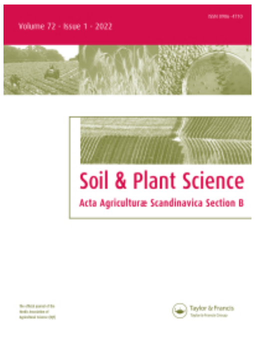

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Low-temperature hay biochar and acid treatment increases fertiliser value of slurry digestate

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ABSTRACT

This study aimed to evaluate the effect of low-temperature-produced (torrefied at 300°C) hay biochar (BC) on the fertilisation effectiveness of digestate (SD) and the loss of nitrogen (N) and carbon (C). We also examined whether acidification could increase the effect of BC on SD effectiveness and compared the effects of untreated and acidified BCs with that of sulphuric acid (H_2SO_4). The treatments included unamended SD (SD_{UT}) and SD amended with untreated BC (BC_{UT}), H_3PO_4-acidified BC, (BC_{H3PO4}) H_2SO_4-acidified BC (BC_{H2SO4}), or pure H_2SO_4. These treatments were compared in a pot experiment with ryegrass (*Lolium perenne*, L.) and evaluated based on N, phosphorus (P), potassium (K), calcium (Ca), and magnesium (Mg) taken up by plants and remaining in the soil. The N input to the soil was doubled for all applied amendments compared with SD_{UT}. However, the fertilisation effectiveness of SD amended with BC_{UT} alone was comparable to that of SD treated with pure H_2SO_4, offering the additional benefits of higher C input and reduced C loss from the soil. We concluded that the effect of BC on the effectiveness of SD as a fertiliser is similar to, or even better than, that of H_2SO_4.

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KEYWORDS
Acidified biochar; biochar; liquid manure digestate; organic fertiliser; N and C loss; nutrient uptake

Introduction

Slurry digestate (SD) is a by-product of biogas production that is frequently used as organic fertiliser for arable crops worldwide. However, gaseous losses during SD storage and application to soil can negatively impact the environment and reduce its value as an effective fertiliser through the emission of ammonia (NH_3), nitrous oxide (N_2O), carbon dioxide (CO_2), and methane (CH_4) (Sørensen and Amato 2002; Ndegwa et al. 2008; Zhang et al. 2021). Depending on the conditions and duration of storage and handling of SD, more than 50% of added N can be lost as NH_3 emissions, while up to 2% of the added carbon (C) may be lost as CH_4 (Sommer et al. 2003; Fangueiro et al. 2015). In recent years, considerable attention has been given to slurry acidification, which effectively reduces NH_3 emissions from storage and also after slurry incorporation into soil (Fangueiro et al. 2015). However, acidified slurry lowers the soil pH and increases the sulphur (S) concentration, making this amendment unsuitable for acidic soils. Loide et al. (2020) showed that the use of acidified SD in acidic soils resulted in increased S, calcium (Ca), and potassium (K) leaching, and decreased N and phosphorus (P) leaching. Therefore, acidification has been shown to have several disadvantages alongside its desirable aspects.

Biochar (BC) is a solid C-rich material obtained from the thermochemical conversion of biomass in an oxygen-limited environment during the pyrolysis and gasification process (Lehmann and Joseph 2015). As an SD amendment, BC has several advantages over acidification. BC can adsorb NH_3 and ammonium nitrogen (NH_4^+-N) from aqueous solutions and the liquid–gas interface of the slurry stores (Taghizadeh-Toosi et al. 2012; Park et al. 2018), which can then become available to plants in soil (Keskinen et al. 2021). Because of its alkaline nature, BC-amended SD may be more favourable than acidified SD for application to acidic soils, where it could increase nutrient availability for plants, especially P availability. Moreover, BC also contains various macro- and micronutrients such as potassium (K), sodium (Na), Mg, Ca, copper (Cu), zinc (Zn), and

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iron (Fe) (Ding et al. 2016; Tomczyk et al. 2020), which also increase the value of SD as a fertiliser. Furthermore, BC's recalcitrant nature allows to persist in the soil without significant C loss (Weng et al. 2022). This longevity ensures that the sequestered C remains stored in the soil, providing a reliable and long-term solution for C sequestration (Li and Tasnady 2023).

Abbas et al. (2020) reported that BC applied with biogas SD reduced N losses and improved plant growth by increasing N use efficiency, microbial growth, and water-holding capacity, particularly for less fertile soils. The success of combined application of BC with organic fertiliser amendments was not only in retaining higher amounts of nutrients in the soil but also in making these nutrients available to plants. However, several studies have shown that BC had a weak or non-significant effect on crop yield and N uptake (Lentz et al. 2014; Foereid et al. 2020). It is frequently reported that the application of BC to soil to ameliorate soil properties and enhance crop productivity is more successful in well-weathered (degraded), low-fertility, and acidic soils, than in fertile healthy soils (Hazman et al. 2022; Khan et al. 2022).

Studies focusing on how BC-amended SD affect N and C losses from the soil and the crop yield have shown wide variation and high uncertainty (Liu et al. 2016; Xu et al. 2021). It has been reported that BC reduces (Angst et al. 2014; Antonangelo et al. 2021), have no significant effect on (Karhu et al. 2011), or even increase soil N_2O emissions (Escuer-Gatius et al. 2020). BC also decreases CH_4 emissions from soil, potentially because of inhibition of methanogenic activity or lack of CO_2 availability (Liu et al. 2011). BC also can increase the potential of the soil as a CH_4 sink as a result of increased methanotrophy (Karhu et al. 2011; Azzi et al. 2024).

The acidification of BC can have positive effects on BC physicochemical properties by decreasing its alkalinity and increasing the amount of surface functional groups (carboxyl, carbonyl, and phenol) (Tsang et al. 2007; Antor et al. 2023). Because of these changes, acidified BC may be more effective than untreated BC in preventing N losses in manure storage by improving BC sorption capacity (Rasse et al. 2022). However, acidified BC can have negative effects on the soil similarly to pure acid.

Our previous research examined the extent to which untreated and acid-treated low-temperature hay BC could reduce NH_3 emissions and increase N retention in SD (Covali et al. 2021). The results indicated that the effect of untreated BC applied mixed into the SD and acidified BC applied on the digestate surface was not different from that of pure H_2SO_4 and H_3PO_4 acids (Covali et al. 2021).

The current study therefore aimed to examine the effect of SD amended with pure H_2SO_4, untreated BC and acidified BC amendments on biomass yield and uptake of N, P, K, Ca, and Mg by perennial ryegrass. A secondary objective of this study was to investigate the effect of the same treatments on soil N, P, K, Ca, Mg, and C concentrations, and the N and C balance in the soil.

The following hypotheses were proposed: (i) BC amendment increases the effect of SD on plant growth and nutrient uptake and reduces N and C losses in the soil; (ii) acidified BC amendment has a greater effect on SD than untreated BC in terms of plant growth, nutrient uptake, and N and C losses in the soil; and (iii) BC amendments exert a greater influence than H_2SO_4 on plant growth and nutrient uptake, while also mitigating N and C losses from the soil.

To the best of our knowledge, this is the first study to directly compare the effects of pure acid and BC on the fertiliser value of cattle SD material. Moreover, the impact of acidification on the efficacy of BC as an SD amendment has not been previously investigated. The findings of this study may help farm managers, industry practitioners, and policymakers to improve crop productivity and mitigate/adapt to climate change.

Materials and methods

Study design

A pot experiment with perennial ryegrass (*Lolium perenne* L.) was conducted in a growth chamber at the Estonian University of Life Sciences (58°22′48.5"N, 26°43′20.9"E) from March 2019 to late May 2019 in Tartu, Estonia. The study had six treatments in total: (i) control (without any amendment), (ii) SD with no amendment (SD_{UT}), (iii) SD with sulphuric acid amendment ($SD + H_2SO_4$), (iv) SD with untreated biochar amendment ($SD + BC_{UT}$), (v) SD with biochar treated with phosphoric acid amendment ($SD + BC_{H3PO4}$), and (vi) SD with biochar treated with sulphuric acid amendment ($SD + BC_{H2SO4}$). Each treatment had eight replicates, resulting in a total of 48 pots.

The first growth period lasted 40 days, after which shoot biomass was determined for all eight replicates, and root biomass for four of the replicates. The second growth period lasted an additional 35 days (a total of 75 days) after which shoot and root biomass were determined for the remaining four replicates that did not receive the root biomass analysis in the first growth period. This experimental design aimed to investigate the durability of the effects of the treatments on the growth of different plant parts (shoots and roots biomass). Soil samples were taken at the end of both growth periods from the replicates from which roots were collected.

PVC tubes with a surface area of 87 cm^2 and a length of 30 cm were used as growing pots. A 3-cm-thick layer consisting of expanded polystyrene foam cap and a second layer of geotextile was fixed at the bottom of each pot. The cap had a hole in the middle allowing excess water drainage. Each pot was filled with 3640 g of air-dried *Mollic Cambisol* (IUSS Working Group WRB 2015) and sand in a 1:1 ratio. The soil pH_{KCl} (pH in 1 M KCl solution) was 6.87, and plant-available P_{AL} (where AL indicates that the ammonium lactate method was used for analysis (Section 2.2) (Egnér et al. 1960)), K_{AL}, Ca_{AL}, and Mg_{AL} were 189, 111, 1863, and 257 mg kg^{-1}, respectively. The soil total nitrogen (N_{tot}) and total carbon (C_{tot}) were 0.11% and 1.35%, respectively.

The BC was produced by torrefaction at 300°C from reed canary grass (*Phalaris arundinacea* L.) hay pellets. This specific low-temperature hay BC was chosen because it was an effective slurry amendment in reducing N and C gas emissions from slurry digestate storage in our previous study (Covali et al. 2021). The BC was ground manually to a particle size of less than 1.25 mm. The BC acidification procedure is described in Covali et al. (2021).

The SD originated from the Ilmatsalu biogas plant (Tartu County, Estonia), with a feedstock consisting of 85% cattle slurry; 10% mixed pig slurry, manure (straw bedding), and green biomass; and less than 5% food industry by-products. Additional information about the physicochemical properties of SD, untreated, and acid-treated BC can be found in Covali et al. (2021).

Before sampling, the SD was homogenised with a stirring rod. One hundred grams were poured into 125-mL Berzelius beakers for control and BC treatments and into 200-mL beakers for the acid treatment. The application rate of BCs and H_2SO_4 (96%) to SD was 5.0 and 0.5 g, respectively. After amendment application, the SD was stored at room temperature (20–22°C) for seven consecutive days. After this period, the dry matter content of SD_{UT} was 7.11%, the N and C concentrations in dry matter were 1.76% and 28.44% in SD_{UT}, 3.56% and 31.36% in $SD + H_2SO_4$, 2.28% and 40.35% in $SD + BC_{UT}$, 2.24% and 42.03% in $SD + BC_{H2SO4}$, and 2.32% and 44.22% in $SD + BC_{H3PO4}$.

The SD + BC amendments were applied mixed into the soil at 0–7 cm depth using the amount of 28.5 g of wet weight per pot. The application rate was based on the SD rate (30,000 kg ha^{-1}) typically applied by farmers in Estonia. Immediately after fertiliser application, the perennial ryegrass ('Raite' cultivar) was sown at a seed rate of 0.2 g per pot. Fifty grams of fine sand and 25 mL of tap water were added on top of the seeds in each pot.

Before fertiliser application and seeding, the soil was moistened three times during a week until field capacity was reached. Starting on the fifth day after sowing, each pot was watered with 100 mL for the first two weeks and with 125 mL for the next two weeks, at a frequency of twice per week. From the fourth week, the water application rate was 150 mL, applied 4–6 times per week.

Plant and soil analyses

The shoot biomass was harvested at 2 cm above the soil surface and dried for 24 h at 40°C in a CFD 1400 SS dryer-dehydrator machine (Alpfrigo, Logatec, Slovenia). For root biomass determination, after shoot removal, the soil was poured out of the growing pots, and the roots were separated by shaking. The roots were air-dried at room temperature (22°C) for two weeks, and final soil and root separation was performed by sieving with a 2-mm sieve. The dry shoot and root biomass was ground with a knife mill Grindomix GM 200 (Retsch, Haan, Germany) to a particle size of <1 mm and weighed using a precision balance PS 450/X (RADWAG, Radom, Poland).

Soil was sampled with an auger (diameter 1 cm) from the upper (0–10 cm) and lower (10–30 cm) layers of a pot. Samples were dried at room temperature for one week and sieved through a 2-mm sieve. The soil pH was measured with a pH meter SevenCompact S210 (Mettler-Toledo, Schwerzenbach, Switzerland) in a soil suspension with distilled water and 1 M potassium chloride (KCl) solution (soil weight:solution volume ratio of 1:5).

Soil plant-available P, K, Ca, and Mg concentrations were analysed by the ammonium lactate method (Egnér et al. 1960). In the biomass, the P, K, Ca, and Mg concentrations were examined using the acid digestion method (Rosopulo et al. 1976). The concentrations of these elements in the extraction solvent were determined with a microwave plasma atomic emission spectrometer (MP-AES 4200, Agilent, Santa Clara, CA, USA). Total N and C concentrations in plant biomass, soil, and SD were determined by the dry combustion method with a varioMAX CNS elemental analyser (Elementar Analysensysteme GmbH, Langenselbold, Germany).

Calculations

The dry matter yield (DMY) (g pot^{-1}) was determined by weighing the combined mass of both the harvested shoot and root biomasses after they were air dried to remove moisture.

The N, P, K, Mg, and Ca uptake was determined as follows:

$$Uptake_E\ (g\ pot^{-1}) = (DMY \times C_E)/100$$

where $Uptake_E$ is nutrient uptake (g pot^{-1}), DMY is dry matter yield (g), and C_E is the element concentration in the biomass (%). The nutrient uptake was calculated separately and cumulatively for shoot and root biomasses. The N and C contents in soil at the beginning (N_{start} and C_{start}) and at the end of experiment (N_{end} and C_{end}) were determined as follows:

$$A_E\ (g\ pot^{-1}) = (S \times C_E)/100$$

where A_E is the element content in the soil (g); S is the soil amount in a pot (g); and C_E is the element concentration in soil (%), where the index E shows the element variant.

The N_{end} and the C_{end} was calculated based on the initial soil weight because the soil contained plant roots at the end of the experiment. The $N_{balance}$ of the soil was calculated as follows:

$$N_{balance}\ (g\ pot^{-1}) = N_{end} - (N_{start} + N_{applied} - N_{uptake})$$

where N_{end} is the N amount in soil at the end of experiment (g), N_{start} is the N amount in soil at the beginning of the experiment (g), $N_{applied}$ is the amount of N applied to the soil as SD (g), and N_{uptake} is the cumulative N amount taken up by the plants (g). All N amounts are expressed as g pot^{-1}. A negative N balance indicates net N loss (g pot^{-1}) during the experiment.

The $C_{balance}$ in soil was calculated as follows:

$$C_{balance}\ (g\ pot^{-1}) = C_{end} - (C_{start} + C_{applied})$$

where C_{end} is the C content of the soil at the end of the experiment (g), C_{start} is the C content of the soil at the beginning of the experiment (g), and $C_{applied}$ is the C amount applied to the soil (g) with SD. All C amounts are expressed as g pot^{-1}. A negative C balance indicates C loss (g pot^{-1}) during the experiment.

Statistical analyses

Two-way analysis of variance (ANOVA) was used to assess the effect of treatments on shoot and root biomass, nutrient uptake, plant-available nutrient concentrations in soil, and N and C losses. Before analysis, normality of all data was checked using the Shapiro–Wilk test. The significance of the results was estimated by the post-hoc Tukey test at the probability level $p < 0.05$. The relationships between nutrient uptake with soil agrochemical parameters was tested with Pearson correlation analysis. A paired *t*-test ($\alpha < 0.05$) was used to test C_{tot} differences over time for the same treatment. Statistical analysis was conducted using R programming software (R Development Core Team 2019).

Results

Shoot dry matter yield

Shoot biomass was influenced by treatment only during the first growth period. Relative to the control, a higher DMY was obtained using SD treated with H_2SO_4 or BC_{UT} (Figure 1). Shoot biomass was higher in

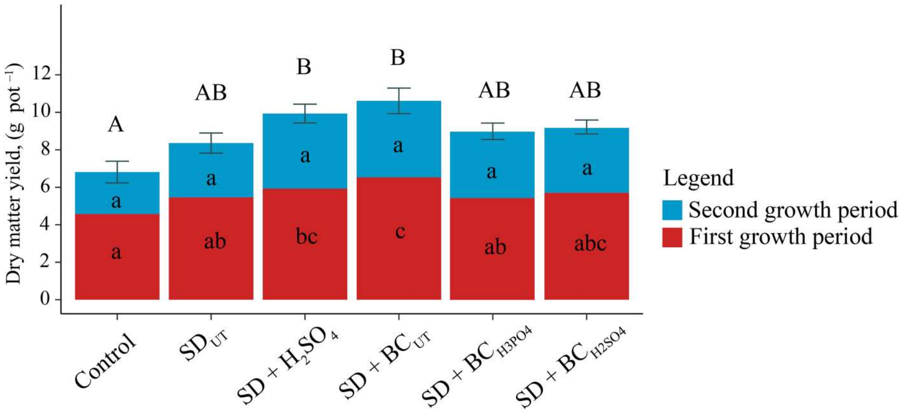

Figure 1. Shoot dry matter yield (DMY) of perennial ryegrass for the two growth periods (mean ± standard error, $n = 8$ for the first growth period and $n = 4$ for the second growth period). Significant differences between the DMY of the first and second growth periods are marked with different lowercase letters. Significant differences in total DMY (sum of DMY of the first and second growth periods) are marked with different capital letters. The differences were significant according to the Tukey's HSD post-hoc test. Control, treatment without any fertiliser; SD_{UT}, untreated slurry digestate; $SD + H_2SO_4$, slurry digestate amended with sulphuric acid; $SD + BC_{UT}$, slurry digestate amended with untreated biochar; $SD + BC_{H3PO4}$, slurry digestate amended with biochar treated with phosphoric acid; $SD + BC_{H2SO4}$, slurry digestate amended with biochar treated with sulphuric acid.

$SD + BC_{UT}$ than SD_{UT}; however, no significant differences were observed between the other BC treatments and control and SD_{UT}.

Over the two growth periods combined, none of the experimental treatments significantly increased the effect of SD on shoot biomass relative to SD_{UT} (Figure 1). However, it was observed that shoot biomass was slightly higher in the $SD + H_2SO_4$ and $SD + BC_{UT}$ treatments compared with SD_{UT}.

Root biomass was not influenced by any treatment during either of the growth periods ($p > 0.05$) (Figure S1).

Nutrient concentrations in the shoot and root biomass

Of all the tested nutrients in the shoot biomass (Table 1), the treatments significantly influenced only the P concentration. After the first growth period, the P concentration was significantly higher in SD_{UT} and $SD + BC_{H3PO4}$ treatments than in the $SD + H_2SO_4$ or $SD + BC_{UT}$ treatments. After the second growth period, the P concentration was highest in the shoots of the control treatment. There was no significant difference in shoot P concentration among the SD treatments.

In the root biomass, the treatments significantly affected the concentrations of N, P, and Ca (Table 2). After the first growth period (roots had been growing for 40 days), the N concentration was significantly higher in all SD treatments compared with the control. After the second growth period (roots had been growing for 75

Table 1. Nutrient concentrations in the shoot biomass after the first and second growth periods.

Treatment	N (%)	P (%)	K (%)	Mg (%)	Ca (%)
			First growth period		
Control	2.18 ± 0.19 a	0.40 ± 0.02 ab	3.33 ± 0.06 a	0.36 ± 0.01 a	0.66 ± 0.01 a
SD_{UT}	3.09 ± 0.21 a	0.44 ± 0.01 a	3.84 ± 0.10 a	0.38 ± 0.02 a	0.58 ± 0.02 a
$SD + H_2SO_4$	2.91 ± 0.26 a	0.33 ± 0.03 b	3.78 ± 0.16 a	0.38 ± 0.02 a	0.59 ± 0.03 a
$SD + BC_{UT}$	2.93 ± 0.20 a	0.31 ± 0.01 b	3.78 ± 0.12 a	0.41 ± 0.02 a	0.78 ± 0.15 a
$SD + BC_{H3PO4}$	3.00 ± 0.04 a	0.45 ± 0.02 a	4.14 ± 0.09 a	0.42 ± 0.02 a	0.72 ± 0.04 a
$SD + BC_{H2SO4}$	3.14 ± 0.27 a	0.39 ± 0.03 ab	3.96 ± 0.25 a	0.40 ± 0.03 a	0.87 ± 0.02 a
			Second growth period		
Control	1.03 ± 0.09 a	0.32 ± 0.01 a	1.94 ± 0.22 a	0.42 ± 0.01 a	0.74 ± 0.02 a
SD_{UT}	0.89 ± 0.05 a	0.24 ± 0.01 b	1.88 ± 0.12 a	0.42 ± 0.02 a	0.64 ± 0.01 a
$SD + H_2SO_4$	0.86 ± 0.05 a	0.23 ± 0.02 b	1.75 ± 0.15 a	0.37 ± 0.02 a	0.60 ± 0.06 a
$SD + BC_{UT}$	0.89 ± 0.08 a	0.22 ± 0.02 b	1.76 ± 0.13 a	0.39 ± 0.01 a	0.77 ± 0.09 a
$SD + BC_{H3PO4}$	0.88 ± 0.04 a	0.27 ± 0.01 ab	2.00 ± 0.11 a	0.43 ± 0.01 a	0.76 ± 0.15 a
$SD + BC_{H2SO4}$	0.82 ± 0.06 a	0.24 ± 0.02 ab	2.00 ± 0.18 a	0.41 ± 0.02 a	0.73 ± 0.06 a

Note: The values are presented as mean ± standard error (n = 4). Results marked with different lowercase letters indicate a significant difference among treatments according to the Tukey's HSD post-hoc test ($p < 0.05$).

Table 2. Nutrient concentrations in the root biomass after the first and second growth period.

Treatment	N (%)	P (%)	K (%)	Mg (%)	Ca (%)
First growth period					
Control	0.54 ± 0.16 b	0.21 ± 0.01 a	1.40 ± 0.05 a	0.33 ± 0.02 a	0.59 ± 0.04 a
SD_{UT}	1.21 ± 0.08 a	0.29 ± 0.01 a	1.82 ± 0.07 a	0.36 ± 0.04 a	0.57 ± 0.08 a
SD + H_2SO_4	1.18 ± 0.09 a	0.24 ± 0.02 a	1.82 ± 0.07 a	0.35 ± 0.02 a	0.47 ± 0.03 a
SD + BC_{UT}	1.27 ± 0.07 a	0.21 ± 0.01 a	1.81 ± 0.22 a	0.31 ± 0.04 a	0.51 ± 0.09 a
SD + BC_{H3PO4}	1.13 ± 0.08 a	0.28 ± 0.02 a	1.65 ± 0.11 a	0.37 ± 0.08 a	0.50 ± 0.08 a
SD + BC_{H2SO4}	1.38 ± 0.09 a	0.28 ± 0.03 a	1.75 ± 0.15 a	0.44 ± 0.03 a	0.46 ± 0.03 a
Second growth period					
Control	0.56 ± 0.05 a	0.18 ± 0.01 b	0.95 ± 0.02 a	0.34 ± 0.07 a	0.67 ± 0.09 ab
SD_{UT}	0.53 ± 0.04 a	0.20 ± 0.01 b	1.02 ± 0.02 a	0.39 ± 0.03 a	0.47 ± 0.02 b
SD + H_2SO_4	0.56 ± 0.04 a	0.16 ± 0.01 b	0.94 ± 0.03 a	0.34 ± 0.02 a	0.40 ± 0.02 b
SD + BC_{UT}	0.50 ± 0.03 a	0.19 ± 0.02 b	1.01 ± 0.05 a	0.37 ± 0.06 a	0.50 ± 0.04 b
SD + BC_{H3PO4}	0.47 ± 0.02 a	0.33 ± 0.05 a	1.09 ± 0.04 a	0.47 ± 0.03 a	1.07 ± 0.27 a
SD + BC_{H2SO4}	0.54 ± 0.02 a	0.21 ± 0.01 b	1.04 ± 0.05 a	0.49 ± 0.05 a	0.45 ± 0.04 b

Note: The values are presented as mean ± standard error ($n = 4$). Results marked with different lowercase letters indicate a significant difference among treatments according to the Tukey's HSD post-hoc test ($p < 0.05$).

days), the P and Ca concentrations were higher in the roots of the SD + BC_{H3PO4} treatment compared with all other treatments, except Ca in the control.

Nutrient uptake

In the first growth period, significant differences in nutrient uptake among treatments were primarily observed for N and K (Figure 2). The N uptake exhibited a significant increase in all SD treatments compared with the control, except in the SD + BC_{H3PO4} treatment. However, the SD amendment did not have a statistically significant impact on N uptake. The K uptake was greater than that of the control in the SD + H_2SO_4, SD + BC_{UT}, and SD + BC_{H3PO4} treatments. Differences in K uptake among the SD treatments were not significant.

In the second growth period (Figure 3), compared with the control, N uptake was significantly greater in the SD + H_2SO_4 or SD + BC_{UT} treatments and K uptake was higher across all treatments in which the SD amendment was used. Only Ca uptake differed significantly among the amended SD treatments, with the highest values found in SD + BC_{UT} or SD + BC_{H3PO4} treatments.

There were significant correlations between assimilation of N, P, K, Ca, and Mg by plants (Table 3). This correlation was stronger during the second growth period, in which significant relationships were observed among all the measured nutrients. During the first growth period, significant correlations were observed among N, K, and Ca uptakes and among K, Ca and Mg uptakes.

Soil chemical characteristics

The experimental treatments had a significant impact on soil pH and the concentrations of N, P, K, and Ca during both growth periods (Tables 4 and 5). The effects were most pronounced in the 0–10 cm soil layer. In the SD + BC_{UT} treatment, soil pH was significantly higher compared with the control. At the end of the first growth period, soil pH in the SD + BC_{UT} treatment was also significantly higher compared with SD + H_2SO_4. In the SD treatments with acidified BC, soil pH did not differ significantly from that in the SD + H_2SO_4 treatment but remained more stable throughout the experiment. The SD + BC_{UT} treatment also had a higher soil N_{tot} concentration than that of the other treatments. After the first growth period, this difference was statistically significant compared with the control, SD_{UT}, and SD + H_2SO_4 treatments, and after the second growth period compared with all other treatments except SD + H_2SO_4. Significant differences in the soil P concentration among the treatments were observed only at the end of the first growth period, with the highest levels observed in the SD + H_3PO_4 treatment. However, by the end of the second growth period, soil P concentrations had equalised across treatments and no significant differences were observed. The soil Ca and K concentrations varied only at the end of the second growth period, with the highest levels observed in the SD + BC_{H2SO4} and SD_{UT} treatments, respectively. The highest soil C_{tot} concentrations were observed in the treatments in which BC was added to SD, with BC_{UT} having the greatest effect. The effect of acidified BC on soil C content was slightly weaker and shorter-lasting than that of BC_{UT}, particularly in the case of BC acidified with H_3PO_4.

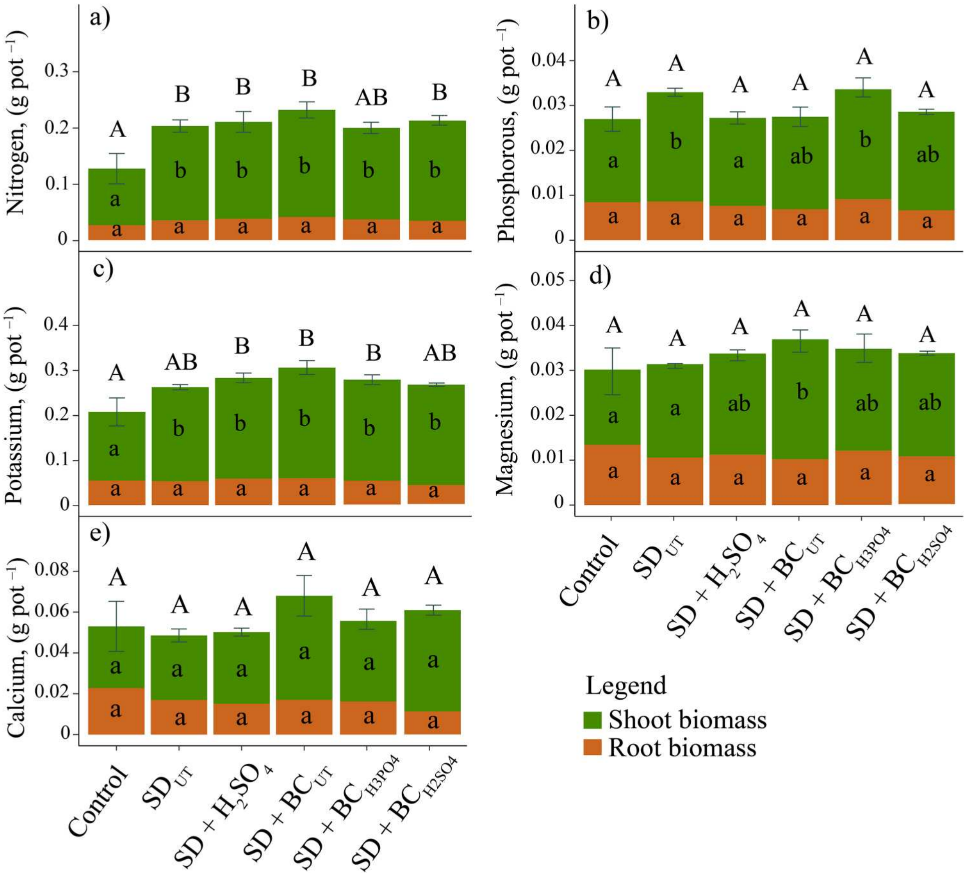

Figure 2. Uptake of (a) nitrogen, (b) phosphorous, (c) potassium, (d) magnesium, and (e) calcium during the first growth period (mean ± standard error, *n* = 4). Significant differences in uptakes by shoots and roots are marked with different lowercase letters. Significant differences in total nutrient uptake (sum of uptake by shoots and roots) are marked with different capital letters. The differences were significant according to the Tukey's HSD post-hoc test. Control, treatment without any amendment; SD_{UT}, untreated slurry digestate; SD + H_2SO_4, slurry digestate amended with sulphuric acid; SD + BC_{UT}, slurry digestate amended with untreated biochar; SD + BC_{H3PO4}, slurry digestate amended with biochar treated with phosphoric acid; SD + BC_{H2SO4}, slurry digestate amended with biochar treated with sulphuric acid.

Soil N balance

In most treatments, the net N balance in the soil was negative, indicating N loss during the experiment (Table 6). The greatest N loss was observed in the control treatment, where it was significantly larger than in most soils receiving SD. There was no significant difference in soil N deficit between SD_{UT} and amended SD treatments after the first growth period. However, the N deficit tended to be smaller for the treatments in which SD was amended with BC_{UT} or acidified BC. A significant difference did emerge after the second growth period. The

Table 3. Pearson correlation coefficients (*r*) of the linear relationship between nutrient uptakes (α = 0.05, *n* = 23). ns, not significant.

Nutrient	N	P	K	Ca	Mg
		First growth period			
N	–				
P	ns	–			
K	0.68	ns	–		
Ca	ns	ns	0.51	–	
Mg	0.58	ns	0.72	0.47	–
		Second growth period			
N	–				
P	0.83	–			
K	0.87	0.91	–		
Ca	0.57	0.49	0.58	–	
Mg	0.89	0.87	0.92	0.73	–

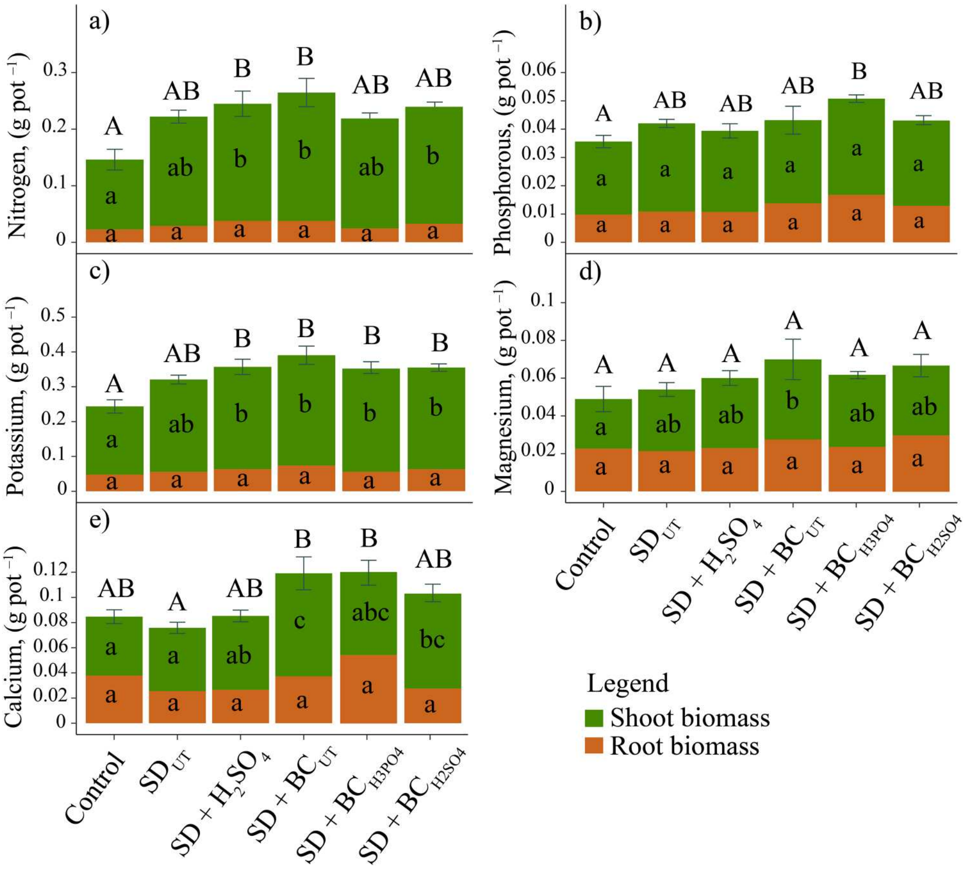

Figure 3. Uptake of (a) nitrogen, (b) phosphorous, (c) potassium, (d) magnesium, and (e) calcium during the second growth period (mean ± standard error, $n = 4$). Significant differences in uptakes by shoots and roots are marked with different lowercase letters. Significant differences in total nutrient uptake (sum of uptake by shoots and roots) are marked with different capital letters. The differences were significant according to the Tukey's HSD post-hoc test. Control, treatment without any amendment; SD_{UT}, untreated slurry digestate; SD + H_2SO_4, slurry digestate amended with sulphuric acid; SD + BC_{UT}, slurry digestate amended with untreated biochar; SD + BC_{H3PO4}, slurry digestate amended with biochar treated with phosphoric acid; SD + BC_{H2SO4}, slurry digestate amended with biochar treated with sulphuric acid.

Table 4. The pH and concentrations of N_{tot}, C_{tot}, P, K, Mg, and Ca in the soil after the first growth period.

Treatment	pH	C_{tot} (%)	N_{tot} (mg kg^{-1})	P (mg kg^{-1})	K (mg kg^{-1})	Mg (mg kg^{-1})	Ca (mg kg^{-1})
			0–10 cm				
Control	7.21 ± 0.09 b	1.35 ± 0.08 b	990 ± 10 b	163 ± 3 ab	52 ± 4 a	247 ± 11 a	1767 ± 16 a
SD_{UT}	7.38 ± 0.06 ab	1.39 ± 0.11 b	1025 ± 85 b	144 ± 5 bc	68 ± 2 a	272 ± 15 a	1899 ± 157 a
SD + H_2SO_4	7.30 ± 0.05 b	1.40 ± 0.02 b	1048 ± 29 b	144 ± 4 bc	61 ± 4 a	259 ± 2 a	1651 ± 39 a
SD + BC_{UT}	7.51 ± 0.03 a	1.68 ± 0.08 a	1300 ± 41 a	155 ± 4 ab	65 ± 4 a	279 ± 13 a	1746 ± 68 a
SD + BC_{H3PO4}	7.36 ± 0.03 ab	1.48 ± 0.05 ab	1075 ± 48 ab	166 ± 1 a	67 ± 6 a	276 ± 8 a	1638 ± 25 a
SD + BC_{H2SO4}	7.45 ± 0.03 ab	1.63 ± 0.08 ab	1202 ± 41 ab	133 ± 2 c	73 ± 7 a	270 ± 5 a	1673 ± 56 a
			10–30 cm				
Control	7.01 ± 0.02 a	1.13 ± 0.02 b	750 ± 50 b	157 ± 3 a	75 ± 3 a	244 ± 28 a	1751 ± 111 a
SD_{UT}	7.16 ± 0.07 a	1.21 ± 0.03 b	1025 ± 25 ab	155 ± 4 a	65 ± 5 a	255 ± 5 a	1706 ± 52 a
SD + H_2SO_4	7.06 ± 0.02 a	1.30 ± 0.10 ab	925 ± 25 ab	146 ± 9 a	64 ± 2 a	259 ± 25 a	1592 ± 89 a
SD + BC_{UT}	7.19 ± 0.03 a	1.42 ± 0.03 a	1100 ± 58 a	160 ± 3 a	66 ± 4 a	262 ± 10 a	1947 ± 203 a
SD + BC_{H3PO4}	7.05 ± 0.02 a	1.35 ± 0.04 ab	1050 ± 50 ab	166 ± 5 a	68 ± 4 a	254 ± 13 a	1795 ± 95 a
SD + BC_{H2SO4}	7.15 ± 0.04 a	1.36 ± 0.06 ab	975 ± 104 ab	167 ± 5 a	54 ± 4 a	256 ± 5 a	1758 ± 156 a

Notes: The values are presented as mean ± standard error ($n = 4$). Results marked with different lowercase letters are statistically different among treatments.

amount of missing N increased significantly in the SD_{UT}, SD + BC_{H2SO4}, and SD + BC_{H3PO4} treatments and was no longer significantly different from the control. The SD + BC_{UT} and SD + H_2SO_4 treatments were distinguished from all other treatments by having significantly lower N loss values. Nonetheless, the SD + H_2SO_4 treatment showed variable results. The N deficit in this treatment in the first growth period was the second largest after the control, but it was the smallest of all experiment treatments by the end of the second period.

Table 5. The pH and concentrations of N_{tot}, C_{tot}, P, K, Mg, and Ca in the soil after the second growth period.

Treatment	pH	C_{tot} (%)	N_{tot} (mg kg^{-1})	P (mg kg^{-1})	K (mg kg^{-1})	Mg (mg kg^{-1})	Ca (mg kg^{-1})
			0–10 cm				
Control	7.37 ± 0.01 b	1.28 ± 0.01 b	950 ± 150 b	171 ± 1 a	57 ± 1 ab	284 ± 12 a	1935 ± 107 ab
SD$_{UT}$	7.52 ± 0.03 ab	1.34 ± 0.02 b	1000 ± 10 b	155 ± 5 a	63 ± 6 a	284 ± 10 a	1813 ± 44 ab
SD + H$_2$SO$_4$	7.53 ± 0.03 ab	1.43 ± 0.13 ab	1125 ± 75 ab	149 ± 1 a	54 ± 2 ab	281 ± 6 a	1786 ± 53 b
SD + BC$_{UT}$	7.56 ± 0.03 a	1.58 ± 0.06 a	1325 ± 25 a	159 ± 7 a	57 ± 6 ab	281 ± 8 a	1853 ± 37 ab
SD + BC$_{H3PO4}$	7.45 ± 0.04 ab	1.33 ± 0.05 b	1025 ± 25 b	158 ± 2 a	44 ± 1 b	301 ± 28 a	1707 ± 43 b
SD + BC$_{H2SO4}$	7.43 ± 0.02 ab	1.48 ± 0.10 ab	1075 ± 48 b	159 ± 11 a	61 ± 2 ab	295 ± 7 a	2163 ± 161 a
			10–30 cm				
Control	7.05 ± 0.00 b	1.11 ± 0.02 b	825 ± 155 b	150 ± 13 a	61 ± 3 a	221 ± 20 a	1503 ± 192 a
SD$_{UT}$	7.18 ± 0.05 ab	1.20 ± 0.04 b	900 ± 41 b	155 ± 4 a	65 ± 7 a	249 ± 12 a	1725 ± 29 a
SD + H$_2$SO$_4$	7.18 ± 0.04 ab	1.25 ± 0.06 ab	888 ± 38 b	162 ± 4 a	64 ± 4 a	249 ± 14 a	1681 ± 88 a
SD + BC$_{UT}$	7.29 ± 0.02 a	1.38 ± 0.02 a	1100 ± 41 a	153 ± 4 a	63 ± 2 a	261 ± 10 a	1714 ± 46 a
SD + BC$_{H3PO4}$	7.16 ± 0.04 ab	1.28 ± 0.05 ab	1000 ± 41 ab	161.94 ± 4 a	59 ± 3 a	257 ± 15 a	1819 ± 109 a
SD + BC$_{H2SO4}$	7.16 ± 0.06 ab	1.31 ± 0.03 ab	1012 ± 31 ab	166.92 ± 3 a	57 ± 3 a	254 ± 5 a	1667 ± 68 a

Notes: The values are presented as mean ± standard error ($n = 4$). Results marked with different lowercase letters are statistically different among treatments.

Table 6. Nitrogen input and output values and net N balance in the soil.

Treatment	N_{tot} of soil (g pot^{-1})	N_{tot} applied with SD (g pot^{-1})	N uptake (g pot^{-1}) (shoots + roots)	N_{tot} amount in soil after biomass harvesting (g pot^{-1})	N balance in soil (g pot^{-1})
			First growth period		
Control	4.00 ± 0.01	0.00 ± 0.00	0.13 ± 0.025 b	3.03 ± 0.12 c	−0.84 ± 0.09 c
SD$_{UT}$	4.00 ± 0.01	0.036 ± 0.005 b	0.21 ± 0.01 a	3.62 ± 0.078 ab	−0.21 ± 0.06 ab
SD + H$_2$SO$_4$	4.00 ± 0.01	0.072 ± 0.003 a	0.21 ± 0.017 a	3.52 ± 0.051 b	−0.35 ± 0.04 b
SD + BC$_{UT}$	4.00 ± 0.01	0.079 ± 0.003 a	0.23 ± 0.013 a	3.87 ± 0.038 a	0.02 ± 0.03 a
SD + BC$_{H3PO4}$	4.00 ± 0.01	0.082 ± 0.006 a	0.20 ± 0.011 ab	3.81 ± 0.1 ab	−0.07 ± 0.10 ab
SD + BC$_{H2SO4}$	4.00 ± 0.01	0.082 ± 0.002 a	0.23 ± 0.016 a	3.91 ± 0.19 a	0.06 ± 0.02 a
			First + second growth period		
Control	4.00 ± 0.01	0.00 ± 0.00	0.15 ± 0.05	3.34 ± 0.060 c	−0.51 ± 0.055 a
SD$_{UT}$	4.00 ± 0.01	0.036 ± 0.005 b	0.21 ± 0.016	3.37 ± 0.078 c	−0.46 ± 0.073 a
SD + H$_2$SO$_4$	4.00 ± 0.01	0.072 ± 0.003 a	0.25 ± 0.022	3.78 ± 0.084 a	−0.11 ± 0.052 c
SD + BC$_{UT}$	4.00 ± 0.01	0.079 ± 0.003 a	0.26 ± 0.033	3.70 ± 0.025 ab	−0.13 ± 0.050 c
SD + BC$_{H3PO4}$	4.00 ± 0.01	0.082 ± 0.006 a	0.23 ± 0.008	3.43 ± 0.075 bc	−0.42 ± 0.072 a
SD + BC$_{H2SO4}$	4.00 ± 0.01	0.082 ± 0.002 a	0.22 ± 0.018	3.64 ± 0.049 abc	−0.21 ± 0.038 ab

Notes: The values are presented as mean ± standard error ($n = 4$). Results marked with different lowercase letters are statistically different among treatments. N/A, not applicable.

Soil C balance

After the first growth period, the C balance was slightly positive in most of the soils ($p > 0.05$, Table 7). However, the C balance was negative in the control and the SD + H$_2$SO$_4$ treatment, suggesting C losses in those treatments.

Table 7. Carbon input and net C balance in the soil.

Treatment	C_{tot} of soil (g pot^{-1})	C_{tot} amount applied with SD (g pot^{-1})	C_{tot} amount in soil after SD incorporation (g pot^{-1})	C_{tot} amount in soil after biomass harvesting (g pot^{-1})	C_{tot} balance in soil (g pot^{-1})
			First growth period		
Control	49.14	0.00 ± 0.00 d	49.14	41.98 ± 2.79 b	−7.16 ± 2.79 Aa
SD$_{UT}$	49.14	0.57 ± 0.01 c	49.72	50.62 ± 1.84 ab	+0.91 ± 1.84 a
SD + H$_2$SO$_4$	49.14	0.64 ± 0.04 c	49.82	47.43 ± 0.56 ab	−2.39 ± 0.56 Aa
SD + BC$_{UT}$	49.14	1.41 ± 0.03 b	50.55	55.78 ± 2.51 a	+5.23 ± 2.51 a
SD + BC$_{H3PO4}$	49.14	1.57 ± 0.05 a	50.66	51.86 ± 1.50 ab	+1.21 ± 1.50 a
SD + BC$_{H2SO4}$	49.14	1.53 ± 0.02 ab	50.68	51.90 ± 3.90 ab	+1.22 ± 3.90 a
			First + second growth period		
Control	49.14	0.00 ± 0.00 d	49.14	44.89 ± 0.61 b	−4.25 ± 0.61 Ab
SD$_{UT}$	49.14	0.57 ± 0.01 c	49.72	45.89 ± 0.99 b	−3.83 ± 0.99 Ab
SD + H$_2$SO$_4$	49.14	0.64 ± 0.04 c	49.82	48.71 ± 1.19 ab	−1.11 ± 1.19 ab
SD + BC$_{UT}$	49.14	1.41 ± 0.03 b	50.55	53.18 ± 1.61 a	+2.63 ± 1.61 a
SD + BC$_{H3PO4}$	49.14	1.57 ± 0.05 a	50.66	46.23 ± 1.65 b	−4.43 ± 1.65 b
SD + BC$_{H2SO4}$	49.14	1.53 ± 0.02 ab	50.68	50.20 ± 0.59 ab	−0.48 ± 0.59 ab

Notes: The values are presented as mean ± standard error ($n = 4$). Results marked with different lowercase letters are statistically different among treatments (two-way ANOVA, $p < 0.05$). The amount of C in the soil at the end of the growth period was also compared with the amount of C in the soil after SD incorporation within each treatment. Capital letters indicate a significant difference in soil C content within that single treatment (paired t-test, $p < 0.05$).

After the second growth period, the C balance in soil was negative in most treatments, with significant differences in the extent of the C deficit between treatments. The only exception to this was the SD + BC_{UT} treatment, for which the C balance continued to be positive, but the C amount in the soil had decreased compared with the previous measurement. The greatest C deficit was observed in the control and in the SD_{UT} and SD + BC_{H3PO4} treatments. The C loss was significantly correlated with N loss during both growth periods. In the first growth period, the correlation coefficient was 0.90 ($p < 0.05$), whereas it was 0.87 ($p < 0.05$) in the second growth period.

Discussion

Impact of SD amendments on nutrient uptake and soil N status

Use of SD with amendments almost doubled the N input to the soil but its effect on N uptake and DMY was small (Table 6, Figures 1–3). The SD + BC_{UT} treatment was the only one in which DMY was significantly higher than in $SD_{UT,}$ but this effect occurred only during the first growth period. The N uptake was not significantly affected by SD amendment; however, during the second growth period, it tended to be not significantly higher in SD + BC_{UT} (13–23.8%) and SD + H_2SO_4 (8.7–19.0%) treatments compared with the others (Figure 3). These results support Foereid et al. (2021) and Frick et al. (2023), who found no significant difference in N uptake when digestate was amended with BC. Moreover, Lentz et al. (2014) added BC produced from hardwood via fast pyrolysis at 500°C to solid dairy cattle manure and did not observe improved manure effects on corn yield and N uptake. Frick et al. (2023) added BC (2.2% w/w) produced from tree and shrub cuttings at 500–600°C to SD. They hypothesised that insufficient BC absorption capacity, or an excessively high dry matter content of the SD may have led to the observed lack of improvement.

The low effect of amended SD on DMY and N uptake in this experiment could be attributed to the fact that the ryegrass sward was newly established at the beginning of the experiment, resulting in a lower N requirement compared with a mature sward. In addition, the considerably higher N uptake compared with the N input with SD suggests a high inorganic N content in the soil, likely because of soil preparation activities such as drying, sieving, and mixing with sand. Soil aeration is known to enhance microbial activity and stimulate net N mineralisation (Gao et al. 2019; Xu et al. 2019).

However, it was also found that at the end of the second growth period, the soil of the SD + BC_{UT} treatment had a significantly higher N concentration than that of all other treatments, except for SD + H_2SO_4 (Table 5). The N balance calculations revealed that the N deficit in these two treatments was significantly lower than in the others. In particular, it had a 3–4 times lower N deficit than that of the SD_{UT} treatment (Table 6), suggesting that SD amended with both BC_{UT} and H_2SO_4 effectively minimised N loss in the soil.

One prevailing hypothesis is that BC impacts on soil N are primarily driven by adsorption of NH_4 (Cao et al. 2023; Vieira Firmino et al. 2024). Sarkhot et al. (2013) found that BC, produced from a mixture of hardwood shavings via slow pyrolysis at 300°C and mixed with filtered liquid dairy manure, significantly reduced net nitrification, ammonification, and N_2O and CO_2 fluxes from the soil, likely due to N adsorption.

In our experiment, N loss in the pots occurred predominantly via gaseous emissions, as water drainage following irrigation was minimal. In this study, the only measured N parameter was N_{tot}, which limits our ability to determine the precise mechanisms by which BC influenced N loss. However, we argue that adsorption was unlikely to be the dominant mechanism through which BC_{UT} affected the soil N cycle. This finding is supported by the hydrophobic nature of BC_{UT} and its low content of the oxygen-containing functional groups necessary for effective NH_4^+ adsorption (Covali et al. 2021). Instead, the results suggest that BC influenced microbial processes in the soil. This interpretation is supported by the fact that the SD amendment altered not only N but also C losses. The BC used in this study was produced at 300°C, yielding partially carbonised compounds containing readily bioavailable C for microbial metabolism (Chen et al. 2015; Wang et al. 2015). Furthermore, milling of BC pellets is likely to have increased the availability of labile C, as the less carbonised interior of the pellets was exposed and the finer particles facilitated microbial interaction. Labile C serves as a substrate and energy source for soil microbes, thereby stimulating microbial biomass production and promoting both N immobilisation and the enhancement of mineralisation and nitrification processes (Cayuela et al. 2014; Nelissen et al. 2015; Liu et al. 2019; Li et al. 2020)

The acidification of BC is one way to increase its cation-exchange capacity and thereby improve the retention of both NH_4^+ and NH_3 (Asada et al. 2006; Hestrin et al. 2019). In addition, acidification significantly lowers the pH of BC, which helps reduce NH_3 losses by making the environment – in this experiment, the digestate and soil – more acidic (Sommer et al. 2017; Andersson et al. 2023; Zireeni et al. 2025). Contrary to our hypotheses, the N loss in treatments in which acidified BC was applied was greater than in the BC_{UT} treatment (Table 6), with the difference becoming apparent only after the second growth period. At the end of the first growth period, no significant differences were observed between the effects of BC_{UT} and the acidified BC treatments. This suggests that the acidified BC reduced N loss at the beginning of the experiment, but this effect diminished over time. A particularly pronounced N deficit in the soil was recorded at the end of the experiment in the SD + BC_{H3PO4} treatment, where it was nearly twice as high as in the SD + BC_{H2SO4} treatment. Although this difference was not statistically significant, it indicates that the type of acid used for BC acidification may influence its effect in soil. The mechanisms underlying the differing impacts of untreated and acidified BC warrant further investigation.

The use of SD with amendments also had a slight positive effect on the uptake of K, P, and Ca (Figures 2 and 3). Similar to N, this effect was more apparent in the second growth period. There was no significant difference in uptake of these nutrients and their soil concentrations between BC-amended SD and H_2SO_4-amended SD, indicating that the BC itself was not a significant source of these elements, contrary to our hypothesis. Several studies have shown that BC enriches the soil with essential plant nutrients (P, K, Mg, and Ca), the composition of which depends on the BC raw material and the production temperature (Gaskin et al. 2010; Enders et al. 2012; Kloss et al. 2012). In our previous experiment, we observed that pellets of the same hay BC significantly increased K concentration in soil (Raave et al. 2020). In that experiment, the amount of BC applied to the soil was almost six times greater than the amount of BC applied to the soil with SD in the current experiment, which may explain the lack of impact of BC on the nutrient uptake and concentration in the soil. In the current experiment, the uptake of K, P, and Ca was positively correlated with N uptake, indicating that SD amendments primarily affected N uptake, which in turn influenced the uptake of other nutrients.

Plant P uptake in the SD + BC_{H3PO4} treatment was higher (non-significant) than in the other SD treatments (Figure 3). This suggests that residual P remaining on the surface of BC_{H3PO4} during acidification (the quantity of which was not quantified) played an important role in enhancing P uptake. This result was unexpected, as the concentration of plant-available P remained high across all treatments throughout the experiment (Tables 4 and 5) and was not a limiting factor for uptake. Moreover, the soil N/P ratio (N/P ratio of 5.5–8.3) was within the optimal range for nutrient uptake (4.5–9, according to Sadras (2006)) at the end of second growth period, indicating that plants in the other treatments should also have been capable of taking up a comparable amount of P from the soil as in the SD + BC_{H3PO4} treatment.

To the best of our knowledge, this is the first time the effect of BC acidified with H_3PO_4 on P uptake has been investigated, so there are no results with which to compare ours. Our findings indicate that BC_{H3PO4} addition to SD increases P uptake when the readily available P content in soil is high. Further research is required to understand the mechanisms behind this increased P uptake.

Impact of SD amendments on soil C status

As shown for the N balance, the C balance in soil also was affected by SD fertilisation and the choice of amendment (Table 7). Compared with the control soil at the beginning and at the end of experiment, which experienced a significant decrease in soil C (Table 7), the C changes in soils among SD treatments were observed later in the experiment. The C loss was lower in soils receiving amended SD; however, the impact of the tested amendments on C loss varied widely. Notably, the SD + BC_{UT} treatment showed a clearly greater amount of C in the soil at the end of the experiment than at the beginning compared with the other treatments. This could be due to our inability to remove all the fibrous roots from the soil in all treatments, and it is likely that masked any C decrease within this specific treatment; however, the changes were smaller than in the other treatments. We hypothesised that the addition of large amounts of labile C to the soil with BC_{UT} would have resulted in an increase in microbial biomass and a decrease in the decomposition of soil organic matter and the applied SD. This hypothesis is supported by the finding that C loss in soil amended with SD + BC_{UT} was lower (non-significant) than that observed in SD +

H_2SO_4, where N input was similar, but no additional C was added to SD. This is in agreement with Lentz and Ippolito (2012), who showed that the loss of organic C from the soil was lower when manure was applied together with BC, leading them to hypothesise that BC may inhibit the mineralisation of manure C.

The soil C losses from the SD + BC_{H3PO4} treatment were significantly higher than those observed in the SD + BC_{UT} treatment by the end of experiment (Table 7). At the same time, the C input to the soil in this treatment was also higher, suggesting that during the storage period, C loss was significantly lower in the acidified BC treatment (Table 7). This indicates that the BC_{UT} was probably more resistant to microbial degradation compared to acidified BC, which may explain the lower soil C and N losses from this treatment. This hypothesis is supported by the finding that a significant difference in N and C loss between these treatments occurred only after the second growth season. However, Hestrin et al. (2020) reported increased C loss during composting when H_2O_2-treated BC was added to manure, hypothesising that this may result from microbial mineralisation of C in oxidised BC or from changes to the composting environment that enhance microbial activity. In the present study, significant C loss was observed only in the treatment with H_3PO_4-acidified BC, whereas C loss associated with H_2SO_4-acidified BC was approximately nine times lower. This suggests that P compounds remaining on the BC surface following H_3PO_4 acidification may have facilitated BC degradation. The increased P uptake observed in this treatment further indicates that these elements were attractive to plant roots. Plant roots support microbial communities by releasing easily available C through root exudates, thereby stimulating microbial activity and potentially accelerating the decomposition of organic matter (Kuzyakov 2010; Adamczyk et al. 2019).

Conclusions

All SD amendments doubled the N input to the soil compared with SD_{UT}; however, their effects on biomass yield and N uptake were minimal, which is probably due to the high inorganic N content in the soil. Among the amendments tested, only BC_{UT} significantly enhanced the effect of SD on plant growth and also reduced N and C losses from soil. Therefore, BC_{UT} is the only amendment suitable to replace H_2SO_4, which is currently used to reduce N losses during slurry storage. Its effect on SD fertilisation properties is comparable to that of H_2SO_4, with the added benefit of increasing C input to the soil and reducing C losses from the soil. However, further tests under farm conditions are still needed to confirm this conclusion.

Contrary to our hypothesis, BC acidification did not enhance its effect on the fertilisation properties of SD. Instead, the results indicated the opposite, with slightly lower DMY and N uptake, as well as higher N and C losses in these treatments.

However, BC acidification may facilitate the uptake of specific nutrients, as evidenced by the significantly higher P uptake in the SD + BC_{H3PO4} treatment. Further long-term research is needed to clarify the mechanisms behind this effect and to assess whether BC acidification can enhance the uptake of other nutrients as well.

Disclosure statement

No potential conflict of interest was reported by the author(s).

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References

Abbas A, Naveed M, Azeem M, Yaseen M, Ullah R, Alamri S, Farooq QUA, Siddiqui MH. 2020. Efficiency of wheat straw biochar in combination with compost and biogas slurry for enhancing nutritional status and productivity of soil and plant. Plants. 9(11):1–19. doi:10.3390/plants9111516.

Adamczyk B, Sietiö OM, Straková P, Prommer J, Wild B, Hagner M, Pihlatie M, Fritze H, Richter A, Heinonsalo J. 2019. Plant roots increase both decomposition and stable organic matter formation in boreal forest soil. Nat Commun [Internet]. 10(1):1–9. doi:10.1038/s41467-019-11993-1.

Andersson K, Delin S, Pedersen J, Hafner SD, Nyord T. 2023. Ammonia emissions from untreated, separated and digested cattle slurry – effects of slurry type and application strategy on a Swedish clay soil. Biosyst Eng [Internet]. 226(January):194–208. doi:10.1016/j.biosystemseng.2023.01.012.

Angst TE, Six J, Reay DS, Sohi SP. 2014. Impact of pine chip biochar on trace greenhouse gas emissions and soil nutrient dynamics in an annual ryegrass system in California. Agric Ecosyst Environ [Internet]. 191:17–26. doi:10.1016/j.agee.2014.03.009.

Antonangelo JA, Sun X, Zhang H. 2021. The roles of co-composted biochar (COMBI) in improving soil quality, crop productivity, and toxic metal amelioration. J Environ Manage. 277(September 2020):1–13. doi:10.1016/j.jenvman.2020.111443.

Antor NH, Mia S, Hasan MM, Lipi NJ, Jindo K, Sanchez-Monedero MA, Rashid MH. 2023. Chemically and biologically activated biochars slow down urea hydrolysis and improve nitrogen use efficiency. Pedosphere [Internet]. 33(4):659–669. doi:10.1016/j.pedsph.2022.06.050.

Asada T, Ohkubo T, Kawata K, Oikawa K. 2006. Ammonia adsorption on bamboo charcoal with acid treatment. J Heal Sci. 52(5):585–589. doi:10.1248/jhs.52.585.

Azzi ES, Li H, Cederlund H, Karltun E, Sundberg C. 2024. Modelling biochar long-term carbon storage in soil with harmonized analysis of decomposition data. Geoderma [Internet]. 441(October 2023):116761. doi:10.1016/j.geoderma.2023.116761.

Cao X, Reichel R, Wissel H, Brüggemann N. 2023. Improving nitrogen retention of cattle slurry with oxidized biochar: an incubation study with three different soils. J Environ Qual. 52(1):1–12. doi:10.1002/jeq2.20424.

Cayuela ML, van Zwieten L, Singh BP, Jeffery S, Roig A, Sánchez-Monedero MA. 2014. Biochar's role in mitigating soil nitrous oxide emissions: a review and meta-analysis. Agric Ecosyst Environ [Internet]. 191:5–16. doi:10.1016/j.agee.2013.10.009.

Chen T, Zhou Z, Han R, Meng R, Wang H, Lu W. 2015. Adsorption of cadmium by biochar derived from municipal sewage sludge: impact factors and adsorption mechanism. Chemosphere [Internet]. 134:286–293. doi:10.1016/j.chemosphere.2015.04.052.

Covali P, Raave H, Escuer-Gatius J, Kaasik A, Tõnutare T, Astover A. 2021. The effect of untreated and acidified biochar on NH3-N emissions from slurry digestate. Sustain. 13(2):1–20. doi:10.3390/su13020837.

Ding Y, Liu Y, Liu S, Li Z, Tan X, Huang X, Zeng G, Zhou L, Zheng B. 2016. Biochar to improve soil fertility. A review. Agron Sustain Dev [Internet]. 36(2):1–18. doi:10.1007/s13593-016-0372-z.

Egnér H, Riehm H, Domingo WR. 1960. Untersuchungen uber die chemische Bodenanalyse als Grundlage fur die Beurteilung des Nährstoffzustandes der Böden. II. Chemische Extraktionsmethoden zur Phosphor – und Kaliumbestimmung. K Lantbrukshögskolans Ann. 26:199–215.

Enders A, Hanley K, Whitman T, Joseph S, Lehmann J. 2012. Characterization of biochars to evaluate recalcitrance and agronomic performance. Bioresour Technol [Internet]. 114:644–653. doi:10.1016/j.biortech.2012.03.022.

Escuer-Gatius J, Shanskiy M, Soosaar K, Astover A, Raave H. 2020. High-temperature hay biochar application into soil increases N 2 O fluxes. Agronomy. 10(1):1–17. doi:10.3390/agronomy10010109.

Fangueiro D, Pereira J, Bichana A, Surgy S, Cabral F, Coutinho J. 2015. Effects of cattle-slurry treatment by acidification and separation on nitrogen dynamics and global warming potential after surface application to an acidic soil. J Environ Manage. 162:1–8. doi:10.1016/j.jenvman.2015.07.032.

Foereid B, Alvarenga E, Szocs J, Makadi M. 2020. Uptake and leaching of sorbed ammonium during early growth of wheat. Int J Recycl Org Waste Agric. 9(2):221–228. doi:10.30486/ijrowa.2020.1885774.1013.

Foereid B, Szocs J, Patinvoh RJ, Horváth IS. 2021. Effect of anaerobic digestion of manure before application to soil – benefits for nitrogen utilisation? Int J Recycl Org Waste Agric. 10(1):89–99. doi:10.30486/IJROWA.2020.1897538.1055.

Frick H, Efosa N, Oberson A, Krause HM, Nägele HJ, Frossard E, Bünemann EK. 2023. Nitrogen dynamics after slurry application as affected by anaerobic digestion, biochar and a nitrification inhibitor. Soil Use Manag. 40(July):1–18. doi:10.1111/sum.12953.

Gao S, DeLuca TH, Cleveland CC. 2019. Biochar additions alter phosphorus and nitrogen availability in agricultural ecosystems: a meta-analysis. Sci Total Environ [Internet]. 654:463–472. doi:10.1016/j.scitotenv.2018.11.124.

Gaskin JW, Speir RA, Harris K, Das KC, Lee RD, Morris LA, Fisher DS. 2010. Effect of peanut hull and pine chip biochar on soil nutrients, corn nutrient status, and yield. Agron J [Internet]. [accessed 2018 Oct 5] 102(2):623. doi:10.2134/agronj2009.0083.

Hazman MY, El-Sayed MEA, Kabil FF, Helmy NA, Almas L, McFarland M, El Din AS, Burian S. 2022. Effect of biochar application to fertile soil on tomato crop production under saline irrigation regime. Agronomy. 12(7):1–17. doi:10.3390/agronomy12071596.

Hestrin R, Enders A, Lehmann J. 2020. Ammonia volatilization from composting with oxidized biochar. J Environ Qual. 49(6):1690–1702. doi:10.1002/jeq2.20154.

Hestrin R, Torres-Rojas D, Dynes JJ, Hook JM, Regier TZ, Gillespie AW, Smernik RJ, Lehmann J. 2019. Fire-derived organic matter retains ammonia through covalent bond formation. Nat Commun [Internet]. 10(1):1–8. doi:10.1038/s41467-019-08401-z.

IUSS Working Group WRB. 2015. World Reference Base for Soil Resources 2014, update 2015 International soil classification system for naming soils and creating legends for soil maps. [place unknown]. doi:10.1038/nnano.2009.216.

Karhu K, Mattila T, Bergström I, Regina K. 2011. Biochar addition to agricultural soil increased CH4 uptake and water holding capacity – results from a short-term pilot field study. Agric Ecosyst Environ [Internet]. [accessed 2018 Oct 5] 140(1–2):309–313. doi:10.1016/j.agee.2010.12.005.

Keskinen R, Nikama J, Kaseva J, Rasa K. 2021. Feasibility of nitrogen-enriched chars as circular fertilizers. Waste Biomass Valorization [Internet]. 12(12):6823–6833. doi:10.1007/s12649-021-01471-5.

Khan M, Basir A, Fahad S, Adnan M, Saleem M, Iqbal A, Amanullah P, Al-Huqail A, Alosaimi A, Saud S, et al. 2022. Biochar optimizes wheat quality, yield, and nitrogen acquisition in low fertile calcareous soil treated with organic and mineral nitrogen fertilizers. Front Plant Sci. 13(May):1–13. doi:10.3389/fpls.2022.879788.

Kloss S, Zehetner F, Dellantonio A, Hamid R, Ottner F, Liedtke V, Schwanninger M, Gerzabek MH, Soja G. 2012. Characterization of slow pyrolysis biochars: effects of feedstocks and pyrolysis temperature on biochar properties. J Environ Qual [Internet]. [accessed 2018 Oct 10] 41(4):990. doi:10.2134/jeq2011.0070.

Kuzyakov Y. 2010. Priming effects: interactions between living and dead organic matter. Soil Biol Biochem [Internet]. 42(9):1363–1371. doi:10.1016/j.soilbio.2010.04.003.

Lehmann J, Joseph S. 2015. Biochar for environmental management: Science, technology and implementation. (2nd ed.). London: Routledge. doi:10.4324/9780203762264.

Lentz RD, Ippolito JA. 2012. Biochar and manure affect calcareous soil and corn silage nutrient concentrations and uptake. J Environ Qual [Internet]. [accessed 2018 Oct 5] 41(4):1033–1043. doi:10.2134/jeq2011.0126.

Lentz RD, Ippolito JA, Spokas KA. 2014. Biochar and manure effects on net nitrogen mineralization and greenhouse gas emissions from calcareous soil under corn. Soil Sci Soc Am J. 78(5):1641–1655. doi:10.2136/sssaj2014.05.0198.

Li S, Tasnady D. 2023. Biochar for soil carbon sequestration: current knowledge, mechanisms, and future perspectives. C-Journal Carbon Res. 9(3):1–30. doi:10.3390/c9030067.

Li X, Wang T, Chang SX, Jiang X, Song Y. 2020. Biochar increases soil microbial biomass but has variable effects on microbial diversity: a meta-analysis. Sci Total Environ [Internet]. 749:141593. doi:10.1016/j.scitotenv.2020.141593.

Liu S, Zhang Y, Zong Y, Hu Z, Wu S, Zhou J, Jin Y, Zou J. 2016. Response of soil carbon dioxide fluxes, soil organic carbon and microbial biomass carbon to biochar amendment: a meta-analysis. GCB Bioenergy. 8(2):392–406. doi:10.1111/gcbb.12265.

Liu X, Zhou J, Chi Z, Jufeng Z, Li L, Zhang X, Jinwei Z, Cheng K, Bian R, Pan G. 2019. Biochar provided limited benefits for rice yield and greenhouse gas mitigation six years following an amendment in a fertile rice paddy. CATENA. 179:20–28. doi:10.1016/J.CATENA.2019.03.033.

Liu Y, Yang M, Wu Y, Wang H, Chen Y, Wu W. 2011. Reducing CH4 and CO2 emissions from waterlogged paddy soil with biochar. J Soils Sediments. 11(6):930–939. doi:10.1007/s11368-011-0376-x.

Loide V, Saue T, Võsa T, Tamm K. 2020. The effect of acidified slurry on crop uptake and leaching of nutrients from a loamy topsoil. Acta Agric Scand Sect B Soil Plant Sci [Internet]. 70(1):31–38. doi:10.1080/09064710.2019.1665705.

Ndegwa PM, Hristov AN, Arogo J, Sheffield RE. 2008. A review of ammonia emission mitigation techniques for concentrated animal feeding operations. Biosyst Eng. 100(4):453–469. doi:10.1016/j.biosystemseng.2008.05.010.

Nelissen V, Rütting T, Huygens D, Ruysschaert G, Boeckx P. 2015. Temporal evolution of biochar's impact on soil nitrogen processes – a 15N tracing study. GCB Bioenergy. 7(4):635–645. doi:10.1111/gcbb.12156.

Park SH, Lee BR, Jung KH, Kim TH. 2018. Acidification of pig slurry effects on ammonia and nitrous oxide emissions, nitrate leaching, and perennial ryegrass regrowth as estimated by15N-urea flux. Asian-Australasian J Anim Sci. 31(3):457–466. doi:10.5713/ajas.17.0556.

Raave H, Escuer-Gatius J, Kauer K, Shanskiy M, Tõnutare T, Astover A. 2020. Permanent grassland hay-derived biochar increases plant N, P and K uptake on an acidic soil. Zemdirbyste. 107(3):227–234. doi:10.13080/z-a.2020.107.029.

Rasse DP, Weldon S, Joner EJ, Joseph S, Kammann CI, Liu X, O'Toole A, Pan G, Kocatürk-Schumacher NP. 2022. Enhancing plant N uptake with biochar-based fertilizers: limitation of sorption and prospects. Plant Soil [Internet]. 475(1–2):213–236. doi:10.1007/s11104-022-05365-w.

R Development Core Team. 2019. R: A language and environment for statistical computing [Internet]. http://www.r-project.org.

Rosopulo A, Hahn M, Stark H, Fiedler J. 1976. Vergleich verschiedener Veraschungsmethoden für die nasschemische Bestimmung von Mengen und Spurenelementen in Kulturpflanzen. Landwirtsch Forsch 29 Sonderh.:199–209.

Sadras VO. 2006. The N:P stoichiometry of cereal, grain legume and oilseed crops. F Crop Res. 95(1):13–29. doi:10.1016/j.fcr.2005.01.020.

Sarkhot DV, Ghezzehei TA, Berhe AA. 2013. Effectiveness of biochar for sorption of ammonium and phosphate from dairy effluent. J Environ Qual. 42(5):1545. doi:10.2134/jeq2012.0482.

Sommer SG, Clough TJ, Balaine N, Hafner SD, Cameron KC. 2017. Transformation of organic matter and the emissions of methane and ammonia during storage of liquid manure as affected by acidification. J Environ Qual. 46(3):514–521. doi:10.2134/jeq2016.10.0409.

Sommer SG, Génermont S, Cellier P, Hutchings NJ, Olesen JE, Morvan T. 2003. Processes controlling ammonia emission from livestock slurry in the field. Eur J Agron. 19(4):465–486. doi:10.1016/S1161-0301(03)00037-6.

Sørensen P, Amato M. 2002. Remineralisation and residual effects of N after application of pig slurry to soil. Eur J Agron. 16(2):81–95. doi:10.1016/S1161-0301(01)00119-8.

Taghizadeh-Toosi A, Clough TJ, Sherlock RR, Condron LM. 2012. Biochar adsorbed ammonia is bioavailable. Plant Soil. 350(1–2):57–69. doi:10.1007/s11104-011-0870-3.

Tomczyk A, Sokołowska Z, Boguta P. 2020. Biochar physicochemical properties: pyrolysis temperature and feedstock kind effects. Rev Environ Sci Biotechnol [Internet]. 19(1):191–215. doi:10.1007/s11157-020-09523-3.

Tsang DCW, Hu J, Liu MY, Zhang W, Lai KCK, Lo IMC. 2007. Activated carbon produced from waste wood pallets: Adsorption of three classes of dyes. Water Air Soil Pollut. 184(1–4):141–155. doi:10.1007/s11270-007-9404-2.

Vieira Firmino M, Trémier A, Couvert A, Szymczyk A. 2024. New insights into biochar ammoniacal nitrogen adsorption and its correlation to aerobic degradation ammonia emissions. Waste Manag. 178(February):257–266. doi:10.1016/j.wasman.2024.02.032.

Wang B, Lehmann J, Hanley K, Hestrin R, Enders A. 2015. Adsorption and desorption of ammonium by maple wood biochar as a function of oxidation and pH. Chemosphere [Internet]. [accessed 2019 Feb 11] 138:120–126. doi:10.1016/j.chemosphere.2015.05.062.

Weng Z, Van Zwieten L, Tavakkoli E, Rose M, Singh B, Joseph S, Macdonald L, Kimber S, Morris S, Rose T, et al. 2022. Microspectroscopic visualization of how biochar lifts the soil organic carbon ceiling. Nat Commun [Internet]. 13(1):1–12. doi:10.1038/s41467-022-32819-7.

Xu H, Cai A, Wu D, Liang G, Xiao J, Xu M, Colinet G, Zhang W. 2021. Effects of biochar application on crop productivity, soil carbon sequestration, and global warming potential controlled by biochar C:N ratio and soil pH: a global meta-analysis. Soil Tillage Res. 213(June):1–10. doi:10.1016/j.still.2021.105125.

Xu Y, Liu K, Han Y, Jiang H, Yao S, Zhang B. 2019. A soil texture manipulation doubled the priming effect following crop straw addition as estimated by two models. Soil Tillage Res [Internet]. 186(September 2018):11–22. doi:10.1016/j.still.2018.09.011.

Zhang Z, Liu D, Qiao Y, Li S, Chen Y, Hu C. 2021. Mitigation of carbon and nitrogen losses during pig manure composting: a meta-analysis. Sci Total Environ [Internet]. 783:147103. doi:10.1016/j.scitotenv.2021.147103.

Zireeni Y, Jones DL, Chadwick DR, Mäenpää MI, Sørensen P. 2025. Low-dose acidification of cattle slurry with sulfuric acid enhances oilseed-rape yield but risks elevated sulfide formation during prolonged storage. Sci Total Environ. 970:1–11. doi:10.1016/j.scitotenv.2025.179014.