5.5 ef	1.5 a	2.2 abc	4.7 a	112.5 b	24.0 a	18.7 b	7.9 a
CM100	9.2 b	1.4 ab	9.3 ab	1.3 a	1.8 c	4.3 a	106.2 bc	26.1 a	19.2 ab	7.9 a
CM200	10.4 ab	1.5 ab	10.5 a	1.2 a	1.9 bc	5.4 a	77.3 d	24.6 a	19.1 b	7.2 a
Prob.	0.0028	0.0049	< 0.0001	0.4483	0.0066	0.6985	< 0.0001	0.0770	0.0357	0.2033
SE	0.41	0.07	0.38	0.11	0.18	0.54	5.41	1.46	1.72	0.26

In columns, means followed by the same letters are not statistically different by the Tukey HSD test (α = 0.05)

Table 5 Nitrogen recovery, apparent N recovery (ANR), and stalk nitrate concentration as a function of sewage sludge (SS) and cow manure (CM) applied at rates of 50 (SS50, CM50), 100 (SS100, CM100), and 200 (SS200, CM200) kg N ha^{-1}, along with a non-amended control (N0)

	2022	2022	2023	2023	2023
	N recovery	ANR	N recovery	ANR	Stalk NO_3^-
	kg ha^{-1}	%	kg ha^{-1}	%	mg kg^{-1}
N0	129.9 d	---	131.5 c	---	263.0 d
SS50	158.0 cd	56.1	181.4 bc	99.8	524.5 d
SS100	169.8 bc	39.9	222.3 ab	90.8	1899.8 bc
SS200	222.2 a	46.1	280.7 a	74.6	3964.7 a
CM50	167.7 bc	75.5	169.9 bc	76.7	633.0 d
CM100	183.7 bc	53.8	206.9 abc	75.4	1093.1 cd
CM200	196.6 ab	33.3	233.1 ab	50.8	2984.5 ab
Prob.	< 0.0001		0.0003		< 0,0001
SE	7.21		15.77		246.72

In columns, means followed by the same letters are not significantly different by the Tukey HSD test (α = 0.05). Apparent N recovery (%) = (N recovered in amended plots– N recovery in unamended plots)/N applied as amendment × 100)

in the CM200 treatment. Mg concentrations in plant tissues did not consistently correlate with the type or rate of organic amendments applied. Regarding micronutrients, a consistent trend was observed for Fe, with higher concentrations detected in plant tissues from the control treatment and lower concentrations associated with treatments receiving the highest rates of organic amendments.

3.3 Nitrogen Use efficiency and Residual Inorganic Nitrogen in the Soil at the End of The Study

N recovery in aboveground biomass varied significantly among treatments in 2022 and 2023 experiments (Table 5). Due to the cumulative effect of DMY (Fig. 3) and N concentration in plant tissues (Tables 3 and 4), N recovery values consistently increased with higher rates of organic amendments (Table 5). In both years, the highest values were observed in the SS200 treatment, while the lowest values were recorded in the control treatment.

ANR tended to decrease with increasing rates of organic amendments, with this decline being more pronounced in cow manure treatments (Table 5). However, at higher amendment rates, the ANR values for sewage sludge (46.1% and 74.6%) were substantially higher than those for cow manure (33.3% and 50.8%).

Stalk nitrate concentration, a commonly used index of N nutritional status in maize at the end of the growing season, increased significantly with the applied rate of organic amendments (Table 5). The highest mean value for this index was also observed in the SS200 treatment.

Soil nitrate levels at the end of the study varied significantly among treatments, showing a consistent increase with higher rates of organic amendments applied (Fig. 4). The effect of treatments on soil ammonium content was less consistent than for nitrate levels, although significant differences among treatments were also observed. Total inorganic N in the soil, calculated as the sum of nitrate-N and ammonium-N, exhibited a clear and significant increasing trend from the control to the highest rates of organic amendments. Although not statistically significant, the mean value for the SS200 treatment (30.6 mg kg^{-1}) was higher than that of the CM200 treatment (27.1 mg kg^{-1}).

3.4 Soil Properties at the End of the Experiment

At the end of the experiment, soil pH varied significantly among treatments, with the lowest value (6.2) recorded in the SS200 treatment (Table 6). Soil organic C tended to increase with higher rates of organic amendments, with the highest mean values observed in the SS200 (24.9 g kg^{-1}) and CM200 (24.6 g kg^{-1}) treatments. Extractable P and K in the soil also showed significant increases with higher rates of organic amendments. The highest P content was recorded in the SS200 treatment (167.7 g kg^{-1}, P_2O_5), while the highest K content was observed in the CM200 treatment (178.3 g

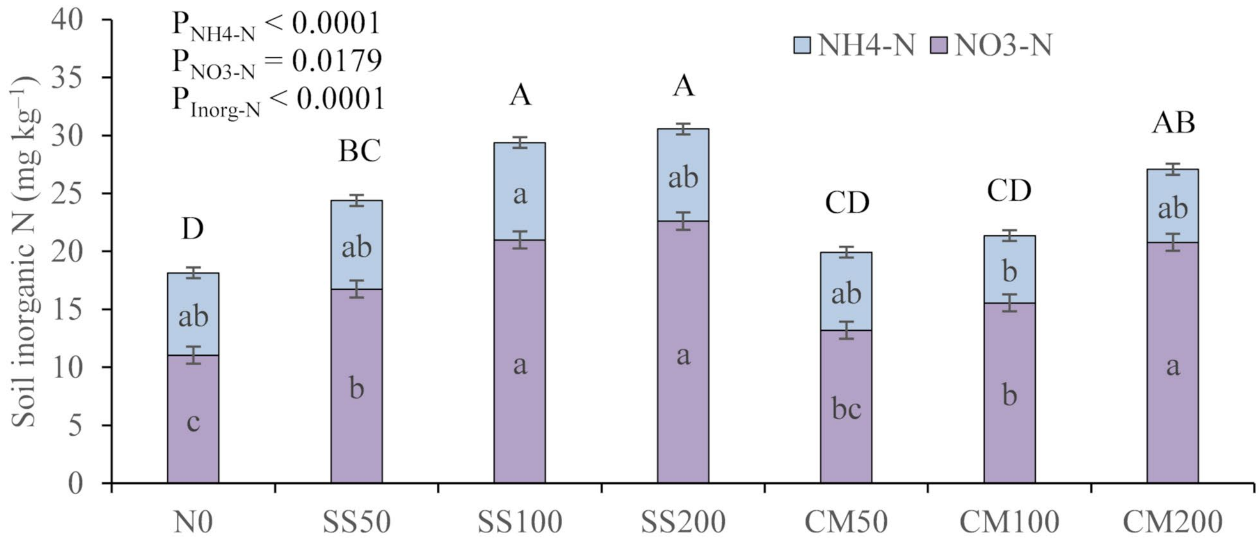

Fig. 4 Soil nitrate (NO_3^--N), ammonium (NH_4^+-N), and total inorganic nitrogen at the end of the second growing season as a function of sewage sludge (SS) and cow manure (CM) applied at rates of 50 (SS50, CM50), 100 (SS100, CM100), and 200 (SS200, CM200) kg N ha^{-1}, along with a non-amended control (N0). For each form of inorganic N (lowercase) and total inorganic N (uppercase), means followed by the same letter are not significantly different by the Tukey HSD test (α = 0.05). The error bars represent the standard errors

Table 6 pH (H_2O), organic carbon (OC), extractable phosphorus (as P_2O_5) and potassium (as K_2O), exchangeable calcium (Ca^{2+}), magnesium (Mg^{2+}), potassium (K^+), and sodium (Na^+), and cation exchange capacity (CEC) as a function of sewage sludge (SS) and cow manure (CM) applied at rates of 50 (SS50, CM50), 100 (SS100, CM100), and 200 (SS200, CM200) kg N ha^{-1}, along with a non-amended control (N0)

	pH	OC	Extractable (mg kg^{-1})		Exchangeable (cmol$_+$ kg^{-1})				
	(H2O)	g kg^{-1}	P (P_2O_5)	K (K_2O)	Ca^{++}	Mg^{++}	K^+	Na^+	CEC
N0	6.4 ab	19.4 bc	118.6 bc	80.5 d	11.9 c	6.6 b	0.2 b	0.2 a	19.0 bc
SS50	6.5 a	18.6 c	104.7 c	87.7 cd	11.6 c	6.3 b	0.2 b	0.1 a	18.3 c
SS100	6.4 bc	19.7 abc	153.4 ab	108.3 bcd	14.3 b	7.0 ab	0.3 b	0.2 a	21.8 ab
SS200	6.2 c	24.9 a	167.7 a	115.3 bc	16.2 a	7.6 ab	0.3 b	0.2 a	24.4 a
CM50	6.3 bc	18.0 c	103.3 c	85.7 cd	10.9 c	6.3 b	0.2 b	0.2 a	17.6 c
CM100	6.3 bc	19.4 abc	113.2 bc	123.0 b	11.7 c	6.7 b	0.2 b	0.1 a	18.8 bc
CM200	6.3 bc	24.6 ab	135.7 abc	178.3 a	15.0 ab	8.2 a	0.6 a	0.2 a	24.0 a
Prob.	0.0001	0.0021	0.0003	< 0.0001	< 0.0001	0.0013	< 0.0001	0.0539	< 0.0001
SE	0.03	1.15	8.22	5.88	0.29	0.27	0.03	0.02	0.54

In columns, means followed by the same letters are not significantly different by the Tukey HSD test (α = 0.05)

kg^{-1}, K_2O). Exchangeable bases increased significantly with the rate of organic amendments applied. The CEC values were 24.4 and 24.0 cmol$_+$ kg^{-1} for the SS200 and CM200 treatments, respectively, compared to 19.0 cmol$_+$ kg^{-1} in the control treatment.

4 Discussion

The maize DMY increased significantly with the applied dose of the organic amendment, from the N0 treatment to the SS200 and CM200 treatments in both growing seasons. No significant differences were observed between cow manure and sewage sludge at the same application rate. Overall, these findings are consistent with most international studies, which have reported enhanced crop productivity following the application of organic amendments (e.g., AL-Huqail et al. 2024; Arrobas et al. 2022; Kilinçoğlu et al. 2024; Niu et al. 2024; Rodrigues et al. 2024), and this underlies the enduring popularity of the organic amendments over time.

When organic amendments are applied to the soil, they serve as a food substrate for heterotrophic microorganisms, which partially convert the organic material into carbon dioxide while releasing minerals into the soil solution through a process known as mineralization (Weil and Brady 2017). One of the key benefits of using organic amendments is the release of nutrients that become available to plants. The rate of organic substrate decomposition depends on environmental conditions, such as temperature and moisture, as well as the composition of the organic material, particularly its C/N ratio. Mineralization tends to occur more rapidly when the N content is higher (Weil and Brady 2017). This may explain why many studies identify N release as one of the most significant effects of organic amendments on crop productivity (Afonso et al. 2021; Arrobas et al. 2022; Dimande et al. 2023; Rodrigues et al. 2024; Zandvakili et al. 2019). In the present study, the substantial amount of N released by organic amendments is likely the primary driver of the increased productivity observed with higher application rates.

However, the beneficial effects of organic amendments on agro-systems cannot be solely attributed to the release of nutrients during mineralization. Organic compounds also influence the soil's physical, chemical, and biological properties, improving water retention capacity, drainage, and aeration, as well as enzymatic activity. These changes impact nutrient cycling in ways that extend beyond the nutrients originally present in the amendment itself, contributing to the increased sustainability of farming systems (Afonso et al. 2021; Arrobas et al. 2022; Bhanwaria et al. 2022; Raza et al. 2024). Organic amendments are often credited with a "manuring effect," a term used to describe their unique positive impact on crop performance that cannot be fully replicated using mineral fertilizers alone (Dimande et al. 2023; Weil and Brady 2017). This concept arises, in part, from the challenges of isolating the specific contributions of organic amendments to crop productivity, as they improve a range of soil properties simultaneously.

Although DMY did not show significant differences between equivalent rates of sewage sludge and cow manure, certain indices of N availability in the soil and plants exhibited interesting trends. The mineral N content in the soil at the time of side-dressing application was consistently higher in the sewage sludge treatments compared to the cow manure treatments. This index, initially developed as the pre-side-dress soil nitrate test (Magdoff et al. 1990), is widely used because it provides reliable information on N availability during the growing season and helps determine appropriate N application rates for side-dressing (Krusekopf et al. 2002; Rodrigues et al. 2021; Rütting et al. 2018). Similarly, the N concentration in the leaves on the same date was consistently higher in the sewage sludge treatments compared to the cow manure treatments, particularly in 2023. Considering the initial composition of the two organic amendments, the sewage sludge has a higher N concentration and a lower C/N ratio (Table 1). Therefore, the lower C/N ratio of sewage sludge likely contributed to a higher initial net mineralization rate, a result commonly reported in the literature (Afonso et al. 2021; Dimande et al. 2023; Weil and Brady 2017). This was sufficient to reveal differences between the two organic manures in those indices, which proved to be more sensitive to variations in soil nitrogen availability than dry matter yield.

A similar trend of higher N concentration in the aboveground biomass of maize was observed in the sewage sludge treatments compared to the cow manure treatments at harvest, although no significant differences were found. The contrast between sewage sludge and cow manure became more pronounced when comparing the N recovered in the treatments with the two organic amendments, as this variable results from the combined effects of DMY and N concentration in the tissues. Additionally, the nitrate concentration in the stalks at harvest, a highly sensitive index of N availability in the soil (Blackmer and Mallarino 1996; Isla et al. 2015; Rodrigues et al. 2021), exhibited a similar pattern, with consistently higher average values in the sewage sludge treatments compared to cow manure. Nitrate is the primary non-metabolized form of N in plants, as ammonium is toxic and predominantly assimilated in the roots (Hawkesford et al. 2023). The amount of nitrate that accumulates in the vacuoles of vascular bundles is typically the best indicator of the N nutritional status of plants (Bryson et al. 2014). Therefore, in both the short and medium term, including the entire growing season, sewage sludge released more N to the plants than cow manure due to its higher N concentration and lower C/N ratio. Although this relationship between the C/N ratio and N release is not always consistent (as substrates rich in lignin, cellulose, and hemicellulose, which have a high C/N ratio but low metabolic energy for microorganisms, may not immobilize N) (Weil and Brady 2017), in the case of sewage sludge, the C/N ratio appeared to be a reliable predictor of N release. Thus, similarly to the indices determined at the time of side-dressing N application, the stalk nitrate test proved to be more sensitive than dry matter yield to variations in soil nitrogen availability, making it highly useful for interpreting the results.

The ANR, a widely used index of N use efficiency in studies of N fertilization and organic amendment application (Bouchet et al. 2016; Rodrigues et al. 2021; Weil and Brady 2017), also indicated high N mineralization from the organic amendments, particularly from sewage sludge. Increasing the rate of N fertilizers typically reduces N use efficiency due to nutrient losses to the environment (Bouchet et al. 2016; Rodrigues et al. 2021; Weil and Brady 2017). However, with organic amendments, lower N use efficiency, compared to mineral fertilizers, may not necessarily reflect N loss from the soil but rather biological immobilization. In other words, the organic amendments did not mineralize all the N during the growing season, leaving a portion immobilized in the soil in organic form (Mallory et al. 2010; Rodrigues et al. 2018). The degradation of organic amendments in the soil is governed by "decay series," which outline the fraction of nutrients, particularly N, released each year following application (Daudén et al. 2004; Dimande et al. 2023; Mallory et al. 2010; Rodrigues et al. 2018). In this study, the amount of N mineralized from the sewage sludge was exceptionally high. The ANR in the second year reached 99.8% in the SS50 treatment and 74.6% in the SS200 treatment, values significantly higher than those typically observed with other organic amendments (Rodrigues et al. 2018). These values reflect the cumulative effect of the decay series over two years, including the mineralization of the residual amendment applied in the first year and the mineralization of the amendment applied in the second year.

The concentration of other macronutrients in the tissues provided fewer clear indications in the treatment comparison than N concentration. However, in the second year, significant differences emerged between treatments for P and K, with a trend toward higher values in treatments with higher rates of organic amendments. A consistent pattern was observed, and sewage sludge resulted in higher P concentrations in the tissues, while cow manure led to higher K concentrations. This consistency has also been noted in previous studies (Arrobas et al. 2024; Cardarelli et al. 2023; Kandil et al. 2020; Rodrigues et al. 2024; Zandvakili et al. 2019). The results appear to reflect the initial composition of the organic amendments, with sewage sludge being particularly rich in P and cow manure significantly richer in K (Table 1).

P is a key component of organic molecules such as nucleic acids, phospholipids in biomembranes, and energy-rich phosphates (Hawkesford et al. 2023). For P to be available to plants, the action of phosphatases is required to degrade the organic substrate (Cesco et al. 2010; Chen et al. 2023; Lambers 2022). Since sewage sludge is a rapidly mineralizing organic amendment with a high P content, this likely contributed to the high availability of P for the plants. In contrast, K is not incorporated into organic structures but exists as a free ion in plant tissues, where it plays a crucial role in maintaining electrical and osmotic homeostasis, activating enzymes and nucleic acids, and supporting cell turgor and related processes (Hawkesford et al. 2023). As a free ion, K is present in low concentrations in sewage sludge, as soluble salts are eliminated with the liquid effluent (Kirchmann et al. 2016; Ndoung et al. 2023). Therefore, its availability to plants is not dependent on the mineralization rate of the organic substrate but rather on its concentration, which is higher in cow manure in this study.

Some attention could also be given to the concentration of Fe in the tissues. The values indicate a clear decrease with increasing rates of organic amendment, a trend observed in both years and for both types of organic amendments applied. The solubility of Fe in the soil is influenced by pH but is primarily governed by the soil's redox potential. Under reducing conditions, the availability of metallic cations, such as Fe, increases due to the dissolution of Fe oxides (Rengel 2023; Weil and Brady 2017). Higher doses of organic amendments led to greater DMY in maize, which is typically associated with increased photosynthetic activity and higher water transpiration during the growing season (Hernández et al. 2021; Wu et al. 2011). Since the plants were irrigated by sprinkler systems, receiving the same amount of water across all treatments, the higher transpiration rates in plants receiving higher doses of organic amendments likely resulted in consistently lower soil moisture levels. This, in turn, would have led to lower Fe concentrations in the soil solution, which contributed to reduced Fe concentrations in the plant tissues.

One of the commonly reported concerns associated with the use of sewage sludge is its potential concentration of heavy metals and the potential for accumulation in soils and contamination of trophic chains (Dhanker et al. 2021; Swain et al. 2021). However, the sewage sludge used in this study originates from a WWTP that does not receive industrial wastewater. Previous studies have shown that it contains low levels of heavy metals and that the risk of contamination is no greater than that associated with the use of farmyard manures (Arrobas et al. 2024; Rodrigues et al. 2024).

The application of organic amendments produced measurable beneficial effects on soil properties. A significant increase in soil organic C content was observed with higher doses of organic amendments, along with increases in the primary cation exchange bases and CEC. Extractable P and K also showed increases, with P exhibiting slightly higher values in treatments with sewage sludge and K in those with cow manure. These findings align with the widely reported benefits of organic amendments (Afonso et al. 2021; Arrobas et al. 2022; Bhanwaria et al. 2022; Rodrigues et al. 2024) and provide insight into why organic fertilizers are so widely recommended.

The application of organic amendments, particularly the SS200 treatment, resulted in a slightly lower soil pH compared to the N0 treatment. The initial pH of the organic amendments was higher than that of the soil, a condition that would typically lead to an increase in soil pH. This result warrants an alternative explanation. Based on the N availability results, it is evident that the sewage sludge underwent significant mineralization during the growing season. The mineralization of the organic substrate, coupled with the subsequent nitrification of ammonium ions, produces an excess of H^+ ions, thereby contributing to soil acidification (Neumann and Ludewig 2023; Sparks et al. 2024). In this instance, this acidifying effect likely outweighed the influence of the higher initial pH of the organic amendment relative to the soil. Nonetheless, this effect is likely temporary, diminishing as the initial substrate continues to decompose through microbial activity. Over time, the enhanced CEC associated with organic matter incorporation may exert a more pronounced influence, potentially mitigating the acidification.

5 Conclusions

The rapid mineralization of sewage sludge, due to its low carbon/nitrogen ratio, leading to the release of essential nutrients such as nitrogen and phosphorus, is likely the most significant finding of this study, offering valuable insights into its agricultural use. Its low potassium content is the main limitation from an agronomic fertilizer perspective. Additionally, there are legal considerations regarding the use of sewage sludge, which were not addressed in this study, notably the requirement to ensure heavy metal concentrations are within legal limits and that it undergoes microbiological disinfection treatment. In this case, the use of sewage sludge does not pose concerns regarding heavy metals, as this wastewater treatment plant does not receive industrial effluents. Nevertheless, companies managing wastewater treatment plants are required to have a sewage sludge management plan in place prior to distributing the organic amendment to farmers. This plan must ensure the chemical and microbiological safety of the material. Future scientific studies should also consider these aspects whenever the origin of the sewage sludge warrants such precautions.

The results for cow manure were not negative. It is simply clear that it is a less reactive organic amendment. A slower effect on vegetation is expected, which could be a determining factor in establishing the optimal conditions for its use.

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Author Contributions Paulo Dimande: investigation; data curation; writing– original draft. Margarida Arrobas: methodology; resources; supervision; writing– review and editing. Manuel Ângelo Rodrigues: conceptualization; funding acquisition; project administration; data curation; writing– review and editing.

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Declarations

Conflict of interest The authors declare no conflict of interest.

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