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Soil enzymatic activity in Acrisols as affected by forest-pasture conversion in northern Amazonia

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Abstract

The objective of this study was to investigate changes in soil enzymatic activities within the carbon (C), nitrogen (N), phosphorus (P), and sulfur (S) cycles following the conversion of forest to pasture. Soil samples classified as Acrisols were collected from seven pasture paddocks and one native forest area (as a reference). The experimental design employed a randomized block arrangement with eight land-use systems: five with *Brachiaria brizantha* (BB), two with *Brachiaria humidicola* (BH), and one with Native Forest (FN), evaluated at two soil depths: 0–10 cm and 10–20 cm. Soil chemical and biological variables were analyzed in conjunction with hydrolase enzyme activities associated with the C, N, P, and S cycles. Statistical analysis data were processed using R Studio (v. 4.2.2). The FN system exhibited the highest β-glucosidase and acid phosphatase activities. The BBQR system (BB on burned and mountainous areas) exhibited elevated activities of arylsulfatase, cellulase, urease, and protease. The BHQC system (BH on burned and limed areas) also showed high cellulase and invertase activities. Overall, conversion to pasture significantly altered enzymatic activities, although BBQR maintained enzymatic activities comparable to those of FN in certain nutrient cycles. These findings underscore the importance of management strategies that preserve soil biological function and quality in savanna-forest transition regions.

1. Introduction

Soils play an essential role in the ecosystem, influencing climate change, sustainability, and food production. They are the foundation of ecosystem services, promoting nutrient cycling and supporting biodiversity, as well as human survival. However, this critical resource is increasingly threatened by deforestation, pollution, and unsustainable agricultural practices (Telo da Gama, 2023).

Large-scale human intervention in these ecosystems, especially in the Amazon, has significantly altered interactions between climate and ecosystems. Pasture expansion remains the main driver of native forest loss, with 44.53 million hectares converted between 1985 and 2020 – 38.10 million hectares to pasture, and 6.06 million hectares to agriculture (Silveira et al., 2022).

Deforested areas in the Amazon are predominantly used for extensive cattle ranching, where pastures serve as the main feed source (Lange et al., 2019). Despite increasing livestock stocking rates, most pastures remain extensive and poorly managed (Garret et al., 2017). Inadequate pasture management leads to soil degradation, reduced productivity, and continued deforestation, perpetuating a cycle of environmental degradation (Silveira et al., 2022). The replacement of natural vegetation with pastures disrupts soil balance and reduces the structural stability of soil organic matter (SOM) in these converted areas (Araújo et al., 2011), which can be exacerbated by the absence of proper management practices.

Soil degradation following forest-to-pasture conversion is often reflected in changes in soil biochemical properties, such as enzymatic activity, which is a sensitive indicator of soil quality and ecosystem health (Mendes et al., 2019). Enzymatic activity represents the cumulative actions of enzymes produced by plants, macrofauna, and microorganisms, and is highly responsive to environmental fluctuations (Sobucki et al., 2021). Poor management and excessive use of inputs, whether synthetic or organic, can impair enzyme efficiency and reduce soil quality (Sobucki et al., 2021).

In soils, enzymes are proteins that catalyze biochemical reactions, accelerating processes essential for life (Cooper, 2000; Balota et al., 2013). Enzymes such as β-glucosidases, phosphatases, and urease, produced by soil microbiota, are essential for maintaining soil fertility, carbon sequestration, and overall soil quality (Daunoras et al., 2024). They facilitate the decomposition of SOM and the cycling of essential nutrients, including C, N, P, and S. Soil enzymatic activity is influenced by various factors such as soil texture, SOM content, and moisture availability (Kotzé et al., 2017).

Given the essential role of enzymes in ecosystem functioning, sustainability, and food security, understanding how agricultural activities affect these biological processes is crucial. In the Amazon, the conversion of forests to pastures has significantly impacted soil enzymatic activity – a key indicator of soil quality – reflecting management-induced changes in ecosystem functioning. Therefore, his study aimed to investigate changes in soil enzymatic activities in Acrisols following forest-to-pasture conversion within the savanna-forest transition region of the Northern Amazon.

2. Materials and Methods

The study was conducted at Fazenda Novo Paraíso, located in the municipality of Alto Alegre, Roraima, Brazil. The site is situated near Vila São Silvestre, approximately 94 km from the capital, Boa Vista, at 2°55'47" N and 61°21'46" W (Fig. 1). According to the Köppen-Geiger classification, the regional climate is Am (tropical monsoon) with a short dry season. The mean annual rainfall is about 1 762 mm, and the mean temperature is 28.2°C, with the rainy season occurring from April to September (Araújo et al., 2024).

The soil in the study area is classified as an Acrisol, according to the Brazilian Soil Classification System (Santos et al., 2025) and the World Reference Base for Soil Resources (IUSS WRB, 2022). Seven pasture paddocks and one native forest area were evaluated (Table 1). The land-use systems were designated based on vegetation, management practices, and fertilization history: FN (Native Forest); BBAR (*B. brizantha* in sandier soil); BBC (*B. brizantha* with liming); BB (*B. brizantha)*; BBQR (*B. brizantha* burned on a sloped area); BBQM (*B. brizantha* burned and mechanized); BHQC (*B. humidicola* burned and limed); BHV (*B. humidicola* near a floodplain).

Reports of burning events were due to accidental fires rather than intentional management. Grazing management in the

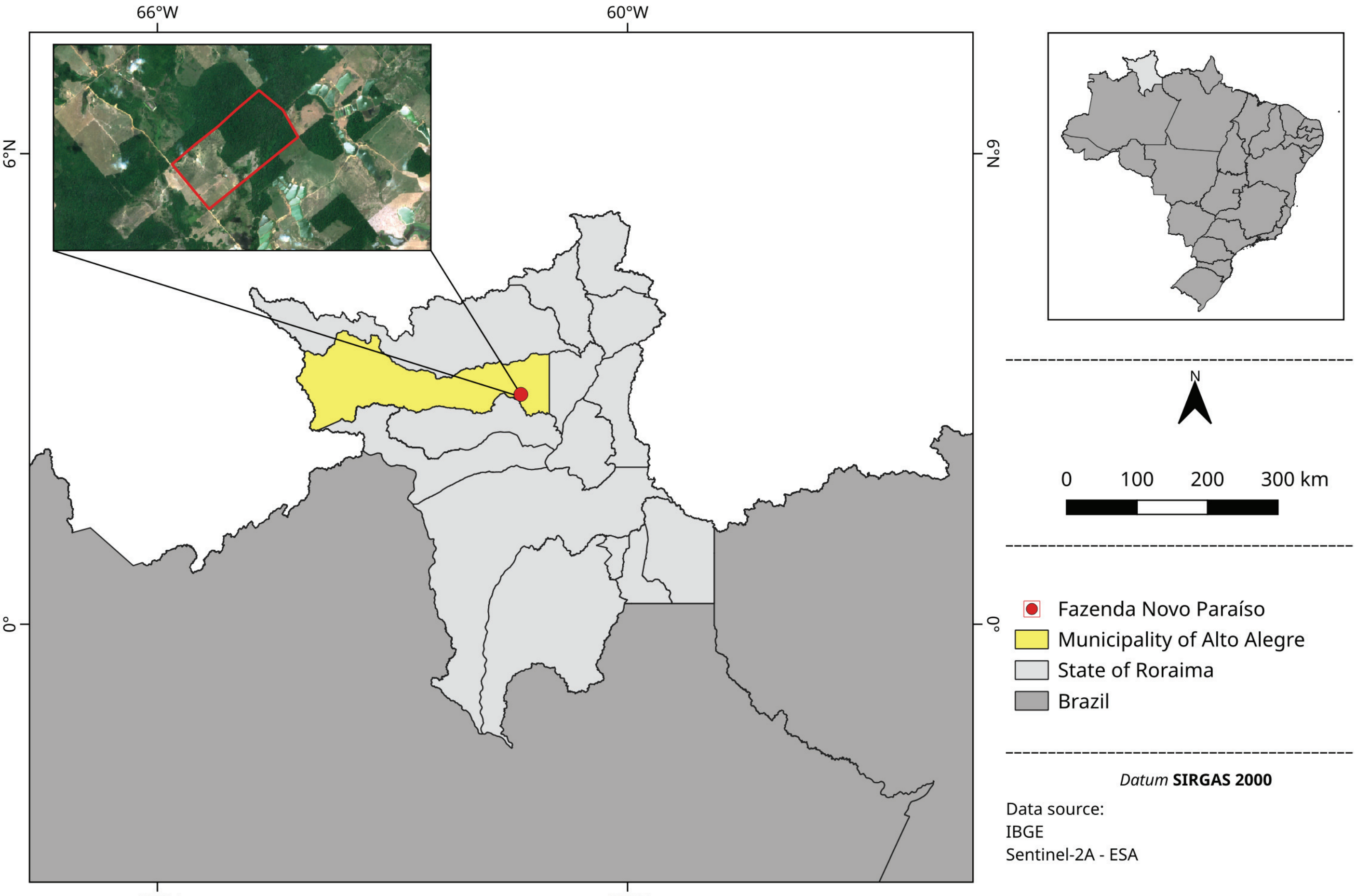

Fig. 1. Location of the study area

Table 1
Land-use history of the different management systems in Alto Alegre, Roraima, Brazil

System	Vegetation	Characteristics	Time Since Conversion to Pasture	Fertility Management
			Years	
FN	FN[1]	Indication of fire[4]	–	–
BBAR	BB[2]	Very sandy soil	25	–
BBC	BB[2]	–	25	Fertilization and liming
BB	BB[2]	–	22	–
BBQR	BB[2]	Burned[4], undulating terrain	15	–
BBQM	BB[2]	Burned[4], mechanized	14	–
BHQC	BH[3]	Burned[4]	26	Fertilization and liming
BHV	BH[3]	Near the floodplain area	16	–

Vegetation ([1]Native forest; [2]*B. brizantha*; [3]*B. humidicola*); ([4]Indication of fire/burned: report of accidental fire); Systems (FN – Native Forest; BBAR – *B. brizantha* in sandier soil; BBC – *B. brizantha* with liming; BB – *B. brizantha*; BBQR – *B. brizantha* burned in a sloped area; BBQM – *B. brizantha* burned and mechanized; BHQC – *B. humidicola* burned and limed; BHV – *B. humidicola* near the floodplain area).

paddocks involved a 30-day grazing period followed by 60 days of rest, with an average stocking rate of 1.5 animal units per ha. Mineral supplementation was applied uniformly across all paddocks, and animal manure contributed to the nutrient cycling process. Some pasture systems experienced productivity decline, prompting liming and fertilization to promote recovery.

Soil sampling took place during the dry season (February and March 2012). A representative 1-ha area was delineated within each system, subdivided into four quadrants. Within each quadrant, 12 mini-trenches (20 cm deep) were excavated, yielding samples from two soil layers: 0–10 and 10–20 cm. The experimental design employed a randomized block arrangement with four replications, utilizing a split-plot scheme. The main plots consisted of the eight land-use systems, and the subplots corresponded to soil depths.

Sampling points were selected using the free walking method along a representative diagonal within each quadrant. Each point corresponded to a 1 dm^3 simple sample, which was combined by depth to form a composite sample. The samples were air-dried for up to 72 hours, and then quartered, crushed, and sieved (< 2 mm). A 500 g subsample was retained for soil enzymatic activity determinations, and another 500 g for soil chemical analyses. Soil chemical analyses included pH (H_2O), available P, exchangeable K^+, Ca^{2+}, Mg^{2+}, Al^{3+}, H+Al, sum of bases (SB), effective cation exchange capacity (t), cation exchange capacity at pH 7.0 (T), base saturation (V), aluminum saturation (m), and SOM, according to the soil analysis methods manual (Teixeira et al., 2017).

For enzymatic activity determination, air-dried and sieved soils (< 2 mm mesh) were moistened to 60% of field capacity and incubated at 25°C in the dark for eight days, then stored at 4°C (Tabatabai, 1994). Moisture content was determined gravimetrically by oven-drying a subsample at 105°C for 24 hours. Activities of hydrolase enzymes from the C (cellulase, invertase, β-D-glucosidase), N (casein-protease, BAA-protease, urease), P (phosphomonoesterase, phosphodiesterase), and S (arylsulfatase) cycles were analyzed according to methods described by Trasar-Cepeda et al. (2000).

The data were subjected to analysis of variance (ANOVA) at a 5% probability level. When significant differences were detected, means were compared using the Scott-Knott test (< 0.05). Data normality and homogeneity were verified prior to ANOVA. Hierarchical cluster analysis was used to group systems with similar characteristics, employing the complete linkage method and Mojena's threshold (k = 1.25). All analyses were performed in RStudio (v. 4.2.2) for Mac (R Core Team, 2022).

3. Results

Table 2 presents the soil chemical properties across different land-use systems and soil depths, providing key insights into how management practices influence soil fertility and enzymatic activity. The soil in the study area is classified as an Acrisol. The analysis of chemical parameters reveals that management practices have a direct impact on soil chemical quality, with notable differences observed among land-use systems. In managed systems such as BBC and BHQC, soil pH values were higher, ranging from 4.9 to 5.5 and 5.3 to 5.8, respectively, compared to the Native Forest (FN), where pH values remained between 4.6 and 4.7. This suggests that liming and fertilization practices in these systems effectively mitigated soil acidity.

Aluminum saturation (m) exceeded 80% in both layers of the FN and BB systems, indicating high potential Al toxicity. In contrast, managed systems such as BHQC and BBC showed substantially lower aluminum saturation, reaching 0% in the 010 cm layer and 32.46% and 46.8% in the 10–20 cm layer, respectively. This reduction reflects the effectiveness of liming in neutralizing exchangeable and improving soil chemical conditions for plant growth.

Table 2
Soil chemical properties in different land-use systems and soil layers

[1]Systems	Soil depths (cm)	pH	P	K^+	Ca^{2+}	Mg^{2+}	Al^{3+}	H+Al	SB	t	T	V	m	SOM
		H_2O	$mg\ dm^{-3}$		$cmol_c\ kg^-$							%		
FN	0–10	4.68	2.2	11	0.01	0.05	1.07	6.1	0.09	1.16	6.19	1.5	92.2	2.61
	10–20	4.63	1.7	7	0	0	1.07	5	0.02	1.09	5.02	0.4	98.2	2.09
BBAR	0–10	5.38	5.7	11	0.46	0.16	0.20	3.1	0.65	0.85	3.75	17.3	23.5	1.17
	10–20	5.19	3.3	5	0.16	0	0.29	3.1	0.17	0.46	3.27	5.2	63	0.91
BBC	0–10	5.56	9.3	11	0.63	0.24	–	2.7	0.9	0.9	3.6	25	0	1.43
	10–20	4.97	5	9	0.31	0	0.29	3.2	0.33	0.62	3.53	9.3	46.8	1.04
BB	0–10	4.97	2	20	0.05	0.06	0.68	5.8	0.16	0.84	5.96	2.7	81	2.48
	10–20	5.06	1.5	9	0	0	1.17	5.2	0.02	1.19	5.22	0.4	98.3	1.83
BBQR	0–10	4.96	2.6	24	0.33	0.11	0.59	5	0.5	1.09	5.5	9.1	54.1	2.61
	10–20	4.92	1.7	18	0.2	0	0.68	4.2	0.25	0.93	4.45	5.6	73.1	1.7
BBQM	0–10	5.18	3.9	11	0.28	0.16	0.39	3.7	0.47	0.86	4.17	11.3	45.3	1.3
	10–20	4.69	2.9	26	0.09	0.02	0.49	4	0.18	0.67	4.18	4.3	73.1	1.04
BHQC	0–10	5.82	5.3	11	0.96	0.24	–	2.4	1.23	1.23	3.63	33.9	0	1.83
	10–20	5.36	4	7	0.49	0.09	0.29	3.2	0.6	0.89	3.8	15.8	32.6	1.3
BHV	0–10	5.35	3.7	46	0.4	0.23	0.29	6.9	0.75	1.04	7.65	9.8	27.9	1.7
	10–20	5.44	3.1	24	0.31	0.07	0.39	3.4	0.44	0.83	3.84	11.5	47	1.3

[1]Systems (FN – Native Forest; BBAR – *B. brizantha* in sandier soil; BBC – *B. brizantha* with liming; BB – *B. brizantha*; BBQR – *B. brizantha* burned in sloped area; BBQM – *B. brizantha* burned and mechanized; BHQC – *B. humidicola* burned and limed; BHV – *B. humidicola* near the floodplain area).

Phosphorus (P) availability also varied among the systems. The highest P concentrations were found in BHQC and BBC (5.0 to 9.3 mg dm^{-3}), while FN recorded only 2.2 mg dm^{-3}, a value considered low for tropical soils. Similarly, higher calcium (Ca^{2+}) and magnesium (Mg^{2+}), concentrations were observed in managed systems – Ca^{2+} ranging from 0.63 to 0.96 $cmol_c$ kg^{-1} and Mg^{2+} around 0.24 $cmol_c$ kg^{-1} – reflecting liming and nutrient inputs.

Base saturation (V%) further distinguishes managed and unmanaged systems. In BHQC, V% reached 33.9%, whereas in FN it remained below 10%, reinforcing the improvement in soil fertility under corrective management.

Soil organic matter (SOM) also showed significant variation among land-use systems. FN and BBQR soils showed SOM levels above 2.5%, classified as medium (1.5–3%), followed closely by BB (around 2.5%). In contrast, BBAR displayed the lowest SOM levels (1.17%). The higher SOM levels in FN and BBQR may be associated with greater litter input and topographic effects favoring SOM accumulation, whereas the moderate SOM in BB reflects limited but continuous organic replenishment under less intensive management.

3.1. Soil enzymes involved in the carbon cycle

The activity of enzymes involved in the carbon cycle exhibited significant variation across different land-use systems and soil depths (Table 3). In general, enzymatic activities decreased with depth, reflecting reduced substrate availability and microbial activity in subsurface layers. Specifically, cellulase activity was highest in the BB and BHQC systems within the 0–10 cm layer, reaching up to 0.082 µmol glucose g^{-1} h^{-1}. Interestingly, in BHQC, cellulase activity remained stable between soil layers, suggesting favorable conditions for cellulose degradation even at depth. In contrast, FN showed the lowest cellulase activity in the surface layer (0.019 µmol glucose g^{-1} h^{-1}). while in the 10–20 cm layer, FN, BBAR, BBC, and BBQR systems showed similar low values (< 0.015 µmol glucose g^{-1} h^{-1}).

Invertase activity was significantly higher in the FN system at the surface layer (0.415 µmol glucose g^{-1} h^{-1}), followed by the BHQC, BBQM, and BBAR systems, which showed no statistical differences among themselves. These systems also maintained relatively higher invertase activity in the 10–20 cm layer. In contrast, the BBC, BB, and BHV systems exhibited lower invertase activity, with values below 0.269 µmol glucose g^{-1} h^{-1} across both depths.

The β-glucosidase activity followed a similar pattern, being highest in the FN system in both soil layers (0.164 and 0.105 µmol p-nitrophenol g^{-1} h^{-1} in 0–10 cm and 10–20 cm, resepctively). The BBC and BBQR systems also exhibited relatively high β-glucosidase activity in the 0–10 cm layer, comparable to FN. Conversely, BHQC and BHV showed the lowest activities (0.058 and 0.060 µmol p-nitrophenol g^{-1} h^{-1}, respectively), indicating reduced microbial capacity to degrade in these managed systems.

Cluster analysis (Fig. 2) further shows the functional grouping of land-use systems based on the activities of the three C cycle enzymes (cellulase, invertase, and β-glucosidase). In the 0–10 cm layer (Fig. 2A), two main clusters were identified:

Table 3
Average values of hydrolytic enzyme activity involved in C cycling between the studied land-use systems and soil layers

Systems	Cellulase[a]				Invertase[a]				β-Glucosidase[b]			
	0–10 cm		10–20 cm		0–10 cm		10–20 cm		0–10 cm		10–20 cm	
FN	0.019	dA	0.010	cA	0.415	aA	0.300	aB	0.164	aA	0.105	aB
BBAR	0.060	bA	0.015	cB	0.201	cA	0.251	aA	0.118	bA	0.054	cB
BBC	0.043	cA	0.005	cB	0.223	cA	0.112	bB	0.149	aA	0.081	bB
BB	0.069	aA	0.037	bB	0.269	cA	0.131	bB	0.091	cA	0.033	cB
BBQR	0.051	bA	0.005	cB	0.319	bA	0.200	bB	0.150	aA	0.063	cB
BBQM	0.061	bA	0.020	bB	0.306	bA	0.260	aA	0.094	cA	0.049	cB
BHQC	0.082	aA	0.071	aA	0.349	bA	0.333	aA	0.058	dA	0.049	cA
BHV	0.040	cA	0.025	bA	0.266	cA	0.143	bB	0.060	dA	0.039	cA
CV%	28.88				23.16				18.29			

[a] μmol glucose g^{-1}h^{-1}; [b] μmol *p*-nitrophenol g^{-1}h^{-1}; Systems (FN – Native Forest; BBAR – *B. brizantha* in sandier soil; BBC – *B. brizantha* with liming; BB – *B. brizantha*; BBQR – *B. brizantha* burned in sloped area; BBQM – *B. brizantha* burned and mechanized; BHQC – *B. humidicola* burned and limed; BHV – *B. humidicola* near the floodplain area). Lowercase letters indicate that values for the same depth across different land-use systems were not significantly different; uppercase letters indicate that, within each land-use system, values between the two soil layers were not significantly different ($p \leq 0.05$).

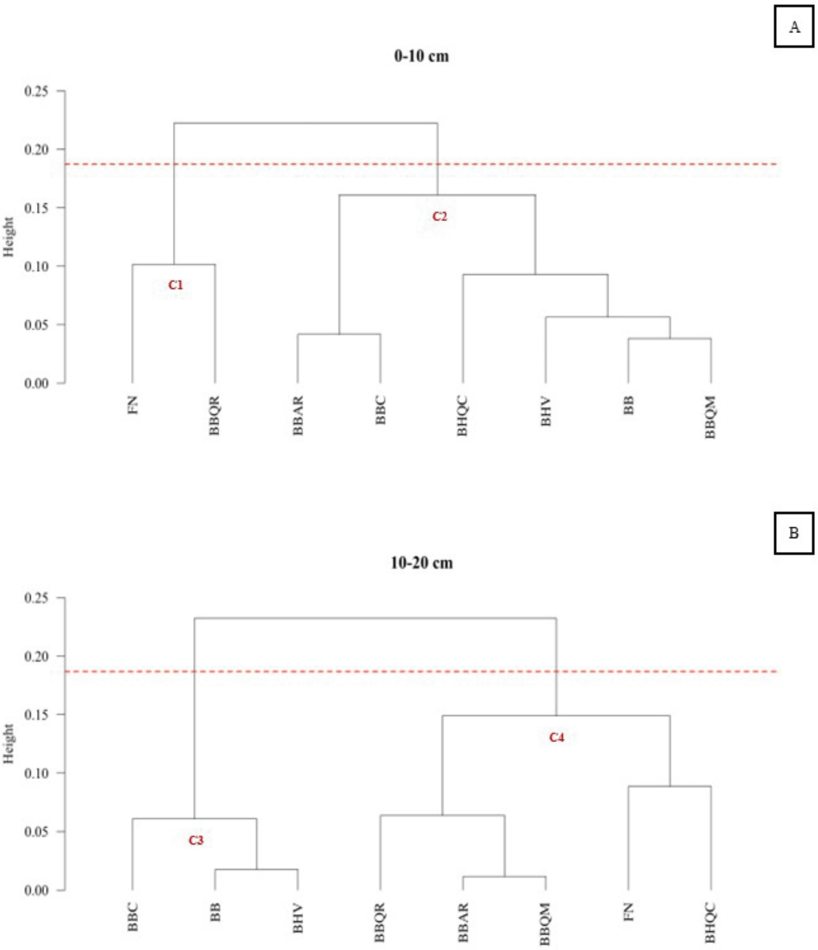

Fig. 2. Cluster analysis of land-use systems based on enzyme activity involved with C cycling in the 0–10 cm (A) and 10–20 cm (B) soil layers

Cluster C1 comprised FN and BBQR, characterized by high invertase (0.367 μmol glucose g^{-1} h^{-1}) and β-glucosidase (0.157 μmol p-nitrophenol g^{-1} h^{-1}), activities. Cluster C2 grouped BBAR, BBC, BB, BBQM, BHQC, and BHV, showing lower averaged enzymatic activities (0.269 and 0.095 μmol p-nitrophenol g^{-1} h^{-1} for invertase and β-glucosidase, respectively).

In the 10–20 cm layer (Fig. 2B), two additional clusters (C3 and C4) were identified. Cluster C3 (BBC, BB, and BHV) showed moderate enzyme activities (0.129 μmol glucose g^{-1} h^{-1} for invertase and 0.051 μmol p-nitrophenol g^{-1} h^{-1} for β-glucosidase, respectively). In contrast, Cluster C4 (BBQR, BBAR, BBQM, FN, and BHQC) exhibited higher enzyme activities (0.270 μmol glucose g^{-1} h^{-1} for invertase and 0.061 μmol p-nitrophenol g^{-1} h^{-1} for β-glucosidase, respectively), suggesting enhanced organic matter decomposition and nutrient availability in these systems.

The distinct behaviors of FN and BBQR systems in cluster C1 is associated with their acidic pH (ranging from 4.6 to 4.9) and high SOM content (2.61%), which supported microbial and enzymatic activities despite soil acidity. Conversely, systems in cluster C2 showed lower SOM levels and greater variability in chemical characteristics, explaining the reduced enzyme activity. In cluster C3, moderate activity levels can be attributed to the absence of aluminum saturation in BBC and moderate SOM content in BB (1.83%). However, the acidic pH and high Al saturation in BB likely limited efficient organic matter decomposition.

3.2. Soil enzymes involved in the nitrogen cycle

The activity of soil enzymes involved in the nitrogen cycle (urease, BAA-protease, and casein-protease) varied significantly among land-use systems and soil depths (Table 4). In general, urease activity was higher in the 0–10 cm layer compared to the 10–20 cm layer, consistent with greater organic substrate and microbial activity near the surface. The BB system showed the highest urease activity among all systems, reaching 8.328 μmol NH_3 g^{-1} h^{-1} in the surface layer and decreasing to 4.254 μmol NH_3 g^{-1} h^{-1} in the 10–20 cm layer. Similarly, the BBQR system showed elevated urease activity in the surface layer (5.828 μmol NH_3 g^{-1} h^{-1}), with a marked decline in the deeper layer (2.611 μmol NH_3 g^{-1} h^{-1}). Other systems, such as FN, BBAR, BBC, BHQC, and BHV, showed comparatively lower urease activities, ranging from 2.097 to 3.549 μmol NH_3 g^{-1} h^{-1} across depths

Unlike urease, proteolytic enzymes (BAA-protease and casein-protease) showed no significant variations between soil layers; thus, their activities were analyzed as depth averages (Table 4). BAA-protease activity was highest in FN (2.082 μmol tyrosine g^{-1} h^{-1}) and BBQR systems (5.438 μmol tyrosine g^{-1} h^{-1}), reflecting strong protein mineralization potential in these systems. The BHQC system exhibited the lowest average BAA-protease activity (0.885 μmol tyrosine g^{-1} h^{-1}), while BB, BBQM, and BHV showed intermediate values. Casein-protease activity followed a similar pattern, with BBQR and BHV displaying the highest mean activities (0.302 and 0.306 μmol tyrosine g^{-1} h^{-1}, respectively), and BB, BBAR, and BBC showing the lowest mean activities.

The elevated urease activity in BB in both soil layers can be explained by the lack of intensive management in this system, coupled with the high SOM content (2.48%) and moderate total cation exchange capacity (5.96 $cmol_c$ kg^{-1}). In FN, although SOM and CEC values were even higher, the occurrence of fire events likely disrupted microbial dynamics, reducing enzymatic activity.

Cluster analysis (Fig. 3) grouped the land-use systems according to their N cycle enzyme activities (urease, BAA-protease, and casein-protease). In the 0–10 cm layer (Fig. 3A), two distinct clusters emerged: Cluster N1, included FN, BHV, BBAR, BBC, and BHQC, characterized by moderate to high urease activity and balanced protease activity. Cluster N2 comprised BB, BBQR, and BBQM, with very high urease activity – particularly in BB and BBQR – and high BAA-protease activity, indicating more active N mineralization processes.

In the 10–20 cm layer (Fig. 3B), cluster N3 exclusively consisted of BBQR, which maintained high BAA-protease and urease activities, while group even at depth. The unique microtopography of this system likely created microenvironments conducive to enzyme stability and function. Cluster N4 grouped the remaining systems such (BB, FN, BBQM, and others) and exhibited

Table 4
Average values of hydrolytic enzyme activity in the N cycle across the land-use systems and soil layers

Systems	Urease[a]		BAA-protease[a]	Casein-protease[b]
	0–10 cm	10–20 cm	Mean [/1]	Mean [/1]
FN	3.549 dA	2.097 bB	2.082 cA	0.227 cA
BBAR	2.416 eA	1.154 bB	2.023 cA	0.155 dA
BBC	1.722 eA	1.656 bA	0.885 dA	0.168 dA
BB	8.328 aA	4.254 aB	1.658 dA	0.153 dA
BBQR	5.828 bA	2.611 bB	5.438 aA	0.302 aA
BBQM	4.724 cA	2.429 bB	2.926 bA	0.206 cA
BHQC	2.237 eA	2.148 bA	0.885 dA	0.248 bA
BHV	3.322 dA	1.736 bB	1.029 dA	0.306 aA
CV%	20.86		22.82	23.20

[a] μmol NH_3 g^{-1} h^{-1}; [b] μmol tyrosine g^{-1} h^{-1}; [/1] Average enzyme activity between soil layers (0–10 and 10–20 cm), statistically equal. Systems: (FN – Native Forest; BBAR – *B. brizantha* in sandier soil; BBC – *B. brizantha* with liming; BB – *B. brizantha*; BBQR – *B. brizantha* burned in sloped area; BBQM – *B. brizantha* burned and mechanized; BHQC – *B. humidicola* burned and limed; BHV – *B. humidicola* near the floodplain area). Similar lowercase letters indicate that values for the same depth across different land-use systems were not significantly different; similar uppercase letters indicate that, within each land-use system, values between the two soil layers were not significantly different (p ≤0.05).

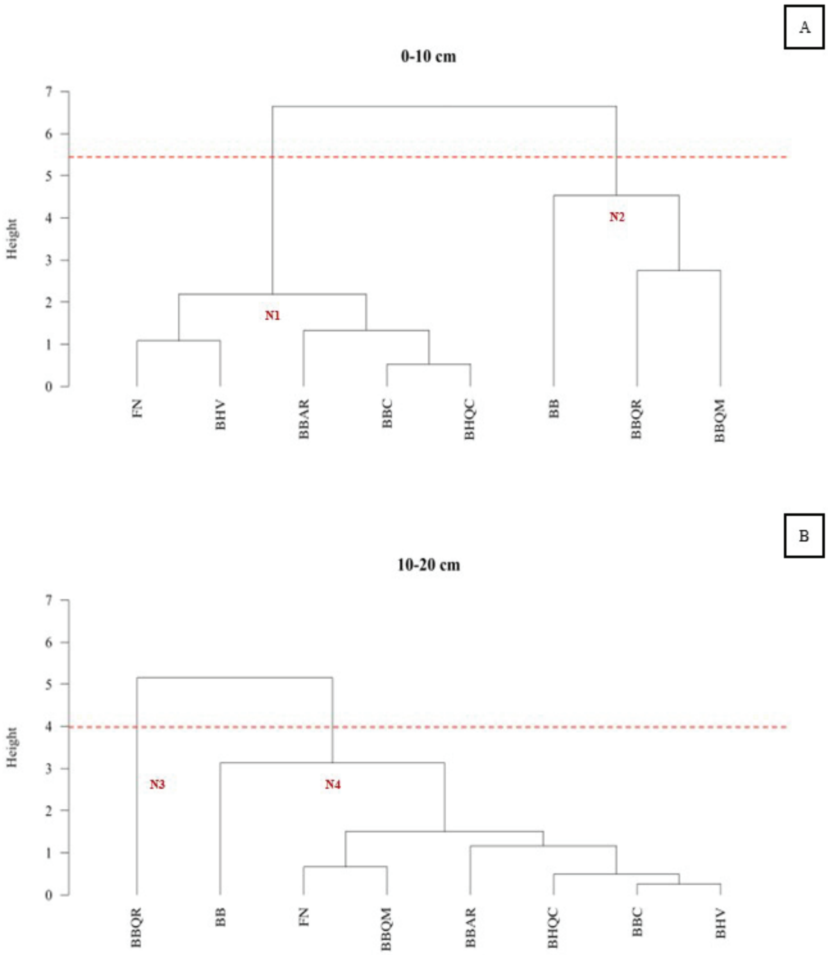

Fig. 3. Cluster analysis of land-use systems based on enzyme activities involved with the N cycle in the 0–10 cm (A) and 10–20 cm (B) soil layers

moderate and more stable enzymatic activity, reflecting a balance between soil management, chemical conditions, and microbial processes.

Overall, N cycle enzyme dynamics reflected both management intensity and SOM status. Systems with minimal disturbance (e.g., BB and BBQR) or natural vegetation (FN) supported greater enzymatic potential for N mineralization, though fire and acidity may constrain activity in some cases.

3.3. Soil enzymes involved in the phosphorus and sulfur cycles

The activities of enzymes associated with the phosphorus (P) and sulfur (S) cycles varied among land-use systems, reflecting diferences in soil fertility and management (Table 5). For phosphatases (phosphomonoesterase and phosphodiesterase), no significant differences were detected between soil depths, suggesting a relatively uniform distribution of these enzymes within the soil profile.

The highest phosphomonoesterase activity occurred in the FN system (2.098 µmol p-nitrophenol g^{-1} h^{-1}), while the lowest activities were observed in BBAR (0.477 µmol p-nitrophenol g^{-1} h^{-1}) and BBC (0.406 µmol p-nitrophenol g^{-1} h^{-1}). Similarly, phosphodiesterase activity followed the same trend, with the FN system again showing the highest mean activity (0.179 µmol p-nitrophenol g^{-1} h^{-1}), and the BHQC system the lowest (0.032 µmol p-nitrophenol g^{-1} h^{-1}). These findings indicate that natural forest conditions support greater potential for organic P mineralization than managed or disturbed systems.

In contrast, arylsulfatase activity, which is involved in the S cycle, varied significantly with soil depth. Enzyme activity was consistently higher in the surface layer (0–10 cm) compared with the subsurface layer (10–20 cm), consistent with greater microbial activity and SOM content near the surface. In the FN system, for instance, arylsulfatase activity reached 0.063 µmol p-nitrophenol g^{-1} h^{-1} at the surface and declined to 0.037 µmol p-nitrophenol g^{-1} h^{-1} at depth, indicating a strong surface stratification pattern typical of undisturbed forest soils.

Cluster analysis (Fig. 4) grouped the land-use systems according to their P (phosphomonoesterase and phosphodiesterase) and S (arylsulfatase) cycle enzymes activities at both depths. In the 0–10 cm (Fig. 4A), cluster PS1 ws composed solely of the FN system, characterized by the highest phosphomonoesterase activity (2.098 µmol p-nitrophenol g^{-1} h^{-1}) and phosphodiesterase (0.179 µmol p-nitrophenol g^{-1} h^{-1}) activities. Cluster PS2, included the remaining systems (BB, BBQR, BHV, BBAR, BBC, BBQM, and BHQC), displaying lower enzymatic activities overall, with phosphomonoesterase values ranging from 0.406 to 1.135 µmol p-nitrophenol g^{-1} h^{-1} and phosphodiesterase values from 0.032 to 0.157 µmol p-nitrophenol g^{-1} h^{-1}.

Table 5
Average values of hydrolytic enzyme activity in the P and S cycles across the land-use systems and soil layers

Systems	Phosphomonoesterase[a]		Phosphodiesterase[a]		Arylsulfatase[a]			
	Mean[/1]		Mean[/1]		0–0.10 m		0.10–0.20 m	
FN	2.098	aA	0.179	aA	0.063	bA	0.037	bB
BBAR	0.477	eA	0.068	dA	0.019	dA	0.011	cA
BBC	0.406	eA	0.061	dA	0.037	cA	0.032	bA
BB	1.135	bA	0.157	bA	0.021	dA	0.010	cA
BBQR	0.928	cA	0.057	dA	0.094	aA	0.057	aB
BBQM	0.712	dA	0.106	cA	0.030	cA	0.031	bA
BHQC	0.623	dA	0.032	eA	0.018	dA	0.019	cA
BHV	0.968	cA	0.066	dA	0.073	bA	0.027	bB
CV%	14.26		21.77		22.42			

[a] µmol *p*-nitrophenol g^{-1} h^{-1}; [/1] Average enzyme activity between soil layers (0–10 and 10–20 cm), statistically equal. Systems (FN – Native Forest; BBAR – *B. brizantha* in sandier soil; BBC – *B. brizantha* with liming; BB – *B. brizantha*; BBQR – *B. brizantha* burned in sloped area; BBQM – *B. brizantha* burned and mechanized; BHQC – *B. humidicola* burned and limed; BHV – *B. humidicola* near the floodplain area). Lowercase letters indicate that values for the same depth across land-use systems were not significantly different; uppercase letters indicate that, within each land-use system, values between the two soil layers were not significantly different ($p \leq 0.05$).

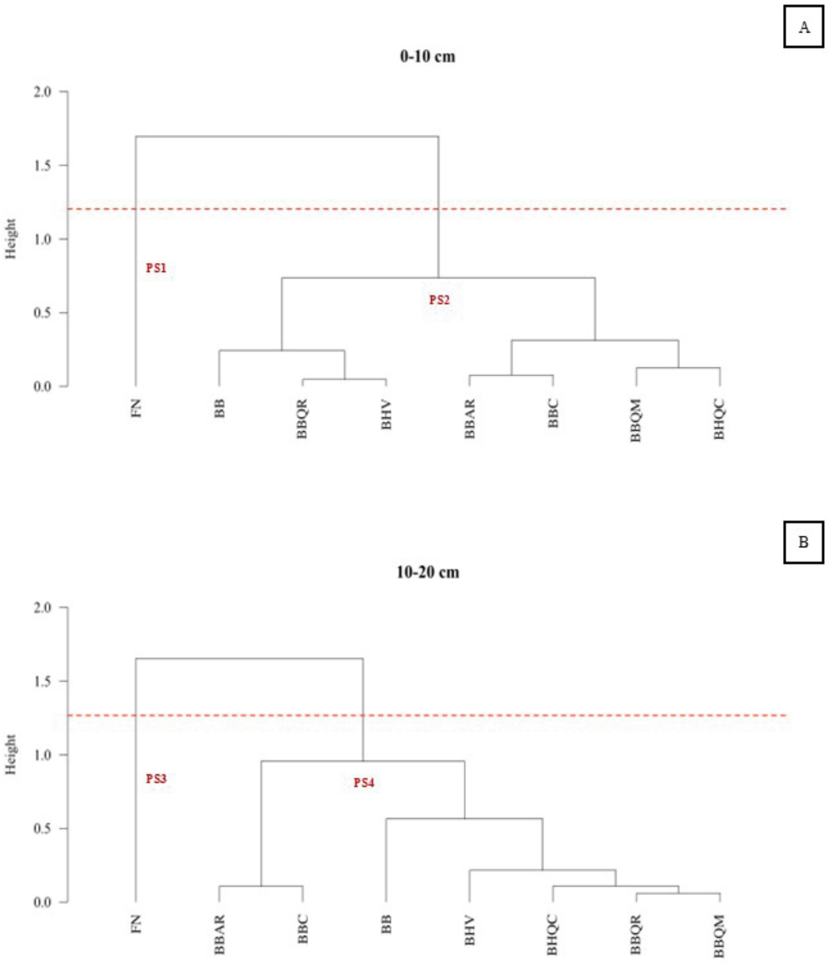

Fig. 4. Cluster analysis of land-use systems based on enzyme activity involved with P and S cycling in the 0–10 cm (A) and 10–20 cm (B) soil layers

A similar pattern was observed in the 10–20 cm layer (Fig. 4B), Where cluster PS3 (FN) remained distinct due to its high enzymatic activity, while cluster PS4 grouped the other systems with lower activities.

The FN system's, distinct behavior is closely associated with its acidic pH (average of 4.6), low P availability (2.2 to 1.7 mg dm^{-3}), and high SOM content (2.61 and 2.09%). These conditions favor phosphatase synthesis as a biochemical response to P limitations, with SOM providing both substrate and habitat for microorganisms producing these enzymes. In contrast, managed systems in cluster PS2 displayed greater variability in chemical properties, partly due to liming and fertilization, which can suppress phosphatase activity by alleviating P limitation or altering microbial community composition.

Overall, the results indicate that phosphorus and sulfur related enzymatic activities are strongly influenced by land-use and management intensity. Undisturbed forest soils (FN) maintain the highest biological potential for P and S cycling, while managed systems exhibit lower and more variable activities, reflecting the combined effects of soil chemistry, disturbance, and nutrient inputs.

4. Discussion

The results for soil chemical properties align with the general characteristics of Acrisols, as described by Feitosa et al. (2016) in a study on soil-landscape relationships in forest islands. These soils are typically oligotrophic, exhibiting low natural fertility. Indicators of soil acidity, such as pH (H_2O), sum of bases (SB), and base saturation (V%), generally ranged from medium to low, while fertility parameters, including available P, exchangeable K, and SOM, also presented low values.

The relatively higher pH observed in managed systems such as BBC and BHQC suggests the application of soil acidity correctives, particularly lime. According to Sobral et al. (2015), liming reduces aluminum toxicity and creates more favorable conditions for root growth and nutrient uptake.

Conversely, the high aluminum saturation observed in FN and BB systems indicates levels that restrict root growth and nutrient uptake. Values above 50% are considered toxic for most crops, reflecting strong soil acidity and the need for management interventions to improve soil fertility.

The elevated P levels found in managed systems such as BHQC and BBC, as reported by Sobral et al. (2015), reflect the use of phosphate fertilization, which contrasts with the low P levels recorded in the Native Forest (FN) system. Similarly, the increased calcium (Ca^{2+}) and magnesium (Mg^{2+}) levels in managed systems such as BBC and BHQC result from liming practices that enhance base cation availability. Although these levels are still considered moderate, they are sufficient to meet the basic requirements of pasture species and contribute to improved base saturation (V%) and the cation exchange capacity (CEC) (Sobral et al., 2015).

Base saturation values above 50% are generally desirable, as they indicate a higher proportion of exchange sites occupied by essential nutrients such as Ca^{2+}, Mg^{2+}, and K^{+}, thereby supporting greater soil fertility and plant productivity. However, systems with low SOM content, such as BBAR, may exhibit limited capacity to retain these nutrients. Low SOM levels are often linked to management intensity and ground cover quality, as well-managed pastures tend to conserve higher levels of SOM through continuous litter input and root turnover (Sobral et al., 2015; Santos et al., 2009).

Overall, these results emphasize that management practices such as liming and fertilization can partially mitigate the chemical limitations inherent to Acrisols, though maintaining adequate SOM remains critical for sustaining soil fertility and resilience in pasture systems.

4.1. Soil enzymes involved in the carbon cycle

The higher cellulase activity observed in the BHQC system can be attributed to its elevated pH (5.8), low levels of exchangeable acidity, and adequate amounts of Ca^{2+} and Mg^{2+}, highlighting the positive effect of liming on enzymatic activity. In contrast, systems with a pH below 5.5 may experience partial cellulase denaturation, which reduces enzyme stability and activity (Gow and Wood, 1988). Although FN has high SOM levels – ranging from 2.6% to 2.1% in the 0–10 and 10–20 cm layers, respectively – the recalcitrant composition of this SOM, rich in lignin and phenolic compounds, hinders degradation and consequently reduces cellulase activity (Tadini et al., 2018).

The history of burning in the BHQC system may also have contributed to higher enzymatic activity, as ash addition promotes greater substrate availability (Wang et al., 2023). Vieira et al. (2021) reported that actinobacteria isolated from post-burn soils exhibit increased cellulolytic activity, suggesting microbial adaptation to stress conditions. In the BBQR system, despite the occurrence of burning, the lower pH (4.9) and the presence of exchangeable H^{+} and Al^{3+} limited the positive effects of ash, resulting in lower cellulase activity compared to BHQC.

Invertase activity was highest in FN, likely due to its higher SOM content (2.61%) and a pH favorable to enzymatic action (Frankenberger and Johanson, 1983). Similar (Uchôa et al., 2024) reported up to an 83% decrease in invertase activity in pasture soils compared to native forests, associated with reduced sucrose availability and a decline in easily degradable SOM. Xiang et al. (2018) further showed that litter decomposition can increase invertase activity by up to 33.2%, reinforcing the importance of SOM as a substrate source for this enzyme.

β-glucosidase, essential for cellulose decomposition, also exhibited higher activity in FN, supported by the abundant SOM and the presence of β-glycosides. Silva-Olaya et al. (2021) reported that forest-to-pasture conversion can reduce β-glucosidase activity by approximately 20%, attributed to lower substrate availability. Native vegetation supports higher enzyme activity by providing protection against enzyme degradation (Balota et al., 2013).

The effect of fire in systems like FN and BBQR is complex. Although burning can increase soil pH and macronutrient levels (Salomão and Hirle 2019), it often suppresses β-glucosidase activity. Evangelista et al. (2012), as observed in the BBQM and

BHQC systems of cluster C2. However, the higher invertase and β-glucosidase activities recorded in cluster C4 appeared to be linked to animal manure deposition, which supplies additional C and essential nutrients that stimulate microbial activity (Antonious et al., 2020). This pattern aligns with the results from systems such as BBQR, BBAR, BBQM, and BHQC, where manure inputs likely enhanced substrate availability.

Conversely, in cluster C3, despite high SOM availability in BB, enzymatic activity was partially inhibited by aluminum saturation. Although aluminum was absent, the acidic pH limited enzyme efficiency, indicating that both pH and aluminum toxicity jointly constrain enzyme performance across systems.

4.2. Soil enzymes involved in the nitrogen cycle

The higher urease activity observed in the BB system aligns with studies, such as those by Barros et al. (2018), which have demonstrated that management practices involving soil cover with spontaneous vegetation or pastures can enhance enzymatic activity and overall soil quality. The elevated urease activity in the BB system can be attributed to the presence of natural vegetation cover and the absence of intensive management, which preserves soil structure and promotes microbial biomass, especially in the surface layer, where urease activity is typically more pronounced. Similarly, Costa et al. (2024) reported that no-tillage systems exhibit higher urease activity due to the presence of surface straw and greater organic carbon inputs, reinforcing the role of vegetation cover in sustaining this enzymatic process.

In contrast, Uchôa et al. (2024) reported lower urease activity in *Brachiaria* pastures compared to native forests, primarily due to reduced SOM and nutrient availability. This observation aligns with the reduced urease activity observed in several pasture systems of this study, indicating that the quality of soil cover and management intensity significantly impact enzymatic performance. According to Dias-Filho and Lopes (2010), appropriate pasture management practices – especially those that maintain SOM – are key to optimizing nutrient cycling and sustaining soil functionality.

Protease activity (BAA-protease and casein-protease) also varied among systems. Trasar-Cepeda et al. (2008) found that these enzymes tend to show higher activity in agricultural soils, where SOM and pH conditions create an environment favorable for enzyme expression. For instance, in the BHV system, for instance, chemical characteristics such as a pH of 5.4 and a Ca^{2+} content of 0.36 $cmol_c\ kg^{-1}$ create a balanced environment conducive to casein-protease activity. These findings suggest that chemically balanced soils facilitate more efficient enzymatic activity, a phenomenon also observed in the BBQR system.

Greenfield et al. (2020) suggested that cultivation can intensify nutrient decomposition and cycling, providing conditions favorable for proteases. Torres et al. (2020) also observed that urease is more active in the surface layers of soils under dense vegetation or fallow, due to organic matter accumulation – corroborating the results obtained in systems with greater vegetation cover in this study.

The high urease activities in BB, BBQR, and BBQM systems appear to be associated with elevated SOM levels and moderately acidic pH. Although such pH conditions are not optimal for urease, the high SOM likely compensates by providing substrates that sustain enzyme activity. Cartes et al. (2009) demonstrated that SOM content and temperature are primary drivers of urease activity, which may explain the observed efficiency in these systems.

Finally, Sieradzki et al. (2023) and Alvear et al. (2006) emphasized the importance of proteases for N availability. In the BBQR system (cluster N3), the high protease activity observed indicates a strong capacity for degrading organic N compounds. This effect is reinforced by the presence of roots and debris, which increase SOM availability and stimulate microbial biomass growth – creating an environment highly conducive to protease activity.

4.3. Soil enzymes involved in the phosphorus and sulfur cycles

The analysis of phosphatase activity (phosphomonoesterase and phosphodiesterase) revealed significant differences among the evaluated land-use systems. In previous studies by Uchôa et al. (2024), higher activities of these enzymes were also reported in native forests compared to pastures, indicating a greater potential for organic P mineralization in forest soils, largely due to the higher SOM content (Conte et al., 2002). The contrasting enzymatic responses between forest and pasture systems suggest that management practices such as liming and fertilization alter SOM composition and availability, thereby influencing phosphatase dynamics. Barros et al. (2018) further noted that, while managed pastures can maintain considerable enzymatic activity, elevated inorganic P levels – such as those observed in the BBC system – may suppress phosphatase production, since excess P reduces the need for enzymatic mineralization.

In the case of arylsulfatase, associated with the S cycle, activity was generally higher in pasture systems such as BBQR and BHV, especially in the 0–10 cm soil layer. Mendes et al. (2021) observed that introducing *Brachiaria* into cropping systems significantly enhanced arylsulfatase activity compared to monocultures, underscoring the role of *Brachiaria* in promoting a favorable microenvironment for soil microorganisms. Similarly, Chaves et al. (2024) reported increased arylsulfatase activity in intercropped systems such as soybean *Brachiaria*, linking this effect to higher SOM levels and improved conditions for microbial proliferation provided by *Brachiaria*. Its extensive root system and continuous exudate release help maintain soil temperature and moisture, supporting both microbial resilience and enzymatic function.

Despite the high SOM content in the FN system, its strong soil acidity (pH 4.6) may have restricted arylsulfatase activity. Chen et al. (2019) and Tabatabai and Bremner (1970) reported that arylsulfatase performs optimally near neutral pH (6.2). Furthermore, Chaves et al. (2024) noted that high S concentrations could also inhibit this enzyme's activity. Supporting this, Kunito et al. (2022) found that arylsulfatase tends to be more active in forest soils with low sulfate availability, whereas in arable soils with higher sulfate levels, activity declines due to substrate inhibition effects.

Acid phosphatase activity was strongly influenced by both pH and SOM. Fernandes et al. (1998) identified a positive relationship between these factors and enzymatic performance, noting that acidic soils, such as those in the FN system (with an average pH of 4.6), favor acid phosphatase catalysis. The high SOM content in such soils also provides abundant organic P, stimulating enzyme production to support P mineralization. Conversely, management practices that modify pH and reduce SOM – such as intensive liming and fertilization – may compromise phosphatase activity, especially in soils with high inorganic P levels, where plants decrease enzyme synthesis due to nutrient sufficiency (Barros et al., 2018).

The structural stability and high SOM content of native forest soils sustain a more diverse bacterial community, which plays an essential role in nutrient cycling (Melo et al., 2021). In contrast, Barros et al. (2018) observed that intensive and long-term management interventions – such as recurrent burning and mechanization – alter soil properties like pH and CEC, thereby reducing enzyme activity, including that of acid phosphatase. These alterations ultimately decrease microbial biomass and diversity, limiting the enzymatic processes essential for maintaining nutrient availability and soil fertility.

5. Conclusions

Pasture systems, especially BHQC, presented elevated cellulase and invertase activities, indicating enhanced decomposition of SOM and more active nutrient cycling. In contrast, β-glucosidase activity was highest in the native forest (FN), suggesting that maintaining natural soil conditions supports greater enzymatic functionality and C turnover.

Multivariate analyses showed that the FN and BBQR systems formed distinct clusters due to their similarly high invertase and β-glucosidase activities. This finding suggests that, despite land-use differs, specific management practices – such as maintaining organic residue cover or balanced soil conditions – can sustain enzymatic activity levels comparable to those in undisturbed forest soils.

Among the N cycle enzymes, urease exhibited he highest activity in the BB system, reflecting the positive influence of natural vegetation cover and the absence of management on SOM preservation and N cycling. BAA-protease and casein-protease showed increased activities in systems affected by burning, indicating that such interventions significantly modify N dynamics. The clustering of BB and BBQR systems, both characterized by elevated urease and protease activities, suggests that long-term management practices and soil chemical properties jointly enhance N availability and enzymatic responses.

Phosphatase activity was greatest in the native forest, associated with high SOM content and low available P, demonstrating the forest soil's inherent ability to mineralize organic P. Conversely, arylsulfatase activity was more pronounced in pasture systems such as BBQR, suggesting that *Brachiaria* management fosters favorable conditions for S cycling through root exudation and microbial stimulation.

Overall, the native forest system (FN) formed a distinct group due to its consistently high enzymatic activity, particularly for phosphatase, reinforcing the ecological importance of conserving natural vegetation. These results highlight that while certain management practices can partially restore enzymatic functionality, the preservation of native forest remains crucial for maintaining the full spectrum of soil biochemical processes essential to ecosystem sustainability.

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

Ethical Approval

Ethical approval was not required for this research.

Data Availability Statement

The data will be made available upon reasonable request.

Conflict of interest

The authors declare the absence of conflicts of interest that could have influenced the development or results of this work. This study was conducted without the involvement of human subjects or animals.

Author Contributions

Ingridy Tavares – Conceptualization, Data curation, Writing – original draft, Writing – review & editing. **Sandra Uchoa** – Funding acquisition, Investigation, Methodology, Supervision, Validation. **José Alves** – Funding acquisition, Investigation, Methodology. **Carlos Matos** – Methodology, Supervision, Validation. **Fernando Gil Sotres** – Conceptualization, Methodology, Supervision. **Elmarie Kotze** – Writing – review & editing. All authors read and approved the final manuscript.

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