RESEARCH ARTICLE



Management systems in the subtropical region of Brazil: aggregation in the assessment of the chemical quality of fragile soils

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(Received 08 August 2024; revised 02 April 2025; accepted 04 April 2025)

Abstract

Soil health is a term used to describe the general state or soil quality in an agroecosystem. The study of aggregate formation pathways has been successfully used to assess soil quality, especially chemistry, particularly in measuring the impact of different forms of use and management on soil health. This study aimed to (i) verify the contribution of biogenic (Bio) and physicogenic (Phy) aggregates to soil fertility; (ii) evaluate the total carbon (TC), nitrogen (TN), phosphorus (TP), and potassium contents and their respective stoichiometric ratios in these aggregates; and (iii) analyse the relationship between the P fractions (labile, moderately labile, and non-labile) and these aggregates. Three management systems were evaluated (permanent pasture, PP; no-tillage system, NT; and no-tillage + Urochloa system, NT+B) as well as a reference area (Atlantic Forest biome vegetation, NF). All the sample areas are located on soils with a sandy texture in the surface horizons. Aggregates were separated, identified, and classified according to their genesis into Bio (biotic factors) and Phy (abiotic factors). Only the PP system had significant increase in the values of TC, TN, TP, TK, and organic and inorganic P. The NT+B system favoured a proportional increase in TC content compared to the aggregates of the NF and NT areas, especially in the subsurface layer (ranging from 31 to 44%). For Bio aggregates, there were increments in TC and TN contents compared to Phy ones, especially in the NT and NT+B systems (8 to 30% for TC and 56 to 239% for TN). Bio aggregates also had the lowest values of C/N ratio in the surface layer (< 30), highest values of C/P ratio in the subsurface layer (> 33), and greater participation of the organic form of P in TP in the surface layer (between 26 and 42%). The chemical attributes in the aggregates were affected differently by the soil management systems, especially PP and NT+B systems. The results verified for Bio aggregates strengthen the hypothesis that these structural units are important sources of nutrients for the soil and reiterate the importance of studying the formation pathways in assessment soil health.

Keywords: Soil health; aggregate origin; nutrient dynamics

Introduction

Efficient use of nutrients in agriculture is vital to the sustainability of production areas (grains, fibres, energy materials, sugarcane, forestry, and pastures) and is closely linked to nutrient cycling. It can be optimized through the adoption of management practices or systems that aim to improve soil fertility, maintenance of biodiversity, increased carbon (C) sequestration in the soil, and

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reduction of negative climate effects (Baptistella *et al.*, 2020). In this context, the nutrient cycling process is favoured in production systems based on the Conservation Agriculture model. This model aims to increase the sustainable development of agriculture in socio-economic aspects, generate competitiveness for agribusiness, ensure food safety and quality, and preserve the environment and land quality (Kassam *et al.*, 2009, 2019; Denardin *et al.*, 2012; Pinto *et al.*, 2021a).

In agricultural areas in regions with tropical and subtropical climates, in Brazil, there is an accelerated cycling of nutrients. In the western region of Paraná (Brazil), most soils in the production areas have low clay content (≤ 200 g kg⁻¹) and high sand contents (Franchini *et al.*, 2016), considered more fragile in terms of texture (Donagemma *et al.*, 2016). In most of these sandy areas, the climate is classified as humid subtropical (Cfa), with hot summers (Alvares *et al.*, 2013), which causes high evapotranspiration. This combination of texture and climatic condition intensifies the transformations of soil organic matter (SOM), reduces the capacity for water and nutrient retention and storage in the soil, reduces biological activity, and increases susceptibility to erosion. In this scenario, plant diversification in the production environment (intercropping and crop rotation) associated with conservation soil management practices or systems (perennial pastures, no-tillage system, crop-livestock or crop-livestock-forest integrations) can mitigate these problems, in addition to reducing production costs without altering yield and mitigating the effects of greenhouse gas emissions (Cerri *et al.*, 2023).

Aggregation is part of the 'potential' of natural and anthropic systems as defined by Holling (2001), a capital that varies along adaptive cycles and that determines ecosystem and agroecosystem evolution. Aggregates experience dynamic processes of creation, stabilization, ageing, destabilization, and disruption (Marquez *et al.*, 2019). A precise description of their spatio-temporal dynamics and turnover is therefore an obligate step in evaluating the effect of aggregation on nutrient storage and conservation in soils (Lavelle *et al.*, 2020). Understanding the state of aggregation in relation to the storage and availability of nutrients requires the integration of concepts about the dynamic functions of plants, soil organisms, and soil, which are important components in assessing soil health (considered 'the frontier of soil science') (Shen and Teng, 2023). In other words, this assessment involves monitoring primary and crucial soil functions for ecosystem health and sustainability (Lal, 2016). To compose the set of indicators used in this assessment, all the key criteria: conceptual, practical, sensitivity, and interpretability (Bünemann *et al.*, 2018), and must be met and they are among the most frequently used in global human health assessments (Poppiel *et al.*, 2025).

The study of the formation pathways (or origins) of aggregates has also been successfully used in the evaluation of soil physical, chemical, and biological quality. The mechanisms of aggregate formation involve physical and chemical (physicogenic aggregates, Phy) and biological (biogenic aggregates, Bio) processes. Differentiation between the morphological classes of aggregates is carried out according to their genesis, origin or formation pathways (Bullock *et al.*, 1985; Pulleman *et al.*, 2005; Velasquez *et al.*, 2007; Batista *et al.*, 2013; Loss *et al.*, 2014; Pereira *et al.*, 2021). Bio aggregates can be physically and chemically differentiated from Phy aggregates and are defined as structural units formed by the action of SOM, roots and microbial exudates, in addition to the mechanical stress (compression) of roots and hyphae, and the activity of soil fauna (Melo *et al.*, 2019). These aggregates influence the functionality of important processes in the soil, such as increased water repellency, increased capacity to retain and store water, nutrients, and contaminants, and increased habitat supply (Guhra *et al.*, 2022).

Different studies have shown that Bio aggregates are important points of stabilization and accumulation of SOM (Ferreira *et al.*, 2020; Pinto *et al.*, 2021a,b; Silva Neto *et al.*, 2021; Fonseca Júnior *et al.*, 2023; Rossi *et al.*, 2023, 2024). However, little is known about the relationships between C, nitrogen (N), and other nutrients and the forms of phosphorus (P) in Bio and Phy aggregates, as well as whether they can act as reservoirs of P. C/N, C/P, and N/P stoichiometric ratios are indicators of the composition and quality of organic residues and SOM, especially regarding the efficiency of soil nutrient balance and cycling mechanisms (release, mineralization,

and immobilization) (Cleveland and Liptzin, 2007; Mullen, 2011; Bui and Henderson, 2013; Zinn *et al.*, 2018; Oliveira Filho *et al.*, 2019; Zhang *et al.*, 2019).

The study of P dynamics (lability and availability) is based on the quantification of the forms (inorganic and organic) of the nutrient in the soil, separated through sequential chemical fractionation techniques, in which the element is compartmentalized into fractions according to its lability and the presence or absence of organic compounds in the bond (Hedley *et al.*, 1982; Gatiboni *et al.*, 2021; Gatiboni and Condron, 2021). However, it is essential to clarify that P fractionation should not be interpreted as a form of chemical speciation (spectroscopic analysis), but rather as an assessment of the quantity and accessibility of nutrient reservoirs to plants (Gu *et al.*, 2020; Gu and Margenot, 2020; Zhao *et al.*, 2023).

Based on the above, the following hypotheses were formulated: a) the diversity of plant species in soil management systems can differentially increase the nutrient content in Bio and Phy aggregates; b) Bio aggregates can be found to have a greater accumulation and availability of nutrients than Phy; and c) Bio aggregates can promote improvements in the chemical quality of the soil's arable layer and contribute to a more efficient diagnosis of soil health, especially in soils with greater textural fragility. To test the hypotheses, the objectives of the study were to i) verify the contribution of Bio and Phy aggregates to soil fertility; ii) evaluate the total carbon, nitrogen, phosphorus and potassium contents and their respective stoichiometric ratios in these aggregates; and iii) analyse the relationship between the phosphorus fractions (labile, moderately labile and non-labile) and these aggregates from different soil use and management systems.

Material and methods

Characteristics of the study region

The study was carried out in the municipality of Terra Roxa (24° 11' 34" S and 54° 06' 62" W), located in the west of the Paraná state (South of Brazil) (Figure 1a). The studied areas are located in an agricultural property. The main characteristics of the region are as follows: (i) average altitude of 319 m; (ii) climate classified as humid subtropical (Cfa) with hot summers, according to Köppen's classification (Alvares *et al.*, 2013); (iii) regional relief classified as gently undulating; (iv) natural vegetation consisting predominantly of typical representatives of the Atlantic Forest biome; and (v) predominant soil classified as *Argissolo Vermelho-Amarelo Distrófico*, with sandy texture in the surface horizons (Santos *et al.*, 2018), with equivalence to Paleudalf in the USA Soil Taxonomy (Soil Survey Staff, 2014) and Acrisol in the FAO classification system (IUSS Working Group WRB, 2015).

History, description and location of soil use and management systems

The research in question is based on a case study with three areas under different production systems and one reference area were evaluated, totalling four soil use and management systems, namely: NT - No-tillage system (soybean-maize succession); NT+B - No-tillage + *Urochloa* system (intercropping of maize with *Urochloa ruziziensis* – succession with soybean); PP – Permanent pasture (*Cynodon dactylon*); and NF – Native forest with typical vegetation of the Atlantic Forest biome (Figure 1 b–e). Detailed information on the sampled areas is shown in Table 1. All sample areas are located on the agricultural property, under the same conditions of relief and climate, and inserted in soils with textural fragility due to their high sand content and low clay content in the 'arable layer'.

Table 2 shows the characterization of the physical and chemical attributes of the soil in the first 0.10 m deep. Sampling was carried out in August 2020, with maize (*Zea mays* L.) being the annual crop that preceded the collection period in the NT and NT+B areas. In October 2020, soybean (*Glycine max* L.) was sown in the grain production areas. In the NT+B area, the interval between



Figure 1. Location of the municipality of Terra Roxa (PR), subtropical region of Brazil (a), and land use history of the areas sampled (b–e). NT: No-tillage system (b); NT+B: No-tillage system + *Urochloa* (c); PP: Permanent pasture (d); and NF: Typical vegetation of the Atlantic Forest biome (e).

Areas sampled	Description and location
No-tillage system (NT)	Area of 28 hectares, with 20 years of conventional tillage system + 25 years of NT in succession of soybeans (summer) and corn (winter). Soil correction is carried out through periodic recommendations based on soil analysis, with the application of calcitic or dolomitic limestone and agricultural gypsum every 3 or 4 years, in the amount of 2.0 and 1.0 Mg ha ⁻¹ , respectively. It receives base fertilization of approximately 200 kg ha ⁻¹ at soybean sowing with NPK formulation in the ratio 04–30–10 and with 10–15–15 formulation at corn sowing in the second crop, following recommendations based on soil analysis. Coordinates: 24° 11' 31.34" S and 54° 06' 52.04" O. Altitude: 322 m.
No-tillage + <i>Urochloa</i> (NT+B) system	Area of 28 hectares, with 20 years of conventional tillage system, then 19 years of NT in succession of soybeans (summer) and corn (winter) and in the last six years a consortium of corn and <i>Urochloa ruziziensis</i> in winter crops (25 years of NT + <i>Urochloa</i> in total). The area receives limestone for correction every 4 years, with an application of approximately 2.0 Mg ha ⁻¹ . For sowing soybeans, a formulation of 15–15–15, 200 kg ha ⁻¹ of NPK plus potassium chloride is applied as a top dressing. When sowing second crop corn, the area receives an application of 240 kg ha ⁻¹ of NPK in the 15–15–15 ratio, plus ammonium sulphate as a top dressing. Coordinates: 24° 11' 28.30" S and 54° 06' 50.60" O. Altitude: 322 m.
Permanent pasture (PP) system	Area of 4.1 hectares, with 45 years of permanent pasture with coast-cross (<i>Cynodon dactylon</i>) and continuous stocking of dairy cattle of 2 UA ha ⁻¹ . The area has been under this management for 45 years and is reformed every 15 years with an application of lime of 2.0 Mg ha ⁻¹ . Coordinates: 24° 11' 34.86" S and 54° 06' 49.06" O. Altitude: 312 m.
Typical vegetation of the Atlantic Forest biome (NF)	Area of 28 hectares of vegetation typical of the Atlantic Forest biome – Semideciduous Seasonal Forest, with no signs of anthropogenic action. Coordinates: 24° 11' 15.22" S and 54° 06' 47.46" O. Altitude: 310 m.

Table 1. Detailed information on the areas sampled in the municipality of Terra Roxa (PR), subtropical region of Brazil

Source: Pinto et al. (2022; 2023).

	Sand	Silt	Clay	тос	рН	Ca ²⁺	Mg^{2+}	H+Al	SB	CEC	BS	K^+	Р	
Areas		g k	g-1		H_2O		c	mol _c dm ⁻³			%	mg c	dm⁻³	
NT	750	130	120	6	6.8	3.2	1.10	0.2	4.7	4.9	96	100	42	
NT+B	770	120	110	8	6.6	3.1	1.30	0.5	5.0	5.4	91	171	41	
PP	610	190	200	16	5.8	4.3	2.20	2.3	7.0	9.3	75	203	51	
NF	670	210	120	7	5.5	1.3	0.70	1.8	2.2	3.9	55	50	23	

Table 2. Characterization of the physical and chemical attributes of the soil in the production areas in the municipality of Terra Roxa (PR) in the 0–0.10 m layer, subtropical region of Brazil

NT: No-tillage system; NT+B: No-tillage system + *Urochloa*; PP: Permanent pasture; NF: Typical vegetation of the Atlantic Forest biome; pH: active acidity; Ca^{2+} : Exchangeable calcium; Mg^{2+} : Exchangeable magnesium; H+AI: Potential acidity; SB: Sum of exchangeable bases; CEC: Cation exchange capacity at pH 7.0; BS: Base saturation; K⁺: Exchangeable potassium; P: Available phosphorus; and TOC: Total organic carbon. **Source**: Pinto *et al.*(2023; 2022).

Urochloa desiccation and soybean sowing was approximately 90 days. This procedure prevents exudates containing desiccant molecules from passing from the target plant to the roots of the main crop.

Collection of samples and separation of aggregates

In the study areas, five plots of 400 m^2 were demarcated. Four (single) undeformed samples (clods) were collected from each plot to make up one composite sample per plot, by opening a mini-trench. Each composite sample corresponds to a pseudorepetitions (totalling five

pseudorepetitions per sampled area), collected in the 0–0.05 and 0.05–0.10 m layers, totalling a set of 40 sampling units (four areas evaluated \times five pseudorepetitions \times two layers). After collection, the samples were air-dried and subsequently sieved using 9.7- and 8.0-mm-mesh sieves, and the aggregates retained in the interval were selected for the study. Aggregates obtained in each interval were examined under a binocular stereomicroscope and manually separated according to their genesis based on their morphology (Bullock *et al.*, 1985; Pulleman *et al.*, 2005; Pereira *et al.*, 2021).

The differentiation between the two types of aggregates was based on the visualization of morphological characteristics such as shape, size, presence of roots, porosity, subunit arrangements and junctions (Bullock *et al.*, 1985; Pulleman *et al.*, 2005; Batista *et al.*, 2013; Melo *et al.*, 2019; Pinto *et al.*, 2021a,b; Pereira *et al.*, 2021). Thus, the aggregates were classified as: **biogenic (Bio)** – those in which it is possible to visualize rounded shapes, caused by the intestinal tract of individuals of the soil macrofauna, mainly Oligochaeta (earthworms) or those in which it is possible to visualize the presence and activity of roots; and **physicogenic (Phy)** – defined by those that showed angular shapes resulting from the interaction between carbon, clay, cations and soil wetting and drying cycles. These were pounded to break up clods and passed through a 2.0-mm-mesh sieve, thus obtaining the air-dried fine earth (ADFE) fraction (Teixeira *et al.*, 2017).

Chemical attributes

Exchangeable calcium and magnesium contents (Ca²⁺ and Mg²⁺) were determined by extraction with 1 mol L⁻¹ potassium chloride (KCl) solution (soil:solution ratio of 1:10 m:v). Exchangeable potassium (K⁺) and available phosphorus (P) were determined using 0.05 mol L⁻¹ hydrochloric acid (HCl) and 0.0125 mol L⁻¹ sulphuric acid (H₂SO₄) solution (soil:solution ratio of 1:10 m:v). Soil potential acidity (H+Al) was extracted with 0.5 mol L⁻¹ calcium acetate (C₄H₆CaO₄) solution, adjusted to pH 7.0 (soil:solution of 1:15 m:v) (Teixeira *et al.*, 2017).

Total carbon and total nitrogen contents (TC and TN) were determined by the dry combustion method in a Perkin Elmer 2400 CHN elemental analyser. Analyses were carried out using samples of aggregates macerated in mortar and passed through a 100-mesh (149 μ m) sieve (Nelson and Sommers, 1996; Sato *et al.*, 2014). Total potassium contents (TK) were extracted and determined according to Tedesco *et al.* (1995).

P fractions were extracted according to the method proposed by Gatiboni and Condron (2021). Five P fractions with different degrees of lability and potential availability were extracted sequentially from 0.50 g of ADFS as follows: Soluble P with 0.01 mol L⁻¹ CaCl₂ solution (_{SOL}P) (labile); available P with Mehlich 3 (Mehlich, 1984) extractant solution (_{M3}P) (labile); inorganic P and autoclaved P extracted with 0.5 mol L⁻¹ NaOH solution (_{OH}Pi and _{autoclave}P, respectively) (moderately labile); and P extracted with 1 mol L⁻¹ HCl (_{HCl}P) (moderately labile).

The P concentration in each fraction was determined using colorimetry (Murphy and Riley, 1962). Organic P extracted with NaOH ($_{OH}Po$) was obtained from the difference between $_{autoclave}P$ and $_{OH}Pi$. Occluded P ($_{OCL}P$) (non-labile) was also quantified, consisting of the less available Pi and Po forms that were not extracted by any of the previous extractants (CaCl₂ 0.01 mol L⁻¹, Mehlich 3, NaOH 0.5 mol L⁻¹ and HCl 1 mol L⁻¹). In turn, $_{OCL}P$ was obtained via sulphuric digestion and hydrogen peroxide treatment (Tedesco *et al.*, 1995). The TP contents were calculated by adding up the P contents of all the extracted fractions ($\Sigma = _{SOL}P + _{M3}P + _{OH}Pi + _{OH}Po + _{HCl}P + _{OCL}P$). Subsequently, the stoichiometric ratios C/N, C/P, and N/P were calculated.

Statistical analysis

To statistically analyse the results of the case study in question, a completely randomized design was used in a 4×2 factorial scheme (soil uses and management systems \times aggregate classes). The data were tested for the normality of residuals (Shapiro-Wilk test), and for the homoscedasticity of variances (Bartlett test). Variables that did not meet the assumptions were transformed (Box-Cox

test) and were retested. The data, except for P proportions, were evaluated by analysis of variance followed by Tukey's test when the assumptions of normality and homoscedasticity were met (transformed or untransformed variables).

In cases where data transformation was inefficient, the Kruskal-Wallis test followed by Fisher's least significant difference criterion was used to evaluate the soil use and management systems for each aggregate classes, and the Wilcoxon test was used to compare variables between the aggregates in each soil use and management system. Multivariate analysis of principal components (PCA) and dendrogram (Euclidean Distance and Ward.D2 methods) were also performed, which is a tree diagram that displays the groups formed by hierarchical grouping (clustering, without limiting subdivisions) of observations at each step and in their levels of dissimilarity, both built based on data of attributes in the 0–0.10 m layer. All tests were performed at 5% significance level in R Software (R Core Team, 2020) using the 'ExpDes.pt' and 'Ggplot2' packages.

Results

Influence of management systems and aggregate origin on soil fertility and TC, TN, TP and TK contents

The fertility results showed differences mainly between soil use and management systems, especially the pasture and maize-*Urochloa* intercropping systems. Aggregates of the PP, NT, and NT+B systems had the highest contents of Ca^{2+} , Mg^{2+} and available P, especially in the surface layer (0–0.05 m layer). High values of H+Al were observed in aggregates of the PP and NF areas in the 0–0.10 m layer. For the same layer, the highest contents of K⁺ were observed in aggregates of the PP and NT+B systems (Table 3).

Bio aggregates of the PP system had the highest contents of Mg^{2+} and available P in the 0.05–0.10 m layer and the highest values of H+Al in the 0–0.10 m layer compared to the Phy ones. Among the aggregate classes of the other areas, no differences were observed in the contents of chemical attributes of fertility (Table 3). The contents of TC, TN, TP, and TK were more influenced by the management systems. Only in the pasture system was there a significant increase in the total nutrient contents for both formation pathways, and the highest contents of TC, TN, TP, and TK were quantified in these aggregates (0–0.10 m layer) (Table 4).

Among the areas of NT, NT+B, and NF, the maize-*Urochloa* intercropping favoured the increase of TC contents compared to the aggregates of the reference area and of the system with soybean/maize succession, more specifically in the subsurface layer, notably, the increase was from 36 to 44% (Bio aggregates, 0–0.05 and 0.05–0.10 m layers) and from 39 to 44% (Phy aggregates, 0–0.05 and 0.05–0.10 m layers) compared to the NF area, and 10 to 44% (Bio aggregates, 0–0.05 and 0.05–0.10 m layers) and 29 to 31% (Phy aggregates, 0–0.05 and 0.05–0.10 m layers) compared to the NT system. There was no proportional increase in the contents of TN, TP and TK quantified in the aggregates of the NT+B area in comparison with the NT and NF areas.

Regarding the aggregate classes, the TC and TN contents quantified in Bio aggregates compared to Phy were proportionally higher by 8% (PP), 11% (NT+B), 13% (NF) and 30% (NT) for TC, and 56% (NF), 178% (NT) and 239% (NT+B) for TN (0–0.05 m layer). An inverse pattern was observed in subsurface, where the TN contents in the aggregates of the Phy pathway were proportionally higher by 50% (NT+B) and 55% (NT) compared to those of the Bio pathway (0.05–0.10 m layer).

Influence of management systems and aggregate origin on stoichiometric ratios

The values of the ratios between TC, TN, and TP (C/N, C/P and N/P) showed differences between the soil use and management systems and the aggregate classes, especially through the C/N ratio. In the surface layer, the highest values of C/N ratio were quantified in the Phy aggregates of the NT

Table 3. Exchangeable calcium (Ca^{2+}), magnesium (Mg^{2+}), and potassium (K^+) contents, available phosphorus (P) and potential acidity (H+Al) of biogenic (Bio) and physicogenic (Phy) aggregates from areas under different production systems, subtropical region of Brazil

	Bio	Phy	Bio	Phy	Bio	Phy	Bio	Phy	Bio	Phy		
	C	a ²⁺	M	g ²⁺	H-	⊢Al	K	+		Р		
Areas			cmol _c d	lm ⁻³				mg c	dm⁻³			
	 Laver 0-0.05 m depth											
NT	2.3 aA	1.7 abA	1.0 aA	1.3 abA	0.9 bA	0.8 bA	125 bcA	108 bA	39 abA	38 abA		
NT+B	2.1 aA	1.7 abA	1.3 aA	1.3 abA	1.5 bA	1.2 bA	192 abA	199 aA	53 aA	44 aA		
PP	2.7 aA	2.8 aA	2.9 aA	2.3 aA	4.8 aA	3.8 aB	250 aA	272 aA	40 abA	34 abA		
NF	0.7 bA	0.9 bA	0.8 bA	0.6 bA	3.8 aA	3.5 aA	90 cA	80 bA	15 bA	15 bA		
		Layer 0.05-0.10 m depth										
NT	1.2 abA	1.2 bA	0.9 bA	0.7 bA	1.0 bA	1.3 bA	71 bA	88 abA	42 abA	42 aA		
NT+B	1.3 aA	1.4 bA	1.1 bA	1.2 abA	1.2 bA	1.1 bA	145 aA	134 aA	35 abA	37 aA		
PP	1.9 aA	2.0 aA	2.2 aA	1.7 aB	3.2 aA	2.4 aB	177 aA	140 aA	46 aA	26 aB		
NF	0.8 bA	0.7 cA	0.6 bA	0.7 bA	3.2 aA	2.8 aA	62 bA	56 bA	21 bA	19 aA		

Averages followed by the same lowercase letter in the column do not differ between management systems for the same type of aggregate. Averages followed by the same capital letter in the row do not differ between aggregate types for the same system evaluated (ANOVA + Tukey's test without data transformations; ANOVA + Tukey's test with data transformations; and Kruskal-Wallis test + Fisher's minimum significant difference); NT: No-tillage system; NT+B: No-tillage system + *Urochloa*; PP: Permanent pasture; and NF: Typical vegetation of the Atlantic Forest biome.

Table 4. Total carbon (TC), nitrogen (TN), phosphorus (TP), and potassium (TK) contents of biogenic (Bio) and physicogenic (Phy) aggregates from areas under different production systems, subtropical region of Brazil

	Bio	Phy	Bio	Phy	Bio	Phy	Bio	Phy
]	ГС	T	N		ГР	T	К
Areas				g k	g ⁻¹			
				Layer 0-0	.05 m depth			
NT	21.1 bA	16.2 bA	0.8 bA	0.3 bA	0.4 bA	0.4 bcA	0.3 cA	0.3 cA
NT+B	23.3 bA	21.0 bA	0.8 bA	0.2 bA	0.4 bA	0.4 bA	0.4 bcA	0.3 bcA
PP	38.2 aA	35.3 aA	3.4 aA	3.3 aA	1.3 aA	1.1 aA	0.5 aA	0.5 aB
NF	17.0 bA	15.1 bA	0.9 bA	0.6 bA	0.4 bA	0.4 cA	0.4 bA	0.4 abA
				Layer 0.05-	0.10 m dept	h		
NT	11.5 bA	11.7 bA	0.3 bA	0.5 bA	0.3 bB	0.4 bA	0.2 aA	0.2 bA
NT+B	16.5 bA	15.2 bA	0.5 bA	0.7 bA	0.3 bA	0.5 abA	0.3 aA	0.3 abA
PP	30.0 aA	25.5 aA	3.0 aA	2.4 aB	0.9 aA	0.9 aA	0.3 aA	0.3 abA
NF	11.5 bA	10.5 bA	0.5 bA	0.4 bA	0.3 bA	0.4 bA	0.4 aA	0.4 aA

Averages followed by the same lowercase letter in the column do not differ between management systems for the same type of aggregate. Averages followed by the same capital letter in the row do not differ between aggregate types for the same system evaluated. ANOVA + Tukey's test without data transformations; ANOVA + Tukey's test with data transformations; and Kruskal-Wallis test + Fisher's minimum significant difference. NT: No-tillage system; NT+B: No-tillage system + *Urochloa*; PP: Permanent pasture; and NF: Typical vegetation of the Atlantic Forest biome.

and NT+B systems, regardless of the management systems and aggregate formation pathways (0-0.05 m layer). In the subsurface layer, an inverse pattern was observed in the C/N ratio values, in particular: Bio aggregates of the NT and NT+B systems had the highest values of C/N ratio, also regardless of the factors evaluated (0.05-0.10 m layer). The lowest values of C/N ratio were observed in aggregates of the PP system (0-0.10 m layer) (Table 5).

Among the systems, the highest values of C/P ratio were found in aggregates of the NT, NT+B and NF areas only in the surface layer (0–0.05 m layer). Among the aggregate classes, higher values of the C/P ratio were observed in Bio aggregates, from the areas of NT in the 0–0.10 m layer and NT+B and NF in the 0.05–0.10 m layer. For the N/P ratio, the highest values were found in aggregates of the PP system, followed by aggregates of the NF area in the 0–0.05 m layer. In the 0.05–0.10 m layer, the highest values of the N/P ratio were quantified in aggregates of the PP

	Bio	Phy	Bio	Phy	Bio	Phy
Areas	(C/N		С/Р	N	/P
			Layer 0-	-0.05 m depth		
NT	28 aB	60 aA	57 aA	44 abB	2,0 abA	0,7 cB
NT+B	30 aB	91 aA	58 aA	51 aA	1,9 bA	0,6 cB
PP	11 cA	11 cA	30 bA	33 bA	2,7 aA	3,0 aA
NF	20 bA	27 bA	49 aA	43 abA	2,4 abA	1,6 bA
			Layer 0.0	5-0.10 m depth		
NT	35 aA	23 aB	42 aA	30 aB	1,2 bA	1,30 bA
NT+B	34 aA	21 aB	49 aA	32 aB	1,4 bA	1,50 bA
PP	10 bA	11 aA	33 aA	29 aA	3,2 aA	2,70 aA
NF	26 aA	25 aA	41 aA	30 aB	1,6 bA	1,20 bA

Table 5. Stoichiometric ratios between carbon, nitrogen, and phosphorus (C/N, C/P, and N/P, respectively) in their total concentrations of biogenic (Bio) and physicogenic (Phy) aggregates from areas under different production systems, subtropical region of Brazil

Averages followed by the same lowercase letter in the column do not differ between management systems for the same type of aggregate. Averages followed by the same capital letter in the row do not differ between aggregate types for the same system evaluated. ANOVA + Tukey's test without data transformations; ANOVA + Tukey's test with data transformations; and Kruskal-Wallis test + Fisher's minimum significant difference. NT: No-tillage system; NT+B: No-tillage system + *Urochloa*; PP: Permanent pasture; and NF: Typical vegetation of the Atlantic Forest biome.

system. Regarding the formation pathways, only in the surface layer did the values of the N/P ratio differ, with higher values observed in Bio aggregates of the NT and NT+B systems compared to the Phy of the same areas (0-0.05 m layer) (Table 5).

Influence of management systems and aggregate origin on labile, moderately labile and nonlabile P contents

Higher P contents were found in aggregates of the PP system in comparison to the grain production systems and non-anthropized system (Table 6), especially $_{M3}P$ and $_{HCl}P$ (2 to 4 times higher), $_{OCL}P$ (2 to 5 times higher), $_{OH}Po$ (2 to 6 times higher) and $_{OH}Pi$ (3 to 8 times higher) (0–0.10 m layer). In the Bio and Phy aggregates of the PP area, the $_{OH}Pi$ fraction was the one that showed the highest participation of TP contents in the surface layer (35%) (0–0.05 m layer). In the subsurface layer, the $_{OH}Pi$ (31%) and $_{OCL}P$ (28%) fractions stood out in Bio aggregates and the $_{OCL}P$ (32%) and $_{OH}Po$ (28%) fractions stood out in Phy (0.05–0.10 m layer) (Table 6).

Intermediate contents of the $_{HCI}P$ fraction were quantified in Bio aggregates of the NT+B system in comparison with the same class of aggregates of the PP, NT, and NF areas, and aggregates of the NT and NT+B systems showed higher contents of the $_{OCL}P$ fraction compared to those of the NF area (0–0.10 m layer). The $_{OH}Po$ (27 to 32%) and $_{OCL}P$ (26 to 34%) fractions showed higher proportions of TP contents in Bio and Phy aggregates of the NT and NT+B systems in surface (0–0.05 m layer). In subsurface, the $_{OCL}P$ fraction in both classes of aggregates (34 to 44%) stood out, followed by the $_{OH}Pi$ (20 and 23%) and $_{M3}P$ (18 and 22%) fractions in Bio aggregates and $_{OH}Po$ (27 and 34%) fraction in Phy, all these results relative to aggregates of the NT and NT+B and NT+B systems (0.05–0.10 m layer) (Table 6).

There were no differences in the contents of the _{SOL}P fraction quantified in the aggregates of the evaluated areas (0–0.10 m layer). The _{%SOL}P fraction is the one with the lowest expression in the relative contribution of TP contents in both classes of aggregates, regardless of the management system (1%), followed by the _{HCl}P fraction with a similar pattern (2 to 6%) (0–0.10 m layer). It is worth pointing out that, in aggregates of the NF area, the _{OH}Po fraction showed a higher relative participation of TP contents in the surface layer (42%) (0–0.05 m layer). A similar pattern was observed in aggregates of Phy origin of the same area in the subsurface layer (_{OH}Po, 37%) (0.05–0.10 m layer) (Table 6).

Areas	- % 13 16 35			
Areas $s_{0L}P$ M_3P $o_{H}Pi$	- % 13 16 35			
mg kg ⁻¹ % mg kg ⁻¹ NT 1 aA 0.3 1 aA 0.2 75 bA 21 56 bA 15 67 bA 18 49 bA NT+B 2 aA 0.5 1 aA 0.2 96 bA 23 80 bA 20 70 bA 17 65 bA	% 13 16 35			
NT 1 aA 0.3 1 aA 0.2 75 bA 21 56 bA 15 67 bA 18 49 bA NT+B 2 aA 0.5 1 aA 0.2 96 bA 23 80 bA 20 70 bA 17 65 bA	13 16 35			
NT+B 2 aA 0.5 1 aA 0.2 96 bA 23 80 bA 20 70 bA 17 65 bA	16 35			
	35			
PP 5 aA 0.4 5 aA 0.4 220 aA 17 202 aA 18 439 aA 35 387 aA				
NE 2 aA 0.5 2 aA 0.5 87 bA 22 82 bA 21 67 bA 18 61 bA	17			
	oci P			
$m\sigma k\sigma^{-1}$ %	%			
NT 105 bA 29 118 bA 32 16 bA 4 18 aA 5 102 bA 28 126 bA	34			
NT+B 111 bA 27 120 bA 29 22 abA 6 23 aA 6 105 bA 26 123 bA	30			
PP 335 aA 26 250 aA 23 35 aA 3 17 aB 2 223 aA 19 219 aA	22			
NF 135 b4 42 134 ab4 41 8 b4 2 9 ab 3 50 c4 14 58 c4	18			
Areas	10			
$M3^{r}$ mg kg ⁻¹ %	%			
NT 2 a4 10 3 a4 10 57 hA 22 53 hA 15 58 hA 23 62 hA	17			
NT+B 2 aA 10 2 aA 04 61 bA 18 61 bA 14 69 bA 20 64 bA	14			
PP 5 a 10 5 a 10 15 a 15 118 a 12 305 a 31 218 a	25			
NE 2 24 10 2 24 10 68 b4 20 63 b4 14 66 b4 27 59 b4	19			
	-			
$m\sigma$ k σ^{-1} % mg k σ^{-1}	0/6			
NT 36 b 14 120 a 27 13 b 5 11 a 3 92 c 35 137 b 6	38			
NT B 43 bB 12 160 aA 24 16 ab 5 16 aA 3 153 bA 44 155 bA	34			
PD 205 a 24 256 a 28 25 a 2 13 a 2 13 a 2 252 a 28 260 a	37			
$\begin{array}{cccccccccccccccccccccccccccccccccccc$	J2			

Table 6. Phosphorus fraction content (mg kg⁻¹ and %) of biogenic (Bio) and physicogenic (Phy) aggregates from areas under different use and management systems, subtropical region of Brazil

Averages followed by the same lowercase letter in the column do not differ between management systems for the same type of aggregate. Averages followed by the same capital letter in the row do not differ between aggregate types for the same system evaluated. ANOVA + Tukey's test without data transformations; ANOVA + Tukey's test with data transformations; and Kruskal-Wallis test + Fisher's minimum significant difference. Relative contribution of each phosphorus fraction in relation to the total phosphorus concentration (TP). sol.PP: Soluble P fraction with CaCl₂ solution; $_{M3}P$: Available P fraction with Mehlich 3 extractant solution; $_{OH}P$: Inorganic P fraction extracted with NaOH solution; $_{OH}P$: Organic P fraction extracted with NaOH solution; $_{HCIP}$: Fraction of inorganic P extracted with HCl; $_{OCL}P$: Fraction of occluded P; NT: No-tillage system; NT+B: No-tillage system + *Urochloa*; PP: Permanent pasture; and NF: Typical vegetation of the Atlantic Forest biome.

Dissimilarity between management systems and aggregate formation pathways

Regarding the principal component analysis (PCA), cumulative variance of 67.2% was observed for principal components (PC) 1 and 2 (Figure 3). According to the PCA (Figure 2), the soil use and management systems are separated with the formation of three different groups in the 0–0.10 m layer: (1) group formed by aggregates of the pasture system (lower right quadrant); (2) group formed by aggregates of the grain production systems (upper left quadrant); and (3) group formed by aggregates of the non-anthropized system (lower left quadrant). PP and NF areas were individualized from NT and NT+B areas, both by the main axis (PC1, 54.2%) and by the secondary axis (PC2, 13.0%). No pattern of separation between the aggregate classes was observed (Figure 2).

The discriminant variables for PC1 formation (high correlation, $-0.70 \ge r \ge 0.70$) were Ca, Mg, TC, TN, TP, _{SOL}P, _{M3}P, _{OH}Pi, _{OH}Po, and _{OCL}P (Table S1). These indicator attributes (except Ca) are the most associated with the Bio and Phy aggregates of the PP area in the 0–0.10 m layer (Figure 2). As for PC2, available P (0.76) was the variable that most contributed to the construction of this axis (Table S1). Bio and Phy aggregates of the NT and NT+B systems are related to the values of the C/N and C/P ratios in the 0–0.10 m layer (Figure 2), although the correlation values of these variables were not classified as high (PC1, -0.34 and -0.45; and PC2, 0.64 and 0.28, respectively) (Table S1).



Figure 2. Principal component analysis integrating the characterization of chemical attributes, stoichiometric ratios, and phosphorus fractions of biogenic (Bio) and physicogenic (Phy) aggregates from areas under different production systems, layer of 0–0.10 m deep, subtropical region of Brazil. Ca: Exchangeable calcium; Mg: Exchangeable magnesium; K: Exchangeable potassium; P: Available phosphorus; H+Al: Potential acidity; TC: Total carbon; TN: Total nitrogen; TP: Total phosphorus; TK: Total potassium; C/N: Carbon and nitrogen ratio; C/P: Carbon and phosphorus ratio; N/P: Nitrogen and phosphorus ratio; SOLP: Soluble P fraction with CaCl₂ solution; M3P: Available P fraction with Mehlich 3 extractant solution; OHPi: Fraction of inorganic P extracted with NaOH solution; OHPo: Fraction of organic P extracted with NaOH solution; HClP: Fraction of inorganic P extracted with HCl; OCLP: Fraction of occluded P; NT: No-tillage system; NT+B: No-tillage system + *Urochloa*; PP: Permanent pasture; and NF: Typical vegetation of the Atlantic Forest biome.

The dendrogram (cluster analysis) for the chemical attributes of the 0–0.10 m layer initially showed the formation of two main clusters, with the aggregates of the pasture system (most heterogeneous group) being separated from the aggregates of the NF, NT and NT+B areas (most homogeneous group), with \pm 100% dissimilarity. Subsequently, three secondary clusters were formed, mainly individualizing aggregates of the non-anthropized system from aggregates of the grain production systems, with approximately \pm 50% dissimilarity. Finally, the dendrogram also indicated the formation of four tertiary clusters, especially differentiating the aggregate classes within the conservation systems, with \pm 30% dissimilarity (Figure 3).

Discussion

Aggregates of the PP system and their relationship with the chemical quality of the arable layer

The nutrient contents in aggregates of the PP system (45 years) were favoured by the soil conditions inherent to the area. Preponderant factors include SOM contents, variation in texture between areas, and the lowest levels of soil disturbance. Conservation practices, such as well-



Figure 3. Hierarchical cluster analysis considering the characterization of chemical attributes, stoichiometric ratios, and phosphorus fractions of biogenic and physicogenic aggregates from areas under different production systems, layer of 0–0.10 m deep, subtropical region of Brazil. Ca: Exchangeable calcium; Mg: Exchangeable magnesium; K: Exchangeable potassium; P: Available phosphorus; H+Al: Potential acidity; TC: Total carbon; TN: Total nitrogen; TP: Total phosphorus; TK: Total potassium; C/N: Carbon and nitrogen ratio; C/P: Carbon and phosphorus ratio; N/P: Nitrogen and phosphorus ratio; SOLP: Soluble P fraction with CaCl2 solution; M3P: Available P fraction with Mehlich 3 extractant solution; OHPi: Fraction of inorganic P extracted with NaOH solution; OHPo: Fraction of organic P extracted with NaOH solution; HCIP: Fraction of occluded P; NT: No-tillage system; NT+B: No-tillage system + *Urochloa*; PP: Permanent pasture; and NF: Typical vegetation of the Atlantic Forest biome.

managed pastures (Cerri *et al.*, 2023), have been shown to be efficient in increasing nutrient storage capacity, which is generally attributed to the greater supply of organic residues (via litter) to the root system of grasses (via rhizodeposition) and to the lower turning of topsoil.

SOM dynamics in pastures is affected by organic material inputs and C outputs from the soil, as well as by decomposition, mineralization, stabilization, and microbial respiration processes (Cherubin *et al.*, 2023). In tropical regions, in soils with variable loads (dependent on pH), most of the physico-chemical reactions are attributed to SOM. It provides macro and micronutrients, mainly N, P and S, and increases the amount of water retained in the soil, altering the zero load point (Demattê and Demattê, 2024). This is why understanding the health of sandy-textured soils strongly depends on investigating the dynamics of SOM (content, and chemical and structural composition) and the state of aggregation (origin, stability and size of aggregates).

The more clayey texture in this system (sandy loam; Table 2) favours organo-mineral interactions, cation exchange capacity and P adsorption in aggregates, reducing nutrient losses due to leaching and/or erosion. The addition of lime and fertilizers in pastures can increase nutrient contents. In the study, the PP system receives only the application of lime during its renovation every 15 years (2.0 t ha⁻¹; Table 1). However, for N, the practice of increasing the nutrient contents in pastures via N fertilization (increases pasture yield), in addition to being costly, can result in higher losses via N₂O emissions in agricultural soils. Tropical grass pastures are commonly effective in using N from the soil and show low losses of this nutrient (Baptistella *et al.*, 2020), with a direct impact on the soil health in tropical climate regions.

Additionally, the C cycling process in the soil is intense in PP system, and a large part of the nutrient derived from forage biomass ingested by animals' returns to the soil via faeces and urine (Cherubin *et al.*, 2023). Thus, the presence of animals deserves to be highlighted in the system of the present study (continuous stocking of dairy cattle with 2 AU ha⁻¹; Table 1), since fresh cattle manure can activate the soil biota and provide a more favourable environment for the growth and

activity of microbial biomass, as well as for the formation of Bio aggregates. Therefore, it leads to higher rates of decomposition of organic material (Bernardi *et al.*, 2023), consequently providing higher nutrient contents, as seen in aggregates of the PP system.

The values of the C/N ratio observed in aggregates of this system indicate higher N mineralization (C/N < 20), being affected by the high TN contents in the aggregates, regardless of the formation pathway. This reinforces the importance of N for the processes mediated by soil microorganisms and, consequently, for the transformations of C in plant residues, SOM and its fractions. The results point to an accelerated decomposition of SOM and/or loss of the more soluble forms of C in the aggregates of the system. Thus, the remaining SOM in these structural units tends to be more stable and/or humified (Zinn *et al.*, 2018). According to Ramos *et al.* (2023), the biomass of the aerial part of grasses has a higher C/N ratio than that of legume species, but is still very favourable to decomposed (Bui and Henderson, 2013). In general, the C/N ratio is inversely proportional to the rate of decomposition of organic material (Zhang *et al.*, 2019).

As for the values of the C/P and N/P ratios in the aggregates of the soil use and management systems, they indicate P mineralization (C/P \leq 200) and N limitation (N/P < 14), respectively. Both stoichiometric ratios can directly affect the soil chemical quality in agricultural systems, with repercussions on soil health. For the N/P ratio, the lowest potentiality in the N limitation process can be observed in the aggregates of the PP system. The C/P ratio is a parameter of P efficacy, i.e., lower values indicate higher nutrient efficacy in the soil. The N/P ratio is considered indicative of N or P limitation at the plant community level and is evaluated as an important factor in the decomposability of organic residues (Bui and Henderson, 2013).

The superiority contents of the P fractions in the aggregates of the PP system are due to the influence of the soil conditions in the area, especially the clay contents and the higher supply of plant residues, except for _{SOL}P fraction. The higher levels of _{M3}P in these aggregates mean that the area is storing P in forms that are readily available to the forage grass grown. The inorganic (_{OH}Pi and _{HCl}P) and organic (_{OH}Po) forms of P with moderate lability were also favoured by the soil conditions of the pasture, evaluated as important reservoirs of the nutrient. The _{OH}Pi fraction represents the inorganic form of P adsorbed to Fe and Al oxides of low crystallinity (Gatiboni *et al.*, 2021), corroborating the higher clay contents in the PP system (200 g kg⁻¹; Table 2) compared to the NT and NT+B systems (120 and 110 g kg⁻¹; Table 2). Aggregates of this system had the highest TC contents, and the greater physical and chemical protection of SOM offered by the clay fraction and by the aggregate formation pathways are promoting the increase and maintenance of _{OH}Po contents (fraction associated with SOM).

As the labile and moderately labile P fractions are more susceptible to dissolution, it is plausible to infer that the sandy loam texture and the stabilization of SOM in the PP system (either by molecular recalcitrance, organo-mineral complexes, occlusion in aggregates, formation of supramolecular structures and/or adsorption in clay minerals) are the factors most correlated with the stabilization and accumulation of P in fractions with different degrees of lability and availability (Gatiboni and Condron, 2021). In the study by Sandim *et al.* (2023), the authors also observed that the dynamics of P in the soil was mainly influenced by SOM, cation exchange capacity and the presence of Fe and Al oxides.

The multivariate techniques helped to understand the pattern of the results obtained for the attributes of the aggregates of the soil use and management systems in association with the univariate statistical tests. According to the principal component correlation matrix, the first group of highly weighted variables implies that PC1 of the PCA is primarily associated with exchangeable cations, TC, TN and TP contents, and the P fractions with different degrees of lability and availability. Variables that are strongly related to the aggregates of the PP system, regardless of the origin of formation of the structural units, highlighting the greater influence of soil conditions in the area on the indicator attributes associated with aggregation.

Aggregates of the NT+B system and their relationship with the chemical quality of the arable layer

In the Midwest region of Brazil, Oliveira *et al.* (2020) compared the cycling and vertical stratification of K uptake in areas of soybean monoculture, soybean/*Urochloa ruziziensis* succession, soybean/maize succession, and maize-*Urochloa ruziziensis* intercropping in succession with soybean. These authors found that areas with the introduction of *Urochloa ruziziensis* accumulated high contents of K, with most of the nutrient originating from the subsurface layers of the soil, corroborating the results of K in aggregates of the NT+B system (25 years of conservation system, the last six years with intercropping of maize with *Urochloa ruziziensis* in the dry season). These results emphasize that studies on nutrient dynamics in agricultural production areas with *Urochloa* species deserve more attention.

Soils cultivated with *Urochloa* tend to have higher contents of SOM due to the large number of plants, greater residue deposition, inhibition of nitrification, rhizodeposition and greater root mass, and incorporation of N via more lignified straw (Baptistella *et al.*, 2020). This justifies the TC results in aggregates of the NT+B systems, mainly in the subsurface layers. The intercropping of maize with *Urochloa ruziziensis* allows greater grain production, concomitant with the formation of a more vigorous root system and *Urochloa* straw. Therefore, the use of grass is essential to the sustainability of the conservation systems. Considers it a key component in the management of cover crops in different production systems in sandy-textured soils (Demattê and Demattê, 2024). *Urochloa ruziziensis* is the species most used together with maize in the subtropical region of the country, aiming to improve soil quality and to provide high-quality forage in July and August (dry season). This is the period in which perennial pastures show low productivity and quality, considered as a critical moment for cattle production on pasture (Franchini *et al.*, 2016).

Another factor related to the proportional increase of TC contents in the aggregates of the NT+B system is found within the collection interval after the desiccation of the annual forage. *Urochloa ruziziensis* is an excellent grass for mulch formation in conservation management areas and has good palatability for animals (Balbino *et al.*, 2012). However, this species requires a longer time for total desiccation, around 20 days, so desiccation must be performed earlier when samples are to be collected to assess fertility levels and/or sowing of the annual crop (Kluthcouski *et al.*, 2004). This factor reduces soil temperature during the day and water losses by evaporation, in addition to providing more suitable conditions for the survival of soil fauna and maintenance of C stocks in the edaphic environment, contributing to the maintenance or increase of the physical, chemical and biological quality of the soil (Franchini *et al.*, 2016).

It is worth mentioning that analysing the interactions between the management practices carried out in the grain production systems and the soil conditions of the study region is essential to understand the decomposition rates of plant residues and their consequent contribution to the mechanisms of nutrient balance and cycling in the soil. In this case, emphasis must be placed on diversification of plant species in time (crop rotation between grasses and legumes) and space (intercropping); maintenance of plant biomass on soil surface, considered as the main practice adopted in the no-tillage system (Cerri *et al.*, 2023); and the sandier texture of the surface layer, which enhances the transformations of plant residues and SOM. Low values of the N/P ratio may signal that N contents in the soil strongly limit the proper functioning of agroecosystems (Cleveland and Liptzin, 2007; Bui and Henderson, 2013; Oliveira Filho *et al.*, 2019). Based on the results of the stoichiometric ratios C/N and N/P, management strategies for SOM associated with aggregation should be created and adopted to optimize the processes of N gains and minimize its losses by volatilization and leaching in the NT and NT+B areas.

The NT+B system is favouring the increase of $_{HCl}P$ contents in the Bio aggregates. These results are important for monitoring the effects of the introduction of *Urochloa ruziziensis* with regard to the pattern of P distribution among its fractions. Due to saturation of the most labile

compartment, the nutrient is redistributed to the compartment of moderate lability. Regarding the $_{HCl}P$ fraction, the binding of P to Ca reduces the availability and lability of the nutrient, due to the saturation of Ca (added by limestone) and P (added by fertilizer) in the soil solution (Sandim *et al.*, 2023). In the Southeast region of Brazil, Almeida and Rosolem (2016) found that *Urochloa ruziziensis* cultivated in rotation with soybean increased P contents in the labile and moderately labile fractions, reducing its contents in the occluded fraction. According to the authors, these results indicate that the presence of *Urochloa ruziziensis* generated a potential effect on the agronomic efficiency of P utilization, regardless of the phosphate fertilization strategy.

In PC2 of the PCA, the highly weighted variable indicates that the component is mainly related to the available P contents, not being correlated with the aggregates of the evaluated areas. The link between the C/N and C/P ratios and the Bio and Phy aggregates of the NT and NT+B systems signifies the presence or absence of nutrients that limit plant growth and development in these aggregates. These nutrients are essential for the correct functionality of grain production systems. Both principal components are separating the PP, NT, NT+B, and NF areas. In the study, the indicator attributes selected by the PCA are considered more critical and effective to express the soil chemical quality and the subsequent response to the different management systems in soils with high textural fragility.

Biogenic aggregates and their relationship with the chemical quality of the arable layer

The nutrient contents in Bio aggregates (Mg^{2+} , available P, H+Al, TC, and TN) are a clear indication of the high potential that the biological formation pathway has to accumulate, protect, and make nutrients available at the most favourable time for the needs of soil organisms (plant species and soil fauna). Contributing to the maintenance or improvement of the chemical quality of the arable layer, and benefiting the soil health of agricultural production systems. Similar results were observed by Melo *et al.* (2019) in conservation management areas with application of liquid pig manure and chicken manure under climatic conditions similar to those of this study. For the authors, Bio aggregation intensifies the structural and chemical improvement of the soil promoted by the application of organic residues (manure).

In the Southeast region of Brazil, Fonseca Júnior *et al.* (2023) quantified higher C contents and its respective chemical and physical fractions in Bio aggregates of the eucalyptus plantation and forest-eucalyptus ecotone areas. For the authors, the results show the importance of studying the origin of aggregates in the evaluation of soil quality in different forest ecosystems. Also in the Southeast region, Rossi *et al.* (2024) found higher C contents in the physical fractions of SOM in Bio aggregates of agroecological production areas managed in soils with textural fragility. According to the authors, Bio aggregation favoured the increase of C contents, which suggests an improvement in soil quality. In the Northeastern region, Sales et al. (2025) investigated the influence of agroforestry systems and coffee monocultures on the genesis of aggregates, and whether this influence affects C and glomalin levels. The authors found that the coffee systems did not affect the formation and distribution of aggregates and that agroforestry with grevillea proved to be more favourable to maintaining the C and glomalin contents associated with Bio and Phy aggregates.

Considering the increase of TN contents in Bio aggregates compared to the Phy of the surface layer of the grain production systems and non-anthropized system, it is possible to infer that the biological aggregation was more efficient in storing N, contributing to reducing the losses of the nutrient in the conservation management and reference areas in the first 0.05 m deep. N inputs into the soil derive from atmospheric deposition, fertilization, and BNF processes. Also in this biological context, grass root exudates can also act directly on the N cycle, inhibiting undesirable processes (e.g., ammonia volatilization, soil erosion, denitrification, and nitrate leaching) in terms of environmental and production sustainability (Baptistella *et al.*, 2020).

In the surface layer, the values of the C/N ratio in Phy aggregates of the NT and NT+B areas indicate a high potential for N immobilization (C/N > 30), influenced by the low TN contents in these aggregates. For Bio aggregates of the same areas, the C/N ratio values are tending towards equilibrium between the processes of decomposition, release, and availability of nutrients. This is a consequence of the greater capacity of the Bio formation pathway to store and preserve nutrients, especially C and N. For C/N ratio values between 20 and 30, mineralization will be equal to immobilization, and values below 20 favour N mineralization (Mullen, 2011). N supply is essential for C sequestration to occur, being pointed out as a key factor to increase the stabilization and accumulation of C in the soil, notably in mineral-associated organic matter (Cotrufo and Lavallee, 2022).

In the subsurface layer, the values of C/N ratio in Phy aggregates of the grain production systems are within the equilibrium range, i.e., outside the rate required by decomposer microorganisms (30). In turn, in the Bio aggregation of the same systems, the values of C/N ratio signal the immobilization of N, but less intense when compared to the immobilization observed in the surface Phy aggregates. Such results are important in the study of nutrient cycling in aggregates of different origins, as the greater immobilization of N in the Phy aggregates of the surface layer can be considered a highly critical factor in its cycle, negatively affecting N availability in grain production systems.

Immobilization occurs when decomposer microorganisms remove inorganic N from the soil, resulting in a decrease in N availability to plant species. The decomposition of straw from grass species (maize and/or *Urochloa*) results in increased immobilization of inorganic N and greater amounts of plant biomass on soil surface, due to its high C/N ratio. Fertilization is factor that can also influence the values of C/N ratio, as they alter C and N contents in the soil. Thus, the factors presented justify the immobilization of N observed in the aggregates of the grain production systems, especially in the surface Phy aggregates. It is worth pointing out that the crop of economic interest that preceded the sampling period was maize (grain and straw production) in the NT and NT+B systems and that in the NT+B area the sampling of soil clods was carried out 35 days after the desiccation of *Urochloa* (animal feed and soil protection). The values of the C/P ratio in Bio aggregates of the NT, NT+B and NF areas stood out, reflecting a less intense P mineralization.

The $_{OH}Pi$, $_{OH}Po$, and $_{OCL}P$ fractions showed the highest participation of TP contents in the Bio and Phy aggregates. Similar results were verified by Gatiboni and Condron (2021). However, the authors quantified higher values of proportion for the occluded fraction (65%) compared to those observed in the aggregates of this study (26 to 44%). The highest proportional values of the occluded fraction show the high P adsorption capacity in highly weathered soils (Gatiboni *et al.*, 2021). In the literature, only Moura *et al.* (2019) analysed the P contents in the different fractions in Bio and Phy aggregates in agricultural production areas under agroecological management in Southeast Brazil. The authors quantified the highest contents of organic P and inorganic P in the Phy aggregates. For them, the P contents observed in the different classes of aggregates show the important role of the formation pathways in the evaluation of the soil chemical quality.

The cluster analysis, integrating the indicator attributes, grouped the systems and the aggregate formation pathways into three distinct clusters. The greatest dissimilarities were observed between the soil use and management systems in the first two clusters, corroborating the results of the PCA. The third cluster separated Bio aggregates from Phy of the grain production systems. Recent studies have also reported the separation of aggregate formation pathways through hierarchical clustering analysis (Fonseca Júnior *et al.*, 2023; Rossi *et al.*, 2024), mainly in the surface soil layer. The results presented in Figure 3 show that: i) the clustering analysis technique was more efficient in separating and grouping the aggregate classes when compared to PCA; and ii) the contribution of the Bio pathway to the chemical quality of the soil may be more evident in conservation systems.

Conclusions

The chemical attributes analysed in the aggregates were affected differently by the soil management systems, especially PP (lower diversity of plant species) and NT+B (higher diversity of plant species) systems. The better chemical quality of the soil in the pasture, identified through aggregate analysis, was evident, regardless of its origin. This shows the beneficial effect of well-managed pastures on sandy-textured soils.

The positive influence of *Urochloa* along with maize in winter cultivation can be seen mainly in the subsurface layer. After six years of the intercropping, its aggregates mainly showed higher K^+ contents and an increase in moderately labile P and C values. The stoichiometric ratios showed different patterns in the mechanisms of balance and cycling of nutrients in subsurface, especially the C/N ratio in the aggregates of the conservation systems.

We found that Bio aggregation promoted a greater accumulation of certain nutrients, contributing to an increase in soil fertility. It should be noted that this benefit was observed more intensely in the first 0.05 m of soil depth in the conservation systems. The results verified for Bio aggregates strengthen the hypothesis that these structural units are important sources of nutrients for the soil and reiterate the importance of studying the formation pathways in assessment soil health, especially in monitoring the chemical quality of the arable layer of texturally fragile soils.

Supplementary material. The supplementary material for this article can be found at https://doi.org/10.1017/S0014479725000109

Acknowledgements. The authors acknowledge the support from CAPES, CNPq, FAPERJ, PPGA-CS/UFRRJ, UEMS, and AGRISUS (PA 2990/20).

Competing interests. The authors declare no competing interests. There are no financial or personal relationships that could have influenced the work reported in this paper.

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Cite this article: Pinto LASR, Ferreira R, Lemos GCO, Lopes IS, Ozório JMB, Rosset JS, Melo TR, and Pereira MG. Management systems in the subtropical region of Brazil: aggregation in the assessment of the chemical quality of fragile soils. *Experimental Agriculture*. https://doi.org/10.1017/S0014479725000109