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Potential of biochar to restoration of microbial biomass and enzymatic activity in a highly degraded semiarid soil

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Biochar is an effective material for enhancing soil ecosystem services. However, the specific impacts of biochar on microbial indicators, particularly in degraded soils, remain poorly understood. This study aimed to evaluate the effects of biochar produced from cashew residues and sewage sludge, in a highly degraded soil, on microbial indicators. We analyzed soil chemical composition and microbial biomass C and N, enzyme activity, and stoichiometry. Cashew biochar increased soil respiration, indicating a higher availability of C to microorganisms compared to sewage sludge biochar and a better adaptation of soil microbial communities to C-rich organic material obtained from a native plant. Although the biochar differentially impacted microbial biomass C, both significantly increased N in the microbial biomass. Arylsulphatase activity did not respond to biochar application, while β-glucosidase, urease, and phosphatases showed significant changes with biochar treatments. Importantly, stoichiometry and vector analysis revealed that both types of biochar increased P limitation for soil microbes. Conversely, both biochar alleviated C and N limitations for the soil microbes. Thus, biochar applications in highly degraded soils should be supplemented with external P sources to maintain soil functions, mainly for cashew residues. Our results provide evidence that biochar can restore soil biological properties and enhance the availability of C and N to microorganisms. These findings have significant implications for restoration practices in degraded lands of semiarid regions.

Keywords Pyrolysis, Microbial activity, Nutrient cycling, Desertification, Soil health

Soil is an essential resource to support plant growth and human life. However, to maintain the productivity level without increasing the environmental level of degradation has become a global challenge[1]. Nowadays, there is an estimative that ~25% of soils worldwide face several types of degradation affecting, directly, 3 billion people[2]. Soil degradation is especially significant across arid and semiarid regions. This is primarily due to unsuitable land use combined with fragile soils and concentrated rainfall, leading to erosion and soil organic carbon loss[3].

Brazilian semiarid soils are facing a strong degradation process mainly due to intense pasture and livestock production[4], where farmers use native vegetation to support animal feeding[5]. Importantly, overgrazing, which occurs when native plants are subjected to intensive grazing without adequate recovery time, significantly impacts soil health. It reduces vegetation cover and exposes the soil to erosion[6]. Consequently, soil organic C has been lost, affecting the soil attributes, especially the biological properties[5,7]. In the restoration process of these degraded soils, grazing exclusion has been applied in this semiarid region with positive effects on soil[5,8], but this process takes at least two decades to restore microbial properties[9].

Since degradation has decreased soil organic matter (SOM), the use of carbon-rich organic residues, such as biochar, could be an interesting short-term strategy for improving soil properties[10,11] and increase soil C pools[12]. Biochar is a stable material obtained through the pyrolysis of biomass, such as agricultural residues, in a low-oxygen environment[13]. This carbon-rich material has the potential to increase soil organic C[12], enhance soil

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structure[14], nutrient availability[10], and soil productivity. Although previous studies have reported positive effects from biochar application on soil properties, little is known about its potential for restoration of degraded soils, mainly regarding the biological properties. These features are important since biological properties are essential to soil functioning[15] and they can be indicators of soil restoration[16].

Soil microbial biomass (SMB) plays an essential role in soil functioning, being particularly important during soil restoration[17]. The SMB releases extra-cellular enzymes that boost biological activity, mainly related to C, N, and P cycling and availability[18]. Particularly, the assessment of stoichiometry of extra-cellular enzymes (C-, N-, and P-acquiring enzymes) could allow us to understand both limitation and availability of C, N, and P to soil microbial biomass[19]. The understanding of potential enzymatic activity provides information about nutrient turnover and ecosystem metabolism which is important to assess the restoration strategy[9]. While several studies have reported the responses of soil microbial biomass, enzymatic activity, and their stoichiometry, from biochar soil application[20–22] little is known about the potential of applying biochar in degraded soils and its effect on soil microbial biomass, enzymatic activity, and stoichiometry.

Brazil is one of the world's largest cashew producers, with the state of Ceará accounting for the highest share of the country's production. This significant output generates a considerable amount of bagasse from agro-industrial processes. To date, this study represents one of the first attempts to explore the potential of cashew bagasse for restoring microbial properties in tropical soils affected by desertification. Thus, could the application of carbon-rich materials improve the microbial community (biomass and enzyme activity) in highly degraded soil, reducing nutrient limitation?

In this study, we hypothesized that biochar obtained through pyrolysis of pseudo fruit bagasse of cashew - a native plant species from Brazilian semiarid region, could be effective in restoring the soil microbial biomass and enzymatic activity and influencing the enzymatic stoichiometry in degraded soil. More importantly, we hypothesized that cashew biochar stimulates microbial properties faster than the well-known biochar obtained from sewage sludge (which has more recalcitrant compounds). Thus, this study assessed the efficiency of biochar sources (from cashew bagasse and sewage sludge) and doses (0, 5, 10, 20, and 40 Mg ha$-^{1}$) to restore soil microbial indicators (biomass, enzymes, and their stoichiometry) in a highly degraded dryland soil (Fig. 1).

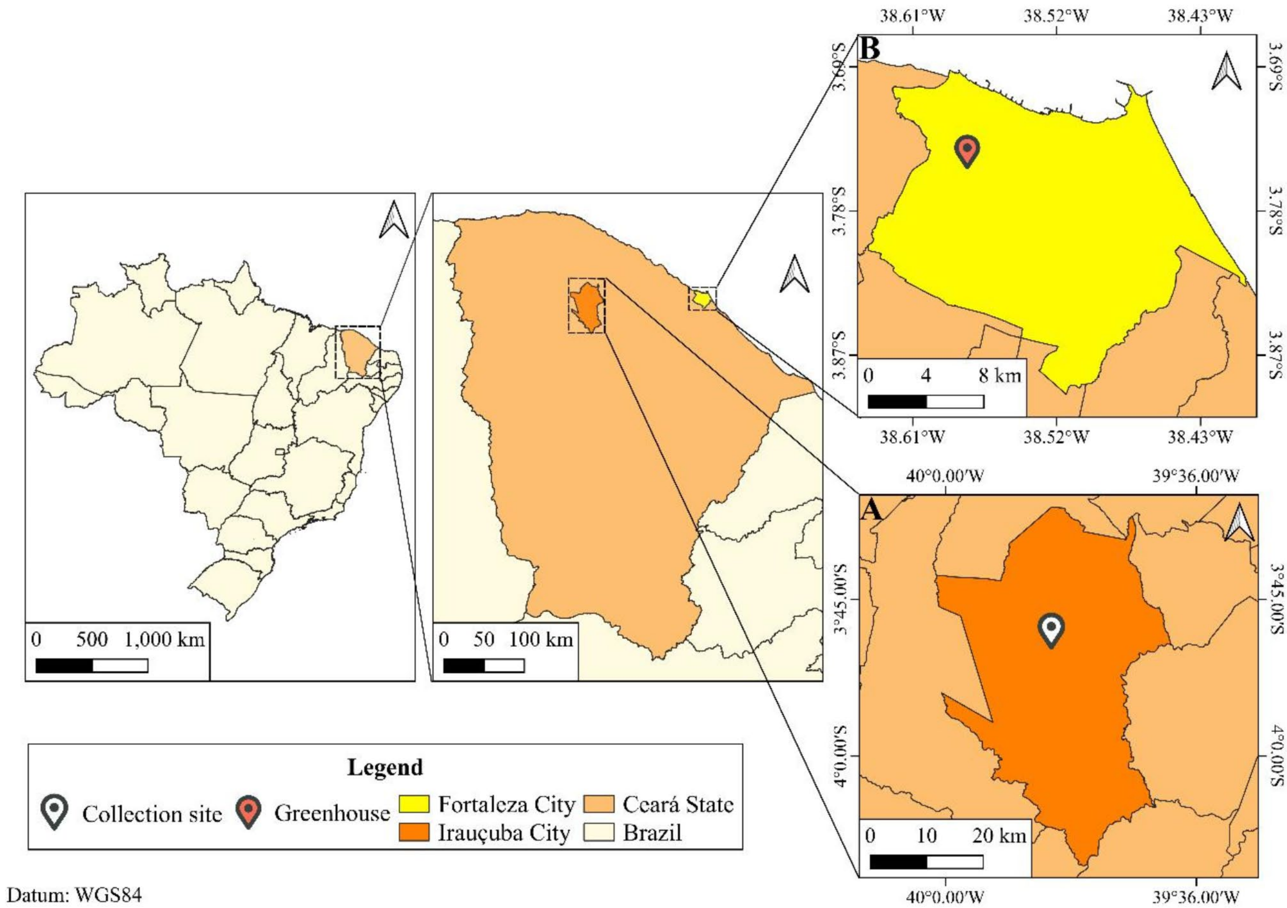

Fig. 1. Geographic location map of (**a**) soil collection site (Irauçuba) and (**b**) greenhouse experiment setup (Fortaleza) in Ceará state, Brazil.

Results

Soil microbial biomass, organic C and N and available P contents

The application of both cashew and sewage sludge biochar increased soil respiration compared to unamended soil (Fig. 2a). However, the most significant effect on soil respiration was found with the application of cashew biochar, where the increase ranged from 50 to 150% at the lowest and highest doses, respectively. The application of 5 and 10 Mg ha^{-1} of cashew biochar, and 10 and 20 Mg ha^{-1} of sewage sludge biochar, increased MBC compared to unamended soil (Fig. 2b). The effect on MBC was more significant with the application of sewage sludge biochar, where the increase was 100% compared to unamended soil. The values of TOC increased ~ 50% with the application of both cashew and sewage sludge biochar at the lowest and highest doses, respectively (Fig. 2c). Both cashew and sewage sludge biochar strongly increased MBN compared to unamended soil (Fig. 2d), showing an increase of ~ 500% with the application of both cashew and sewage sludge biochar. The values of total N increased with the application of both biochar, but the highest value was observed for the highest doses of cashew biochar (~ 100% compared to unamended soil) (Fig. 2e). Available soil phosphorus increased with the application of both biochar, mainly in 20 and 40 Mg ha^{-1} and in sewage sludge treatments (Fig. 2f).

The microbial C quotient increased with the application of 10 and 20 Mg ha^{-1} of both biochar, but the effect was more prominent with the application of sewage sludge biochar (Fig. 3a). The application of sewage sludge biochar increased the values of microbial C quotient by ~ 100% compared to unamended soil. Regarding the microbial N quotient, we observed the highest values with the application of both biochar, with values increased by about 1,000% compared to unamended soil (Fig. 3b). Since the application of both biochar significantly increased MBN, the values of the MBC: MBN ratio decreased significantly (Fig. 3c). On the other hand, the application of 5 and 10 Mg ha^{-1} of cashew biochar, and 10 and 20 Mg ha^{-1} of sewage sludge biochar, increased the values of the organic C: N ratio (Fig. 3d).

Soil enzymatic activity

The soil enzymatic activity showed distinct responses to the application of both cashew and sewage sludge biochar (Fig. 4). The application of both biochar did not change the activity of arylsulfatase in soil (Fig. 4a). The responses of ß-glucosidase were contrasting when compared cashew and sewage sludge biochar (Fig. 4b). The application of cashew biochar decreased the activity of ß-glucosidase in soil but the application of 5 Mg ha^{-1} of sewage sludge biochar while increased the activity of ß-glucosidase, it decreased enzyme activity with the increase of the doses. The acid and alkaline phosphatase showed distinct patterns of response to biochar. The acid phosphatase decreased after the application of cashew biochar at 5, 10, and 20 Mg ha^{-1}, but increased at the highest dose (Fig. 4c). When sewage sludge biochar was applied, acid phosphatase increased with the application of 10 Mg ha^{-1} and 20 Mg ha^{-1}. The alkaline phosphatase increased 150% to the highest doses of both cashew and sewage sludge biochar compared to unamended soil (Fig. 4d). The urease activity increased with the application of both biochar (Fig. 4e). However, the highest values of urease activity were observed with the application of 40 Mg ha^{-1} of cashew biochar and 20 Mg ha^{-1} of sewage sludge biochar. The application of cashew biochar increased urease activity by ~ 400%, while sewage sludge biochar increased it by ~ 600% compared to unamended soil.

Enzymatic stoichiometry

The application of both cashew and sewage sludge biochar decreased the enzymatic C: N ratio (Fig. 5a). In contrast, the enzymatic N: P ratio increased with the application of cashew biochar, with the highest enzymatic N: P ratio observed at 10, 20, and 40 Mg ha^{-1} (Fig. 5c). The application of sewage sludge biochar also increased the enzymatic N: P ratio, but the highest value was found with the application of 20 Mg ha^{-1}. Interestingly, the enzymatic C: P ratio was higher in unamended soil and with the application of 10 Mg ha^{-1} of cashew biochar (Fig. 5b). When sewage sludge biochar was applied, we observed a higher enzymatic C: P ratio with the application of 5 Mg ha^{-1}, while 20 and 40 Mg ha^{-1} promoted a decrease in the enzymatic C: P ratio compared to unamended soil.

The A (angle) and L (unitless) changed when comparing cashew and sewage sludge biochar and their application doses (Table 1). The unamended soil and those with the application of 5 Mg ha^{-1} of cashew biochar showed the highest vector A, while the application of sewage sludge biochar decreased vector A compared to unamended soil. The highest values of vector L were found in unamended soil (0.99) and decreased with increased doses of both biochar, reaching 0.64 and 0.69 at the highest doses of cashew and sewage sludge biochar.

Discussion

Soil microbial biomass and organic C and N

This study used a highly degraded soil by overgrazing to verify the potential of biochar application to restore soil biological properties. The degraded soil evaluated presents low soil microbial biomass due to the high degradation, and losses of vegetation cover and soil organic C[5]. Thus, after the application of both cashew and sewage sludge biochar, it was verified a restoration of soil microbial biomass. These results are in line with the main hypothesis that the application of biochar, mainly cashew biochar obtained through a native plant species, was effective in restoring soil microbial biomass and enzymatic activity.

The application of both biochar significantly increased soil basal respiration which means more biological activity derived from decomposition of organic materials[23]. Particularly, soil respiration was higher with the application of cashew biochar suggesting a better adaptation of microbial community to this organic material obtained from native plant species. In addition, the cashew biochar presents higher content of C as compared to sewage sludge biochar (Table 2). We observed that increased biochar doses provide higher available C to soil microbes, increasing soil respiration. Indeed, previous studies have shown increased soil respiration after the

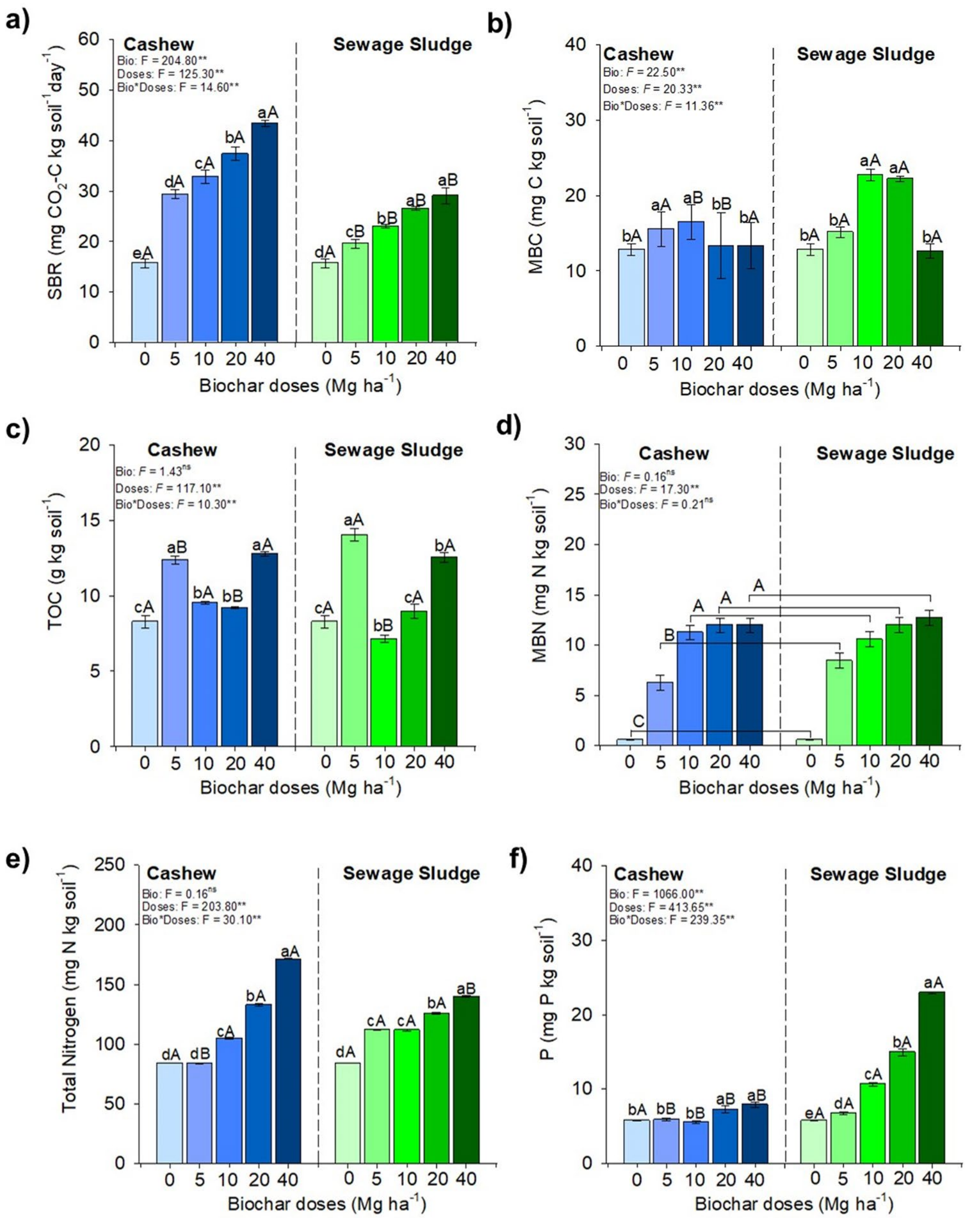

Fig. 2. Soil basal respiration (**a**) and C (**b**, **c**), N (**d**, **e**) and P (**f**) contents in soil and microbial biomass under biochar application (cashew and sewage sludge) in five different doses (0, 5, 10, 20, and 40 Mg ha$-^{1}$). Means followed by the same letter did not differ significantly according to the Scott-Knott test ($p \leq 0.05$). Lowercase letters are used to compare treatments (doses) within each biochar source (i.e., cashew or sludge), while uppercase letters are used to compare treatments (doses) between different biochar sources, $n = 4$. Data variability is represented by error bars.

application of biochar due to higher labile C incorporated into the soil[24,25]. However, the microbial biomass C was higher in low to medium biochar doses which indicates that the highest doses of biochar stimulated soil respiration through an input of available C, but it did not reflect in higher microbial biomass C levels. Both biochar presented low C/N ratio (<20) which promotes lower C immobilization into microbial biomass[26], mainly in higher doses. The microbial biomass N was more influenced by the application of both biochar reflecting the high content of N in both materials compared with unamended soil. Indeed, this contributed to the higher total N found in soil, and to the lower soil C: N ratio (TOC/total N), mainly when cashew biochar was applied in higher doses.

Since the application of sewage sludge biochar increased microbial biomass C (mainly 10 and 20 Mg ha$-^{1}$), it reflected in the increased microbial C quotient (qMic-C) which means a higher fraction of organic matter incorporated as microbial biomass[27]. Regarding microbial N quotient (qMic-N), we observed a higher increase with the application of both biochar, and this reflected the high increase in microbial biomass N. Thus, as N was more incorporated into microbial biomass than C, this decreased the microbial C: N ratio (MBC/MBN),

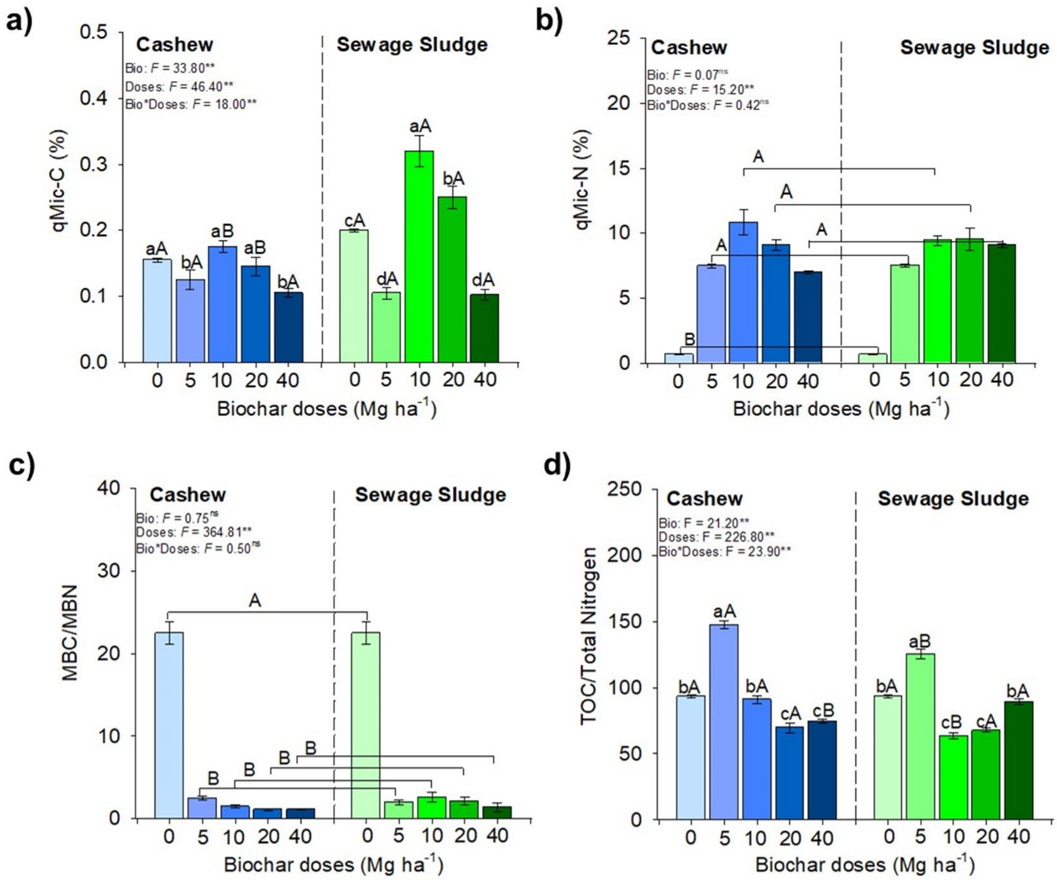

Fig. 3. Metabolic quotient (**a**), microbial use efficiency of C (**b**) and N (**c**), and relationships between C and N of microbial (**d**) biomass under biochar application (cashew and sewage sludge) in five different doses (0, 5, 10, 20, and 40 Mg ha$-^{1}$). Means followed by the same letter did not differ significantly according to the Scott-Knott test ($p \leq 0.05$). Lowercase letters are used to compare treatments (doses) within each biochar source (i.e., cashew or sludge), while uppercase letters are used to compare treatments (doses) between different biochar sources, $n = 4$. Data variability is represented by error bars.

indicating a higher positive effect of both biochar on bacteria than fungi[28]. Thus, it means that the application of both cashew and sewage sludge biochar stimulates more bacterial than fungal communities in the soil. Indeed, previous studies reported that bacterial communities are more active in biochar-treated soil than fungal communities[29,30]. Future studies should aim to comprehensively understand the entire ecology of both bacterial and fungal communities with different biochar doses.

Soil enzymatic activity

The extra-cellular enzymes provide information about the potential mineralization of C (ß-glucosidase), N (urease), P (acid phosphatase), and S (arylsulphatase) which can indicate potential changes in soil biochemical status[31]. The applications of both biochar stimulated soil enzymatic activity, but with distinct responses according to different sources. The application of both biochar did not affect the activity of arylsulfatase which suggests little effect on cycling of S in soil. While no studies were done with cashew biochar yet, a previous study using sewage sludge biochar showed no alterations in arylsulfatase activity and no effect on S cycling[32]. In addition, this no response of arylsulfatase can be related to a lower abundance of fungi than bacteria in soil, as previously reported. It is known that fungi contain ester sulfate that produces sulfatases; thus, the lower abundance of fungi decreases arylsulfatase activity[33]. In contrast, ß-glucosidase activity was more variable as affected by distinct biochar which have different impacts on C cycling. Thus, the application of cashew biochar was not enough to increase the ß-glucosidase in this degraded soil. The addition of labile C-sources (e.g., cashew based-biochar) could influence the ß-glucosidase activity since Wei et al.[34] demonstrated that labile organic C input reduced the related C-acquisition enzyme activities. Interestingly, Foster et al.[35] found that ß-glucosidase reduction may be due to the adsorption of ß-glucosidase substrate in the biochar surface. Similarly, Lehmann et al.[36] suggested that the reduced enzyme synthesis was due to the interaction between microbes and carbon on the biochar surface, where biochar adsorption onto ß-glucosidase led to decreased enzyme production[37]. On the other hand, sewage sludge biochar applied in the lowest doses (5 and 10 Mg ha$-^{1}$) promoted a stimulatory effect on ß-glucosidase possibly due to the addition of C-recalcitrant content[38]. However, higher doses of sewage sludge biochar can increase the content of metals and, thus, affect the ß-glucosidase[39]. A previous study reported β-glucosidase as

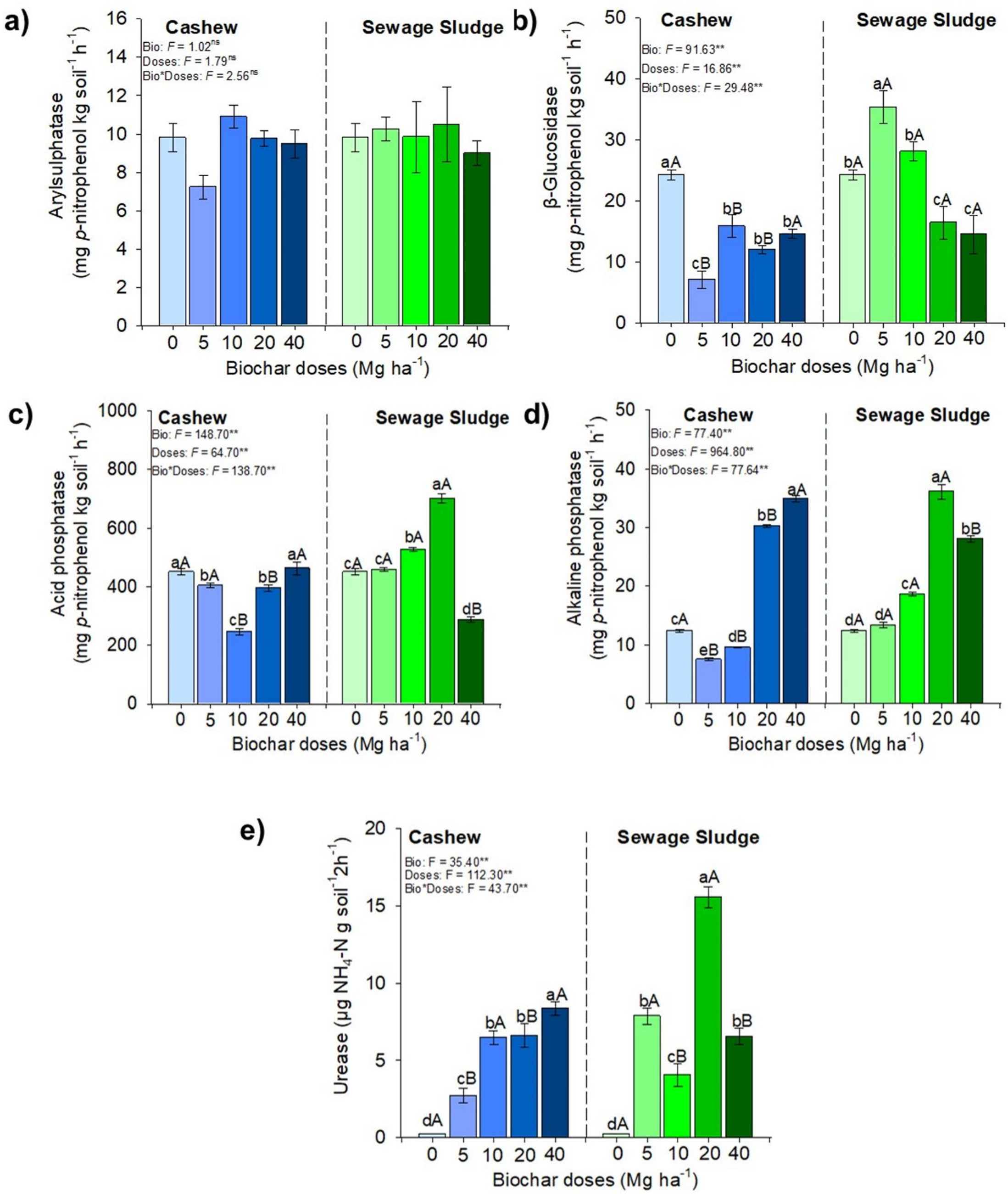

Fig. 4. Soil extracellular enzyme activity related to S (**a**), C (**b**) P (**c**, **d**), and N (**e**) cycles under biochar application (cashew and sewage sludge) in five different doses (0, 5, 10, 20, and 40 Mg ha$-^1$). Means followed by the same letter did not differ significantly according to the Scott-Knott test ($p \leq 0.05$). Lowercase letters are used to compare treatments (doses) within each biochar source (i.e., cashew or sludge), while uppercase letters are used to compare treatments (doses) between different biochar sources, $n = 4$. Data variability is represented by error bars.

the most sensitive indicator of the adverse impact of metals[39]. Several factors could contribute to the inhibition of C-acquiring enzymes, including biomass, pyrolysis temperature, and soil texture. For example, biochars from herbs and wood diminish C-acquiring enzyme activity[40]. This suppression could be related to either the inherent properties of the biochar or alterations in the microbial community following the application of the biochar (e.g., inhibitory compounds in biochar such as metals). Also, the presence of phenolic and lignin compounds can change the chemical composition of soil organic matter, thereby decreasing the bioavailability of carbon compounds that beta-glucosidase can decompose[41] in higher doses of sewage sludge application.

The activity of soil phosphatases showed distinct effects of biochar on biochemical processes related to P cycling. The decrease of acid phosphatase with the application of cashew biochar at 5, 10, and 20 Mg ha$-^1$ could be due to a high P content on biochar and, more importantly, a higher C limitation status observed on

Biochar (Mg ha−1)	Vector A		Vector L	
	Cashew	Sewage sludge	Cashew	Sewage sludge
0	69.87 aA	69.87 aA	0.99 aA	0.99 aA
5	70.35 aA	59.92 cB	0.70 bA	0.73 cA
10	60.82 bB	64.02 bA	0.68 bA	0.80 bA
20	62.80 bA	59.38 cB	0.64 bA	0.59 eB
40	61.45 bA	61.46 cA	0.64 bA	0.69 dA

Table 1. Vectors A (angle) and L (unitless) of soil extracellular enzyme stoichiometry under biochar application (cashew and sludge) in five different doses (0, 5, 10, 20, and 40 mg ha−1). Means followed by the same letter did not differ significantly according to the Scott-Knott test ($p \leq 0.05$). Lowercase letters are used to compare treatments (doses) within each biochar source (i.e., cashew or sludge), while uppercase letters are used to compare treatments (doses) between different biochar sources, $n=4$.

Attributes	Unit	Biochar source	
		Cashew	Sewage Sludge
pH (H_2O)	-	9.6	9.1
C	g kg−1	480.10	348.00
N	g kg−1	27.09	24.45
C/N	-	17.72	14.20
P	g kg−1	11.62	17.70
Na	g kg−1	0.35	4.09
K	g kg−1	7.71	6.10
Ca	g kg−1	1.95	19.30
Mg	g kg−1	4.54	7.30
Cu	mg kg−1	51.0	170.0
Fe	mg kg−1	768	15,300
Mn	mg kg−1	45.0	390.0
Zn	mg kg−1	59.0	1390
Cd	mg kg−1	-	1.0
Cr	mg kg−1	2.0	40.0
Mo	mg kg−1	1.0	10.0
Ni	mg kg−1	4.0	23.0
Pb	mg kg−1	1.0	16.0
Al	g kg−1	1.35	26.8

Table 2. Chemical characterization of biochar from cashew bagasse residues and sewage sludge. - Non detected.

enzymes stoichiometry. Jin et al.[42] demonstrated that manure biochar decreased the acid phosphatase activity, which could be attributed to a higher nutrient P availability in biochar. However, in the highest doses of cashew and sewage sludge biochar, we observed an increased acid phosphatase (and in soil P contents), probably due to the higher values of total N found in soil after applying the biochar. Previous studies have reported that both acid and alkaline phosphatases are produced at the cost of N[43,44], which indicates that the content of total N is a determinant of phosphatase activities. In addition, the increased alkaline phosphatase with the highest doses of each biochar suggests a direct effect of the alkaline pH value found in both tested biochar[45]. It suggests that pyrolytic biochar could enhance soil P contents via both acid and alkaline phosphatases.

The observed increase in urease activity following the application of cashew and sewage sludge biochar demonstrates the significant impact that these amendments can have on N cycling in soil. Additionally, urease activity is significantly modulated by soil pH; applying alkaline biochar to acidic soil can enhance urease activity by improving the soil to a higher pH[46]. Also, in a nutrient-limited system (see below), microbial proliferation could induce enzymatic activity to use C-sources derived from biochar application[47]. Importantly, Zhang et al.[48] demonstrated a positive correlation between urease activity and microbial biomass carbon after the application of wheat straw-derived biochar (8 t ha−1 and 16 t ha−1), indicating that soil N cycling is driving via the potential of the microbial community to increase its biomass.

Enzymatic stoichiometry

The application of biochar changed the soil enzymatic stoichiometry which influenced the availability and limitations of C, N, and P to microbes. Further, the decrease in enzymatic C: N ratios due to biochar application suggests that microbial biomass uses more N than C in its biological processes. This reflected the higher values of N-acquiring enzymes (urease) found with the application of biochar. On the other hand, the degraded soil which was not amended with biochar showed a higher enzymatic C: N ratio, as also observed by Silva et al.[9] for degraded soil. The application of both biochar increased the enzymatic N: P ratio, which confirms the positive effect of applying biochar, i.e., the input of N, promoting the activity of N-acquiring enzymes. This is important since these N-acquiring enzymes improve the cycling of N in the soil[9]. In general, the application of both biochar at high rates decreased the enzymatic C: P ratio, reflecting the lower activity of C-acquiring enzymes (ß-glucosidase).

The values of vectors L (length) and A (angle) are useful to show the degree of C limitation (vector L) and P limitation relative to N (vector A)[49]. The highest values of vector L found in unamended soil suggest more C limitation to soil microbes[50], while when biochar is applied this C limitation decreases. Regarding vector A, the unamended (and 5 Mg ha$-^{1}$ of cashew biochar) soil showed the highest values which indicate more P limitation than N to soil microbes. These results confirm that the application of biochar increases the availability of C and N while promoting a limitation of P to soil microbes. Interestingly, P limitation decreased with higher biochar doses, as indicated by the lower A angle, which could be related to an improvement in soil P content (Fig. 2f). However, despite this increase, the A angle remains higher than 45º (the limit for P limitation[51]) and the rate of P accumulation remains very low for Brazilian semiarid soils[52].

Materials and methods
Soil sampling
The degraded area is located at Irauçuba, Ceará state, Brazil (3°46'16.38"S, 39°49'54.00"W, Fig. 1). This region exhibits highly degraded soil due to overgrazing[6]. The soil is classified as Planosol[53], and the climate is categorized as Bshw – tropical hot semiarid[54], with an annual rainfall of 539 mm, concentrated in January to April, and an average temperature of 26 to 28 ºC[55]. The region presents intensive human activities, which accelerate the soil degradation process.

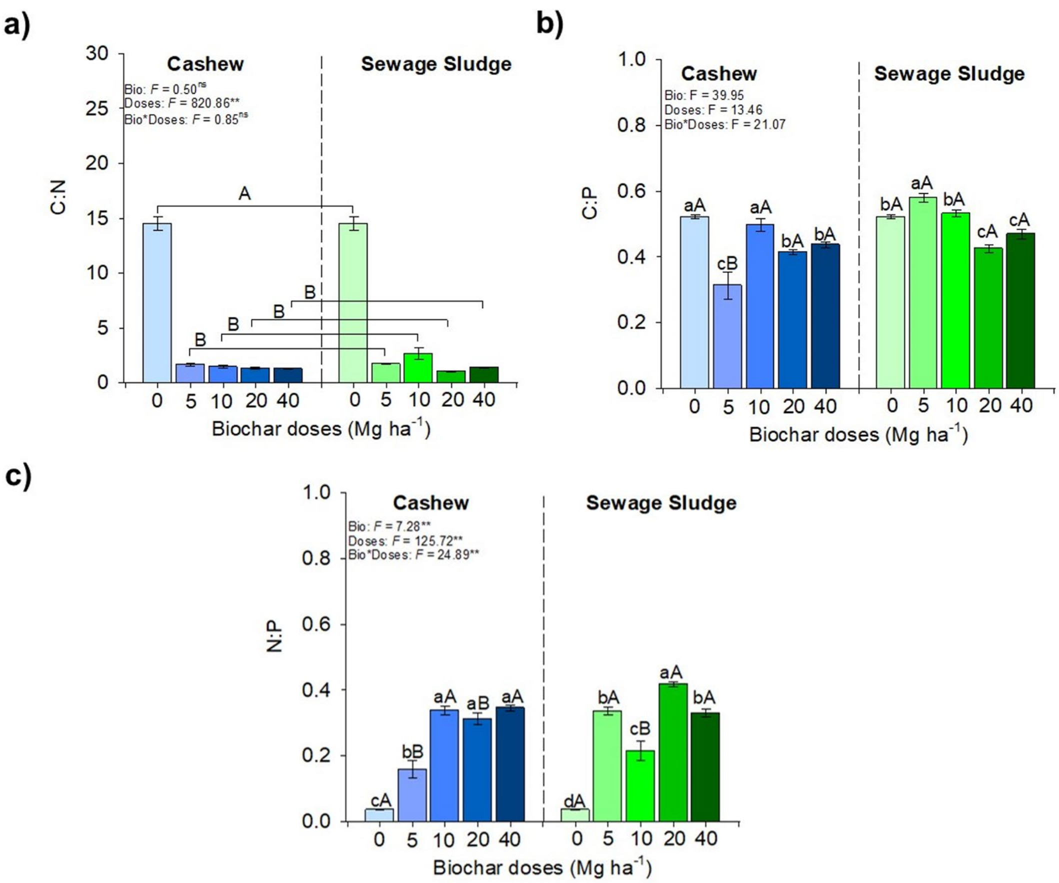

Fig. 5. Soil extracellular enzymes stoichiometry under biochar application (cashew and sewage sludge) in five different doses (0, 5, 10, 20, and 40 Mg ha$-^{1}$). Means followed by the same letter did not differ significantly according to the Scott-Knott test ($p \leq 0.05$). Lowercase letters are used to compare treatments (doses) within each biochar source (i.e., cashew or sludge), while uppercase letters are used to compare treatments (doses) between different biochar sources, $n = 4$. Data variability is represented by error bars.

Soil samples were collected from a depth of 0–10 cm, sieved through a 2 mm mesh to remove large debris, and immediately used in the experiment to ensure the survival of microorganisms. Soil chemical characterization (Table 3) was performed following the methodology described by EMBRAPA[56].

Biochar production and characterization

The biochars were produced by the pyrolysis of cashew (*Anacardium occidentale*) (pseudo fruit) bagasse and by the co-pyrolysis of sewage sludge. Cashew bagasse was collected from a nut farmer in Aracati municipality, while the sludge was obtained from a domestic sewage treatment plant in Fortaleza municipality (Upflow Anaerobic Sludge Blanket), both located in Ceará state, Brazil. The pyrolysis temperature was 500 °C, and a residence time of 190 min for cashew bagasse and 97 min for co-pyrolysis (sewage sludge), with a heating rate of 10 °C min^{-1} under moderate nitrogen flow (SPPT Technological Research Company).

The nutrient content (Ca, Mg, Al, Fe, Mn, and Zn) in the biochar were determined by inductively coupled plasma optical emission spectrometry (ICP-OES), while K and Na were measured by flame photometry where the samples were previously submitted to acid digestion following dry ash method suggested by Enders and Lehmann[57]. After acid digestion, P content was determined by the molybdovanadophosphoric acid (MAPA) colorimetric method, measuring the absorbance at 400 nm (AJX-1600 spectrophotometer, Micronal®). Total nitrogen was obtained according to Mendonça and Matos[58], applying acid digestion with sulfuric acid and following the Kjeldahl method, while carbon was determined via the Walkely-Black method. The chemical characterization of biochar is presented in Table 2.

Experiment setup

This experiment was carried out in greenhouse conditions at the Federal University of Ceará state, Fortaleza municipally, Brazil (3°44’35.51"S, 38°34’33.37"W, Fig. 1). We used a completely randomized design in a 2×5 factorial scheme: two sources of biochar (cashew and sewage sludge) and five doses (0, 5, 10, 20, and 40 Mg ha^{-1}), with four replicates, resulting in 40 experimental units. The tested doses were defined below the dose of 2% (w/w) considered limiting for biochar application in soils[59]. Polyvinyl chloride (PVC) columns (20 cm in diameter and 50 cm in height) were used, and each column was filled with degraded soil. Each biochar was incorporated and followed by a 30-day incubation period. Maize (*Zea mays* L., BRS 2022 cultivar) was the plant species cultivated in this experiment. Soil fertilization was applied to each column with urea (837 mg), simple superphosphate (4433.6 mg), and KCl (418.6 mg) before plant emergence. Additional applications of urea (837 mg) and KCl (209.3 mg) were made at 25 and 45 days after plant emergence, respectively[60].

Each column received tensiometers with mercury manometers at a depth of 0.2 m to measure the matric potential. The matric potential readings were taken twice a day (early morning and early afternoon). The values were converted into moisture using the soil-water retention curve (SWRC) specific to each treatment. Irrigation was based on the available water capacity (AWC) for each treatment, defined as the difference between soil moisture at field capacity (FC) and at the permanent wilting point (PWP) (AWC = FC – PWP). Irrigation with distilled water was initiated whenever it was determined that 30% of the AWC had been depleted, as indicated by soil moisture measurements. When required, the needed water to raise the soil moisture to FC was calculated by considering the difference between the FC and the moisture at the measurement time. The experiment concluded when the plants reached the flowering stage, 60 days after sowing, totaling 90 days.

Soil chemical and microbial activity analysis

Total C, N and P contents

All chemical and microbiological analyses were conducted after the plant harvest. Briefly, total organic carbon (TOC) was measured using the potassium dichromate digestion method in an acidic medium, followed by titration with ferrous ammonium sulfate[61]. Total nitrogen (Total N) was measured according to the method described by Mendonça and Matos[58], which involves extracting nitrogen from the soil with sulfuric acid, performing Kjeldahl distillation with sodium hydroxide, and titrating with boric acid. Soil available phosphorus was determined through the Melich-1 extractor as proposed by EMBRAPA[62].

Soil basal respiration and microbial biomass

Soil basal respiration (SBR) was assessed using the method of Anderson and Domsch[63]. Soil respiration measured the volume of CO_2 released over ten days, with readings recorded every 24 h. Soil microbial biomass C (MBC) and N (MBN) were measured by the fumigation-extraction method[64]. MBC and MBN were calculated by the difference between fumigated and non-fumigated samples, with a conversion factor of 0.33 for MBC[65] and 0.54 for MBN[66]. Microbial C and N quotients (i.e., *q*Mic-C and *q*Mic-N, respectively) were calculated using the relationship between MBC and TOC (MBC/TOC) and MBN and total N (MBN/Total N).

pH	Na+	K+	Ca^{2}+	Mg^{2}+	Al^{3}+	H + Al	C	*P*
-	$cmol_c$ kg^{-1}						g kg^{-1}	mg kg^{-1}
5.1	0.08	0.09	6.97	0.46	0.54	2.52	6.07	8.45

Table 3. Chemical properties (0–10 cm) of the highly degraded soil (Planosol) collected at Irauçuba, Ceará, Brazil.

Soil enzyme activity
The potential activities of β-glucosidase (EC 3.2.1.21), acid (E.C. 3.1.3.2), alkaline (E C 3.1.3.1) phosphatases, arylsulphatase (EC 3.1.6.1) and urease (EC 3.5.1.5), were determined using standard methods. Briefly, β-glucosidase activity was measured with ρ-nitrophenyl β-glucopyranoside as the substrate, incubated for 1 h at 37 °C, and the resulting ρ-nitrophenol was quantified spectrophotometrically at 400 nm[67]. Acid and alkaline phosphatase activity were assessed using disodium ρ-nitrophenyl phosphate as the substrate, incubated for 1 h at 37 °C, and the ρ-nitrophenol produced was measured at 420 nm[68]. Arylsulphatase was measured after the release of ρ-nitrophenol, when the soil was incubated with ρ-nitrophenyl potassium sulfate solution[69]. Urease activity was determined using urea as the substrate, incubated for 2 h at 37 °C, and the ammonium produced was measured spectrophotometrically at 660 nm[70].

Enzymatic stoichiometry
The soil enzymatic stoichiometry was determined following two distinct methodologies: (1) The ratios of enzymatic activities, including C: N (β-glucosidase / urease), C:P (β-glucosidase / acid phosphatase) and N: P (urease / acid phosphatase)[71] ; (2) A vector analysis of enzymatic stoichiometry[49,72]. Vector length and vector angle were calculated following Moorhead et al.[49].

$$\text{Vector length} = \sqrt{X^2 + Y^2} \tag{1}$$

$$\text{Vector angle} = \text{Degrees}\ [\text{ATAN2(X; Y)}] \tag{2}$$

where X is:

$$X = \frac{\ln(\beta\ - glucosidase)}{\ln(\beta\ - glucosidase) + \ln\left(acid\, phospatase\right)} \tag{3}$$

and Y is:

$$Y = \frac{\ln(\beta\ - glucosidase)}{\ln(\beta\ - glucosidase + \ln\left(urease\right)} \tag{4}$$

A longer vector length represents high C limitation (C/energy deficiency to other nutrients), and a vector angle < 45° or > 45° indicates N or P limitation, respectively.

Statistical analysis
The data obtained were subjected to Levene's test for homogeneity and the Shapiro-Wilk test for normality. Subsequently, a two-way analysis of variance (ANOVA) was performed using the F-test ($p \leq 0.05$). Means were compared using the Scott-Knott test ($p \leq 0.05$). We used R Studio software (version 1.3.1093).

Conclusions
Applying cashew and sewage sludge biochar in highly degraded soil significantly changed soil microbial biomass and activity. Thus, applying biochar in degraded soil could be a potential strategy to restore soil microbial biomass and enzymatic activity. However, the responses of extra-cellular enzymes vary according to biochar feedstock and biochar application rate, indicating complex interactions. The results of enzymatic stoichiometry and vector analysis showed an increase in P limitation to soil microbes with the application of both biochar, even sewage sludge increasing soil P contents. In contrast, both biochar reduced the limitation of C to soil microbes. These findings reinforce the potential of biochar to restore soil biological properties and increase the availability of nutrients. These features bring implications to restoration practices in degraded lands of semiarid regions. Although key soil extracellular enzymes have been analyzed, there are additional enzymes crucial for assessing the soil's potential in nutrient cycling, such as N-acetyl-β-glucosaminidase (NAG) for nitrogen. The metabolic processes in soil involve a wide range of enzymes[51], and future studies on biochar-based products should adopt a holistic approach to microbial nutrient cycling.

Data availability
The datasets generated during and/or analyzed during the current study are available from the corresponding author on reasonable request.

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Declarations

Competing interests
The authors declare no competing interests.

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