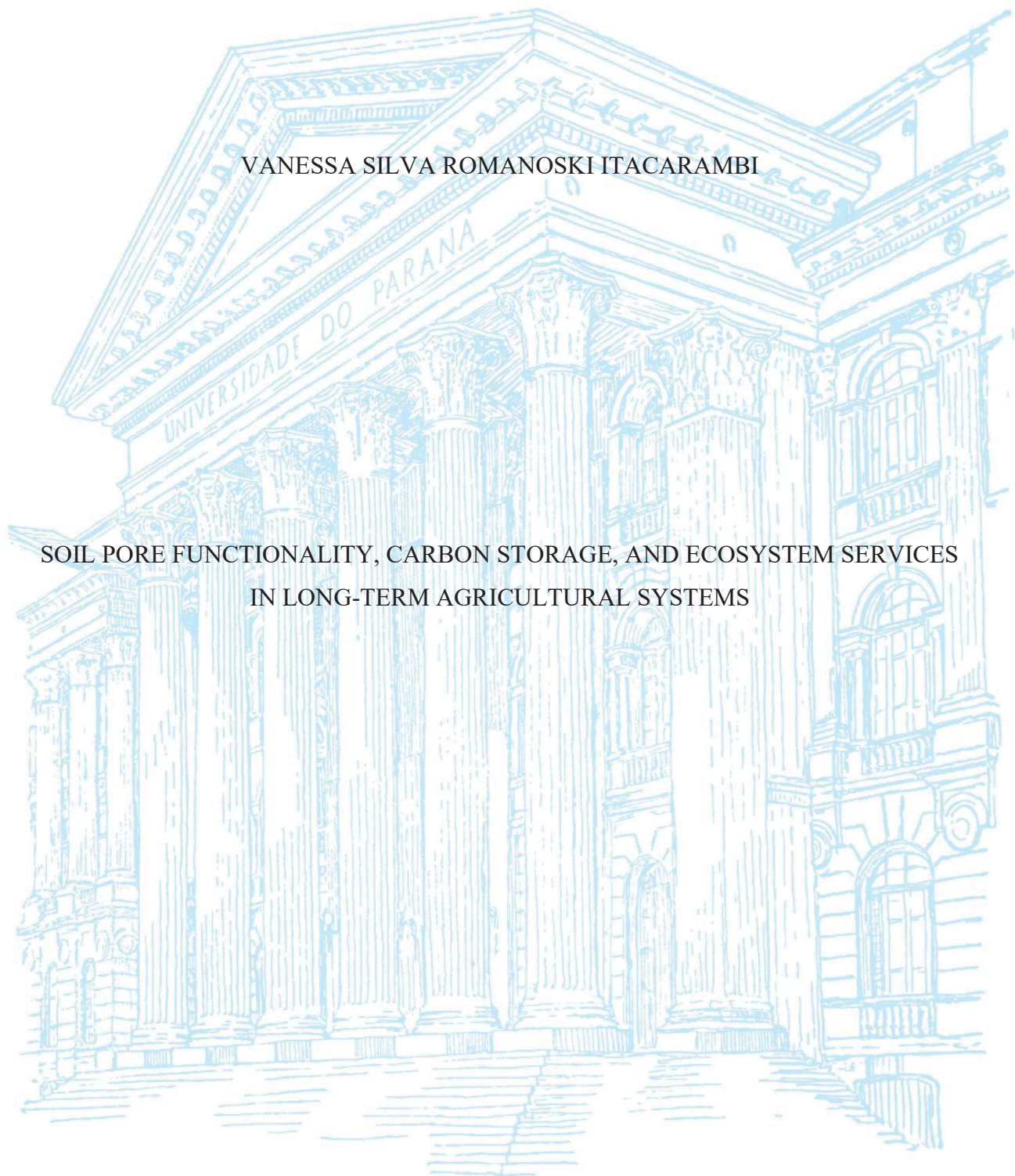


UNIVERSIDADE FEDERAL DO PARANÁ

VANESSA SILVA ROMANOSKI ITACARAMBI

SOIL PORE FUNCTIONALITY, CARBON STORAGE, AND ECOSYSTEM SERVICES
IN LONG-TERM AGRICULTURAL SYSTEMS



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VANESSA SILVA ROMANOSKI ITACARAMBI

SOIL PORE FUNCTIONALITY, CARBON STORAGE, AND ECOSYSTEM SERVICES
IN LONG-TERM AGRICULTURAL SYSTEMS

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RESUMO

A intensificação sustentável da agricultura, através de sistemas integrados e conservacionistas, é fundamental para restaurar a funcionalidade do solo e mitigar as mudanças climáticas. O objetivo geral desta tese foi analisar a interdependência entre a funcionalidade do sistema de poros do solo, os estoques de carbono orgânico do solo e os serviços ecossistêmicos sob estratégias de manejo de conservação de longo prazo. Para atingir esse objetivo, a pesquisa abrangeu a avaliação de sistemas agrícolas integrados no sul do Brasil, bem como o uso de culturas de cobertura (>30 anos) em solos arenosos temperados na Suécia. A pesquisa foi estruturada em dois estudos de longa duração. O primeiro, realizado no Sul do Brasil (clima subtropical) em um Latossolo de textura argilosa, avaliou sistemas integrados de produção agropecuária (Pecuária, Lavoura-Pecuária, Pecuária-Floresta e Lavoura-Pecuária-Floresta) sob plantio direto. O segundo, conduzido no Sudoeste da Suécia (clima temperado) em solos de textura arenosa, investigou o impacto de culturas de cobertura (*Lolium perenne*) versus solo descoberto. No primeiro capítulo (publicado) os sistemas Lavoura-Pecuária e Lavoura-Pecuária-Floresta aumentaram a condutividade hidráulica saturada (Ks) e a permeabilidade ao ar (Ka) em um Latossolo argiloso. No segundo capítulo, calculou-se o Índice de Serviços Ecossistêmicos (ISE) para avaliar a oferta de serviços em sistemas pecuários e pecuários-florestais, visando quantificar, por meio de uma abordagem integrada, os benefícios e os *trade-offs* decorrentes da presença de árvores nas pastagens. A aplicação deste índice demonstrou que a integração do componente arbóreo eleva a provisão global de serviços, aumentando o ISE de 0,52 (Pecuária) para 0,72 (Pecuária-Floresta). Evidenciou-se que eventuais reduções na provisão forrageira provocadas pelo sombreamento são funcional e amplamente compensadas pela produção de madeira e, principalmente, pela adicionalidade no sequestro de carbono, o que impulsiona significativamente os serviços de regulação climática. No âmbito dos processos físicos (Capítulo 3), observou-se uma relação negativa entre o carbono orgânico e os fluxos de água e ar, sugerindo que a aeração excessiva pode acelerar a mineralização da matéria orgânica. Já no estudo sueco (Capítulo 4), em solos arenosos vulneráveis, o manejo com culturas de cobertura (*Lolium perenne*) provou ser favorável ao aporte de carbono orgânico no solo e a retenção de água via microporosidade. Conclui-se que a adoção de sistemas integrados melhora as propriedades físicas do solo e potencializa serviços ambientais, demonstrando que a diversidade biológica atua como reguladora fundamental da estrutura dos poros. Dessa forma, o manejo conservacionista assegura o equilíbrio entre a funcionalidade hidráulica e a preservação do carbono, promovendo a resiliência dos sistemas agrícolas frente aos desafios climáticos globais.

Palavras-chave: Carbono Orgânico do Solo; Estrutura do Solo; Conectividade da Porosidade do Solo; Geometria da Porosidade do Solo; Permeabilidade ao Ar; Condutividade Hidráulica; Agricultura Conservacionista.

ABSTRACT

Sustainable agricultural intensification, through integrated and conservation systems, is fundamental to restoring soil functionality and mitigating climate change. The general objective of this thesis was to analyze the interdependence among soil pore system functionality, soil organic carbon stocks, and ecosystem services under long-term conservation management strategies. To achieve this, the research encompassed the evaluation of integrated agricultural systems in Southern Brazil, as well as the use of cover crops (>30 years) in temperate sandy soils in Sweden. The research was structured into two long-term studies. The first, conducted in Southern Brazil (subtropical climate) on a clayey Ferralsol, evaluated integrated crop-livestock systems (Livestock, Crop-Livestock, Livestock-Forestry and Crop-Livestock-Forestry) under no-tillage. The second, conducted in Southwestern Sweden (temperate climate) on sandy soils, investigated the impact of cover crops (*Lolium perenne*) versus bare soil. In the first chapter (published), the Crop-Livestock and Crop-Livestock-Forestry systems increased saturated hydraulic conductivity (Ks) and air permeability (Ka) in a clayey Ferralsol. In the second chapter, the Ecosystem Services Index (ESI) was calculated to evaluate the provision of services in livestock and livestock-forestry systems, aiming to quantify, through an integrated approach, the benefits and trade-offs arising from the presence of trees in pastures. The application of this index demonstrated that the integration of the tree component enhances the overall provision of services, increasing the ESI from 0.52 (Livestock) to 0.72 (Livestock-Forestry). It was evidenced that any reductions in forage provision caused by shading are functionally and largely offset by wood production and, most notably, by the additionality in carbon sequestration, which significantly boosts climate regulation services. Regarding physical processes (Chapter 3), a negative relationship was observed between organic carbon and water and air fluxes, suggesting that excessive aeration may accelerate organic matter mineralization. In the Swedish study (Chapter 4), on vulnerable sandy soils, management with cover crops (*Lolium perenne*) proved favorable for soil organic carbon input and water retention via microporosity. It is concluded that the adoption of integrated systems improves soil physical properties and enhances ecosystem services, demonstrating that biological diversity acts as a fundamental regulator of pore structure. Thus, conservation management ensures the balance between hydraulic functionality and carbon preservation, promoting the resilience of agricultural systems against global climate challenges.

Keywords: Soil Organic Carbon; Soil Structure; Soil Porosity Connectivity; Soil Porosity Geometry; Air Permeability; Hydraulic Conductivity; Conservation Agriculture.

LIST OF FIGURES

Figure 1 - Timeline of the experiment conducted between 2013 and 2021 to evaluate regenerative systems.

Figure 2 - Principal Component Analysis (PCA) of soil attributes under regenerative systems: Livestock (L), Crop-Livestock (CL), Livestock-Forestry (LF), Crop-Livestock-Forestry (CLF) at depths of 0.00-0.30 m, with cluster ellipses obtained by hierarchical clustering.

Figure 3 - Dendrogram resulting from the hierarchical clustering analysis of soil attributes under regenerative systems: Livestock (L), Crop-Livestock (CL), Livestock-Forestry (LF), and Crop-Livestock-Forestry (CLF), at a depth of 0.00-0.30 m.

Figure 4 - Soil physical indicators at different depths for livestock (solid line) and livestock-forestry (dashed line) systems.

Figure 5 - Corrected carbon stock (Cst, Mg ha⁻¹) in livestock and livestock-forestry systems at three cumulative depths (0.00-0.20 m, 0.00-0.30 m, and 0.00-1.00 m).

Figure 6 - Indicators of ecosystem services assessed in livestock and livestock-forestry systems.

Figure 7 - Integrated assessment of ecosystem services in the livestock and livestock-forestry systems.

Figure 8 - Relationship between saturated hydraulic conductivity (Ks) and soil total porosity, TP (A); Microporosity, MIC (B); Large macroporosity, MAC (C) and Macroporosity, MaP (D), at depths of 0.00-0.05; 0.05-0.15 and 0.15-0.30 m.

Figure 9 - Regression between saturated hydraulic conductivity (Ks, cm h⁻¹) and pore organization (k₁), connectivity (k₂) indexes for pores >100 μm (MAC), >30 μm (MaP) at depths of 0.00-0.05, 0.05-0.15, and 0.15-0.30 m.

Figure 10 - Regression analysis between intrinsic air permeability of soil Ka_(MAC) and Ka_(MaP) (μm²) with large macroporosity (MAC) and macroporosity (MaP), at depths of 0.00-0.05; 0.05-0.15 and 0.15-0.30 m.

Figure 11 - Regression analysis between saturated hydraulic conductivity, Ks; intrinsic air permeability of soil, Ka_(MAC) and Ka_(MaP), in pores with diameters > 100 and > 30 μm, respectively, and the soil organic carbon, SOC (g kg⁻¹), at depths of 0.00-0.05; 0.05-0.15 and 0.15-0.30 m.

Figure 12 - Principal Component Analysis (PCA) of soil attributes for soil management systems with and without cover crops at Fotegården (0.00-0.20 m depth).

Figure 13 - Principal Component Analysis (PCA) of soil attributes for soil management systems with and without cover crops at Fotegården (0.30-0.50 m depth).

Figure 14 - Principal Component Analysis (PCA) of soil attributes for soil management systems with and without cover crops at Mellby (0.00-0.20 m depth).

Figure 15 - Principal Component Analysis (PCA) of soil attributes for soil management systems with and without cover crops at Mellby (0.30-0.50 m depth).

LIST OF TABLES

Table 1 - Weighted mean values ($n = 36, \pm SD$) of general attributes of the systems: Livestock (L), Crop-Livestock (CL), Livestock-Forestry (LF), Crop-Livestock-Forestry (CLF) at depths of 0.00-0.30 m.

Table 2 - Summary of Multivariate Analysis of Variance (MANOVA) using the Pillai test for systems comparison: Livestock (L), Crop-Livestock (CL), Livestock-Forestry (LF), Crop-Livestock-Forestry (CLF) at depths of 0.00-0.30 m.

Table 3 - Summary of Eigenvalues, Eigenvectors, and Cumulative Variance of Principal Component Analysis (PCA) for the systems: Livestock (L), Crop-Livestock (CL), Livestock-Forestry (LF), Crop-Livestock-Forestry (CLF) at depths of 0.00-0.30 m.

Table 4 - Indicators, functional categories, and weights used to calculate the Ecosystem Services Index (ESI).

Table 5 - Indicator thresholds and scoring curves for the assessment of ecosystem services

Table 6 - Sward structure, forage accumulation, and animal performance in livestock and livestock-forestry systems.

Table 7 - General characterization attributes related to carbon stock; water and air flows; soil bulk density, porosity and functionality of the soil pore system in conservationist systems.

Table 8 - Linear regression between saturated hydraulic conductivity, ($K_s, \text{cm h}^{-1}$) and soil total porosity, TP; Microporosity, MIC; Large macroporosity, MAC and macroporosity, MaP.

Table 9 - Regression analysis equation between saturated hydraulic conductivity, ($K_s, \text{cm h}^{-1}$) and pore organization (k_1) and connectivity (k_2) indexes.

Table 10 - Regression analysis equation between intrinsic air permeability of the soil $K_{a(\text{MAC})}$ and $K_{a(\text{MaP})}$ and large macroporosity (MAC) and macroporosity (MaP).

Table 11 - Regression analysis equation between saturated hydraulic conductivity, $K_s (\text{cm h}^{-1})$; intrinsic air permeability... and of the soil organic carbon, SOC (g kg^{-1}).

Supplementary Table 3. Soil texture in (g kg^{-1}) and particle density (ρ_p) in (Mg m^{-3}), for depths of 0.00-0.05; 0.05-0.15 and 0.15-0.30 m.

Table 12 - Mean values \pm standard deviation ($n = 15, \pm SD$) of soil attributes under management systems with and without cover crops at Fotegården (0.00-0.20 m and 0.30-0.50 m depths).

Table 13 - Summary of Multivariate Analysis of Variance (MANOVA) using the Pillai test for soil management systems with and without cover crops at Fotegården (0.00-0.20 m and 0.30-0.50 m depths).

Table 14 - Summary of Eigenvalues, eigenvectors, and cumulative variance from Principal Component Analysis (PCA) of soil management systems with and without cover crops at Fotegården.

Table 15 - Mean values \pm standard deviation ($n = 15, \pm SD$) of soil attributes under management systems with and without cover crops at Mellby (0.00-0.20 m and 0.30-0.50 m depths).

Table 16 - Summary of Multivariate Analysis of Variance (MANOVA) using the Pillai test for soil management systems with and without cover crops at Mellby (0.00-0.20 m and 0.30-0.50 m depths).

Table 17 - Summary of Eigenvalues, eigenvectors, and cumulative variance from Principal Component Analysis (PCA) of soil management systems with and without cover crops at Mellby.

LIST OF ABBREVIATIONS AND ACRONYMS

ADG	Average Daily Gain
AGB	Above-Ground Biomass
ANOVA	Analysis of Variance
BD	Bulk Density
BGB	Below-Ground Biomass
CAPES	Coordination for the Improvement of Higher Education Personnel
CL	Crop-Livestock
CLF	Crop-Livestock-Forest
CNPq	National Council for Scientific and Technological Development
Cst	Soil Carbon Stock
CT	Tree Carbon Stocks
CV	Coefficient of Variation
DBH	Diameter at Breast Height
DWG	Daily Live Weight Gain
ESPI	Ecosystem Services Provision Index
IPCC	Intergovernmental Panel on Climate Change
K	Hydraulic Conductivity
Ka	Intrinsic Air Permeability of Soil
Ks	Saturated Hydraulic Conductivity
L	Livestock
LF	Livestock-Forestry
MAC	Large Macroporosity (pores > 100 μm)
MAI	Mean Annual Increment
MANOVA	Multivariate Analysis of Variance
Ks	Saturated Hydraulic Conductivity
L	Livestock
MaP	Macroporosity (pores > 30 μm)
MIC	Microporosity (pores < 30 μm)
NITA	Center for Technological Innovation in Agriculture
PAWC	Plant-Available Water Capacity
PCA	Principal Component Analysis
PR	Penetration Resistance

RMSE	Root Mean Square Error
SCS	Soil Carbon Stocks
SD	Standard Deviation
SIPA	Integrated Crop-Livestock Systems (Portuguese acronym)
SLU	Swedish University of Agricultural Sciences
SOC	Soil Organic Carbon
TC	Total Carbon
THA	Total Herbage Accumulated
TP	Total Porosity
UFPR	Federal University of Paraná

SUMÁRIO

1 GENERAL INTRODUCTION	19
2 LITERATURE REVIEW	22
2.1 INTEGRATED PRODUCTION SYSTEMS	22
2.2 PORE FUNCTIONALITY AND SOIL PHYSICAL QUALITY	23
2.3 SOIL CARBON STOCKS IN THE SYSTEM	24
2.4 ECOSYSTEM SERVICES AND TRADE-OFF ANALYSIS	25
2.5 REFERENCES	26
3 CHAPTER 1 - DYNAMIC SOIL HYDRAULIC PROPERTIES IN REGENERATIVE AGRICULTURE: EFFECTS OF CROP AND FOREST INTEGRATION IN LIVESTOCK SYSTEMS	31
3.1 ABSTRACT	31
3.2 INTRODUCTION	32
3.3 MATERIAL AND METHODS.....	33
3.3.1 Experimental site and area.....	33
3.3.2 History, experiment management and treatments	33
3.3.3 Soil sampling	35
3.3.4 Soil assessments	35
3.3.5 Soil hydraulic properties.....	36
3.3.6 Intrinsic air permeability of soil	36
3.3.7 Assessment of soil porosity, organization and connectivity indexes, plan-available water capacity, bulk density and soil texture.....	37
3.3.8 Soil Carbon Stock (Cst).....	37
3.3.9 Statistical analyses	38
3.4 RESULTS	39
3.5 DISCUSSION.....	43
3.6 CONCLUSION	47
3.7 REFERENCES	47
4 CHAPTER 2 – A LONG-TERM LIVESTOCK-FORESTRY SYSTEM IMPROVES ECOSYSTEM SERVICES IN A SUBTROPICAL REGION.....	52
4.1 ABSTRACT	52
4.2 INTRODUCTION	53
4.3 MATERIAL AND METHODS.....	54
4.3.1 Study site	54

4.3.2 Experimental design and treatment description.....	55
4.3.3 Soil characterization	55
4.3.4 Quantification of soil organic carbon stock.....	56
4.3.5 Assessment of forage biomass, pasture structure, and animal performance	57
4.3.6 Performance assessment of tree.....	58
4.3.7 Ecosystem services index calculation.....	59
4.3.8 Statistical analyses.....	61
4.4 RESULTS.....	62
4.4.1 Soil physical quality indicators.....	62
4.4.2 Soil and tree carbon stocks	64
4.4.3 Livestock and livestock-forestry system production.....	65
4.4.4 Ecosystem services index	66
4.5 DISCUSSION.....	68
4.5.1 Performance of ecosystem services indicators	68
4.5.2 Ecosystem services synergies and trade-offs between livestock and livestock-forestry systems	69
4.5.3 Challenges, opportunities, and limitations of the study.....	72
4.6 CONCLUSION	73
4.7 REFERENCES	73
5 CHAPTER 3 - THE TRADE-OFF BETWEEN FLUID TRANSPORT PROPERTIES AND ORGANIC CARBON PRESERVATION IN CLAYEY SOILS.....	78
5.1 ABSTRACT	78
5.2 INTRODUCTION	79
5.3 MATERIAL AND METHODS.....	80
5.3.1 Study Area.....	80
5.3.2 Site History, Use and Management	81
5.3.3 Soil Sampling	82
5.3.4 Soil Assessments	82
5.3.5 Statistical analysis	83
5.6 RESULTS.....	84
5.6.1 General Characterization of Soil Attributes	84
5.6.2 Relationship between Soil Hydraulic Properties, Pore System, and Soil Organic Carbon (SOC)	85
5.7 DISCUSSION.....	90

5.8 CONCLUSION	94
5.9 SUPPLEMENTARY MATERIAL	94
5.10 REFERENCES	95
6 CHAPTER 4 - LONG-TERM COVER CROPS IMPROVE SOC AND PHYSICAL PROPERTIES IN SANDY SOILS.....	99
6.1 ABSTRACT	99
6.2 INTRODUCTION	100
6.3 MATERIAL AND METHODS.....	101
6.3.1 Experimental site and area.....	101
6.3.2 History, experiment management, and treatments	102
6.3.3 Soil sampling.....	103
6.3.4 Statistical analyses.....	104
6.4 RESULTS.....	104
6.5 DISCUSSION.....	112
6.6 CONCLUSION	115
6.7 REFERENCES	115
7 GENERAL CONCLUSIONS.....	118
8 REFERENCES	119
9 SOCIAL IMPACT OF RESEARCH.....	129
10 APPENDIX: TECHNICAL PRODUCTION – ORGANIZED EVENT	131
11 ANNEX – PROOF OF EXTENSION ACTIVITIES AND EVENT ORGANIZATION 133	
12 BIOGRAPHICAL SKETCH.....	137

1 GENERAL INTRODUCTION

The sustainable intensification of contemporary agriculture is grounded in the transition from conventional monocultures to more complex and resilient productive arrangements, such as Integrated Crop-Livestock Systems (ICLS) (Lemaire et al., 2014; Moraes et al., 2014). These systems, anchored in the principles of conservation and regenerative agriculture, seek to mimic natural processes through the spatial and temporal diversification of species, integrating agricultural, livestock, and forestry components within the same area and space (Kassam et al., 2019). By promoting synergism among system components, these strategies aim not only at the production of food, fibers, and energy but also at the restoration of ecosystem functionality, reducing dependence on external inputs and mitigating the impacts of global climate change (Lal, 2020).

The adoption of conservation practices in integrated systems plays a crucial role in improving soil structural quality, favoring the formation of a complex and functional pore architecture (Rabot et al., 2018). This physical reorganization is determinant for the dynamics of water and air fluxes, directly influencing the soil's capacity to act as a sink or source of greenhouse gases (Keller et al., 2017). Concomitantly, the continuous biomass input and diversified root activity enhance soil organic carbon (SOC) sequestration (Paustian et al., 2016; Rasse et al., 2005). The interdependence between a preserved physical structure and carbon stock is the basis for the provision of multiple ecosystem services, including hydrological cycle regulation, nutrient cycling, and the maintenance of soil biodiversity, transcending the classical productivist view towards a multifunctionality approach of the rural landscape (Power, 2010; Robinson et al., 2013).

In this context, biological management, especially through the use of cover crops and perennial pastures, emerges as the primary driver of edaphic transformations (Poeplau et al., 2015). The constant presence of living roots acts on soil biostructuring, creating preferential channels for fluid flow and protecting organic matter against excessive mineralization (Meurer et al., 2020). Whether in tropical clayey soils or temperate sandy soils, cover crop management is decisive in modulating the interaction between soil physics and carbon chemistry, defining system resilience against water and mechanical stresses (Basche & DeLonge, 2019; Wittwer et al., 2017).

The central hypothesis of this thesis was that agricultural systems based on conservation practices, such as Integrated Crop-Livestock-Forestry (ICLF) systems in tropical clayey soils or the use of cover crops in temperate sandy soils, promote a functional reorganization of the soil pore architecture. It was hypothesized that this structure acts as the main regulator of the

relationship between fluid transport (water and air fluxes) and soil organic carbon preservation. Consequently, management strategies that prioritize conservation practices not only improve physical properties but also maximize the provision of ecosystem services, creating resilient soil systems capable of sustaining productivity and environmental functions in contrasting edaphoclimatic scenarios. The general objective of this thesis was to analyze the interdependence between soil pore system functionality, soil organic carbon stocks, and the provision of ecosystem services under conservation management strategies in integrated agricultural systems in long-term experiments in Southern Brazil, and the use of cover crops in temperate sandy soils in Sweden.

This thesis is structured into four chapters that investigate these dynamics under different perspectives and scales. The first chapter, published in the journal *Soil and Tillage Research* (Volume 253, 2025, Article 106680), addresses the limited understanding of the effects of saturated hydraulic conductivity (Ks) and intrinsic air permeability in regenerative systems, as well as their relationship with carbon stocks. This study investigates and tests the hypothesis that integrated systems exhibit better soil hydraulic properties compared to an exclusive Livestock system under regenerative practices. Specifically, the aim of this first chapter was to compare three different integrated arrangements (Crop-Livestock, Livestock-Forest, and Crop-Livestock-Forest) with a single Livestock system, evaluating regenerative agricultural practices and investigating how these integrations affect soil structure, with an emphasis on soil hydraulic properties.

Given the physical similarity observed between the exclusive Livestock and the Livestock-Forest systems in the first study, the second chapter sought to investigate whether the introduction of the tree component, even without drastically altering hydraulic properties, would bring benefits in ecosystem services. To this end, the provision of ecosystem services was evaluated through an integrated approach, quantifying the trade-offs and synergies between forage production, timber, and regulation and support services. This chapter expands the purely physical analysis to a holistic view of the sustainability of productive arrangements.

The third chapter returns to the analysis of physical processes, utilizing the same long-term experiment presented in Chapter 1. However, the approach here is distinct and in-depth: it sought to understand the functional mechanisms governing the interaction between soil structure and carbon. Through regression analyses and pore connectivity indices, this chapter investigates the trade-off between increased aeration/flux and organic carbon preservation, elucidating how pore architecture (macro and microporosity) modulates organic matter stability in a subtropical clayey Ferralsol.

The third chapter is grounded in the hypothesis that fluid transport capacity (K_s and K_a) in these soils is primarily governed by the abundance and continuity of large macropores (MAC), rather than total porosity; however, we postulate a functional trade-off where the same organized and connected pore network that favors high permeability to water and air also intensifies oxidative processes, leading to decrease in soil organic carbon (SOC). Thus, the objective of this work was to analyze the interrelationships between soil fluid transport properties (saturated hydraulic conductivity, K_s and air permeability, K_a), pore system attributes (macroporosity, mesoporosity, microporosity and organization and connectivity indexes), as well soil organic carbon content.

The fourth chapter expands the frontier of knowledge to a contrasting edaphoclimatic scenario, evaluating the impact of cover crops (*Lolium perenne*) versus bare soil in coarse-textured soils. This study is the result of a doctoral internship abroad, conducted at the Swedish University of Agricultural Sciences (SLU) in Uppsala, from May to October 2024. Under the supervision of Dr. Lorena Chagas Torres and Professor Dr. Thomas Keller, in the Department of Soil and Environment (Soil Mechanics and Soil Management Group), this chapter investigates how biological management acts to mitigate drainage and aid in physical structure in temperate sandy soils, complementing the thesis's perspective on the universality and specificities of conservation management.

2 LITERATURE REVIEW

2.1 INTEGRATED PRODUCTION SYSTEMS

The transition from conventional agricultural models to integrated production systems represents an essential technological advancement for global sustainability and improved agricultural governance (Rodrigues et al., 2023). These productive arrangements are classified into different modalities, depending on the biological components intentionally inserted into the same area (Vilela et al., 2021). Crop-Livestock Integration (CLI) consists of the intercropping, succession, or rotation of crops with pastures, aiming for nature-based solutions for income diversification and efficient nutrient cycling (Carvalho et al., 2024). In turn, Livestock-Forestry Integration (LFI) combines trees and pastures, offering superior thermal comfort and directly influencing the adaptation and well-being of the herd (Martin et al., 2025). Full mastery of these definitions allows for proper rural landscape planning to maximize efficiency in emission mitigation and land use (Monteiro et al., 2024).

Crop-Forestry Integration (CFI) is structured by the intercropping of tree species with agricultural crops, optimizing tree growth and wood density in the same space as the crops (Kruchelski et al., 2021). The maximum level of ecophysiological and operational complexity is reached in Crop-Livestock-Forestry Integration (CLFI), which simultaneously encompasses agricultural, livestock, and forestry components (Corte et al., 2022). The synergy generated in these systems promotes the recovery of soil multifunctionality and degraded pastures, one of the greatest challenges of modern agriculture in tropical ecosystems (Silva-Olaya et al., 2025). Furthermore, the insertion of the tree component acts as a vital support for natural capital, altering the dynamics of the managed area (England et al., 2020). The adoption of these complex technologies mitigates environmental liabilities and reduces greenhouse gas emissions compared to traditional systems (Amadori et al., 2022).

The complexity of these integrated systems is intensely reflected in the interactions below the soil surface, where the roots of different species explore edaphic profiles and affect structural quality (Cavaliere-Polizeli et al., 2022). The deep and vigorous root system interacts strongly with soil biology, promoting essential modifications in cover and fungal dynamics (Silva et al., 2020). Simultaneously, conservationist systems ensure a constant addition of residues and exudates that stimulate soil organic carbon storage and soil quality in the surface layer (Cavaliere-Polizeli et al., 2024a). This functional complementarity of ecological niches is fundamental for restoring carbon stocks and ensuring agronomic resilience in the face of climate change (Moraes Sá et al., 2025). Therefore, the sustainable intensification enabled by

degradation-free ecosystems ensures productive stability and the mitigation of greenhouse gases (Portugal et al., 2023).

2.2 PORE FUNCTIONALITY AND SOIL PHYSICAL QUALITY

Soil physical quality is frequently monitored through dynamic hydraulic properties, such as saturated hydraulic conductivity (K_s) (Pachepsky & Park, 2015). K_s reflects the ease with which water moves through the pore system under saturated conditions, being strongly dependent on the degree of compaction and soil bulk density (Reichert et al., 2009). In systems with animal presence, continuous trampling and the load exerted by cattle can reduce surface macroporosity and alter soil structure (Negrón et al., 2019). On the other hand, air permeability acts as a sensitive indicator of the continuity of drainable pores, essential for compressive behavior and physical functionality under grazing (Suzuki et al., 2022). The joint evaluation of these properties provides a mechanistic overview of long-term changes in integrated agricultural systems (Nascimento et al., 2019).

To move beyond the isolated evaluation of static porosity and understand fluid dynamics, literature has adopted advanced analyses focused on structural organization and the connectivity of the pore system (Hamamoto et al., 2016). Connectivity quantifies the degree of functional interconnection of the soil air space, being a determining factor for the efficiency of gas transport properties in the edaphic profile under no-till (Silva et al., 2021). Simultaneously, tortuosity describes the complexity of preferential flow paths, directly influencing the resistance to air and water movement in the porous matrix (Poulsen, 2015). Organization indices and pore volume characteristics allow for the differentiation of systems that diverge in their conduction capacity, even when possessing similar densities (Reynolds et al., 2009). Consequently, maintaining a network of functional and dynamic pores ensures structural resilience against long-term mechanical stresses (Cavalieri et al., 2009).

In Livestock-Forestry systems, the roots of the species act directly on the functioning of the pore system, with the input of active biomass creating interconnected biopores (Hernandez Gamboa et al., 2023). After long-term interactions, these dynamics restructure preferential pathways for water infiltration and gas exchange within the scope of conservation agriculture (Romanoski et al., 2025). The simultaneous presence of the tree and forage components works in favor of aggregate stabilization and the preservation of soil quality (Valani et al., 2022). Thus, the benefits of biological structuring and adequate pore distribution far outweigh the pressure exerted by conventional surface management (Cassaro et al., 2011).

2.3 SOIL CARBON STOCKS IN THE SYSTEM

The inclusion of cover crops during off-seasons or rotation cycles is a strategy to effectively increase soil organic carbon stocks in the long term (Poeplau et al., 2015). These plant species produce a high amount of phytomass, which protects the surface and reduces the loss of nutrients such as nitrogen and phosphorus (Aronsson et al., 2016). The root system of these crops can offset residue removal, facilitating the uninterrupted chemical and physical improvement of the edaphic profile (Ruis & Blanco-Canqui, 2017). The physical occlusion of organic matter within well-formed aggregates protects carbon from accelerated microbial decomposition and prevents losses in the form of carbon dioxide (Barreto et al., 2009). Thus, cover crops function as essential biological tools for the continuous optimization of the edaphic environment (Wanic et al., 2019).

The temporal dynamics of carbon sequestration are strongly influenced by residue addition rates and the control generated by species diversity in crop rotation (Liu et al., 2022). In conservationist systems, the dynamic interactions between soil structure and organic matter dictate the capacity for storage and biological renewal (Meurer et al., 2020). Systems that favor the constant input of biomass ensure higher levels of organic carbon and promote remarkable structural quality compared to degraded soils (Cavaliere-Polizeli et al., 2024b). In contrast, management and local edaphoclimatic conditions are decisive factors governing variations in carbon dynamics, especially in pasture areas (Daboit et al., 2025). This balance, when well-managed, is the engine that mitigates environmental risks in scenarios of prolonged climate change (Nimblad Svensson et al., 2026).

In addition to processes restricted to the soil, the tree component of forestry systems immobilizes significant volumes of carbon in its structural aboveground biomass (Machado et al., 2025). The continuous phenological growth of trees is directly impacted by management practices thirty years after interventions, establishing a continuous pathway for atmospheric carbon removal (Vidal et al., 2026). The precise quantification of this stored carbon often requires linking the functional traits of the forest understory and canopy biomass (Teixeira et al., 2025). The integration of these components adds significant aboveground carbon stocks, an effect also evidenced in forest areas of high taxonomic complexity (Antunes et al., 2025). In this way, integration elevates systemic carbon provision to levels significantly higher than those found in simplified systems (Cá et al., 2022).

2.4 ECOSYSTEM SERVICES AND TRADE-OFF ANALYSIS

The modern methodological transition to the quantification of the Ecosystem Services Index (ESI) has allowed for diagnosing the condition of the soil resource and the real environmental provision provided by integration-based systems (Valani et al., 2023). This numerical index groups physical and biological services, comparatively evaluating areas managed under conservation (Valani et al., 2022). Food provision is the central base in purely productive metrics, with conservation agriculture requiring the soil to act as this essential pillar (Schreefel et al., 2020). However, the economic realities of conservation agriculture point out that long-term sustainability requires the valuation of services that go beyond the simple commodity market (Ikerd, 2021). The empirical application of conservationist metrics proves that the adoption of these practices generates effects for the biodiversity of farmlands (Svoboda et al., 2026).

The implementation of silvopastoral models imposes the occurrence of trade-offs resulting from productive interactions, directly affecting the productivity and growth of intercropped trees (Kruchelski et al., 2023). Competition in space promotes changes in weed infestation patterns in the understory, requiring complex rotations for the maintenance of productive temporary pastures (Dominschek et al., 2021). Although shading generates restrictions, the system compensates by promoting comfort and radically improving animal adaptation under grazing (Martin et al., 2025). Despite potential punctual losses in forage provision, there is a simultaneous gain in the harvest and density of the wood produced (Kruchelski et al., 2021). Understanding these sustainability challenges quantitatively is indispensable for promoting governance and innovation in these integrated models (Rodrigues et al., 2023).

The compensation for primary production losses occurs through superior climatic advantages, transforming no-till-based systems into vital solutions for the restoration of carbon stocks (Moraes Sá et al., 2025). The final greenhouse gas tally attests to significant reductions in nitrous oxide and methane emissions in integrated properties in southern Brazil (Amadori et al., 2022). Additionally, practices such as the use of winter cover crops prevent risks of nitrate leaching, preserving the quality of water resources as a supporting service (Vogeler et al., 2023). The synergistic interaction confirms that carbon farming practices positively affect the soil and ensure global economic stability (Pettersson et al., 2025). Ultimately, correct valuation proves that ecosystem processes are the definitive engines for generating and sustaining global conservation agriculture (Garbisu et al., 2025).

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3 CHAPTER 1 - DYNAMIC SOIL HYDRAULIC PROPERTIES IN REGENERATIVE AGRICULTURE: EFFECTS OF CROP AND FOREST INTEGRATION IN LIVESTOCK SYSTEMS ¹

3.1 ABSTRACT

Regenerative agriculture has been enhanced worldwide to improve not only the environmental but also the social and economic dimensions of sustainable food production. The objective of this study was to compare three different integrated livestock systems against a single livestock system under regenerative agriculture practices, investigating how the integrations affect soil structure, with emphasis on soil hydraulic properties. The experiment was established in 2012 on clayey soil at the Canguiri Experimental Station, Pinhais municipality, PR, Brazil. The experimental design was a randomized block with three blocks and four treatments: Livestock (L), Crop-Livestock (CL), Livestock-Forest (LF), and Crop-Livestock-Forest (CLF). All systems are managed pesticide-free and according to conservation tillage principles. The samples were collected in 2022, down to 0.30 m. Soil attributes analyzed included saturated hydraulic conductivity (K_s), intrinsic air permeability of soil (K_a) at matric potentials -3 kPa and -10 kPa, soil bulk density, total porosity, porosity of large macropores (MAC; $\varnothing > 100 \mu\text{m}$), macroporosity (MaP; $\varnothing > 30 \mu\text{m}$), microporosity (MIC; $\varnothing < 30 \mu\text{m}$), plant-available water (PAWC), air-filled porosity, organization and connectivity indexes and soil carbon stock (Cst). Agricultural systems such as Crop-Livestock (CL) and Crop-Livestock-Forest (CLF) exhibit better soil hydraulic properties compared to Livestock (L). The L and LF systems showed similarities and maintained adequate structural characteristics. The analysis revealed the formation of two clusters: the first, consisting of the CL (Crop-Livestock) and CLF (Crop-Livestock-Forest) systems, demonstrated a positive association among K_s , $K_{a(\text{MAC})}$, $K_{a(\text{MaP})}$, Cst and MIC. The second cluster, which includes the L (Livestock) and LF (Livestock-Forest) systems, suggested a possible relationship between BD and MaP. The quality of the cluster analysis was considered robust, with a cophenetic correlation of 0.75. The statistical tests indicated that these differences were significant ($p < 0.05$). These findings highlight that integrating cropland with livestock, as in CL and CLF systems, enhances soil structure and pore functionality while maintaining carbon stocks, contributing to long-term sustainability.

Key words: Soil Pore Functionality; Soil Porosity Connectivity; Soil Porosity Geometry; Conservation Agriculture; Best Soil Management Practices

¹ Paper published in Soil and Tillage Research (Romanoski et al., 2025 - <https://doi.org/10.1016/j.still.2025.106680>)

3.2 INTRODUCTION

Regenerative integrated systems have been claimed to improve conditions of aeration, water storage, and carbon stock in the soil, resulting in higher crop yields (Blanco-Canqui & Lal, 2009; Ruis & Blanco-Canqui, 2017; Silva et al., 2021). Agricultural practices significantly influence the dynamics of air and water flows in soil (Guedes et al., 2012; Çerçioğlu et al., 2019), which are governed by soil hydraulic properties that are in turn affected by soil structure (Ball, 1988; Blackwell et al., 1990a; Groenevelt et al., 1984; Tuli et al., 2005).

Regenerative agriculture is a systemic approach aimed at restoring natural resources by promoting soil health, biodiversity, and social and environmental sustainability (Schreefel et al., 2020; Garbisu et al., 2025). It is based on a productive, climate-resilient, and ethically committed agriculture that contributes to ecosystem services and the safe operating space of the Earth system (Garbisu et al., 2025). According to Rodale (1983), its pillars are: soil health, animal welfare, and social justice. Practices such as no-till farming, soil cover, crop rotation, the use of green manures, management-intensive grazing, and integration crop-livestock are fundamental to supporting these principles and strengthening more balanced production systems (Moyer, 2020; Ikerd, 2021; Ahmed et al., 2024).

Changes in soil physical properties directly impact ecosystem services, including agricultural production, water and air quality, and soil carbon stocks (Blanco-Canqui & Ruis, 2018). Soil structure degradation, often driven by agricultural practices, reduces macroporosity, restricts root growth, and alters water and air dynamics, ultimately compromising carbon retention (Blanco-Canqui & Ruis, 2018). Despite advancements in regenerative integrated systems, significant knowledge gaps remain, particularly concerning soil saturated hydraulic conductivity (K_s) and intrinsic air permeability (K_a) in these systems. For instance, a review by Valani et al. (2022) revealed that only 5% of studies on integrated systems evaluated K_s , and none examined K_a , highlighting critical research gap.

The intrinsic air permeability of soil (K_a) is a sensitive indicator of soil structural changes caused by agricultural practices (Tang et al., 2011). It measures the ability of air to flow through interconnected pore, reflecting pore organization and connectivity (Blackwell et al., 1990b; Tuli et al., 2005). Quantifying K_a is crucial for assessing soil structure and understanding the effects of management practices on soil functionality (Cavaliere et al., 2009; Hamamoto et al., 2016; Martínez et al., 2016). Studies have demonstrated that higher K_a correlates with reduced soil density, increased macroporosity, and enhanced fluid transport efficiency (Suzuki et al., 2022). For example, Brazil's integrated crop-livestock systems cover more than 17 million hectares, playing a crucial role in environmental conservation and rural

growth (Rodrigues, et al., 2023; Vilela et al., 2021). Regenerative integrated systems, such as Crop-Livestock, Livestock-Forest, and Crop-Livestock-Forest, preserve soil structure by avoiding plowing and promoting continuous and connected pore development. These systems enhance water and air distribution and storage, supporting root growth and soil carbon sequestration (Barreto et al., 2009; Fabrizzi et al., 2009; Gamboa et al., 2023). Additionally, conservation practices in agricultural systems increase organic carbon content and improve soil physical attributes over time (Cavaliere-Polizeli et al., 2024a).

Given the limited understanding of the effect of saturated hydraulic conductivity (K_s) and intrinsic air permeability in regenerative systems, as well as their relationship with carbon stock, this study investigates and tests the hypothesis that integrated systems exhibit better soil hydraulic properties compared to an exclusive livestock system, under regenerative practices. Specifically, the aim of this study was to compare three different integrated livestock systems with a single livestock system, evaluating regenerative agricultural practices and investigating how these integrations affect soil structure, with an emphasis on soil hydraulic properties.

3.3 MATERIAL AND METHODS

3.3.1 Experimental site and area

The study was conducted at the Center for Technological Innovation in Agriculture (NITA), located at the Canguiri experimental station of the Federal University of Parana (UFPR) in the municipality of Pinhais, State of Parana, Brazil, (25° 24'02" S, 49° 07'12" W, 930 m a.s.l.). The local climate, according to Koppen's classification, belongs to the Cfb type, subtropical humid without a dry season. This study was part of a long-term experiment, implemented in 2013, covering a total area of approximately 35 hectares, on physically degraded soil conditions (Cavaliere-Polizeli et al., 2022). The soil is classified as Ferralsols (WRB, 2022), and the texture is clayey (52% clay, 10% silt, and 38% sand) (Supplementary material, Table S).

3.3.2 History, experiment management and treatments

Before the experiment installation, between 2002 and 2012, the experimental area was managed under conventional tillage with maize (*Zea mays*) cultivation and was also used for training and testing agricultural machinery traffic. This management resulted in the physical degradation of the soil, primarily caused by compaction and erosion. In 2012, the soil was scarified to a depth of 0.40 m and harrowed (<0.20 m). Treated sewage sludge was incorporated at a rate of 10 Mg ha⁻¹ of N VIRO® onto the soil surface. During the winter of that same year,

black oats (*Avena strigosa*) were sown with 100 kg of P₂O₅ ha⁻¹ in the planting row across the entire experimental area. Between 2013 and 2021, the average annual application of fertilizers across the area was 200 kg ha⁻¹ of nitrogen (urea), 115 kg ha⁻¹ of P₂O₅ (natural phosphate), and 115 kg ha⁻¹ of K₂O (potassium chloride).

In 2013, the experiment was conducted using a randomized block design with four treatments: Livestock (L), Crop-Livestock (CL), Livestock-Forest (LF), and Crop-Livestock-Forest (CLF), under regenerative agriculture practices. The treatments were distributed in three blocks, and soil sampling were collected at depths of 0.00-0.05, 0.05-0.15, and 0.15-0.30 m. The management of areas in this study incorporates regenerative agricultural practices including direct planting using the "planting green" technique, pesticide-free management, mineral fertilization, proper stocking of animals, and reduced traffic load and intensity.

No-till farming using the "planting green" technique was adopted as an alternative to direct planting without desiccation, primarily for weed control (Mirsky et al., 2012; Dominschek et al., 2021). The experimental area has been a pesticide-free site since 1996, in accordance with Decree No. 1753/1996; however, the use of chemical fertilizers is allowed (Cavalieri-Polizeli et al., 2022).

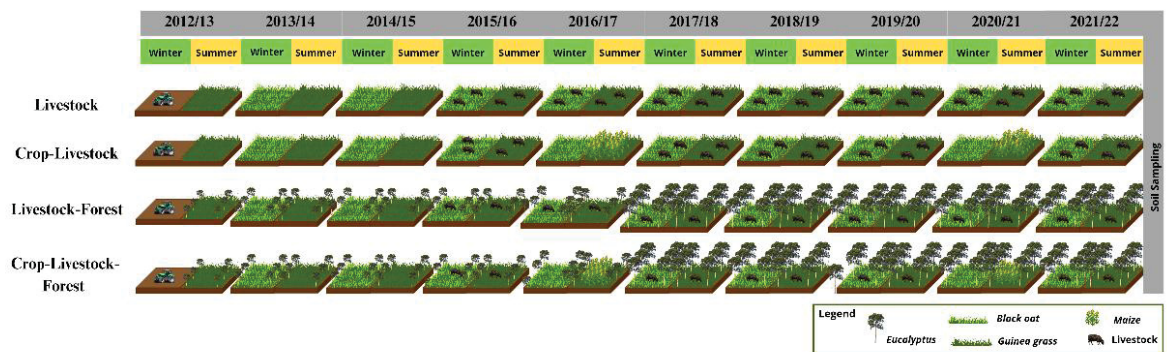
In the Livestock treatment, the perennial pasture used in the summer was *Megathyrus maximus* cv. Aries, seeded in 2013, while black oats (*Avena strigosa*) were cultivated in the winter. Grazing began in 2015 with Angus steers weighing approximately 200 kg. The pastures were managed under continuous stocking, providing animals with forage that maximized their intake rate. During both the cold and warm seasons, the average target pasture height was maintained at 24 cm, as proposed by (Carvalho, 2013). Stocking rate adjustments were made using the "put and take" technique (Mott and Lucas, 1952). The average stocking rates were 866, 718, and 627 kg ha⁻¹ of live weight, respectively, in the Livestock (L), Livestock-Forest (LF) and Crop-Livestock-Forest (CLF), for 10 months each year, with two months reserved for sowing and establishing the pasture.

In the Crop-Livestock (CL) and CLF treatments, a "ley farming" model was followed, where pasture is cultivated for three years, and the following summer, an agricultural crop (maize) is planted using direct seeding into green cover, while oats are sown using the "green planting" technique without herbicides for desiccation.

In 2013, 357 eucalyptus trees (*Eucalyptus benthamii*) were planted per hectare in the Livestock-Forest (LF) and CLF treatments, following the topography of the land. Thinnings occurred at 36, 44, and 78 months. After each thinning, the density of trees per hectare in the LF treatment was reduced to 258, 174, and 52 trees, while in the CLF, the densities were 320,

174, and 52 trees per hectare. The final mean spacing was 28×6 m (for more details, see (Kruchelski et al., 2021)).

Figure 1 - Timeline of the experiment conducted between 2013 and 2021 to evaluate regenerative systems. The treatments include Livestock, Crop-Livestock, Livestock-Forest, and Crop-Livestock-Forest. The experiment was carried out in summer and winter cycles, with soil sampling conducted in 2021. The crops represented include corn or *Megathyrus maximus* cv. Aries grass (summer) and black oat (*Avena strigosa*) (winter). The forest component consisted of eucalyptus (*Eucalyptus benthamii*), planted in 2013. Grazing began in 2015 with Angus steers. Maize was cultivated in 2016, and the area alternated between livestock (2017-2019, 2021) and maize cultivation again in 2020.



SOURCE: Author (2026).

3.3.3 Soil sampling

The sampling occurred in two periods, 2021 (Block I) and 2022 (Blocks II, and III), due to drier soil conditions, that impaired the sampling. To represent the depth of 0.00-0.30 m, 144 undisturbed samples in metal cylinders with volume of 113 cm^3 (0.06×0.04 m – diameter and height), were collected in three layers (0.00-0.05, 0.05-0.15, and 0.15-0.30 m), composing a weighted mean to 0.00-0.30 m of depth. Each system was evaluated by 36 samples, being four sampling points, three layers, and three blocks.

3.3.4 Soil assessments

The soil physical attributes determined were: saturated hydraulic conductivity (Ks), intrinsic air permeability of soil (Ka) at -3 and -10 kPa matric potentials, soil bulk density, total porosity, air-filled porosity, macroporosity, mesoporosity, microporosity, pore continuity index and pore organization index were determined for the identified pore classes. Disturbed soil samples were taken to determine soil texture and soil organic carbon (SOC).

3.3.5 Soil hydraulic properties

The undisturbed soil samples were saturated for 48 hours and then subjected to the constant head method (Klute & Dirksen, 1986), for the analysis of saturated hydraulic conductivity (K_s), which was calculated using the Darcy-Buckingham equation as follows (Eq. 1):

$$K_s = \frac{V \cdot L}{A \cdot t \cdot (h + L)} \quad (1)$$

Where V is the volume (cm^3) of water collected in the graduated cylinder during the time t (h); A is the cross-sectional area, (m^2) of the sample; L is the length (m) of the soil-filled sample; and h corresponds to $\frac{3}{4}$ of the height (m) of the upper ring. The K_s values derived from Eq. (1) were transformed into units of cm h^{-1} .

3.3.6 Intrinsic air permeability of soil

After determining the K_s , the samples were equilibrated to matric potentials of -3 and -10 kPa on a tension table. Once equilibrium was reached, the samples were weighed, and intrinsic air permeability of soil (K_a) was measured. For this, a permeameter proposed by Figueiredo, (2010) was used, which determines the air flow rate through an area perpendicular to the fluid direction (Q) and the resulting potential difference (dP/dz), assuming that air density has a negligible influence. Thus, K_a was calculated using the formula (Eq.2):

$$K_a = \frac{Q \cdot \eta \cdot dz}{A_s \cdot dP} \quad (2)$$

Where Q is the air flow rate ($\text{m}^3 \text{s}^{-1}$); A_s is the cross-sectional area (m^2) of the volumetric ring; η is the dynamic viscosity of air (N s m^{-2}); dz (m) is the distance in the differential of air movement in the soil sample (i.e., the height of the volumetric ring); dP , (Pa), is the differential pressure obtained by subtracting the final pressure generated by the air flow in the soil from the initial atmospheric pressure. The K_a values (m^2) derived from Eq. (2) were transformed into units of μm^2 .

3.3.7 Assessment of soil porosity, organization and connectivity indexes, plan-available water capacity, bulk density and soil texture

Total porosity (TP) was calculated considering the difference between the dry soil mass and the saturated soil mass, divided by the volume of the ring (Grossman & Reinsch, 2002). The air-filled porosity (ϵ) was determined at the matric potentials of -3 kPa and -10 kPa. The soil porosity distribution was based on pore diameter, following the classification by Koorevaar (1983): porosity of large macropores $>100 \mu\text{m}$ (MAC), mesoporosity $100 - 30 \mu\text{m}$, and microporosity $<30 \mu\text{m}$ (MIC). Additionally, macroporosity (MaP) was defined as the fraction of soil volume occupied by pores with a diameter $>30 \mu\text{m}$. The proportion of larger pores, such as soil MAC, was determined by the difference between total porosity (TP) and volumetric soil water content at a matric potential of -3 kPa. The macroporosity (MaP), was determined by the difference between the microporosity (MIC), obtained as volumetric soil water content, at a potential of -10 kPa, and TP.

The data from K_a and ϵ analyses at matric potentials of -3 and -10 kPa were used to indirectly calculate the pore organization index ($k_1 = K_a/\epsilon$) and pore connectivity ($k_2 = K_a/\epsilon^2$), which are geometric characteristics of the soil pore space (Groenevelt et al., 1984). These indexes (k_1 and k_2) were associated with the equivalent pore diameter at each applied matric potential. Therefore, k_1 (for MAC) or k_1 (for MaP) and k_2 (for MAC) or k_2 (for MaP) serve as indicators of how air-filled macroporosity with diameters larger than $100 \mu\text{m}$ or $>30 \mu\text{m}$, respectively, are organized and connected within the soil pore system (Poulsen, 2015).

Plant-available water capacity (PAWC, $\text{m}^3 \text{m}^{-3}$), which is a measure of water storage, was calculated as the difference between the volumetric water content ($\text{m}^3 \text{m}^{-3}$) at field capacity (matric potential -10 kPa) and the permanent wilting point (matric potential -1500 kPa) (Reynolds et al., 2009).

The soil bulk density (BD) was determined using the volumetric ring method. The samples were dried in a forced-air oven ($105 \text{ }^\circ\text{C}$ for 48 hours) to obtain the dry soil mass and establish the relationship between the dry soil mass and the volume of the ring (Grossman & Reinsch, 2002). For soil texture, the Bouyoucos hydrometer method was used (Gee & Or, 2002).

3.3.8 Soil Carbon Stock (Cst)

For the soil organic carbon SOC (%) the colorimetric method was used according to Quaggio and van Raij, 1979. The soil carbon stock (Cst) was calculated with correction for soil

mass (Eq. 3), as suggested by (Ellertl et al., 1995), using a native secondary forest adjacent to the experimental area as a reference:

$$Cst (Mg\ ha^{-1}) = \left[SOC \cdot BD \cdot \left(\frac{BD_{NF}}{BD} \right) \cdot e \right] \quad (3)$$

Where Cst is the soil carbon stock at the layer (0.00-0.30 m), SOC is the soil organic carbon (%), BD is the bulk density of the soil ($Mg\ m^{-3}$), BD_{NF} is the bulk density of the soil ($0.58\ Mg\ m^{-3}$) from the native secondary forest, and e is the thickness of the sampled layer (m).

3.3.9 Statistical analyses

The data from the soil layers (0.00-0.05 m, 0.05-0.15 m, and 0.15-0.30 m) were used to calculate a weighted mean corresponding to the total depth of 0.00-0.30 m. The weighted mean was applied to more accurately represent the mean value of a variable along the profile, considering the proportional contribution of each layer, whose thicknesses vary (0.05 m, 0.10 m, and 0.15 m). Additionally, the standard deviation was calculated for each layer as well as for the overall average of the four systems analyzed.

Based on these data, MANOVA tests (Pillai's trace, $p < 0.05$) were conducted to compare different combinations of systems: Livestock vs. Crop-Livestock, Livestock vs. Livestock-Forest, and Livestock vs. Crop-Livestock-Forest in the 0.00-0.30 m layer. Subsequently, collinearity analysis and principal component analysis (PCA) were performed to identify the main directions of variation among the regenerative systems. To explore clustering patterns in the data, hierarchical clustering analysis was conducted. In this process, the Mahalanobis distance matrix was calculated using standardized variables, and clustering was performed using the Ward linkage method. All statistical calculations were performed using R software (version 4.3.1, R Core Team, 2023) with the support of the following packages: dplyr, ExpDes, MultivariateAnalysis, FactoMineR, factoextra, and corrplot.

The choice of MANOVA (Multivariate Analysis of Variance) over ANOVA (Analysis of Variance) is justified by the need to consider multiple dependent variables simultaneously while accounting for their potential correlations. In this study, soil physical attributes such as saturated hydraulic conductivity (K_s), intrinsic air permeability (K_a), and macroporosity exhibit interdependence, and MANOVA allows for the evaluation of treatment effects on this set of variables collectively. Unlike univariate ANOVA, which analyzes each variable separately, MANOVA reduces the risk of Type I errors by considering the correlation structure among

variables, providing a more comprehensive view of the impacts of the evaluated systems. Furthermore, this multivariate approach enhances the robustness of statistical analysis, enabling the detection of patterns that might be overlooked in isolated univariate tests.

Hierarchical clustering analysis was used to identify similarity patterns among treatments. The Mahalanobis distance was chosen for its ability to account for correlations among variables, ensuring a more accurate measure of dissimilarity. The Ward linkage method was applied to minimize within-group variability, generating more homogeneous, coherent, and meaningful clusters. This approach complements the multivariate analysis, facilitating the interpretation of treatment effects.

3.4 RESULTS

Table 1 presents the weighted mean values of soil attributes evaluated in different regenerative systems, considering the 0.00-0.30 m soil layer. The weighted mean values of saturated hydraulic conductivity (K_s) across the four systems ranged from 7.4 cm h⁻¹ to 11 cm h⁻¹. Livestock had the lowest value (7.4 cm h⁻¹), while the Crop-Livestock-Forestry had the highest (11 cm h⁻¹). High standard deviations in some systems indicate significant variability in the results. The overall weighted average was 9.18 cm h⁻¹, suggesting that integrated systems have the highest values of saturated hydraulic conductivity.

The weighted mean values of intrinsic air permeability of the soil (K_a) for macroporosity with diameters greater than 100 μm $K_{a(\text{MAC})}$, varied across the evaluated systems. Both the Livestock and Crop-Livestock system had similar means of 8.7 (± 2), while the Livestock-Forestry had a mean of 8.4 (± 2), and the Crop-Livestock-Forestry recorded the highest mean of 13.4 (± 2). The overall weighted mean was 9.80 (± 2.40).

Table 1 - Weighted mean values ($n = 36, \pm$ SD) of general attributes of the systems: Livestock (L), Crop-Livestock (CL), Livestock-Forestry (LF), Crop-Livestock-Forestry (CLF) at depths of 0.00-0.30 m

Soil attributes	Systems					Mean ¹
	Livestock	Crop-Livestock	Livestock-Forestry	Crop-Livestock-Forestry		
Ks (cm h ⁻¹)	7.4 (± 7)	9.6 (± 10)	8.7 (± 8)	11 (± 10)	9.18 (± 1.52)	
Ka _(MAC) μm ²	8.7 (± 2)	8.7 (± 2)	8.4 (± 2)	13.4 (± 2)	9.80 (± 2.40)	
Ka _(MaP) μm	44 (± 10)	49 (± 10)	37 (± 10)	36 (± 10)	41.5 (± 6.14)	
PAWC (m ³ m ⁻³)	0.21 (± 0.04)	0.22 (± 0.05)	0.18 (± 0.05)	0.17 (± 0.04)	0.20 (± 0.02)	
BD (Mg m ⁻³)	1.19 (± 0.04)	1.20 (± 0.04)	1.23 (± 0.04)	1.22 (± 0.04)	1.21 (± 0.02)	
TP (m ³ m ⁻³)	0.54 (± 0.01)	0.53 (± 0.01)	0.53 (± 0.01)	0.54 (± 0.01)	0.54 (± 0.01)	
MAC (m ³ m ⁻³)	0.07 (± 0.01)	0.07 (± 0.01)	0.07 (± 0.01)	0.08 (± 0.01)	0.07 (± 0.01)	
MaP (m ³ m ⁻³)	0.12 (± 0.01)	0.08 (± 0.01)	0.08 (± 0.01)	0.07 (± 0.01)	0.09 (± 0.02)	
MIC (m ³ m ⁻³)	0.35 (± 0.01)	0.39 (± 0.01)	0.38 (± 0.01)	0.38 (± 0.01)	0.38 (± 0.02)	
k ₁ (MAC)	1.9 (± 0.1)	1.9 (± 0.1)	1.99 (± 0.1)	2.09 (± 0.1)	1.97 (± 0.09)	
k ₂ (MAC)	3.2 (± 0.2)	3.2 (± 0.2)	3.19 (± 0.2)	3.21 (± 0.2)	3.20 (± 0.01)	
k ₁ (MaP)	2.3 (± 0.1)	2 (± 0.1)	1.81 (± 0.1)	2.05 (± 0.1)	2.04 (± 0.20)	
k ₂ (MaP)	3 (± 0.2)	2.9 (± 0.2)	2.67 (± 0.2)	2.88 (± 0.2)	2.86 (± 0.14)	
ε (m ³ m ⁻³)	0.19 (± 0.06)	0.15 (± 0.05)	0.15 (± 0.04)	0.16 (± 0.05)	0.16 (± 0.02)	
Cst (Mg ha ⁻¹)	39 (± 7)	41 (± 8)	31 (± 6)	30 (± 6)	35.2 (± 5.56)	

¹ Mean values of the four systems. Number of samples per system, n ; standard deviation, SD; saturated hydraulic conductivity, Ks; intrinsic air permeability of soil Ka_(MAC) and Ka_(MaP) respectively, in pores with diameters > 100 and > 30 μm; plant-available water capacity, PAWC; bulk density, BD; soil total porosity, TP; macroporos, MAC; macroporosity, MaP; microporosity, MIC; characterization and functioning of the soil pore system: pore organization index k_1 [μm² (m³ m⁻³)⁻¹], pore connectivity index k_2 [μm² (m³ m⁻³)⁻²] in pores with diameters > 100 (MAC) μm and > 30 μm (MaP) and air-filled porosity (ε) for matric potentials of -10 kPa and carbon stock, Cst.

SOURCE: Author (2026).

The weighted mean values of the pore organization (k_1) and connectivity (k_2) index were analyzed for pores with diameters greater than 100 μm (MAC) and 30 μm (MaP). For k_1 (MAC), the overall mean was 1.97 (± 0.09), with integrated systems showing higher values. For k_1 (MaP), the weighted mean was 2.04 (± 0.20), with the Livestock system standing out with the highest mean. Regarding the k_2 index, the mean for k_2 (MAC) was 3.20 (± 0.01), while for k_2 (MaP), the overall mean was 2.86 (± 0.14), reflecting variation among the evaluated systems.

The multivariate analysis of variance (MANOVA) revealed significant differences between Livestock (L) and the integrated systems, highlighting the influence of integrating agricultural components (Table 2). The Pillai test indicated significant differences between L and the Crop-Livestock (CL) and Crop-Livestock-Forestry (CLF) systems. No statistical differences were observed between L and Livestock-Forestry (LF) and CL with CLF.

Table 2. Summary of Multivariate Analysis of Variance (MANOVA) using the Pillai test for systems comparison: Livestock (L), Crop-Livestock (CL), Livestock-Forestry (LF), Crop-Livestock-Forestry (CLF) at depths of 0.00-0.30 m

Systems	LF	CL	CLF
L	0.110 ^{ns}	0.020*	0.009*
LF		0.020*	0.030*
CL			0.100 ^{ns}

* Significant at the 5% probability level according to Pillai's test; ns - not significant

SOURCE: Author (2026).

The principal component analysis (PCA) explained 88% of the variance, with eigenvalues of 4.46 (PC1) and 1.36 (PC2) (Table 3). In PC1, the most significant contributions came from the factors Cst, Ks, Ka(MAC), Ka(MaP), BD, and MaP. PC2 was predominantly influenced by the contribution of MIC.

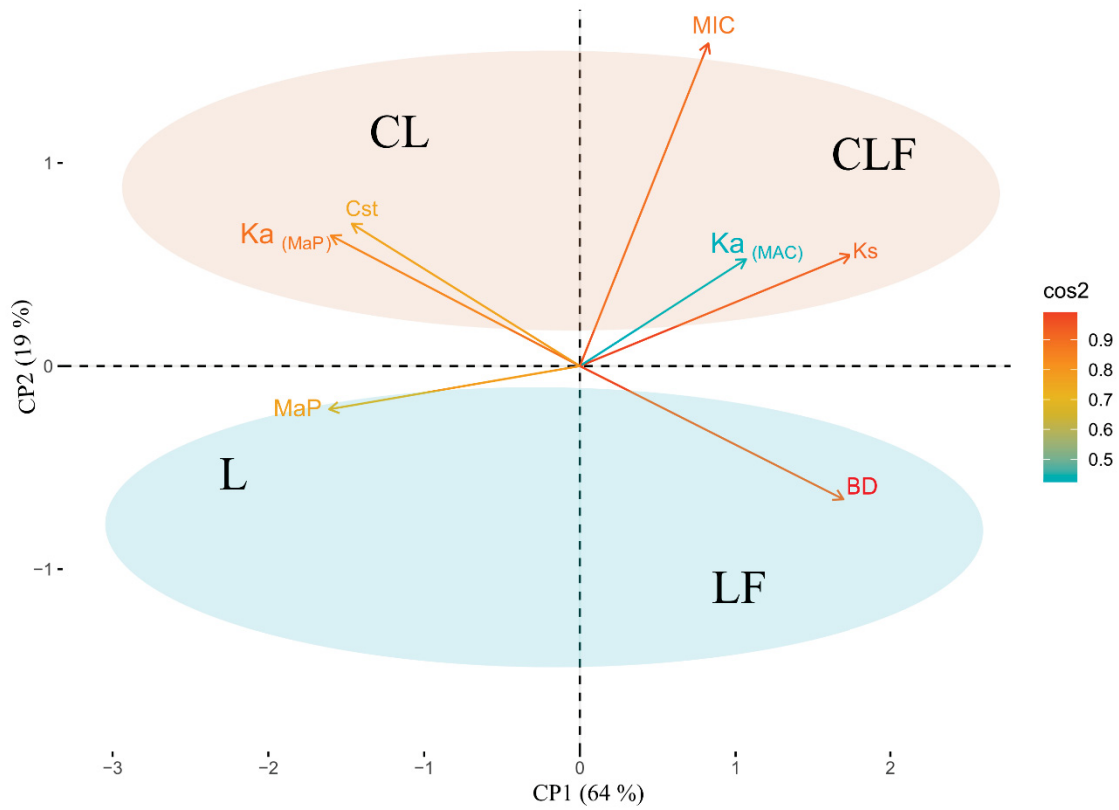
Table 3 - Summary of Eigenvalues, Eigenvectors, and Cumulative Variance of Principal Component Analysis (PCA) for the systems: Livestock (L), Crop-Livestock (CL), Livestock-Forestry (LF), Crop-Livestock-Forestry (CLF) at depths of 0.00-0.30 m

Soil attributes	CP 1		CP2	
	Eigenvectors	Contribution (%)	Eigenvectors	Contribution (%)
Cst (Mg ha ⁻¹)	-0.80	14.3	0.38	10.9
Ks (cm h ⁻¹)	0.95	20.1	0.29	6.6
Ka _(MAC) (μm ²)	0.58	7.5	0.28	6.1
Ka _(MaP) (μm ²)	-0.88	17	0.35	9.2
BD (Mg m ⁻³)	0.92	19.1	-0.35	9.6
MaP (m ³ m ⁻³)	-0.88	17.2	-0.11	1.02
MIC (m ³ m ⁻³)	0.45	4.5	0.86	56.3
Eigenvalues		4.46		1.36
Cumulative variance (%)		64		19

SOURCE: Author (2026).

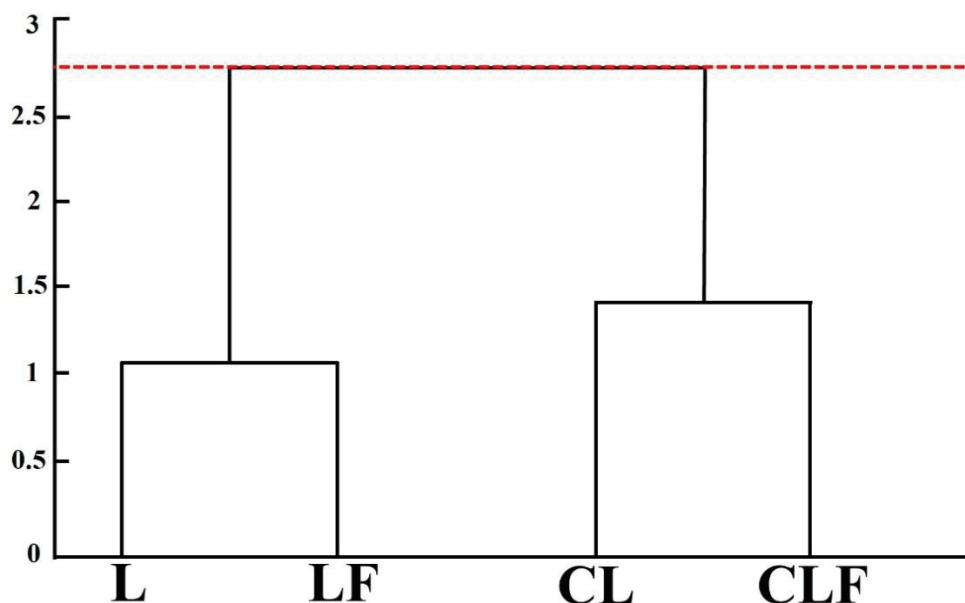
The hierarchical clustering analysis revealed the formation of two clusters, indicating the segmentation of the data based on their similarity: the first, consisting of the CL (Crop-Livestock) and CLF (Crop-Livestock-Forest) systems, showed a positive association between Ks, Ka(MAC), Ka(MaP), MIC, and Cst. The second cluster, which includes the L (Livestock) and LF (Livestock-Forest) systems, indicated a possible relationship between BD and MaP (Figure 2 and 3).

Figure 2 - Principal Component Analysis (PCA) of soil attributes under regenerative systems: Livestock (L), Crop-Livestock (CL), Livestock-Forestry (LF), Crop-Livestock-Forestry (CLF) at depths of 0.00-0.30 m, with cluster ellipses obtained by hierarchical clustering. Carbon stock, Cst; saturated hydraulic conductivity, Ks; intrinsic air permeability of soil $K_{a(MAC)}$ and $K_{a(MaP)}$ respectively, in pores with diameters > 100 and $> 30 \mu\text{m}$; bulk density, BD; macroporosity, MaP; microporosity, MIC.



SOURCE: Author (2026).

Figure 3 - Dendrogram resulting from the hierarchical clustering analysis of soil attributes under regenerative systems: Livestock (L), Crop-Livestock (CL), Livestock-Forestry (LF), and Crop-Livestock-Forestry (CLF), at a depth of 0.00-0.30 m. The red line indicates the dissimilarity cutoff (2.81) used to define the two main clusters.



SOURCE: Author (2026).

The intercluster and intracluster distances provide insights into the dissimilarity between the clusters. The intercluster distance between Cluster 1 and Cluster 2 was 2.81. The intracluster distances for clusters 1 and 2 were 1.19 and 1.45, respectively. The cophenetic correlation was 0.75, indicating good quality in the cluster analysis.

The cutoff line in the dendrogram (Figure 3) reinforces the separation of regenerative systems into two distinct groups, as already evidenced in Figure 2 by the Principal Component Analysis (PCA) with the formation of two clusters. Thus, the consistency between the two analytical approaches strengthens the robustness of the classification of the studied systems.

3.5 DISCUSSION

The results of this study demonstrated that the Crop-Livestock (CL) and Crop-Livestock-Forestry (CLF) treatments showed significant differences compared to Livestock (L). The use of principal component analysis (PCA) allowed the identification of the variables that contributed most to differentiating the systems. Saturated hydraulic conductivity (Ks) made the largest contribution to PC1 (20.1%), with a high eigenvector value (0.95), standing out as a predominant variable in the CLF system. This finding aligns with the results of Singh et al. (2021), which identified Ks as a crucial indicator of soil quality in integrated farming systems,

promoting better water flow and facilitating nutrient movement in the soil. Additionally, the positive contribution of air permeability (K_a) (7.5%) reinforces the role of integrated systems in improving pore connectivity, which is essential for enhancing aeration and biological activity (Negrón et al., 2019).

Microporosity (MIC) made the largest contribution (56.3%) in PC2, and a moderate eigenvector value (0.38), highlighting its importance for water retention and soil stability, especially in the CLF system. The CL system stood out for its association with carbon stock (Cst), which showed significant contributions of 14.3% in PC1 and 10.9% in PC2, evidencing the role of soil carbon stock as a key variable in differentiating this system. The presence of crop in the systems, such as CL and CLF, favors water retention, helps mitigate droughts, and enhances soil stability, while promoting better carbon storage, as demonstrated by Nascimento et al. (2019) and Cavalieri-Polizeli et al. (2024b).

The clusters between the L and LF systems reflect the functional and structural similarities in soil properties within each group, with both systems exhibiting shared characteristics associated with management practices. The L and LF systems showed similarities, particularly regarding soil bulk density, which contributed 19.1% to the first principal component (PC1).

The integration of cropland with livestock, as seen in CL and CLF systems, has a positive impact on soil physical properties and dynamic processes, such as those quantified by saturated hydraulic conductivity (K_s) and air permeability (K_a) (Awal et al., 2019), both of which are responsible for various functional aspects of the soil system. Previous studies also indicate that integrated systems enhance soil properties, including air permeability (K_a) and pore connectivity, enhancing soil structure and the sustainability of agricultural practices (Cassaro et al., 2011; Silva et al., 2021; Dhaliwal & Kumar, 2022).

The results of this study demonstrate that the integrated systems CL and CLF enhance pore system connectivity and organization, as reflected in the values of intrinsic air permeability (K_a). This improvement arises from maintaining a more continuous pore structure, a characteristic of agricultural practices that avoid excessive soil disturbance. Studies such as those by Martínez et al. (2016) and Suzuki et al. (2022) highlight that improved pore organization reduces compaction and enhances aeration, which are essential for biological activity and agricultural sustainability.

Complementarily, the CLF and CL systems exhibited higher values of saturated hydraulic conductivity (K_s), indicating improved pore continuity and interconnection, which are essential for the soil's hydraulic functionality. This pore system connectivity, as highlighted

by Poulsen (2015), optimizes the simultaneous transport of water and air, fostering soil sustainability and healthy plant development.

In summary, our results reinforce the idea that the integration of cropland with livestock, as seen in CL and CLF systems, has the potential to improve soil quality by balancing functional and structural variables such as air permeability, hydraulic conductivity, and carbon stock (Figure 2). Integration of cropland with livestock is more effective in promoting long-term sustainability (Singh et al., 2021), while contributing to the improvement of soil physical properties and carbon preservation, both essential elements for sustainable agricultural management.

Although not all the attributes listed in Table 1 were used in the multivariate analyses due to the effect of collinearity – that is, the presence of highly correlated variables, which may compromise the interpretation of the results – they are important for characterizing the studied systems.

The analyzed systems showed saturated hydraulic conductivity (K_s) values classified as moderately rapid, with an average of 9.18 cm h^{-1} , according to the classification proposed by Reichardt (1978). These values indicate that the soil's ability to conduct water is adequate. The K_s showed high variability, with a coefficient variation of 100 %. Previous studies such as Awal et al. (2019), Pavlů et al. (2021), Jabro et al. (2023), and Wu et al (2024), have reported CVs for K_s ranging from 80 % to 240 %, indicating characteristics variability of the variable. This high variability in K_s is widely acknowledged in the literature (Bormann & Klaassen, 2008).

The studied systems did not present limit values of intrinsic air permeability of soil (K_a), varying between 8.4 to $49 \mu\text{m}^2$, far exceeding the threshold of $1.0 \mu\text{m}^2$ considered critical for root oxygenation (McQueen et al., 2002). The intrinsic air permeability of soil provides key insights into pore continuity, organization, and connectivity, essential for structural quality (Hamamoto et al., 2016; Martínez et al., 2016). Besides that, BD, K_s , and K_a are crucial for the movement and storage of water, air, nutrients, and heat, directly influencing root system growth and development (Wang et al., 2021).

The results of the analysis indicate that the systems exhibit PAWC values $0.20 \text{ m}^3 \text{ m}^{-3}$, considered ideal for root growth and development (Reynolds et al., 2009). These values indicate that the systems are suitable for plant growth. The fact that all systems fall into the ideal category suggests that the management practices in all systems are effective in maintaining soil water retention capacity within a range favorable for plant growth.

The critical soil density (Critical BD) values estimated for the treatments (L, CL and LF = 1.44 Mg m^{-3} ; CLF = 1.42 Mg m^{-3}) are within the expected limits for tropical soils in southern

Brazil, according to the criteria established by Reichert et al. (2009). Furthermore, in the four treatments evaluated, the soil bulk density (BD) values were below 1.26 Mg m^{-3} , a threshold considered safe for clayey-textured soils related to the risk of restricting root growth (Pachepsky & Park, 2015). These results indicate that the physical conditions of the soils in the analyzed treatments are suitable for root development, demonstrating that the management of systems employed contribute to maintaining the structural quality of the soil, avoiding compaction levels that could compromise the soil's ecological and productive functionality.

Air-filled porosity at the -10 kPa matric potential, an essential aspect for proper root growth, was not limiting in the analyzed regenerative systems ($\epsilon \geq 0.10 \text{ m}^3 \text{ m}^{-3}$), according to Grable & Siemer (1968). This condition favored crop development due to increased oxygen availability in the root zone, which is crucial for plant respiration and metabolism (Reynolds et al., 2009). These benefits were further enhanced by practices such as no-till farming, which, 10 years after its implementation, demonstrated a positive impact on soil structural organization, promoting a balance between macroporosity and microporosity, essential for the sustainability of regenerative systems.

The results of this study have important implications for the adoption of regenerative agricultural practices. Integrated systems, such as Crop-Livestock (CL) and Crop-Livestock-Forest (CLF), demonstrated improvements in key soil properties, such as water retention and increased carbon sequestration. Although the results of this study provide valuable insights into the effects of regenerative systems on soil physical properties, some limitations must be acknowledged. The analysis was based on samples collected in 2022, which may not fully reflect seasonal or annual variations in soil properties, which could be influenced by different climatic conditions or management practices over time. Additionally, the study was conducted at a single experimental station, limiting the generalization of the results to other regions with different edaphic and climatic characteristics.

Future research should consider evaluating a broader range of scenarios to enhance the understanding of regenerative practices across diverse environmental conditions. This includes assessing the impact of different climatic conditions and soil textures, which can influence soil health, crop productivity, and the effectiveness of integrated systems. Expanding the scope of research in this way will provide more comprehensive insights, supporting the development of sustainable strategies adaptable to various ecosystems.

Overall, the results obtained indicate that the studied systems exhibit soil physical characteristics highly favorable to plant growth and development. These attributes include a high capacity for available water storage, soil density below critical thresholds for clayey soils,

and adequate air porosity to ensure efficient root oxygenation. The implementation of regenerative management across all systems has played a pivotal role in fostering these beneficial soil conditions.

3.6 CONCLUSION

The results of this study confirmed the hypothesis that integrated regenerative systems, such as Crop-Livestock (CL) and Crop-Livestock-Forest (CLF), exhibit better soil hydraulic properties compared to Livestock (L). The CL and CLF systems demonstrated higher saturated hydraulic conductivity (Ks) and greater air permeability (Ka). On the other hand, the L and LF systems showed adequate soil structural characteristics but had a less pronounced impact on physical properties. These findings emphasize the importance of adopting regenerative management practices to improve soil structure and mitigate environmental impacts.

In this context, the study reinforces that the integration of cropland with livestock, as seen in CL and CLF systems, promotes sustainability by effectively balancing soil functionality and carbon conservation. Future research should evaluate diverse scenarios, considering different climatic conditions, soil textures, and management practices, providing deeper insights into their potential for sustainable agriculture.

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4 CHAPTER 2 – A LONG-TERM LIVESTOCK-FORESTRY SYSTEM IMPROVES ECOSYSTEM SERVICES IN A SUBTROPICAL REGION

4.1 ABSTRACT

The conversion of pastures into integrated systems is advocated as a vital strategy for sustainable intensification and climate change mitigation. The objective of this study was to evaluate and compare the provision of ecosystem services in livestock and livestock-forestry systems, aiming to quantify, through an integrated approach, the benefits and trade-offs derived from the presence of trees in pasture-based animal production. The study was conducted in a long-term experiment located in Southern Brazil, comparing an exclusive livestock system with a livestock-forestry system (*Eucalyptus benthamii*). Using the Ecosystem Services Index (ESI) to compare the systems, indicators of supporting, provisioning, and regulating ecosystems services were analyzed. The results demonstrated the superiority of the livestock-forestry system in the evaluated ecosystem functions. Regarding supporting services, driven by improvements in soil aeration, the livestock-forestry and livestock systems achieved similar scores (0.81 vs. 0.72). In provisioning services, timber production functionally compensated for the reduction in livestock yield, raising the livestock-forestry system's index to 0.69 compared to 0.58 for the livestock system. The most distinct difference was observed in regulating services, where the additionality of tree carbon resulted in an index of 0.66 for the livestock-forestry system versus only 0.27 for the livestock system. Consequently, the overall ESI confirmed the greater multifunctionality of the livestock-forestry system (0.72) compared to the exclusive livestock system (0.52), validating the livestock-forestry system as a robust agricultural tool for reconciling agronomic production and environmental conservation.

Keywords: Integrated Crop-Livestock-Forestry; Carbon Sequestration; Climate Regulation; Multifunctionality.

4.2 INTRODUCTION

Sustainable agricultural intensification is one of the most pressing global challenges, requiring strategies that reconcile food security with climate change mitigation (Monteiro et al., 2024). In this scenario, the conversion of monocultures or exclusive pastures into integrated pastures, such as livestock-forestry has been widely advocated as a Nature-based Solution (NbS) face this global challenge (Miralles-Wilhelm, 2021; Carvalho et al. 2024). The productive arrangements of integrated systems have the potential to reverse soil degradation processes and increase the provision of ecosystem services (Villela et al., 2026), soil organic carbon storage in both tree biomass and soil, while maintaining food production (Daboit et al., 2025; Moraes Sá et al., 2025).

From the perspective of ecosystem services, integrating trees into pastures promotes landscape multifunctionality (England et al., 2020). In addition to provisioning services (timber and meat), the forestry component is expected to enhance regulating services, such as nutrient cycling and carbon storage, and supporting services, such as the maintenance of soil physical structure and health (Bieluczyk et al., 2025; Ortiz et al., 2023). In addition, trees are the primary offset for the enteric methane emission in livestock-based systems that enhance the climate regulation (Valani et al., 2023). However, the interception of solar radiation by the tree canopy can limit forage accumulation and animal performance, creating a productive trade-off (Mayer et al., 2026). Consequently, the simultaneous assessment of these benefits presents significant methodological challenges. Most current approaches still evaluate production components and environmental variables in isolation, failing to capture the trade-offs and synergies that occur in complex systems (Silva-Olaya et al., 2025)

The main research problem lies in the inherent complexity of livestock-forestry systems and the scarcity of standardized accounting methods capable of quantifying the integrated contribution of their components. Although data exist on carbon sequestration in forests or animal production in pastures, few studies have quantified how the introduction of the tree component alters the overall balance of ecosystem services compared to consolidated livestock systems in subtropical regions. The lack of unified metrics hinders the understanding of the tree's real environmental value within the system and limits the formulation of public policies regarding Payment for Ecosystem Services (PES) (Chamorro-Vargas et al., 2025).

The justification for this study relies on the need to validate tools that integrate soil physical properties, animal and tree productivity, and environmental variables into a single sustainability index for ecosystem services provision. While recent initiatives, such as the comprehensive frameworks developed by Silva-Olaya et al. (2022) in the Colombian Amazon

and Bieluczyk et al. (2025) in the Caatinga biome, have successfully assessed soil health indicators, they were conducted in distinct climatic contexts and focused primarily on soil attributes. Consequently, these studies did not encompass tree components or animal productivity data. Therefore, understanding the dynamics of forage biomass, pasture structure, and animal performance in conjunction with soil organic carbon and tree biomass in subtropical livestock-forestry systems is crucial to legitimize their role as carbon sinks (Machado et al., 2025; Teixeira et al., 2025) and as an agricultural practice that ensures the sustainability of simultaneous food and fiber production in the face of global climatic demands.

Therefore, the study hypothesis is that the integration of pastures with *Eucalyptus benthamii* improves ecosystem services compared to exclusive livestock systems, offsetting potential local productive losses with gains in climate regulation and soil conservation. Based on this hypothesis, the objective of this study was to evaluate soil physical quality, carbon stocks and meat and timber production and consequently ecosystem services in livestock and livestock-forestry systems, aiming to quantify, through an integrated approach, the benefits and trade-offs derived from the presence of trees in pasture-based animal production.

4.3 MATERIAL AND METHODS

4.3.1 Study site

This study was conducted within a long-term experiment established in 2013 in the municipality of Pinhais, Paraná, Brazil (25° 24'02" S, 49° 07'12" W, 930 m) located at the Canguiri Experimental Station (Federal University of Parana, UFPR). The region's climate is classified as Köppen type Cfb (humid subtropical with no dry season). The soil is a Ferralsol (IUSS Working Group WRB, 2022) with a clay texture, composed of 52% clay, 10% silt, and 38% sand. The experiment was established on physically degraded soil (Cavaliere-Polizeli et al., 2022).

It is noteworthy that the experimental site has been maintained free of pesticides since 1996, in compliance with State Decree No. 1753/1996 (Paraná, 1996). The management strategies adopted align with the principles of conservation agriculture (Cavaliere-Polizeli et al., 2024), integrating rational mineral fertilization, adjusted animal stocking rates, and minimized machinery traffic. Instead of chemical desiccation, the system uses the "planting green" technique, sowing crops directly into living cover crops, as an ecological strategy for weed suppression and soil cover maintenance (Mirsky et al., 2012; Dominschek et al., 2021).

Prior to the experiment's establishment, from 2002 to 2012, the area was under a conventional tillage system for maize (*Zea mays*) cultivation. This decade of management,

combined with its use for testing agricultural machinery, resulted in significant soil physical degradation, primarily compaction and erosion. In 2012, a recovery process was initiated, involving soil subsoiling to a 0.40 m depth and harrowing of the top 0.20 m. Treated sewage sludge (VIRO®) was surface-applied at a rate of 10 Mg ha⁻¹. Since then, annual fertilization has been maintained at an average rate of 300 kg ha⁻¹ of urea (\approx 135 kg ha⁻¹ N), 200 kg ha⁻¹ of natural phosphate (\approx 70 kg ha⁻¹ P₂O₅), and 100 kg ha⁻¹ of potassium chloride (\approx 60 kg ha⁻¹ K₂O).

In 2021, soil analysis of the 0-10 cm layer showed the following: pH (CaCl₂) of 5.1; organic matter content of 4.7 %, estimated by multiplying total organic carbon (measured via dry combustion) by 1.724; exchangeable Ca = 4.9 cmolc dm⁻³, Mg = 2.9 cmolc dm⁻³, and Al³⁺ extracted with 1.0 mol L⁻¹ KCl; available K = 0.44 cmolc dm⁻³ (172 mg dm⁻³) and P = 7.6 mg dm⁻³, both extracted using Mehlich-1; and base saturation of 68 %, with cation exchange capacity (CEC) at pH 7.0 calculated as the sum of H⁺, Al³⁺, Ca²⁺, Mg²⁺, and K⁺.

4.3.2 Experimental design and treatment description

The experiment established following a randomized block design with three replications. It comprised two treatments: a full-sun pasture system, designated as Livestock System (LS), and a Livestock-Forestry System (LFS).

In 2013, 357 eucalyptus (*Eucalyptus benthamii*.) trees were planted in single rows on silvopastoral systems (SS). These lines were established by leveling the central tree line on the hillslope, with adjacent lines placed in parallel up and down the slope – a practice aimed at reducing runoff and promoting soil conservation. The predominant row orientation was northwest to southeast, despite slight variations due to terrain curvature. Thinning operations were carried out at 36, 44, and 78 months after planting, reducing tree density to 258, 174, and finally 52 trees per hectare, respectively (for more details, see (Kruchelski et al., 2021)). At the time of the study, *E. benthamii* trees reached an average height of 26.6 ± 3.5 m.

In the livestock system, the forage system was based on a perennial summer pasture of Guinea grass (*Megathyrsus maximus* cv. Aries), sown in 2013, overseeded with an annual winter pasture of black oats (*Avena strigosa*). Grazing began in 2015, for an average of 280 days per grazing cycle (year) with a continuous stocking method in a moderate grazing intensity.

4.3.3 Soil characterization

Soil sampling was conducted in January 2023, during which three replicate trenches were excavated in each land use system. To assess physical properties, a total of 240

undisturbed samples were collected using 113 cm³ metal cylinders (6 cm diameter × 4 cm height). These were taken from five distinct depths: 0.00-0.10, 0.10-0.20, 0.20-0.40, 0.40-0.60, and 0.60-1.00 m, and were designated for the determination of soil bulk density, total porosity, macroporosity, microporosity and penetration resistance. Concurrently, 30 disturbed soil samples were collected (3 trenches × 5 depths × 2 systems) for the analysis of soil organic carbon (SOC) and texture.

The samples were saturated, weighed, and then placed on the tension table at a matric potential of -10 kPa. After equilibrium was reached at -10 kPa, soil penetration resistance (PR) was measured in the laboratory using a bench penetrometer. For each sample, three measurements of PR were taken, and the average value was subsequently calculated. From this procedure, total porosity (TP) was determined using the method described by (Grossman & Reinsch, 2002), based on the difference between saturated and dry soil mass per cylinder volume. Porosity was then partitioned into microporosity (MIC) and macroporosity (MAC). MIC (pores <30 µm) was determined as the volumetric water content at -10 kPa matric potential. MAC (pores >30 µm) was subsequently calculated by subtracting microporosity from total porosity (MAC = TP - MIC). Soil bulk density (BD) was measured using the core method, calculated as the ratio of oven-dried soil mass (105 °C for 48 hours) to the cylinder's volume (Grossman & Reinsch, 2002). The Bouyoucos hydrometer method was employed to determine the soil's texture (Gee & Or, 2022). The 30 disturbed samples were air-dried and sieved through a 2 mm mesh and then submitted to soil organic carbon analysis.

4.3.4 Quantification of soil organic carbon stock

The SOC was determined using a Vario El III CHNS Elementar analyzer. Prior to the analysis, soil samples were sieved through a 0.2 mm mesh. The quantification followed the dry combustion method outlined by Skejmstad and Taylor (1999), where samples were combusted at 950 °C under a constant oxygen flow to ensure complete oxidation. The SOC concentration was then calculated from the analysis of the released gases.

Following the methodology of Sisti et al. (2004), soil carbon stocks were calculated and corrected on an equivalent soil mass basis, using a native vegetation profile as a reference to compensate for variations in soil bulk density between systems. The quantification was performed for the depths of 0.00-20 cm, reflecting the management layer; 0.00-30 cm, as the standard for IPCC inventories; and down to 1.00 m, to represent the total stock in the soil profile.

The corrected carbon stock (Cst) was calculated using the following Eq. (1):

$$Cst (Mg\ ha^{-1}) = \sum_{i=1}^{n-1} CTi + [MTn - (\sum_{i=1}^n MTi - \sum_{i=1}^n MSi)] * CTn \quad (1)$$

Where Cst corrected soil carbon stock, (Mg C ha⁻¹), $\sum_{i=1}^{n-1} CTi$ sum of the total carbon (Mg ha⁻¹) in depth 1 (surface) to n-1 (penultimate) of the treatment profile. MTn total soil mass (Mg ha⁻¹) in all depth of the reference profile (native vegetation). $\sum_{i=1}^n MTi$ total soil mass (Mg ha⁻¹) in all depths of the treatment profile. $\sum_{i=1}^n MSi$ mass of soil in the deepest layer in the treatment profile, and CTn carbon concentration (Mg C per Mg of soil) in the deepest layer (n) of the treatment profile.

4.3.5 Assessment of forage biomass, pasture structure, and animal performance

For this study the first grazing cycle occurred from July 2021, to April 2022 (Year 1), and the second from July 2022, to April 2023 (Year 2), totaling 276 and 281 grazing days, respectively. In both years, three crossbred Angus steers (average initial weights of 170 ± 24 kg in Year 1 and 142 ± 34 kg in Year 2; 7-10 months of age) were allocated per paddock, totaling 18 tester animals per grazing cycle ($n = 36$, 3 test steers per paddock \times 6 paddocks \times 2 years).

Pastures were managed under continuous stocking with variable stocking rate (put-and-take technique; Mott and Lucas, 1952) to maintain a target canopy height of 24 cm. Sward height was monitored every 15 days by measuring 150 points per paddock with a sward stick, and stocking rate was adjusted accordingly. Tester animals remained full-time in the experimental units. Put-and-take animals of similar weight and age were added or removed as needed to regulate sward height and ensure consistent grazing intensity across paddocks. Animals had ad-libitum access to fresh water and mineral salt in their paddocks. No supplemental feeding was provided, all intake derived exclusively from pasture.

The herbage mass (HM) and herbage accumulation rate (HAR) were measured at a 28-day interval (stocking period). On each stocking period, the HM [kg of dry matter (DM) ha⁻¹] was estimated by randomly locating three metal frames (0.5 \times 0.5 m) on each paddock and cutting at ground level all the herbage inside. The herbage samples were oven-dried at 55 °C for 72 h and weighed on a precision scale (0.001 g) to estimate DM. Additionally, on each stocking period, herbage cuts were performed inside and outside of three grazing exclusion cages in each paddock to estimate the HAR (kg DM ha⁻¹ day⁻¹) according to Klingman et al.

(1943). By summing the daily accumulation rates over the entire grazing cycle, the total herbage accumulated was calculated, representing the gross primary productivity of the pasture.

The herbage allowance was estimated following Sollenberger et al. (2005), calculated as the ratio between the mean herbage mass and the stocking rate. Additionally, the leaf allowance was determined by ratio average stoking rate per leaf blades mass, obtained through the morphological separation of the cut samples.

The stocking rate was calculated based on the sum of the body weight of all animals (testers and put-and-take) per area. Animal performance was assessed by weighing the tester animals every 28 days after a fasting period. The average daily gain (ADG) was derived from the weight difference between periods, and the daily live weight gain (DWG) per hectare was calculated by multiplying the ADG of the tester animals by the average stocking rate, providing a measure of the system's overall animal productivity.

4.3.6 Performance assessment of tree

To estimate the carbon stock in the arboreal component (*Eucalyptus benthamii*), an allometric growth equation was developed specifically for the site conditions (Kruchelski et al., 2022). A destructive sampling was carried out between 2019 and 2022, where 34 trees were felled in the livestock-forestry arrangements (Kruchelski et al., 2022). For each individual, the diameter at breast height (DBH) and total height (h) were measured, and the fresh mass of the stem (wood and bark), branches, and leaves was quantified following the definitions by Picard et al. (2012). Dry biomass was determined by drying samples of each component at 65 °C until constant weight.

The Gompertz model (Tjørve and Tjørve, 2017) was selected to predict above-ground biomass (AGB) due to its previously reported high performance in forest modeling for this species (Kruchelski et al., 2022; Vidal et al., 2026). The model was fitted using the Levenberg-Marquardt method (Elzhov et al., 2016) in the R software environment (R Core Team, 2025), utilizing the combination of DBH^2 and h as independent variables the allometric equation is described as follows (Eq. 2):

$$y = \beta_0 \cdot e^{(-e^{(\beta_1 - \beta_2 \cdot X)})} + \varepsilon \quad (2)$$

Where: y is the total above-ground biomass (AGB, in kg); β_0 , β_1 , and β_2 are the coefficients of the non-linear regression model; X is the combined variable (DBH^2h); and ε is the residual error.

The total tree carbon stock was calculated following three steps: (i) estimation of AGB using the adjusted Gompertz equation; (ii) estimation of below-ground biomass (BGB) applying the root-to-shoot ratio of 0.205 (Mokany et al., 2006; IPCC, 2006); and (iii) conversion of total biomass (AGB + BGB) to carbon using the factor of 0.47 (IPCC, 2006). The individual tree carbon values were summed and extrapolated to the area level to obtain the total tree carbon stock (Mg C ha^{-1}).

To quantify timber production, the stem volume of individual trees was estimated using a fifth-degree polynomial tapering function (Schöepfer, 1966) adjusted from 31 harvested trees. The detailed methodology regarding the tapering function adjustment, destructive sampling for volume, and the validation statistics are provided in Supplementary Material S1. The Mean Annual Increment (MAI) was then determined by multiplying the average annual volumetric increment per tree ($\text{m}^3 \text{ year}^{-1}$) by the tree density (52 trees ha^{-1}) at the time of evaluation.

4.3.7 Ecosystem services index calculation

The ecosystem services in the livestock and livestock-forestry systems was evaluated using an Ecosystem Services Index (ESI). The calculation framework followed a three-step procedure outlined in the literature (Cherubin et al., 2016; Rinot et al., 2019; Bieluczyk et al., 2025). Step I (selection of indicators): (i) Supporting services, assessed through soil physical quality functions including support for plant growth (Bulk Density - BD), soil water storage (Microporosity - MIC), soil aeration (Macroporosity - MAC), and soil structural support (Penetration Resistance - PR); (ii) Regulating services, represented by carbon storage capacity, comprising soil carbon retention (Soil Carbon Stocks - SCS) and tree biomass storage (Tree Carbon Stocks - CT); and (iii) Provisioning services, measured by productivity indicators including animal performance (Daily Live Weight Gain - DWG), forage availability (Total Herbage Accumulated - THA), and timber yield (Mean Annual Increment - MAI). The indicators comprising the minimum data set were grouped into three ecosystem services categories to evaluate the integrated performance of the systems (Table 4).

Table 4 - Indicators, functional categories, and weights used to calculate the Ecosystem Services Index (ESI)

Ecosystem Services	Weight I	Indicators	Weight II
Supporting Service	0.33	Bulk density - BD	0.25
		Macroporosity - MAC	0.25
		Microporosity - MIC	0.25
		Penetration Resistance - PR	0.25
Regulating Service	0.33	Soil Carbon Stock - SCS	0.50
		Tree Carbon Stocks - TCS	0.50
Provisioning Service	0.33	Daily Live Weight Gain - DWG	0.33
		Total Herbage Accumulated - THA	0.33
		Mean Annual Increment - MAI	0.33

SOURCE: Author (2026).

Step II (Interpretation of measured indicators): The values of each indicator were normalized into dimensionless scores ranging from 0 to 1. The transformation followed a "more is better" or "less is better" approach depending on the indicator's relationship with the soil function or ecosystem service regarding production and environmental protection (Cherubin et al., 2016). The more is better and less is better scoring curves were determined by literature review and consensus of study co-authors (Table 5). Therefore, for "more is better" indicators (i.e., MAC, MIC, SCS, TCS, ADG, THA and MAI), the score was calculated by dividing the observed value by the highest value in the dataset (Eq. 3). For "less is better" indicators (i.e., BD and PR), the score was obtained by dividing the lowest value in the dataset by each observed value (Eq. 4), as follows:

$$S = \frac{X}{X_{max}} \quad (3)$$

$$S = \frac{X_{min}}{X} \quad (4)$$

Where S is the normalized score (0-1), X is the observed value, X_{max} is the maximum observed value, and X_{min} is the minimum observed value.

Table 5 - Indicator thresholds and scoring curves for the assessment of ecosystem services

Indicator	Unit	Xmax ¹	Xmin	Scoring curve
Bulk density - BD	Mg m ⁻³	1.47	0.95	Less is better
Macroporosity - MAC	m ³ m ⁻³	0.12	0.05	More is better
Microporosity - MIC	m ³ m ⁻³	0.51	0.42	More is better
Penetration Resistance - PR	MPa	2.74	0.92	Less is better
Soil Carbon Stock - SCS	Mg ha ⁻¹	247.10	51.50	More is better
Tree Carbon Stocks - TCS	Mg ha ⁻¹	49.08	32.25	More is better
Daily live weight gain - DWG	kg ha ⁻¹ day ⁻¹	2.81	1.57	More is better
Total herbage accumulated - THA	kg ha ⁻¹	21,476	10,630	More is better
Mean Annual Increment - MAI	m ³ ha ⁻¹ year ⁻¹	35.19	25.99	More is better

¹ Xmax and Xmin: maximum and minimum values, respectively.

SOURCE: Author (2026).

Step III (Integration of indicators in an overall index): Finally, a weighted additive integration strategy was employed to calculate the ESI (Rinot et al., 2019; Bieluczyk et al., 2025). The normalized scores were aggregated to represent their associated ecosystem service. Assuming equal importance for indicators within each service, the weights were distributed as detailed in Table 5. The final ESI was calculated as the sum of the weighted scores, according to Eq. 5:

$$ESI = \sum_{i=1}^n Si * Wi \quad (5)$$

Where Si is the normalized score of the *i*-th indicator and Wi is the weight assigned to that indicator.

4.3.8 Statistical analyses

The data were initially tested for normality of residuals using the Shapiro-Wilk test and for homogeneity of variances using the Bartlett test. Once the assumptions were met, the results were subjected to Analysis of Variance (ANOVA) according to the randomized block design to compare the livestock and livestock-forestry systems. Specific criteria were applied depending on the variable type: soil physical indicators were evaluated separately for each sampled layer, while Soil Carbon Stocks (SCS) were analyzed considering three cumulative depths (0-0.20 m, 0-0.30 m, and 0-1.00 m). Productive variables (sward height, herbage mass, herbage accumulation rate, total herbage accumulated, herbage allowance, leaf allowance, stocking rate, average daily gain, and daily live weight gain) were compared directly using the F-test at a 5% significance level ($p < 0.05$). The individual ecosystem services Provisioning (P),

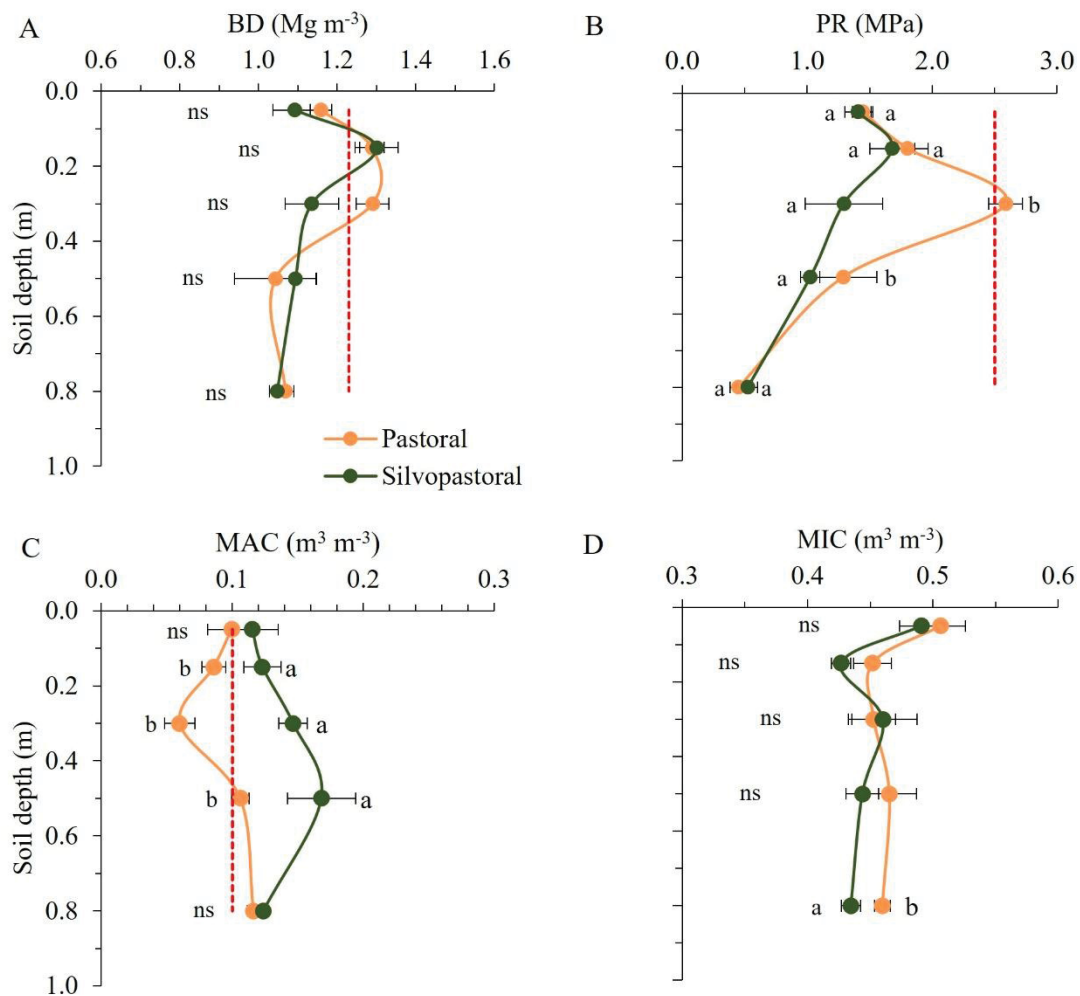
Supporting (S), and Regulating (R) and the overall Ecosystem Services Index (ESI) were also compared between the two systems, with the soil components of the index considering the average of the 0.00-1.00 m depth. All statistical analyses were performed using the ExpDes package within the R statistical software environment.

4.4 RESULTS

4.4.1 Soil physical quality indicators

The soil physical attributes were influenced by the systems (Figure 4). BD did not differ significantly between the livestock and livestock-forestry systems at any evaluated depth ($p > 0.05$; Figure 4A). However, in relation to a critical limit of 1.23 Mg m^{-3} , the average BD for both systems at the 0.1-0.2 m depth was slightly above this threshold, at 1.29 Mg m^{-3} . In the subsequent 0.2-0.4 m layer, only the livestock system exhibited a BD value exceeding this critical level (Figure 4A).

Figure 4 - Soil physical indicators at different depths for livestock (solid line) and livestock-forestry (dashed line) systems. The panels show: (A) Bulk Density (BD, Mg m^{-3}), (B) Penetration Resistance (PR, MPa), (C) Macroporosity (MAC, $\text{m}^3 \text{m}^{-3}$), and (D) Microporosity (MIC, $\text{m}^3 \text{m}^{-3}$). Points represent mean values and error bars indicate the standard error. For each depth, different lowercase letters denote a significant difference between systems, while 'ns' indicates no significant difference (F-test, $p \leq 0.05$). The vertical dashed red lines indicate the critical limits for root growth regarding bulk density (BD; 1.23 Mg m^{-3} , Pachepsky and Park, 2025) in (A), and penetration resistance (PR; 2.5 MPa , Cavalieri-Polizeli et al., 2022) in (B). In (C), the line represents the critical limit for soil aeration ($0.10 \text{ m}^3 \text{m}^{-3}$, Grable & Siemer, 1968).



SOURCE: Author (2026).

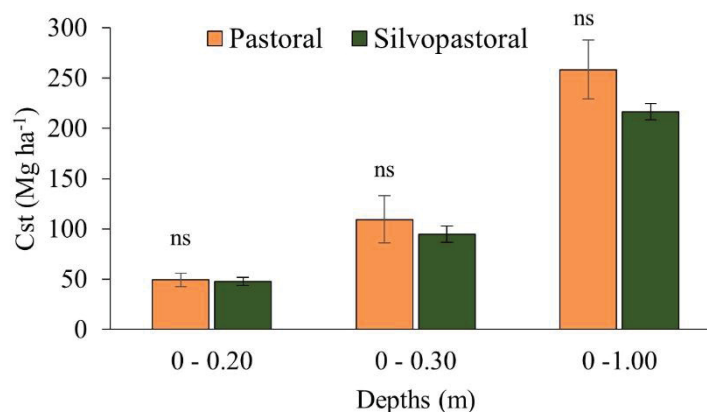
The PR revealed more pronounced differences. This indicator was significantly higher in the livestock-forestry system at the 0.2-0.4 m and 0.4-0.6 m depths, exceeding the livestock-forestry system by 50% and 21%, respectively ($p \leq 0.05$; Figure 4B). Notably, only the livestock system surpassed the critical PR for root growth of 2.5 MPa in the 0.2-0.4 m layer. Consequently, soil aeration, indicated by macroporosity, was significantly improved in the livestock system, which showed values 43%, 144%, and 58% greater than the livestock system

in the 0.1-0.2 m, 0.2-0.4 m, and 0.4-0.6 m depths, respectively (Figure 4C). This allowed the livestock-forestry system to maintain macroporosity at or above the critical threshold of $0.10 \text{ m}^3 \text{ m}^{-3}$ down to a depth of 0.4 m. In contrast, microporosity was largely unaffected by the systems, showing statistical similarity at all depths except for the 0.6-1.0 m layer, where the livestock system had a microporosity 5% higher than the livestock-forestry system ($p \leq 0.05$; Figure 4D).

4.4.2 Soil and tree carbon stocks

The Cst did not differ between the livestock and livestock-forestry systems across all evaluated depths ($p > 0.05$; Figure 5).

Figure 5 - Corrected carbon stock (Cst, Mg ha^{-1}) in livestock and livestock-forestry systems at three cumulative depths (0-0.20 m, 0-0.30 m, and 0-1.00 m). Bars represent the mean stock (Mg ha^{-1}) and error bars indicate the standard error. For each depth, 'ns' denotes no significant difference between the systems (F-test, $p > 0.05$).



SOURCE: Author (2026).

In the 0-0.20 m layer, the Cst was 49.4 Mg ha^{-1} in the livestock system and 47.7 Mg ha^{-1} in the livestock-forestry system. For the 0-0.30 m depth, relevant for IPCC inventories, the stocks were 109.4 Mg ha^{-1} and 94.8 Mg ha^{-1} for the livestock and livestock-forestry systems, respectively. Considering the entire soil profile down to 1.00 m, the total Cst was 258 Mg ha^{-1} in the livestock system and 216 Mg ha^{-1} in the livestock-forestry system.

To estimate the above-ground tree biomass and adjust the allometric equation, a total of 34 trees were harvested from the Integrated Livestock-Forestry and Integrated Crop-Livestock-Forestry (16 and 18 trees, respectively). The Gompertz model proved to be efficient for estimating the biomass of *Eucalyptus benthamii*, presenting a high coefficient of determination ($R^2 = 0.976$) and a low Root Mean Square Error (RMSE = 9.58 %; 57.14 kg) and a consistent

Akaike Information Criterion (AIC = 379.6). Furthermore, the White test (2.19^{ns}) confirmed the homoscedasticity of the residuals, indicating the robustness of the model for biomass prediction. The adjusted allometric equation used was (Eq. 6):

$$\text{Biomass} = 2962 \cdot e^{(-e^{(1.236-0.2308.X)})} \quad (6)$$

Based on these estimates, the Mean Annual Increment (MAI) of the timber component was 31.33 m³ ha⁻¹ year⁻¹. Regarding carbon storage, the mean Tree Carbon Stocks (TCS) in the livestock-forestry system reached 44.26 Mg ha⁻¹. Of this total, 36.73 Mg ha⁻¹ corresponded to above-ground carbon (AGC) and 7.53 Mg ha⁻¹ to below-ground carbon (BGC), highlighting the significant contribution of the forestry component to the system's carbon pool.

4.4.3 Livestock and livestock-forestry system production

Significant differences were observed in pasture structural and production variables between systems (Table 6). Sward height was 0.03 m greater in the livestock system (0.24 m) compared to the livestock-forestry system (0.21 m) ($p = 0.003$). Consequently, herbage mass (1,902 vs 1,508 kg ha⁻¹), the herbage accumulation rate (64 vs 44 kg ha⁻¹ day⁻¹), and the total herbage accumulated (19,310 vs 13,122 kg ha⁻¹) were all significantly higher in the livestock system ($p < 0.001$ for all variables).

Table 6 - Sward structure, forage accumulation, and animal performance in livestock and livestock-forestry systems

Variables		Systems		SE ¹	p-value
		Livestock	Livestock-Forestry		
Sward height	m	0.24	0.21	0.0052	0.003
Herbage Mass	kg ha ⁻¹	1902	1508	53	<0.001
Herbage accumulate rate	kg ha ⁻¹ day ⁻¹	64	44	2.67	<0.001
Total herbage accumulated	kg ha ⁻¹	1,9310	13,122	481	<0.001
Herbage allowance	kg kg ⁻¹	2.50	2.50	0.084	0.754
Leaf allowance	kg kg ⁻¹	0.85	0.90	0.040	0.410
Stocking rate	kg ha ⁻¹	817	694	30	<0.001
Average daily gain	kg an ⁻¹ day ⁻¹	0.819	0.840	0.034	0.695
Daily live weight gain	kg ha ⁻¹ day ⁻¹	2.40	1.80	0.09	<0.001

¹ SE: standard error of the mean

Despite the differences in forage production, the herbage allowance (2.5 kg kg^{-1}) and leaf allowance (0.85 kg kg^{-1}) were kept statistically similar between systems ($p = 0.754$ and $p = 0.410$, respectively). To achieve this, the stocking rate was significantly lower ($p < 0.001$, Table 6) in the livestock-forestry system (694 kg ha^{-1}) compared to the livestock system (817 kg ha^{-1}).

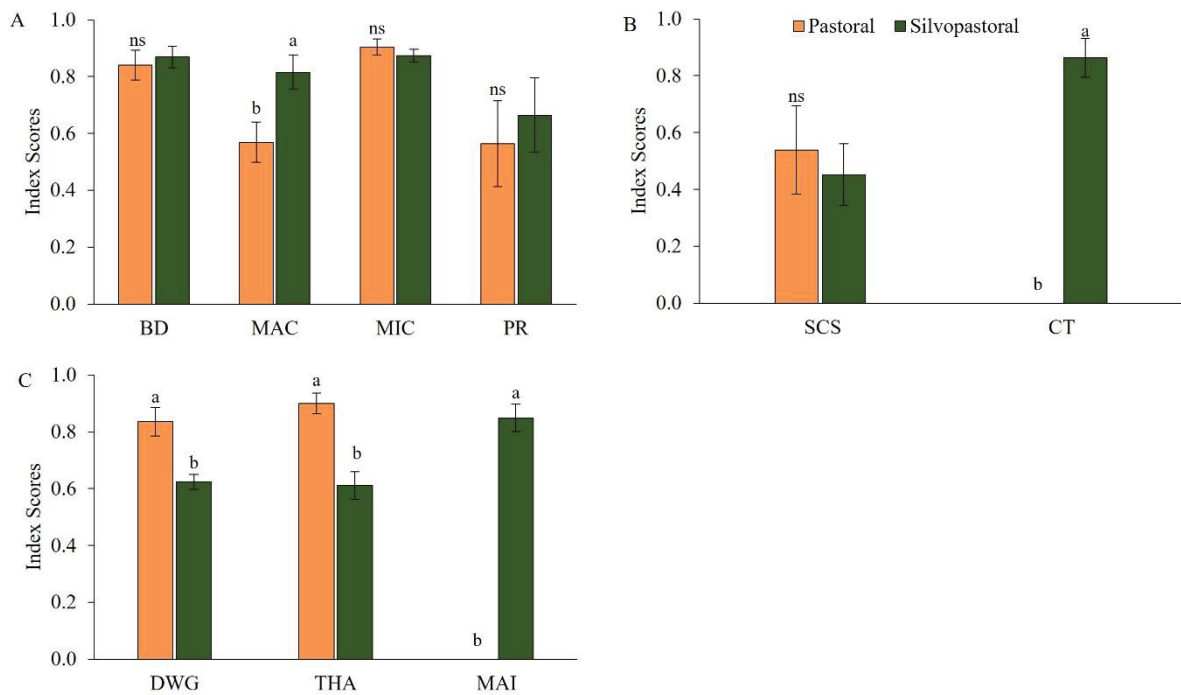
The average daily gain per animal showed no statistical difference between systems, being $0.830 \text{ kg animal}^{-1} \text{ day}^{-1}$ in the average of systems ($p = 0.695$, Table 6). However, the live weight gain per area was 25% significantly higher in the livestock system when compared to the livestock-forestry system ($p < 0.001$).

4.4.4 Ecosystem services index

In the support services, individual index scores for bulk density (BD; 0.84 in the livestock system vs. 0.87 in the livestock-forestry system), microporosity (MIC; 0.90 vs. 0.87), and penetration resistance (PR; 0.56 vs. 0.66) were statistically similar ($p > 0.05$) between the systems (Figure 6A). In contrast, the livestock-forestry system exhibited a significantly higher macroporosity (MAC, Figure 6A) score than the livestock system (0.82 vs. 0.57, respectively).

For regulating services, SCS was similar between systems ($p > 0.05$), while CT was specific to the livestock-forestry system (Figure 6B). In the provisioning services, livestock system presented higher index scores for DWG and THA, whereas the livestock-forestry system exclusively recorded the timber MAI (Figure 6C).

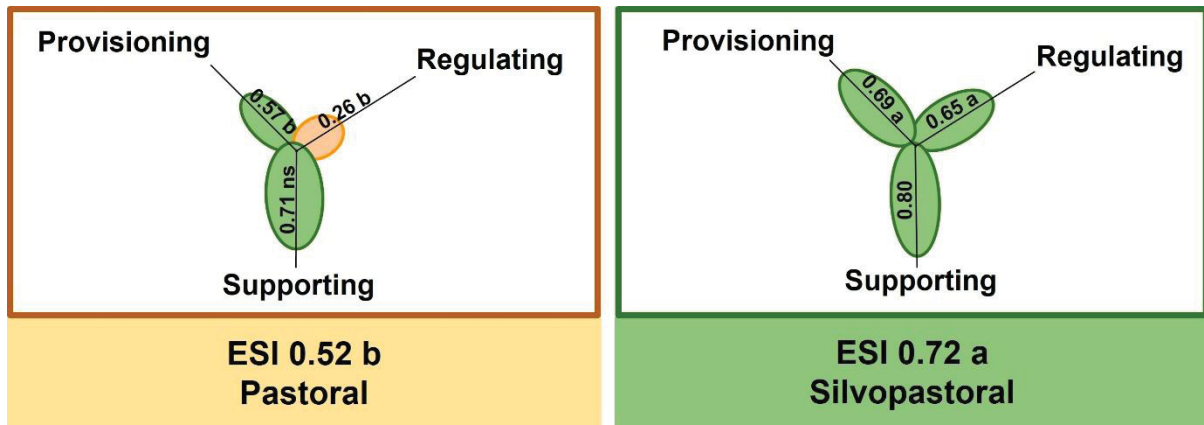
Figure 6 - Indicators of ecosystem services assessed in livestock and livestock-forestry systems. The normalized scores represent: **(A)** Supporting services (soil physical quality): Bulk Density (BD), Macroporosity (MAC), Microporosity (MIC), and Penetration Resistance (PR); **(B)** Provisioning services (productive performance): Daily Live Weight Gain (DWG), Total Herbage Accumulated (THA), and timber Mean Annual Increment (MAI); and **(C)** Regulating services (climate regulation potential): Soil Carbon Stocks (SCS) and Tree Carbon Stocks (CT). Bars with different letters indicate significant differences between systems according to F-test ($p < 0.05$). ns: not significant



SOURCE: Author (2026).

When aggregated into ecosystem services categories (Figure 7), the Supporting service scores did not differ significantly between the systems (0.71 in the livestock system vs. 0.80 in the livestock-forestry system; $p > 0.05$). However, the livestock-forestry system demonstrated significantly higher scores for both Provisioning (0.69 vs. 0.57) and Regulating (0.65 vs. 0.26) functions compared to the livestock system (Figure 7).

Figure 7 - Integrated assessment of ecosystem services in the livestock and livestock-forestry systems. The diagram illustrates the specific scores for (i) Supporting (soil physical quality), (ii) Regulating (soil and tree carbon stocks), and (iii) Provisioning (animal production, forage, and timber) services. The boxes below indicate the overall Ecosystem Services Index (ESI), which aggregates the three specific functions. Means followed by different lowercase letters indicate significant differences between the systems (Livestock vs. Livestock-Forestry) for the same ecosystem service and for the overall ESI, according to F-test ($p < 0.05$). ns: no significant difference between systems.



SOURCE: Author (2026).

The overall performance, measured by the Ecosystem Services Index (ESI, Figure 7), confirmed the superiority of the livestock-forestry system, which achieved a significantly higher ESI score compared to the livestock system (0.71 vs. 0.52, respectively).

4.5 DISCUSSION

4.5.1 Performance of ecosystem services indicators

The assessment of individual indicators revealed that soil physical quality was generally maintained across systems. The lack of significant differences in BD and PR between the livestock-forestry and livestock systems demonstrate the structural resilience of the systems (Figure 4). This physical stability is likely underpinned by the intrinsic properties of the local Ferralsol, particularly its high clay content and stable micro-aggregation, which dissipate mechanical stress (Sá et al., 2025; Teixeira et al., 2025). Additionally, the continuous root activity of the perennial pasture, associated with grazing management under moderate grazing intensity, plays a fundamental role in reinforcing soil structure, promoting aggregation that counteracts the effect of hoof pressure, maintaining the soil's capacity to support plant growth in both systems (Daboit et al., 2025).

However, a distinct pattern emerged regarding soil aeration, where the livestock-forestry system exhibited superior macroporosity scores compared to the livestock system

(Figure 6). This significant improvement suggests a positive synergistic effect of the arboreal component on pore size distribution. The deep and coarse root system of *Eucalyptus benthamii* promotes the formation of preferential flow paths and biopores, enhancing air and water movement through the soil profile (Bieluczyk et al., 2025). This mechanism is crucial in livestock-forestry systems, as it improves the soil's hydraulic conductivity and resistance to erosion, providing a superior supporting service compared to the livestock-forestry system (Coblinski et al., 2019).

Furthermore, the maintenance of these physical attributes is essential for the sustainability of the system's biological functioning. As highlighted by Bieluczyk et al. (2025), physical integrity is a precursor to soil health, facilitating root exploration and microbial activity. The presence of trees contributes to a more complex litter layer and varying root depths, which stimulates soil biota and further stabilizes soil aggregates (Silva et al., 2020).

The results confirm that the introduction of trees into the pasture did not compromise soil structure, validating the system's viability for long-term intensification (Figure 4). While the livestock system relied heavily on the fibrous root system of grasses to maintain porosity, the livestock-forestry system benefited from the complementary action of woody roots. This duality enhances the resilience of the supporting services, ensuring that the soil remains functional even under the pressure of combined animal and forestry production (Monteiro et al., 2025).

4.5.2 Ecosystem services synergies and trade-offs between livestock and livestock-forestry systems

Regarding provisioning services, a clear trade-off was identified between the forage-livestock axis and the forestry component. The livestock system achieved significantly higher scores for livestock-related indicators, specifically Daily Live Weight Gain (DWG) and Total Herbage Accumulated (THA). This reduction in the livestock-forestry system is a typical ecophysiological response to light competition, as shading restricts the photosynthetic rate of the C₄ forage grass (*Megathyrsus maximus*), reducing biomass accumulation per unit area (Ortiz et al., 2023; Portugal et al., 2023). Consequently, the stocking rate must be adjusted downwards in the livestock-forestry systems to match the lower carrying capacity, which explains the lower total animal output per hectare compared to the full-sun pasture.

However, it is crucial to interpret this trade-off not as a loss, but as a shift in production strategy. While the livestock-forestry system scored lower on specific forage-animal metrics, it was exclusively responsible for timber provision, represented by the high Mean Annual

Increment (MAI). The rapid growth of *Eucalyptus benthamii* demonstrates the species' adaptation to the system, effectively occupying the vertical stratum and generating a high-value product that is absent in the livestock (Kruchelski et al., 2023). This timber production compensates for the reduced livestock revenue, creating a "safety net" for the producer. As noted by Chamorro-Vargas et al. (2025), this product diversification is a key enabler for the adoption of livestock-forestry systems, as it reduces exposure to market volatility associated with a single commodity.

Moreover, the quality of the animal provisioning service should be considered alongside the quantity. Although the total gain per area was lower in the livestock-forestry system, individual animal performance was maintained at competitive levels. The shade provided by trees improves animal thermal comfort, potentially increasing feed conversion efficiency and animal welfare (Martin et al., 2025). In this context, the livestock-forestry systems integrated system optimizes the efficiency of the biological machinery (the cattle) while simultaneously harvesting solar energy through a second canopy layer (the trees), maximizing the overall resource use efficiency of the land (Alves et al., 2025).

Ultimately, the significant enhancement of the Provisioning function observed in Figure 7 confirms that the timber output effectively compensated for the reduction in forage and animal production in the livestock-forestry system. This shift indicates a transition from a specialized system to a multifunctional one. While the livestock system maximizes a single output (meat), the livestock-forestry system optimizes the combined output of meat and wood (Table 6). This productive complementarity is a distinctive feature of livestock-forestry systems, offering a pathway to increase the overall productive and economic yield of agricultural landscapes without expanding into new areas (Monteiro et al., 2025).

In terms of regulating services, distinct patterns emerged regarding carbon dynamics. While SCS did not differ significantly between systems, the presence of trees introduced a crucial carbon sink component (CT), which was absent in the livestock system (Figure 5). The stability of SCS indicates that the conversion to a livestock-forestry system did not deplete soil organic matter, maintaining the baseline stock established by the pasture (Cá et al., 2022). This persistence of soil carbon is vital, as grassland soils in Southern Brazil are significant reservoirs, and their preservation relies on the physical protection afforded by soil aggregation and clay content (Sá et al., 2025; Teixeira et al., 2025).

Consequently, the distinctive contribution of the livestock-forestry system to climate regulation lies in the "additionality" of the tree biomass. While the pasture component cycles carbon relatively quickly through grazing and decomposition, the woody biomass of *Eucalyptus*

benthamii provides long-term storage, acting as a persistent drain for atmospheric CO₂ (Antunes et al., 2025; Machado et al., 2025). This arboreal component significantly elevated the Regulating function score in the livestock-forestry system (Figure 7), driven by the substantial carbon storage in tree trunks, branches, and roots. This finding aligns with global assessments identifying agroforestry as a high-potential solution for natural climate solutions (Huang et al., 2023).

Furthermore, the integration of trees may enhance the quality and stability of the carbon stored. Although total soil stocks were similar, the input of recalcitrant litter from eucalyptus and the turnover of woody roots contribute to more stable soil organic matter fractions over time (Alves et al., 2025). This qualitative improvement, combined with the quantitative accumulation in biomass, creates a robust regulating service that goes beyond simple sequestration. The system effectively couples the fast-cycling C pool of the pasture with the slow-cycling C pool of the trees, maximizing the residence time of carbon within the ecosystem (Amadori et al., 2022).

Therefore, the livestock-forestry system represents a strategy for climate change mitigation compared to livestock system – an open pastureland. By combining the stable soil C stock with the rapidly accumulating tree biomass, the livestock-forestry systems maximizes the regulating service per unit area. This capacity to sequester carbon while maintaining agricultural production is critical for meeting national and international climate goals, positioning livestock-forestry systems as a key technology for low-carbon agriculture (Corte et al., 2022; Monteiro et al., 2024).

The visual integration of ecosystem services through radar charts (Figure 7) illustrates the enhanced multifunctionality of the livestock-forestry system. While the livestock system relied heavily on supporting and livestock-provisioning services, the livestock-forestry system exhibited a more balanced and expanded distribution of services, covering supporting, provisioning (wood and meat), and regulating ecosystem services. This broad distribution of benefits is the visual representation of resilience; a system that depends on multiple pillars is less vulnerable to the collapse of any single function (Carvalho et al., 2024; Silva-Olaya et al., 2025). The expansion of the "service area" in the radar chart confirms that integration does not merely trade one service for another but expands the total ecosystem service envelope of the land.

The overall performance, measured by the ESI, confirmed the statistical superiority of the livestock-forestry system (Figure 7). The livestock-forestry system achieved a significantly higher ESI score compared to the livestock system. This aggregated metric highlights that the

synergistic effects of integrating trees with pasture, specifically the gains in climate regulation and timber provision, outweighed the local trade-offs associated with reduced pasture yield. This aligns with the concept of sustainable intensification, where the goal is to increase the efficiency of resource use and the delivery of environmental benefits simultaneously (Bieluczyk et al., 2025).

4.5.3 Challenges, opportunities, and limitations of the study

Beyond the specific results of this study, the proposal of the ESI represents a methodological advancement for evaluating livestock-forestry systems. While traditional approaches often analyze production and environmental variables in isolation, the ESI allows for a holistic understanding of the system's multifunctionality including environmental and agricultural components. As highlighted by Bieluczyk et al. (2025), integrated frameworks facilitate stakeholders' perceptions and guide public policy better than analyzing diverse disconnected properties. By incorporating supporting (soil physical quality), regulating (carbon stocks), and provisioning (forage, animal and tree biomass production) services into a single metric, this methodology reveals trade-offs that simple productivity indicators might miss. For instance, while the livestock-forestry system may have different stocking rates compared to livestock, the ESI demonstrates how the arboreal component compensates through climate regulation and soil protection services. Therefore, this index serves as a potential standardized tool for future research and for designing Payment for Ecosystem Services (PES) schemes, shifting the focus from specific yield maximization to overall system sustainability.

Despite the methodological advancements provided by the ESI, some limitations of this study must be acknowledged. First, the assessment represents a “snapshot” taken 10 years after the experiment's establishment, capturing a specific mature stage of the livestock-forestry system. Because the temporal dynamics were not evaluated year by year, the index reflects the cumulative effects over a decade rather than the continuous fluctuations and early-stage trade-offs in ecosystem services provision during the system's development. Furthermore, the findings are inherently linked to the specific local conditions a subtropical climate and clayey Ferralsol and the use of *Eucalyptus benthamii*, a fast-growing species with a distinct canopy architecture. Therefore, while the ESI framework itself is adaptable, extrapolating these specific index scores to other biomes, soil types, or integrated systems with different tree species should be done with caution, as different arrangements may yield distinct ecosystem service synergies and trade-offs.

In conclusion, this study demonstrates that livestock-forestry systems are a viable strategy to enhance ecosystem services in the subtropic climate region. The trade-off in beef cattle production is effectively neutralized by the substantial gains in timber provision and carbon sequestration, resulting in a system with greater overall environmental and productive value. The ESI framework proved to be a robust tool for capturing these complex interactions, offering a clear metric to value the multifunctionality of integrated agricultural landscapes. These findings support the expansion of livestock-forestry systems as a pathway to reconcile food security with climate change mitigation and soil conservation.

4.6 CONCLUSION

The livestock-forestry system proves to be an effective strategy for sustainable intensification, validating the hypothesis that integration promotes an overall ecosystem services improvement compared to the livestock (ESI: 0.72 vs. 0.52). Although shading imposed an inevitable trade-off resulting in reduced forage and livestock production, this decline was functionally offset by timber production, raising the Provisioning Ecosystems Services compared to in the livestock system. Furthermore, the significant additionality of carbon sequestration in tree biomass resulted in an increase in Regulating Ecosystems Services. Simultaneously, the livestock-forestry system maintained soil structural quality, achieving a supporting service score similar to the open system. Therefore, the livestock-forestry arrangement not only diversifies the farm's economic output but also acts as a robust tool to reconcile food security with climate change mitigation in subtropical landscapes.

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5 CHAPTER 3 - THE TRADE-OFF BETWEEN FLUID TRANSPORT PROPERTIES AND ORGANIC CARBON PRESERVATION IN CLAYEY SOILS

5.1 ABSTRACT

The relationship between soil hydraulic properties and soil organic carbon (SOC) is complex. The objective of this study was to analyze the relationship between soil hydraulic properties and soil organic carbon, seeking to understand how the architecture and functional properties of the pore system affect this interaction. This study was conducted within a long-term experiment (initiated in 2012) at the Canguiri Experimental Station in Pinhais, Brazil, on a clayey Ferralsol managed under four pesticide-free conservation systems: Livestock (L), Crop-Livestock (CL), Livestock-Forest (LF), and Crop-Livestock-Forest (CLF). In 2022, soil samples were collected down to a depth of 0.30 m. Soil attributes analyzed included saturated hydraulic conductivity (K_s), intrinsic air permeability of soil (K_a) at matric potentials -3 kPa and -10 kPa, soil bulk density, total porosity, large macroporosity (MAC; $\varnothing > 100 \mu\text{m}$), macroporosity (MaP; $\varnothing > 30 \mu\text{m}$), microporosity (MIC; $\varnothing < 30 \mu\text{m}$), air-filled porosity, organization and connectivity indexes and soil organic carbon. Results indicated that SOC was negatively correlated with saturated hydraulic conductivity (K_s) and intrinsic air permeability (K_a). This suggests that enhanced flux transport, particularly gas exchange, may accelerate organic matter mineralization. The pore system architecture, governed by large macroporosity (MAC) and its functionality, was the main driver of these fluxes, with K_s showing a strong positive correlation with the organization (k_1) and connectivity (k_2) index. The most pronounced inverse relationship was observed between SOC and intrinsic air permeability in macropores $K_{a(\text{MAC})}$, reinforcing that a highly connected pore network facilitates oxygen diffusion. Consequently, increases in pore connectivity and permeability were associated with variations in SOC content, highlighting a dynamic trade-off between physical functionality and carbon preservation in these systems.

Key words: Soil Structure, Pore Functionality; Porosity Connectivity; Air Permeability.

5.2 INTRODUCTION

Soil organic carbon (SOC) is an essential component for the health of terrestrial ecosystems, influencing soil fertility, soil water retention, and the mitigation of climate change (Kumar et al., 2019; Wang et al., 2020). Soil organic matter directly affects physical properties by promoting the formation and stabilization of aggregates, this contributes to the development of a more stable and functional porous structure (Tisdall & Oades, 1982; Meurer et al., 2020; Blanco-Canqui et al., 2023). Understanding these relationships is fundamental to guiding management practices that balance carbon conservation with maintenance of the soil's physical functionality.

Soil physical properties, particularly saturated hydraulic conductivity (K_s) and air permeability (K_a), are indicators of the functionality of soil's porous structure (Poulsen et al., 2013; Blanchy et al., 2023; Tobella et al., 2024). Pore network functionality depends on the degree of organization, connectivity and continuity of the pores. Together these attributes regulate fluid flow, including processes such as infiltration and aeration. In turn these processes critical for sustaining root growth, soil respiration, and soil microbial activity (Poulsen, 2013; Martínez et al., 2016). (Silva et al., 2021; Suzuki et al., 2022).

Within the soil pore architecture, pore size is a key determinant of soil hydraulic and aeration behaviour, in which macropores function as preferential pathways for the rapid transport of water and air, while micropores are predominantly associated with water retention (Tisdall & Oades, 1982). According to the classic definition by Tisdall & Oades (1982), macropores are characterized as pores $> 75 \mu\text{m}$, a threshold primarily defined to allow root penetration, functioning as preferential pathways for the rapid transport of water and air, while micropores are associated with water retention. However, this study followed the classification by Koorevaar (1983), specifically distinguishing large macroporosity as pores $> 100 \mu\text{m}$. The choice of this higher threshold is justified because these large macropores represent the most conductive fraction of the pore network, acting as the primary conduits for rapid water infiltration and gas exchange. However, the total pore volume does not fully describe its functionality. For this reason, indexes of the pore network's organization and connectivity have been used to evaluate its geometry and performance. High values for these indexes indicate a more continuous and efficient structure for the transport of fluids and gases (Hamamoto et al., 2016; Poulsen, 2013; Gamboa et al., 2023).

In general, studies report that an increase in SOC favors the formation of stable structures with a greater presence of functional macropores, which tends to increase K_s (Tisdall & Oades, 1982; Tobella et al., 2024). However, more recent evidence has challenged this

generalization, pointing to negative correlations under certain conditions. Field studies and large-scale database analyses show that, in some soils, high levels of organic matter can be associated with a reduction in hydraulic conductivity (Nemes et al., 2005; Wang et al., 2009; Jarvis et al., 2013). Thus, the relationship between SOC and transport soil properties, such as K_s and K_a , can be considered complex. For example, hydrophobicity induced by organic compounds can restrict water flow in sandy soils (Wang et al., 2009; Jarvis et al., 2013). Furthermore, the physical and biogeochemical processes that control carbon changes occur at the microscale: pore geometry defines the flow paths and creates reactive zones (hotspots), where the availability of water and oxygen modulates the rates of carbon decomposition (Yan et al., 2018). Thus, the same structure that protects carbon from decomposition can, under certain conditions, favor its mineralization (Meurer et al., 2020).

Despite the recognized importance of soil structure, the specific influence of pore network architecture on organic carbon dynamics in tropical soils remains poorly understood, often lacking a comprehensive integration of how different pore size fractions and their connectivity simultaneously regulate fluid fluxes and carbon persistence. This study is based on the hypothesis that fluid transport capacity (K_s and K_a) in these soils is primarily governed by the abundance and continuity of large macropores (MAC), rather than total porosity; however, we postulate a functional trade-off where the same organized and connected pore network that favors high permeability to water and air also intensifies oxidative processes, leading to decrease in soil organic carbon (SOC). Thus, the objective of this work was to analyze the interrelationships between soil fluid transport properties (saturated hydraulic conductivity, K_s and air permeability, K_a), pore system attributes (macroporosity, mesoporosity, microporosity, and organization and connectivity indexes), as well soil organic carbon content.

5.3 MATERIAL AND METHODS

5.3.1 Study Area

This research was carried out at the Agricultural Technological Innovation Center (NITA), situated within the Canguiri experimental farm of the Federal University of Paraná (UFPR), in Pinhais, Paraná, Brazil (25°24'02" S, 49°07'12" W; elevation 930 m). According to the Köppen classification, the climate is categorized as Cfb, humid subtropical with no distinct dry season. The study is part of a long-term field trial initiated in 2012 on a site totaling roughly 35 hectares, characterized by prior physical soil degradation (Cavaliere-Polizeli et al., 2022).

The soil, classified as Ferralsol (IUSS Working Group WRB, 2022), has a clayey texture, composed of approximately 52% clay, 10% silt, and 38% sand (see Supplementary Table 3).

5.3.2 Site History, Use and Management

Pesticides have not been used at the Canguiri experimental farm since 1996 due to Decree No. 1753/1996, although mineral fertilizers remain permitted (Cavalieri-Polizeli et al., 2022). Between 2002 and 2011, the area was subjected to conventional maize (*Zea mays*) cultivation and intensive use for agricultural machinery training, which led to severe soil compaction and erosion. In 2012, subsoiling was performed to 0.40 m depth, followed by harrowing up to 0.20 m. That same year, treated sewage sludge (N VIRO®) was incorporated at a rate of 10 Mg ha⁻¹. Black oat (*Avena strigosa*) was sown during the winter, along with 100 kg ha⁻¹ of P₂O₅ applied in-furrow across the entire area. From 2012 to 2021, the mean annual fertilizer inputs included 200 kg ha⁻¹ of nitrogen (urea), 115 kg ha⁻¹ of P₂O₅ (natural phosphate), and 115 kg ha⁻¹ of K₂O (potassium chloride).

In 2012, the field trial was established using a randomized block design with four systems of soil use: Livestock (L), Crop-Livestock (CL), Livestock-Forest (LF), and Crop-Livestock-Forest (CLF), each replicated in three blocks. The systems follow principles of conservation agriculture, now often described as conservationist practices. Management strategies included no-tillage with “planting green,” applied as an alternative to herbicide desiccation for weed control (Mirsky et al., 2012; Dominschek et al., 2021), pesticides-free, mineral fertilization, adequate animal stocking, and reduced machinery traffic.

In the Livestock (L) system, perennial summer pasture was established with *Megathyrus maximus* cv. Aries, and black oat was used as a winter cover. Grazing began in 2015 with Angus steers (~200 kg), under continuous stocking. Forage height was maintained around 24 cm year-round, based on recommendations by Carvalho (2013), and stocking rates were adjusted using the “put-and-take” approach (Mott & Lucas, 1952). The mean live weight stocking densities were 866, 718, and 627 kg ha⁻¹ for the L, LF, and CLF systems, respectively, maintained for 10 months annually, with two months allocated for pasture reestablishment.

The CL and CLF systems followed a ley farming approach, alternating three years of pasture with a summer maize crop, sown directly into the standing winter cover. Oats were also established without herbicide application.

In the forest-integrated systems (LF and CLF), 357 eucalyptus trees (*Eucalyptus benthamii*) per hectare were planted in 2013 following the terrain's contour lines. Thinning operations occurred at 36, 44, and 78 months, reducing tree densities to 258, 174, and 52 trees

ha⁻¹ in LF and 320, 174, and 52 trees ha⁻¹ in CLF, respectively. Final spacing averaged 28 × 6 m (for further details, see Kruchelski et al., 2021).

5.3.3 Soil Sampling

Due to an unusually dry season in 2021, only one block was sampled that year. The other two blocks were sampled in 2022, once soil moisture conditions were more favorable for structural analysis. In total, 144 undisturbed soil samples were collected using metal cylinders (113 cm³; 0.06 m diameter × 0.04 m height), at three depths: 0.00-0.05 m, 0.05-0.15 m, and 0.15-0.30 m. A weighted average was calculated for the 0.00-0.30 m layer. Each system was characterized by 36 samples from four sampling points per plot across three depths and three blocks.

5.3.4 Soil Assessments

Measured soil physical parameters included saturated hydraulic conductivity (Ks), air permeability (Ka) at -3 and -10 kPa, bulk density, total porosity, air-filled porosity, macroporosity, mesoporosity, microporosity, pore connectivity, and pore organization indexes, defined below. Disturbed samples were analyzed for soil texture and organic carbon content (SOC).

Undisturbed soil samples were saturated for 48 hours and analyzed using the constant head method (Klute & Dirksen, 1986). The saturated hydraulic conductivity (Ks) was computed using the Darcy-Buckingham equation (Eq. 1):

$$Ks = \frac{V * L}{A * t * (h + L)} \quad (1)$$

Where V is the collected water volume (cm³), L is the sample length (m), A is the cross-sectional area (m²), t is the time (h), and h is ³/₄ the height of the upper ring (m) with water. Results were converted to cm h⁻¹.

Following Ks measurements, samples were equilibrated to -3 and -10 kPa using a tension table. Thereafter, the soil samples were weighed and the intrinsic air permeability of soil (Ka) was measured using the permeameter proposed by Figueiredo (2010). The following equation was used (Eq. 2):

$$Ka = \frac{Q * \eta * dz}{As * dP} \quad (2)$$

Where Q is air flow rate ($\text{m}^3 \text{s}^{-1}$), η is air dynamic viscosity (N s m^{-2}), dz is sample height (m), A_s is sample area (m^2), and dP is the pressure differential (Pa). K_a was expressed in μm^2 .

Total porosity (TP) was calculated considering the difference between the saturated soil mass and the dry soil mass, divided by the volume of the ring (Grossman & Reinsch, 2002). Air-filled porosity (ϵ) was determined at both matric potentials. Pore size classification followed Koorevaar (1983), which large macroporosity (MAC, $>100 \mu\text{m}$ in diameter); was calculated as the difference between total porosity (TP) and volumetric water content at -3 kPa . Macroporosity (MaP; $> 30 \mu\text{m}$ in diameter) was determined by the difference between TP and microporosity (MIC, $\leq 30 \mu\text{m}$ in diameter), the latter obtained as volumetric water content at -10 kPa . The organization ($k_1 = K_a/\epsilon$) and connectivity ($k_2 = K_a/\epsilon^2$) indexes were derived from K_a and ϵ values and relate to pore geometry (Groenevelt et al., 1984; Poulsen, 2015). Bulk density was obtained via the volumetric ring method after drying samples at $105 \text{ }^\circ\text{C}$ for 48 hours (Grossman & Reinsch, 2002). Soil texture was obtained using the hydrometer method (Gee & Or, 2002).

Soil organic carbon (SOC) content was determined via the colorimetric method (Quaggio & van Raij, 1979).

5.3.5 Statistical analysis

The data were subjected to the Shapiro–Wilk normality test (Shapiro & Wilk, 1965) and Bartlett’s test for homogeneity of variances, adopting a significance level of $p > 0.05$. Mean values were calculated to characterize the soil attributes at each depth layer (0.00-0.05 m, 0.05-0.15 m, and 0.15-0.30 m).

Regression analyses were performed to assess the relationships among soil physical attributes at each depth. The models were selected based on the significance of the parameters ($p < 0.05$) and the highest coefficient of determination (R^2). The following relationships were evaluated: (i) saturated hydraulic conductivity (K_s , cm h^{-1}) as a function of total porosity (TP), microporosity (MIC), large macroporosity (MAC, $>100 \mu\text{m}$), and macroporosity (MaP, $>30 \mu\text{m}$); (ii) K_s as a function of pore organization (k_1) and connectivity (k_2) indexes for MAC and MaP; (iii) intrinsic air permeability (K_a , μm^2), based on MAC and MaP, as a function of the respective macroporosity attributes; and (iv) Soil physical properties such as saturated hydraulic conductivity (K_s) and intrinsic air permeability (K_a) as a function of soil organic carbon content (SOC). All statistical analyses were performed using the *ExpDes* package in R software, version 4.3.1 (R Core Team, 2023).

5.6 RESULTS

5.6.1 General Characterization of Soil Attributes

Soil physical properties and soil organic carbon (SOC) were characterized for different depths and management systems (Table 7). Soil bulk density (BD) values ranged from 1.08 to 1.25 Mg m⁻³ across the profile, while total porosity and macroporosity varied from 0.52 to 0.59 m³ m⁻³, with the most porous conditions generally observed in the uppermost layer.

Table 7 - General characterization attributes related to carbon stock; water and air flows; soil bulk density, porosity and functionality of the soil pore system in conservationist systems. Livestock (L), Crop-Livestock (CL), Livestock-Forestry (LF), Crop-Livestock-Forestry (CLF) at depths of 0.00-0.05; 0.05-0.15 and 0.15-0.30 m

Soil attributes	SOC	Ks	Ka _(MAC)	Ka _(MaP)	BD	TP	MAC	MaP	MIC	k_1 MAC	k_2 MAC	k_1 MaP	k_2 MaP
	g kg ⁻¹	cm h ⁻¹	μm ²	μm ²	Mg m ⁻³	----- cm ³ cm ⁻³ -----							
0 – 0.05 m													
L	31.3	11.8	13.1	57.7	1.11	0.58	0.04	0.09	0.45	2.2	3.6	2.4	3.3
CL	39.0	10.5	4.3	19.3	1.08	0.59	0.07	0.10	0.42	1.6	2.8	1.7	2.5
LF	28.0	12.0	9.4	14.5	1.17	0.53	0.08	0.09	0.37	2.0	3.1	1.7	2.6
CLF	28.0	13.0	15.1	35.1	1.15	0.56	0.09	0.07	0.41	2.1	3.2	2.0	2.8
0.05 – 0.15 m													
L	24.5	6.5	4.7	56.8	1.22	0.52	0.07	0.14	0.32	1.80	3.08	2.2	3.0
CL	27.8	7.3	7.3	23.7	1.23	0.53	0.05	0.09	0.38	2.06	3.5	1.8	2.7
LF	26.8	7.1	8	25.6	1.25	0.52	0.07	0.10	0.35	1.81	3	2.3	2.4
CLF	25.1	9.9	12.4	24.5	1.27	0.52	0.08	0.07	0.37	2.05	3.1	2.0	2.8
0.15 – 0.30 m													
L	29.5	6.6	10	63.3	1.19	0.54	0.08	0.11	0.35	2.0	3.1	2.2	3.0
CL	27.5	11.2	11.2	41.4	1.25	0.52	0.08	0.07	0.37	2.05	3.2	2.2	3.1
LF	19.5	7.6	8.4	25.7	1.23	0.53	0.06	0.06	0.41	2.1	3.3	2.2	3.0
CLF	18.5	11.3	13.7	35.8	1.21	0.54	0.08	0.08	0.38	2.1	3.2	2.0	2.9

¹ Mean values of the four systems. Soil organic carbon, SOC; Saturated hydraulic conductivity, Ks; intrinsic air permeability of soil Ka_(MAC) and Ka_(MaP) respectively, in pores with diameters > 100 and > 30 μm; bulk density, BD; soil total porosity, TP; macroporos, MAC; macroporosity, MaP; microporosity, MIC; characterization and functioning of the soil pore system: pore organization index k_1 [μm² (m³ m⁻³)⁻¹], pore connectivity index k_2 [μm² (m³ m⁻³)⁻²] in pores with diameters > 100 (MAC) μm and > 30 μm (MaP) and air-filled porosity (ε) for matric potentials of -10 kPa and carbon stock, Cst.

SOURCE: Author (2026).

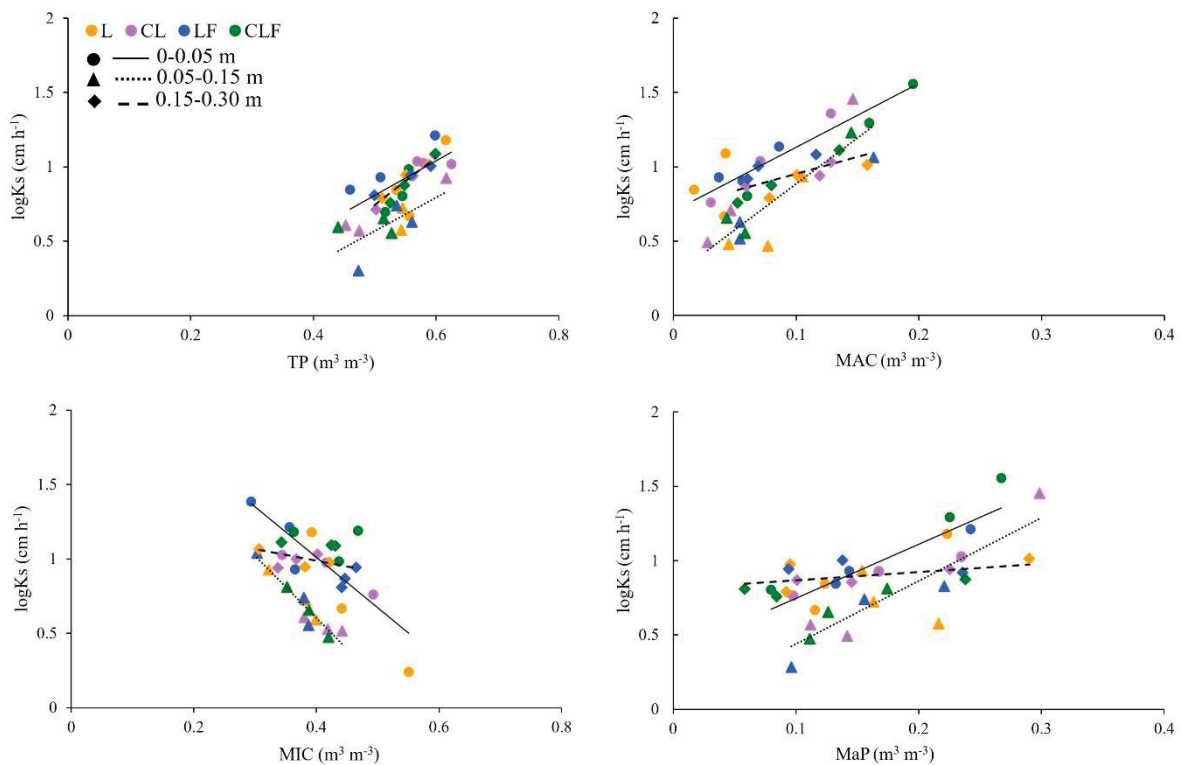
The SOC content varied from 18.5 to 39.0 g kg⁻¹ across the soil profile. For the fluid transport properties, saturated hydraulic conductivity (Ks) values ranged from 10.5 to 13.0 cm

h^{-1} while intrinsic air permeability $K_{a(\text{MAC})}$ varied considerably, with values between 4.3 to 15.1 μm^2 , in the surface layer. The pore organization index (k_1) ranged from 1.6 to 2.2 for large macroporosity (MAC) and 1.7 to 2.4 for macroporosity (MaP). The pore connectivity index (k_2) presented higher ranges, varying from 3.0 to 3.6 for MAC and 2.4 to 3.3 for MaP.

5.6.2 Relationship between Soil Hydraulic Properties, Pore System, and Soil Organic Carbon (SOC)

Regression analyses revealed that the soil physical properties were governed by the structure of the pore system. Saturated hydraulic conductivity (K_s) showed a positive and significant correlation with total porosity (TP), large macroporosity (MAC), and macroporosity (MaP) (Figure 8, Table 8). The relationship with MAC and MaP was particularly strong in the 0.00-0.05 m layer ($R^2 = 0.80$ and $R^2 = 0.81$, respectively). In contrast, K_s showed a negative relationship with microporosity (MIC), especially in the layers of 0.00-0.05 and 0.05-0.15 m of depth.

Figure 8 - Relationship between saturated hydraulic conductivity (K_s) and soil total porosity, TP (A); microporosity, MIC (B); large macroporosity, MAC (C) and macroporosity, MaP (D), at depths of 0.00-0.05; 0.05-0.15 and 0.15-0.30 m.



SOURCE: Author (2026).

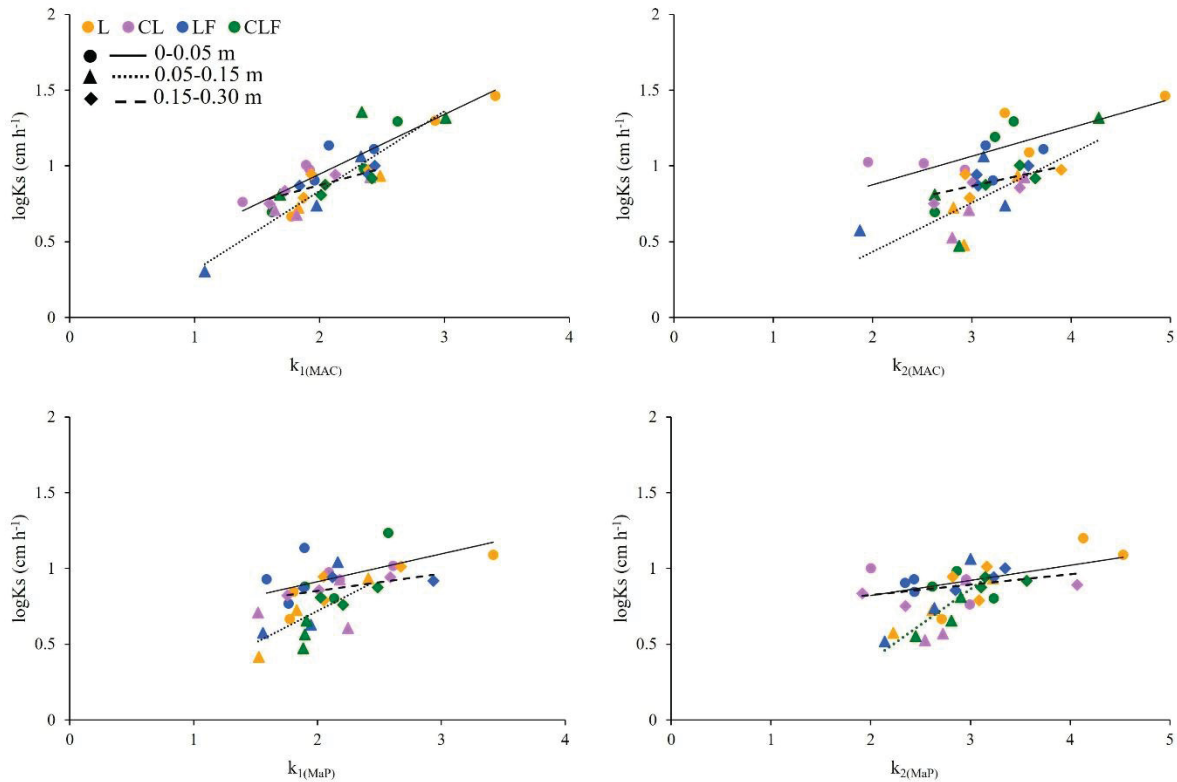
Table 8 - Linear regression between saturated hydraulic conductivity, (K_s , cm h^{-1}) and soil total porosity, TP; microporosity, MIC; Large macroporosity, MAC and macroporosity, MaP at depths of 0.00-0.05, 0.05-0.15, and 0.15-0.30 m.

Soil attributes	Equation	R^2	Probability
logKs vs. TP	$\log K_{s(1)} = -0.35 + 2.33TP$	0.40	<0.001
logKs vs. TP	$\log K_{s(2)} = -0.55 + 2.25TP$	0.45	<0.001
logKs vs. TP	$\log K_{s(3)} = -0.87 + 3.23TP$	0.71	<0.001
logKs vs. MIC	$\log K_{s(1)} = 2.36 - 3.39MIC$	0.62	<0.001
logKs vs. MIC	$\log K_{s(2)} = 2.27 - 4.17MIC$	0.80	<0.001
logKs vs. MIC	$\log K_{s(3)} = 1.29 - 0.78MIC$	0.17	<0.001
logKs vs. MAC	$\log K_{s(1)} = 0.7 + 4.3MAC$	0.80	<0.001
logKs vs. MAC	$\log K_{s(2)} = 0.2 + 6.1MAC$	0.77	<0.001
logKs vs. MAC	$\log K_{s(3)} = 0.7 + 2.3MAC$	0.52	<0.001
logKs vs. MaP	$\log K_{s(1)} = 0.4 + 3.6MaP$	0.81	<0.001
logKs vs. MaP	$\log K_{s(2)} = 0.1 + 4.2MaP$	0.68	<0.001
logKs vs. MaP	$\log K_{s(3)} = 0.8 + 0.5MaP$	0.26	<0.001

(1), (2), and (3) correspond to the depths of 0.00-0.05 m, 0.05-0.15 m, and 0.15-0.30 m, respectively
SOURCE: Author (2026).

Pore geometry was also a determining factor (Figure 9 and 10). The correlation between K_s and the large macropore organization index $k_{1(MAC)}$ was strong in the upper layers ($R^2 = 0.84$ at 0.00-0.05 m and $R^2 = 0.81$ at 0.05-0.15 m), decreasing with depth ($R^2 = 0.66$ at 0.15-0.30 m) (Table 9). The relationship with the large macropore connectivity index $k_{2(MAC)}$ was moderate in all layers, while the relationship with the macropore connectivity index $k_{2(MaP)}$ was stronger in the intermediate layer ($R^2 = 0.69$). Intrinsic air permeability of soil (K_a , Table 10) also showed a strong positive correlation with MAC and MaP, with the $K_{a(MaP)}$ vs. MaP relationship showing the highest coefficient of determination ($R^2 = 0.95$) in the 0.05-0.15 m layer.

Figure 9 - Regression between saturated hydraulic conductivity (K_s , cm h^{-1}) and pore organization (k_1), connectivity (k_2) indexes for pores $>100 \mu\text{m}$ (MAC), $>30 \mu\text{m}$ (MaP) at depths of 0.00-0.05, 0.05-0.15, and 0.15-0.30 m.



SOURCE: Author (2026).

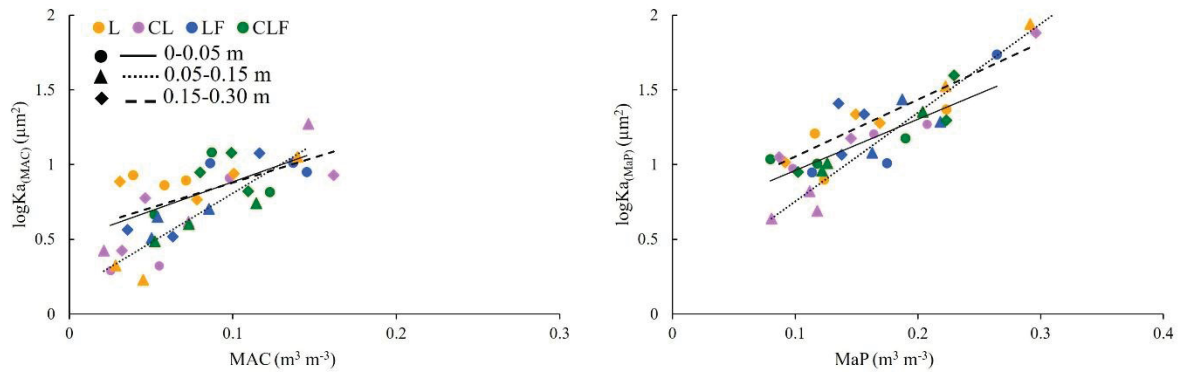
Table 9 - Regression analysis equation between saturated hydraulic conductivity, (K_s , cm h^{-1}) and pore organization (k_1) and connectivity (k_2) indexes for pores $>100 \mu\text{m}$ (MAC) and $>30 \mu\text{m}$ (MaP) at depths of 0.00-0.05, 0.05-0.15, and 0.15-0.30 m.

Soil attributes	Equation	R ²	Probability
logK _s vs. $k_{1(\text{MAC})}$	$\log K_{s(1)} = 0.1 + 0.4k_{1(\text{MAC})}$	0.84	<0.001
logK _s vs. $k_{1(\text{MAC})}$	$\log K_{s(2)} = -0.2 + 0.5k_{1(\text{MAC})}$	0.81	<0.001
logK _s vs. $k_{1(\text{MAC})}$	$\log K_{s(3)} = 0.4 + 0.2k_{1(\text{MAC})}$	0.66	<0.001
logK _s vs. $k_{2(\text{MAC})}$	$\log K_{s(1)} = 0.5 + 0.2k_{2(\text{MAC})}$	0.45	<0.001
logK _s vs. $k_{2(\text{MAC})}$	$\log K_{s(2)} = -0.2 + 0.3k_{2(\text{MAC})}$	0.54	<0.001
logK _s vs. $k_{2(\text{MAC})}$	$\log K_{s(3)} = 0.4 + 0.15k_{2(\text{MAC})}$	0.44	<0.001
logK _s vs. $k_{1(\text{MaP})}$	$\log K_{s(1)} = 0.5 + 0.1k_{1(\text{MaP})}$	0.33	<0.001
logK _s vs. $k_{1(\text{MaP})}$	$\log K_{s(2)} = -0.1 + 0.4k_{1(\text{MaP})}$	0.40	<0.001
logK _s vs. $k_{1(\text{MaP})}$	$\log K_{s(3)} = 0.6 + 0.1k_{1(\text{MaP})}$	0.28	<0.001
logK _s vs. $k_{2(\text{MaP})}$	$\log K_{s(1)} = 0.6 + 0.09k_{2(\text{MaP})}$	0.26	<0.001
logK _s vs. $k_{2(\text{MaP})}$	$\log K_{s(2)} = -0.5 + 0.4k_{2(\text{MaP})}$	0.69	<0.001
logK _s vs. $k_{2(\text{MaP})}$	$\log K_{s(3)} = 0.6 + 0.06k_{2(\text{MaP})}$	0.22	<0.001

(1), (2), and (3) correspond to the depths of 0.00–0.05 m, 0.05–0.15 m, and 0.15–0.30 m, respectively

SOURCE: Author (2026).

Figure 10 - Regression analysis between intrinsic air permeability of soil $K_{a(MAC)}$ and $K_{a(MaP)}$ (μm^2) with large macroporosity (MAC) and macroporosity (MaP), at depths of 0.00-0.05; 0.05-0.15 and 0.15-0.30 m.



SOURCE: Author (2026).

Table 10 - Regression analysis equation between intrinsic air permeability of the soil $K_{a(MAC)}$ and $K_{a(MaP)}$ and large macroporosity (MAC) and macroporosity (MaP), at depths of 0.00-0.05; 0.05-0.15 and 0.15-0.30 m.

Soil attributes	Equation	R ²	Probability
$\log K_{a(MAC)}$ vs. MAC	$\log K_{a(MAC)(1)} = 0.5 + 3.9MAC$	0.34	<0.001
$\log K_{a(MAC)}$ vs. MAC	$\log K_{a(MAC)(2)} = 0.15 + 6.6MAC$	0.85	<0.001
$\log K_{a(MAC)}$ vs. MAC	$\log K_{a(MAC)(3)} = 0.54 + 3.3 MAC$	0.40	<0.001
$\log K_{a(MaP)}$ vs. MaP	$\log K_{a(MaP)(1)} = 0.6 + 3.4MaP$	0.70	<0.001
$\log K_{a(MaP)}$ vs. MaP	$\log K_{a(MaP)(2)} = 0.16 + 5.9MaP$	0.95	<0.001
$\log K_{a(MaP)}$ vs. MaP	$\log K_{a(MaP)(3)} = 0.6 + 3.8MaP$	0.79	<0.001

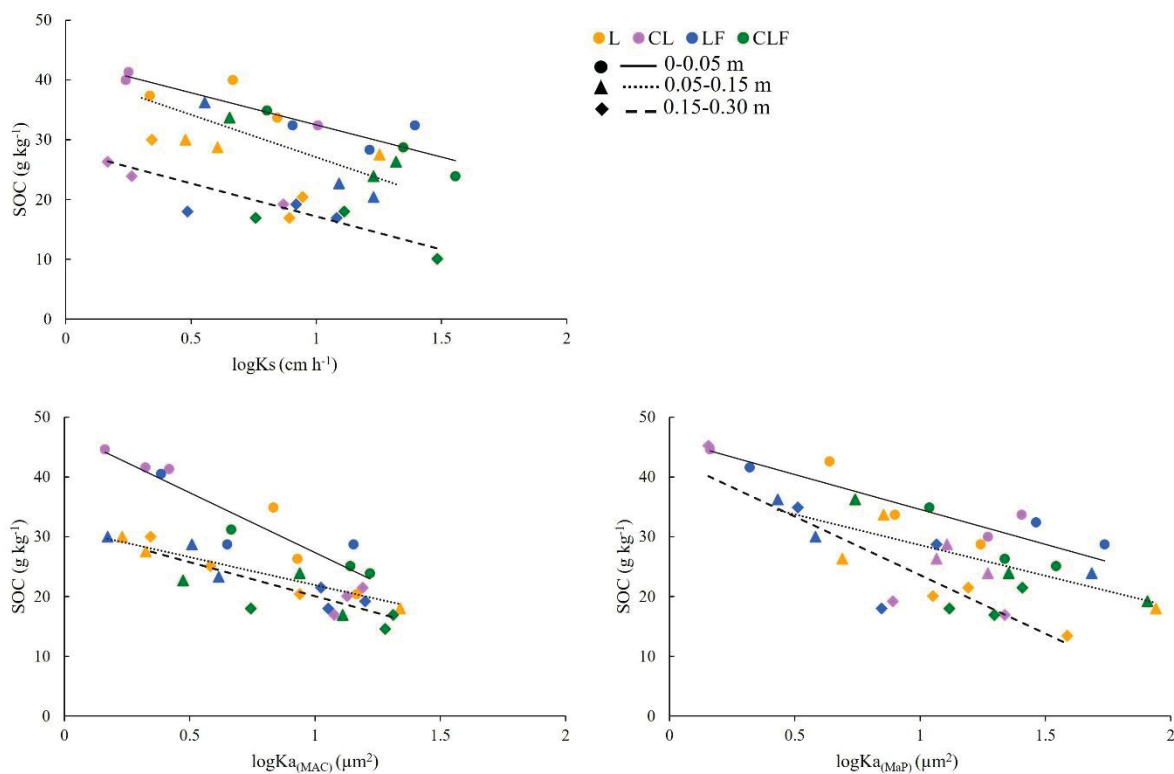
(1), (2), and (3) correspond to the depths of 0.00–0.05 m, 0.05–0.15 m, and 0.15–0.30 m, respectively

SOURCE: Author (2026).

The relationship between the soil's hydraulic properties and the soil organic carbon (SOC) content was negative and statistically significant at all analyzed depths (Figure 11). Complementing this analysis, Table 11 presents the corresponding linear regressions alongside R² and p-values. Evaluating the angular coefficients (b) and determination coefficients (R²) is crucial for understanding the influence of each property on SOC at different soil depths.

The linear regression between SOC and Ks showed coefficients of determination (R²) ranging from 0.67 to 0.82 (Table 11). The slope coefficients (b) were negative at all depths, indicating that increasing the Ks causes decreasing in SOC. The slope of the line was steeper in the 0.05-0.15 m layer (b = -1.4), suggesting a stronger influence of Ks on SOC in this intermediate layer.

Figure 11 - Regression analysis between saturated hydraulic conductivity, K_s ; intrinsic air permeability of soil, $K_{a(MAC)}$ and $K_{a(MaP)}$, in pores with diameters > 100 and $> 30 \mu\text{m}$, respectively, and the soil organic carbon, SOC (g kg^{-1}), at depths of 0.00-0.05; 0.05-0.15 and 0.15-0.30 m.



SOURCE: Author (2026).

Table 11 - Regression analysis equation between saturated hydraulic conductivity, K_s (cm h^{-1}); intrinsic air permeability of the soil $K_{a(\text{MAC})}$ and $K_{a(\text{MaP})}$ (μm^2) in pores with diameters > 100 and $> 30 \mu\text{m}$, respectively, and of the soil organic carbon, SOC (g kg^{-1}), at depths of 0.00-0.05; 0.05-0.15 and 0.15-0.30 m.

Soil attributes	Equation	R ²	RMSE	Probability
SOC vs. logKs	SOC ₍₁₎ = 43.2 - 10.7 logKs	0.82	2.05	<0.001
SOC vs. logKs	SOC ₍₂₎ = 41.2 - 14.2 logKs	0.68	3.19	<0.001
SOC vs. logKs	SOC ₍₃₎ = 28.2 - 11.1 logKs	0.67	2.63	<0.001
SOC vs. logKa _(MAC)	SOC ₍₁₎ = 59.6 - 31.8 logKa _(MAC)	0.80	5.85	<0.001
SOC vs. logKa _(MAC)	SOC ₍₂₎ = 31.2 - 9.3 logKa _(MAC)	0.64	2.48	<0.001
SOC vs. logKa _(MAC)	SOC ₍₃₎ = 31.4 - 11.4 logKa _(MAC)	0.65	2.30	<0.001
SOC vs. logKa _(MaP)	SOC ₍₁₎ = 58.7 - 20.8 logKa _(MaP)	0.81	4.67	<0.001
SOC vs. logKa _(MaP)	SOC ₍₂₎ = 38.8 - 10.3 logKa _(MaP)	0.76	2.80	<0.001
SOC vs. logKa _(MaP)	SOC ₍₃₎ = 43.1 - 19.5 logKa _(MaP)	0.73	4.50	<0.001

(1), (2) and (3) at depths of 0.00-0.05; 0.05-0.15 and 0.15-0.30 m.

SOURCE: Author (2026).

The negative relationship between SOC and $K_{a(\text{MAC})}$ was the strongest among those analyzed, with R^2 ranging from 0.61 to 0.84. The most notable negative impact occurred in the surface layer of 0.00-0.05 m, which had the steepest slope coefficient ($b = -2.0$). The influence of $K_{a(\text{MAC})}$ on SOC gradually decreased with depth. For $K_{a(\text{MaP})}$, the relationship with SOC was also significant, with less steep slope coefficients compared to $K_{a(\text{MAC})}$ in the surface layers, but with a value of $b = -2.0$ in the 0.15-0.30 m layer.

5.7 DISCUSSION

The evaluated management systems presented mean K_s values between 6.5 and 13.0 cm h^{-1} , falling within ranges considered high for clayey soils, according to the classification presented by Ottoni et al. (2025). Although this textural class is frequently associated with low permeability, Tobella et al. (2024) highlight that Ferralsols exhibit a distinct hydraulic behavior, remaining highly permeable due to the natural stability of their granular microstructure. Our results corroborate this pattern, indicating the preservation of functional macropores typical of well-managed oxidic soils. The maintenance of this hydraulic functionality is supported by Rauber et al. (2025), who, under similar conditions in Southern Brazil, demonstrated that the infiltration rate is governed by biomass input and the absence of soil disturbance. Therefore, the high values found here reflect the effectiveness of long-term conservation management in preserving pore continuity, contrasting with scenarios of severe surface compaction induced by intensive grazing described by Suzuki et al. (2022).

Simultaneously, high soil organic carbon (SOC) contents, exceeding 30 g kg^{-1} at the surface, characterize these systems as carbon sinks, driven by the continuous biomass input in

integrated systems (Oliveira et al., 2018). These results corroborate the values observed by Cavalieri-Polizeli et al. (2024) in the same experiment. According to Cotrufo (2022), such accumulation improves fertility and acts as an agent in structural stabilization via microaggregates, reinforcing the adopted management as efficient strategy for greenhouse gas mitigation and the maintenance of soil physical quality.

The saturated hydraulic conductivity (K_s) was governed by the volume of macropores, exhibiting a significant positive correlation with both large macroporosity (MAC) and macroporosity (MaP), while showing a negative relationship with microporosity (MIC) (Figure 8). This behavior confirms that water flux in these Ferralsols is functionally dependent on the presence of larger, drainage-capable pores, rather than total porosity per se. This aligns with findings by Tobella et al. (2024), who observed that in tropical soils, K_s is primarily driven by structural porosity rather than textural pores. The negative correlation with MIC reinforces that a predominance of small, water-retaining pores restricts hydraulic conductivity, a trade-off often observed when soil compaction transforms macropores into micropores, limiting fluid transport (Reichert et al., 2018).

The strong positive correlation found between saturated hydraulic conductivity (K_s) and the macropore organization index $k_{1(MAC)}$ indicates that the geometric arrangement and continuity of the larger pores, rather than simply the total pore volume, governed fluid transport in this soil. The high values of k_1 and k_2 associated with high K_s indicate that the macropore network in these conservation systems is not only abundant but also geometrically continuous and well-organized. Silva et al. (2021) emphasize that pore connectivity is a more sensitive indicator of soil functional quality than porosity total. The association found here suggests that the long-term absence of soil disturbance, combined with biological activity (roots and fauna), has preserved the continuity of the pore system, allowing for efficient water transport and the structural functionality described by Mentges et al. (2016).

The intrinsic air permeability (K_a) demonstrated a dependence on the volume of macropores, particularly for large macroporosity (MAC) (Figure 3). This relationship underscores that K_a is a highly sensitive proxy for detecting changes in the structural porosity responsible for soil aeration. Suzuki et al. (2022) noted that air permeability decreases exponentially with the loss of macropores due to compaction, reinforcing that the high K_a values observed in our study are directly linked to the preservation of these large, air-conducting pores. Furthermore, the correlation between K_a and MAC corroborates the findings of Silva et al. (2021), who established that intrinsic air permeability parameters are excellent predictors of

structural pore attributes, serving as a reliable metric for the structural integrity of the soil surface layers.

The most significant finding was the strong negative relationship between SOC and the intrinsic air permeability of large macropores $K_{a(MAC)}$, which explained up to 84% of the carbon variation at the soil surface. This inverse correlation suggests that a highly efficient and well-connected pore network facilitates oxygen transport to the soil matrix. According to Keiluweit et al. (2017) and Yang et al. (2018), increased aeration in soil microsites alleviates anaerobic constraints, thereby potentially aerobic microbial decomposition and organic matter mineralization.

Our results corroborate research that has shown a negative association between hydraulic properties and SOC, especially in temperate regions (Nemes et al., 2005; Jarvis et al., 2013; Peth et al., 2008; Schlüter et al., 2011). For example, Larsbo et al. (2016) demonstrated that soils with low SOC were characterized by a porosity dominated by large macropores. On the other hand, the literature also presents positive relationships between SOC and K_s (Lehmann et al., 2021; Hodnett et al., 2002). This dual behavior highlights the multifaceted role of organic matter as a substrate for decomposition and as an agent for soil structure formation. The direction of the relationship, whether positive or negative, appears to be highly dependent on the intrinsic properties of the soil, particularly its texture and clay mineralogy, as well as climatic and management conditions.

The divergence between these two results can also be explained by the soil's mineralogical composition. While in soils with 2:1 clay minerals (such as smectites) SOC is crucial for the formation of aggregates and porosity, in kaolinitic and oxidic soils, like the one in this study, the microaggregated structure is naturally stable due to the electrostatic interaction of oxides (Hodnett et al., 2002; Lado et al., 2004). In these soils, the increase in macroporosity favors air permeability and oxygen supply, which can alleviate anaerobic constraints in microsites and intensify carbon mineralization, according to mechanisms described by Keiluweit et al. (2017) and Yang et al. (2018).

Improving soil structure is one of the key objectives of conservation agriculture; practices must balance the need for adequate soil infiltration and aeration with the need to protect and sequester soil carbon.

The implementation of these systems promoted high biomass input and structural stability, increasing carbon sequestration (Cavaliere-Polizeli et al., 2024). However, the improvement in pore architecture increases intrinsic air permeability, intensifying oxygen diffusion into the soil matrix (Martínez et al., 2016). Consequently, aerobic microbiota

maximize organic matter mineralization, particularly during periods of lower soil moisture, when rapid drainage through macropores eliminates the protective anoxia of microsites (Keiluweit et al., 2017). Thus, the enhanced transport functionality accelerates decomposition cycles, limiting local carbon accumulation and explaining the observed negative correlation.

Our results, along with others (Tobella et al., 2024; Blanchy et al., 2023), demonstrate that management practices significantly influence soil hydraulic properties. The challenge lies in increasing organic matter to a level that improves physical quality without creating excessively permeable conditions that lead to a net loss of carbon in the long term. Beyond total content, different carbon fractions (e.g., particulate versus mineral-associated) play distinct roles in pore formation and fluid dynamics (Cotrufo et al., 2022; Cavalieri-Polizeli et al., 2024). This suggests that management should focus not only on increasing carbon inputs but also on managing the resulting hydraulic properties to regulate microscale oxygen availability and mitigate decomposition, a functional interdependence supported by recent studies on stabilization mechanisms (Keiluweit et al., 2017; Yang et al., 2018).

It is essential to acknowledge the limitations of this investigation. The conclusions are derived from a single experimental site with only one textural class (clayey), managed under conservationist principles. The direct applicability of these findings to soils with different textures, such as sandy or silty soils, or those under different climatic regimes or conventional management, requires further investigation (Tobella et al., 2024). The relationships observed here may differ significantly in soils where the structure is more fragile or where a different mineralogy dictates the physical properties (Scheffler et al., 2011; Ottoni et al., 2025). Therefore, caution should be exercised when extrapolating these specific results to other environments.

This study provides evidence that in structurally stable, subtropical clay soils, optimized soil permeability is negatively correlated with organic carbon contents, likely due to increased aeration-driven mineralization. Future research should aim to validate these findings across a wider range of soil types and climates. Directly measuring carbon mineralization rates alongside physical properties would provide definitive confirmation of the proposed mechanisms. Additionally, future work should employ advanced techniques such as X-ray computed tomography to characterize the pore network and validate the proposed mechanisms.

The main result of this study is the negative relationship between saturated hydraulic conductivity and intrinsic air permeability, and soil organic carbon (SOC) in a clayey Ferralsol under a subtropical climate. This evidence supports the hypothesis that a higher efficiency in

fluid transport, especially of gases, is associated with a reduction in carbon stocks due to the intensification of aerobic microbial activity (Keiluweit et al., 2017; Yan et al., 2018).

5.8 CONCLUSION

Fluid transport properties in a clayey Ferralsol under conservationist livestock systems were primarily governed by the architecture of the macropore network rather than total porosity. Saturated hydraulic conductivity (K_s) and intrinsic air permeability (K_a) exhibited strong positive correlations with large macroporosity (MAC) and pore connectivity indices (k_1 and k_2), particularly in surface layers, confirming that a continuous and organized pore system drives water and gas fluxes. However, this structural efficiency presented a functional trade-off. The strong negative relationship observed between soil organic carbon (SOC) and the intrinsic air permeability of large macropores $K_{a(MAC)}$, which explained up to 84% of carbon variation, supports the hypothesis that a highly connected pore network enhances oxygen supply, thereby intensifying organic matter mineralization. Therefore, conservation management maintains high physical functionality, creating a functional trade-off where the efficient pore network supports essential aeration, thereby favoring organic matter turnover alongside accumulation.

5.9 SUPPLEMENTARY MATERIAL

Table S - Soil texture in (g kg^{-1}) and particle density (ρ_p) in (Mg m^{-3}), for depths of 0.00-0.05; 0.05-0.15 and 0.15-0.30 m

Conservationist Systems	ρ_p (Mg m^{-3})	Sand (g kg^{-1})	Silt (g kg^{-1})	Clay (g kg^{-1})	Class
0 - 0.05 m					
Livestock (L)	2.51	395	100	504	Clayey
Crop-Livestock (CL)	2.54	320	166	512	
Livestock-Forestry (LF)	2.53	358	104	537	
Crop-Livestock-Forestry (CLF)	2.48	304	100	595	
0.05 - 0.15 m					
Livestock (L)	2.54	391	83	525	Clayey
Crop-Livestock (CL)	2.53	412	100	487	
Livestock-Forestry (LF)	2.54	362	83	554	
Crop-Livestock-Forestry (CLF)	2.57	408	83	508	
0.15 - 0.30 m					
Livestock (L)	2.36	395	100	504	Clayey
Crop-Livestock (CL)	2.50	321	133	546	
Livestock-Forestry (LF)	2.56	462	108	429	
Crop-Livestock-Forestry (CLF)	2.56	408	83	508	

SOURCE: Author (2026).

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6 CHAPTER 4 - LONG-TERM COVER CROPS IMPROVE SOC AND PHYSICAL PROPERTIES IN SANDY SOILS

6.1 ABSTRACT

Carbon farming strategies have been promoted worldwide to reconcile climate mitigation with the maintenance of soil functionality in agricultural landscapes. This study aimed to compare long-term experiments (>30 years) with and without cover crops to investigate how these strategies affect soil structure, emphasizing soil physical properties and soil organic carbon (SOC). The experiments were conducted on coarse-textured soils (sandy loam and loamy sand) at Mellby and Fotegården, southwestern Sweden. The experimental design compared two main management systems: With Cover Crops (*Lolium perenne*) and Without Cover Crops (bare soil during winter). Samples were collected down to 0.50 m (layers 0.00-0.20 m and 0.30-0.50 m). Soil attributes analyzed included hydraulic conductivity (K), soil bulk density (BD), penetration resistance (PR), macroporosity (MaP), microporosity (MIC), plant-available water capacity (PAWC), and soil organic carbon (SOC). The multivariate analysis (MANOVA) revealed significant differences between management systems ($p < 0.05$), highlighting a systemic effect of the biological management. The Principal Component Analysis (PCA) revealed the formation of two distinct patterns: the first, consisting of the With Cover Crops system, demonstrated a positive association among SOC, MIC, PAWC, and PR. Specifically, this system maintained hydraulic conductivity within a balanced range (31-38 cm h⁻¹), favoring water retention. In contrast, the Without Cover Crops system was driven by macroporosity and exhibited excessive hydraulic conductivity in subsurface layers (> 41 cm h⁻¹ at Fotegården and > 66 cm h⁻¹ at Mellby). These findings highlight that long-term adoption of cover crops modifies the pore network functionality, preventing the formation of excessive flow pathways observed in bare soils, while maintaining carbon stocks contributes to the long-term sustainability of coarse-textured soils.

Keywords: Soil Organic Carbon; Soil Physical Quality; Hydraulic Conductivity.

6.2 INTRODUCTION

The current paradigm of European and global agriculture is rapidly shifting towards "Carbon Farming" strategies, where soil management aims not only at food production but also at climate mitigation through carbon sequestration and the preservation of functional biodiversity (Svoboda et al., 2026). In intensive cropping systems, the simplification of crop rotations and frequent soil tillage have reduced soil organic carbon (SOC) stocks, compromising the physical and chemical resilience of agroecosystems against extreme climatic events (Petersson et al., 2025). The adoption of conservation practices, such as crop diversification, emerges as an essential tool to promote soil recarbonization and the maintenance of ecosystem services (Liu et al., 2022). However, the effectiveness of these practices varies significantly according to local edaphoclimatic conditions, requiring robust regional validations (Shah et al., 2021).

Agricultural intensification on coarse-textured soils, such as the sandy loams and loamy sands of southern Scandinavia, presents unique challenges due to the inherent structural fragility and low water and nutrient retention capacity of these environments (Aronsson et al., 2016). In these regions, the humid temperate climate, characterized by frequent precipitation outside the growing season and freezing-thawing cycles, exacerbates the risk of physical degradation and environmental losses when the soil remains unprotected (Svensson et al., 2026). The predominance of macropores and the low cohesion between sand particles make these soils highly permeable yet susceptible to rapid leaching of nitrates and phosphorus into groundwater and surface waters (Lindén et al., 1999; Norberg & Aronsson, 2024). Therefore, soil cover management becomes a critical factor not only for environmental sustainability but also for maintaining long-term agrarian functionality (Torstensson et al., 2006).

The traditional practice of keeping the soil under bare fallow during autumn and winter disrupts the continuity of biological cycles, exposing the surface to erosive processes and the natural reconsolidation of the soil matrix, especially in textures with low inter-particle cohesion (Wanic et al., 2019). The absence of an active root system to anchor particles and the cessation of exudate inputs lead to the degradation of functional porosity, frequently resulting in surface compaction and reduced natural permeability (Pieper et al., 2015). Recent studies indicate that warmer and wetter winters accelerate organic matter mineralization and drainage flow, rendering conventional bare soil management unsustainable, as the loss of physical integrity reduces the productive capacity of subsequent crops (Hanrahan et al., 2021; Vogeler et al., 2023).

As mitigation strategy, the use of cover crops, notably perennial ryegrass (*Lolium perenne* L.), has been widely adopted and studied in long-term experiments in Sweden to reduce nitrogen leaching (Svensson et al., 2026). Beyond the environmental function, in sandy soils where the formation of stable aggregates is limited by clay scarcity, soil organic carbon (SOC) assumes the role of the main stabilizer of the porous matrix (Poeplau et al., 2015). The introduction of these plants allows the soil to remain biologically active during critical months, acting simultaneously as nutrient traps and carbon pumps, a mechanism considered one of the most efficient ways to recover carbon stocks in degraded arable lands (Petersson et al., 2025).

However, although the chemical and environmental benefits (leaching reduction and C stocks) are well documented in these long-term experiments, there is a gap in understanding how these alterations impact soil physical properties, such as hydraulic conductivity and penetration resistance (Svensson et al., 2026). It is crucial to determine whether the observed carbon increment is sufficient to translate into measurable physical improvements in coarse-textured soils (Liu et al., 2022).

Given this context, the hypothesis of this study is that the long-term adoption of cover crops (*Lolium perenne*) promotes the increase of soil organic carbon, resulting in improved soil physical properties compared to bare soil. The objective was to compare long-term experiments (>30 years) with and without cover crops to investigate how these strategies affect soil structure, specifically soil physical properties and soil organic carbon.

6.3 MATERIAL AND METHODS

6.3.1 Experimental site and area

The study was conducted at two long-term experimental sites managed by the Swedish University of Agricultural Sciences (SLU) in southwestern Sweden. The first site, Mellby (R0-8403), is located near Laholm, Halland County (56° 29' N, 13° 00' E, ~16 m a.s.l.), while the second site, Fotegården (R0-8410), is located near Lidköping, Västra Götaland County (58° 30' N, 13° 09' E, ~45 m a.s.l.). According to Köppen's classification, the local climate belongs to the Cfb type (temperate oceanic), characterized by humid conditions without a dry season. This study was part of the Swedish long-term soil fertility experiments, implemented in 1983 at Mellby and 1993 at Fotegården, covering distinct pedoclimatic conditions (Aronsson et al., 2016). At Mellby, the soil texture is classified as sandy loam (61% sand, 25% silt, and 9% clay) (Torstensson et al., 2006; Svensson et al., 2026). At Fotegården, the soil presents a coarser texture, classified as loamy sand (78% sand, 10% silt, and 6% clay) (Aronsson et al., 2003).

6.3.2 History, experiment management, and treatments

Two long-term field experiments located in southwestern Sweden were used in this study: Mellby (R0-8403) and Fotegården (R0-8410). Both sites are part of the Swedish long-term soil fertility experiments and have been managed to evaluate the effects of agricultural practices, such as cover crops and tillage timing, on nutrient leaching and soil quality (Aronsson et al., 2016).

The Mellby (R0-8403) experiment is located near Laholm, Halland (56° 29' N, 13° 00' E, ~16 m a.s.l.) and was established in 1983, receiving its current design in 1989 (Stenberg et al., 1999; Svensson et al., 2026). The soil is classified as a sandy loam (61% sand, 25% silt, 9% clay) with approximately 5% organic matter in the topsoil (Torstensson et al., 2006; Poeplau et al., 2015). The site is characterized by sand deposits (90-130 cm depth) underlain by a nearly impermeable glacio-fluvial clay layer, facilitating the measurement of nutrient leaching (Svensson et al., 2026).

The experimental design consists of separately tile-drained plots of 40 x 40 m (1600 m²), allowing for the direct measurement of discharge and nitrogen (N) leaching from individual treatments (Stenberg et al., 1999; Svensson et al., 2026). The crop rotation is dominated by spring cereals, mainly oats (*Avena sativa* L.), barley (*Hordeum vulgare* L.), and spring wheat (*Triticum aestivum* L.), with occasional cultivation of potatoes (*Solanum tuberosum* L.).

The treatments evaluated in this study included systems with and without under-sown cover crops (primarily perennial ryegrass, *Lolium perenne* L.) combined with different nitrogen fertilization rates and tillage strategies. In plots with cover crops, the ryegrass was typically under-sown in spring together with the main crop and incorporated into the soil during spring ploughing in the following year, whereas plots without cover crops were stubble cultivated in early autumn (Poeplau et al., 2015; Svensson et al., 2026).

The Fotegården (R0-8410) experiment, established in 1993, is located near Lidköping, Västra Götaland (58° 30' N, 13° 09' E, ~45 m a.s.l.) (Lindén et al., 1999). The soil texture is a loamy sand (78% sand, 10% silt, 6% clay) with an organic matter content of 5% in the topsoil (Aronsson et al., 2016). This site represents a coarser-textured soil compared to Mellby, making it highly susceptible to nutrient leaching.

The experimental plots are 30 x 28 m (840 m²) (Lindén et al., 1999). The management history at Fotegården has focused on mitigating nitrogen leaching through the use of catch crops and optimized tillage timing.

6.3.3 Soil sampling

The experimental design consists of three randomized blocks. The treatments are: with ryegrass cover crop and without cover crop. Undisturbed soil samples were collected in 2023 at depths of 0.00-0.20 m and 0.30-0.50 m. In each block, five undisturbed soil core samples were collected, totaling 120 samples. The soil attributes analyzed included penetration resistance, hydraulic conductivity, soil bulk density, total porosity, microporosity, macroporosity, plant-available water and soil organic carbon.

Initially, the undisturbed soil samples were saturated by capillarity, with the water level maintained at three-quarters of the ring height for 48 hours. Subsequently, the samples were equilibrated at a matric potential of -10 kPa using a tension table. At this potential, measurements of hydraulic conductivity (K) and soil penetration resistance (PR) were performed using two independent sets of samples.

The hydraulic conductivity was measured using a mini tension disc infiltrometer (Mini TDI) with a 4.5 cm diameter porous steel disc. The samples were initially subjected to a tension table with a suction of -10 kPa (i.e., to reach an appropriate tension point for analysis). This suction adjustment is necessary to ensure that the samples are in proper conditions before conducting hydraulic conductivity measurements. The sample was prepared with a soil core, polyamide (PA) mesh, fine quartz sand, and a stencil to level the sand. The Mini TDI was set to a tension of -5 cm, applying negative pressure to the soil, and the measurement was carried out until the infiltration rate reached a steady-state after 10-30 minutes.

The penetration resistance was measured at the following matric potential -10 kPa, using a micro-penetration resistance method. The insertion velocity was 4 mm min⁻¹, reaching a depth of 25 mm. The needle used for measurement had a 1 mm cone diameter, a 0.8 mm shaft diameter, and a 30° cone angle. Each insertion took approximately 8 minutes, with a total of 32 hours of measurements.

The total porosity (TP) was calculated based on the particle density and the bulk density of the soil. Microporosity (MIC) was obtained from the water content retained at a potential of -60 hPa, following Brewer's (1964) classification of pores. Macroporosity (MaP) was determined as the difference between TP and microporosity (Flint & Flint, 2002b). Plant-available water capacity (PAWC, m³ m⁻³), a measure of water storage, was calculated as the difference between the volumetric water contents (m³ m⁻³) at field capacity (matric potential -10 kPa) and the permanent wilting point (-1500 kPa) (Reynolds et al., 2009).

Soil bulk density (BD) was determined using the core method, where the samples, were dried in a forced-air circulation oven (105 °C for 48 hours) to obtain the dry mass of the soil,

and the relationship between dry mass and core volume was established (Grossman & Reinsch, 2002).

6.3.4 Statistical analyses

The dataset was analyzed separately for each soil layer (0.00-0.20 m and 0.30-0.50 m). Initially, the data were subjected to univariate Analysis of Variance (ANOVA). However, given the high interdependence among soil physical properties and the limited sensitivity of univariate tests to detect systemic changes in this study, a multivariate approach was prioritized to identify integrated patterns of variation.

To compare the cropping systems (With Cover Crops vs. Without Cover Crops), a Multivariate Analysis of Variance (MANOVA) was performed using Pillai's trace statistic ($p < 0.05$), which is robust to deviations from multivariate normality. The MANOVA was chosen to simultaneously evaluate the set of correlated dependent variables: soil penetration resistance (PR), hydraulic conductivity (K), soil bulk density (BD), microporosity (MIC), macroporosity (MaP), plant-available water capacity (PAWC), and soil organic carbon (SOC).

To further elucidate the structure of variance and the relationships between variables and treatments, Principal Component Analysis (PCA) was conducted based on the correlation matrix to address multicollinearity. In the PCA biplots, ellipses representing the 95% confidence interval were constructed using the Mahalanobis distance to visualize the separation between groups. All statistical procedures were performed using the R software (version 4.3.1; R Core Team, 2023), employing the packages *dplyr* for data manipulation, *ExpDes* for ANOVA, *MultivariateAnalysis* for MANOVA, and *FactoMineR*, *factoextra*, and *corrplot* for PCA and visualization.

6.4 RESULTS

At Fotegården, the univariate analysis (Table 12) indicated that most soil physical attributes (BD, TP, MaP, MIC, PAWC) and organic carbon (SOC) did not differ significantly between management systems in either soil layer. However, significant differences ($p < 0.05$) were observed for penetration resistance (PR) and hydraulic conductivity (K). In the surface layer (0.00-0.20 m), the presence of cover crops resulted in significantly higher PR and lower K compared to the treatment without cover crops. In contrast, at the 0.30-0.50 m depth, the cover crop system exhibited significantly lower PR (1.60 MPa) compared to the without cover crops (2.46 MPa), while hydraulic conductivity remained lower in the with cover crops.

Table 12 - Mean values \pm standard deviation ($n = 15, \pm$ SD) of soil attributes under management systems with and without cover crops at Fotegården (0.00-0.20 m and 0.30-0.50 m depths).

Soil attributes	Systems											
	With Cover Crops			Without Cover Crops			With Cover Crops			Without Cover Crops		
	0 - 0.20 m						0.30 - 0.50 m					
BD	1.36	(± 0.11)	ns	1.31	(± 0.12)	ns	1.49	(± 0.11)	ns	1.54	(± 0.12)	ns
TP	0.48	(± 0.02)	ns	0.55	(± 0.17)	ns	0.42	(± 0.02)	ns	0.41	(± 0.17)	ns
MaP	0.25	(± 0.03)	ns	0.32	(± 0.17)	ns	0.24	(± 0.03)	ns	0.21	(± 0.17)	ns
MIC	0.23	(± 0.04)	ns	0.23	(± 0.03)	ns	0.18	(± 0.04)	ns	0.20	(± 0.03)	ns
PAWC	0.18	(± 0.04)	ns	0.18	(± 0.03)	ns	0.15	(± 0.04)	ns	0.17	(± 0.03)	ns
PR	2.17	(± 0.63)	a	1.11	(± 0.20)	b	1.60	(± 0.63)	b	2.46	(± 0.20)	a
K	23.28	(± 5.35)	b	29.50	(± 8.66)	a	31.52	(± 5.35)	b	41.62	(± 8.66)	a
SOC	31.04	(± 2.43)	ns	30.33	(± 2.89)	ns	25.87	(± 2.43)	ns	22.38	(± 2.89)	ns

SOURCE: Author (2026).

Multivariate analysis of variance (MANOVA) revealed significant differences between the management systems, highlighting the influence of cover crop adoption (Table 13). Pillai's trace test indicated significant differences between With Cover Crops and Without Cover Crops systems in both 0.00-0.20 m and 0.30-0.50 m soil layers.

Table 13 - Summary of Multivariate Analysis of Variance (MANOVA) using the Pillai test for soil management systems with and without cover crops at Fotegården (0.00-0.20 m and 0.30 - 0.50 m depths).

Systems	With Cover Crops 0.00 - 0.20 m	With Cover Crops 0.30 - 0.50 m
Without cover crops 0.00 - 0.20 m	$6.47 \times 10^{-5} ***$	
Without cover crops 0.30 - 0.50 m		$9.56 \times 10^{-8} ***$

*** Significant at the 1% probability level according to Pillai's test; ns - not significant

SOURCE: Author (2026).

In the 0.00-0.20 m layer, principal component analysis (PCA) explained 76.3% of the variance, with eigenvalues of 1.20 (PC1) and 1.82 (PC2) (Table 14). For PC1, the most significant contributions came from BD, MaP, PR, K, and SOC. MIC and PAWC predominantly influenced PC2.

In the 0.30-0.50 m layer, PCA explained 89.3% of the variance, with eigenvalues of 1.29 (PC1) and 1.84 (PC2) (Table 14). For PC1, the most significant contributions came from BD, MIC, PR, K, and SOC. MaP and PAWC predominantly influenced PC2.

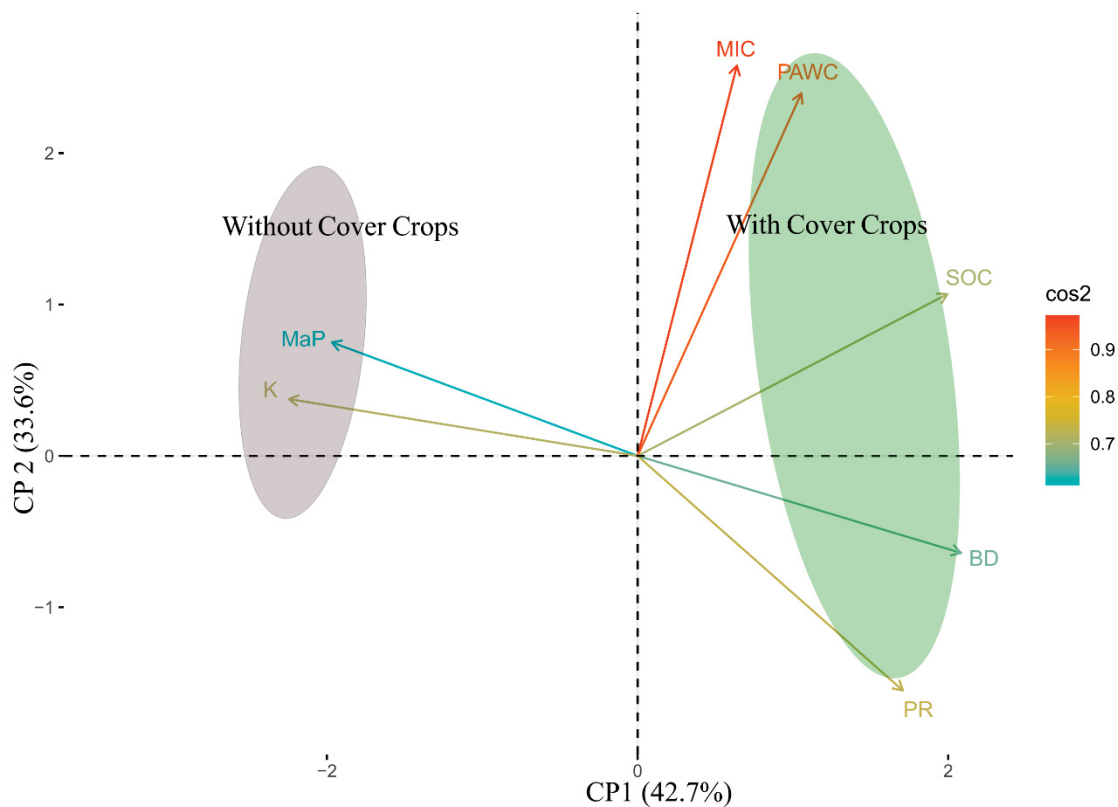
Table 14 - Summary of Eigenvalues, eigenvectors, and cumulative variance from Principal Component Analysis (PCA) of soil management systems with and without cover crops at Fotegården (0.00-0.20 m and 0.30-0.50 m depths).

Soil attributes	CP 1		CP 2	
	Eigenvectors	Contribution (%)	Eigenvectors	Contribution (%)
0.00 - 0.20 m				
BD (Mg m ⁻³)	0.77	20.02	-0.23	2.41
MaP (m ³ m ⁻³)	-0.73	17.88	0.27	3.30
MIC (m ³ m ⁻³)	0.23	1.88	0.95	38.99
PAWC (m ³ m ⁻³)	0.39	5.12	0.88	33.66
PR (MPa)	0.63	13.47	-0.57	14.09
K (cm h ⁻¹)	-0.83	23.24	0.13	0.82
SOC (g kg ⁻¹)	0.74	18.36	0.39	6.70
Eigenvalues	1.20		1.82	
Cumulative variance (%)	42.7		33.6	
0.30 - 0.50 m				
BD (Mg m ⁻³)	0.93	20.78	-0.00	0.00
MaP (m ³ m ⁻³)	0.10	0.24	0.99	47.81
MIC (m ³ m ⁻³)	-0.95	21.92	-0.14	1.02
PAWC (m ³ m ⁻³)	-0.03	0.03	0.98	46.86
PR (MPa)	0.96	22.16	-0.23	2.79
K (cm h ⁻¹)	0.98	22.99	0.12	0.73
SOC (g kg ⁻¹)	-0.70	11.85	0.12	0.77
Eigenvalues	1.29		1.84	
Cumulative variance (%)	59.9		29.4	

SOURCE: Author (2026).

At Fotegården (0.00-0.20 m depth), the separation between groups is clearly visualized by the confidence ellipses in the PCA biplots, indicating data segmentation based on similarity (Figure 12). The Without Cover Crops treatment showed a positive association with K and MaP. Conversely, the With Cover Crops system was characterized by the relationship among MIC, PAWC, SOC, BD, and PR.

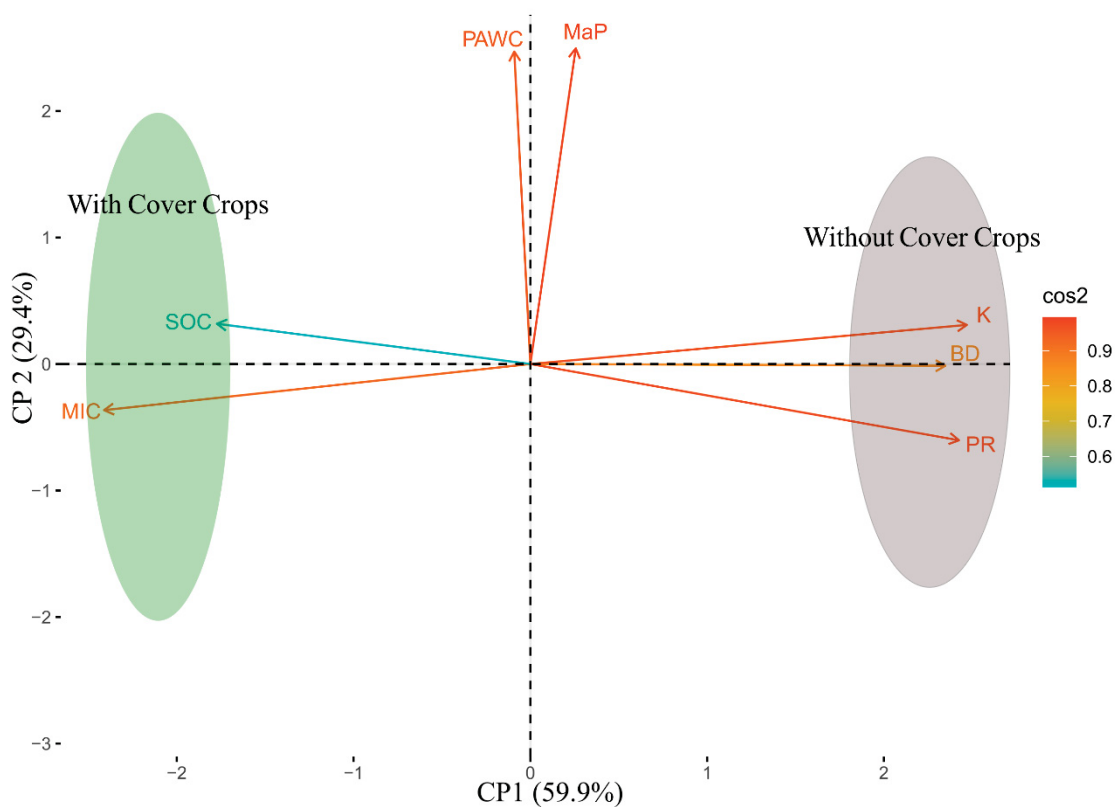
Figure 12 - Principal Component Analysis (PCA) of soil attributes for soil management systems with and without cover crops at Fotegården (0.00-0.20 m depth). Ellipses represent the 95% Confidence interval. Soil organic carbon, SOC; hydraulic conductivity, K; penetration resistance, PR; plant-available water, PAWC; bulk density, BD; macroporosity, MaP; microporosity, MIC.



SOURCE: Author (2026).

At Fotegården (0.30-0.50 m depth), the separation between groups is clearly visualized by the confidence ellipses in the PCA biplots, evidencing data segmentation based on management (Figure 13). The With Cover Crops treatment showed a positive association with SOC and MIC. Conversely, the Without Cover Crops system was characterized by the interrelationship among K, BD, and PR.

Figure 13 - Principal Component Analysis (PCA) of soil attributes for soil management systems with and without cover crops at Fotegården (0.30-0.50 m depth). Ellipses represent the 95% Confidence interval. Soil organic carbon, SOC; hydraulic conductivity, K; penetration resistance, PR; plant-available water, PAWC; bulk density, BD; macroporosity, MaP; microporosity, MIC.



SOURCE: Author (2026).

At the Mellby site, univariate analysis (Table 15) demonstrated that most soil physical properties (BD, TP, MaP, MIC, PAWC) and soil organic carbon (COS) remained statistically similar across management systems. However, significant changes were observed in PR and K. Regardless of the depth evaluated (0.00-0.20 m or 0.30-0.50 m), the system With Cover Crops consistently presented significantly higher PR values and lower hydraulic conductivity (K) compared to the treatment Without Cover Crops.

Table 15 - Mean values \pm standard deviation ($n = 15, \pm$ SD) of soil attributes under management systems with and without cover crops at Mellby (0.00-0.20 m and 0.30-0.50 m depths).

Soil attributes	Systems											
	With Cover Crops			Without Cover Crops			With Cover Crops			Without Cover Crops		
	0 - 0.20 m						0.30 - 0.50 m					
BD	1.51	(± 0.11)	ns	1.50	(± 0.08)	ns	1.70	(± 0.09)	ns	1.68	(± 0.04)	ns
TP	0.44	(± 0.04)	ns	0.45	(± 0.03)	ns	0.35	(± 0.05)	ns	0.34	(± 0.02)	ns
MaP	0.20	(± 0.04)	ns	0.21	(± 0.04)	ns	0.21	(± 0.06)	ns	0.21	(± 0.06)	ns
MIC	0.24	(± 0.04)	ns	0.24	(± 0.02)	ns	0.14	(± 0.06)	ns	0.12	(± 0.06)	ns
PAWC	0.18	(± 0.04)	ns	0.18	(± 0.02)	ns	0.12	(± 0.05)	ns	0.10	(± 0.06)	ns
PR	1.73	(± 0.57)	a	1.29	(± 0.34)	b	3.11	(± 1.23)	a	2.01	(± 0.7)	b
K	8.32	(± 1.63)	b	12.71	(± 9.76)	a	38.48	(± 23.2)	b	66.85	(± 34.14)	a
SOC	29.3	(± 3.04)	ns	27.64	(± 3.04)	ns	12.31	(± 7.08)	ns	15.35	(± 10.89)	ns

¹ Mean values of the two systems. Number of samples per system, n ; standard deviation, SD; bulk density, BD; soil total porosity, TP; macroporosity, MaP; microporosity, MIC; plant-available water, PAWC; penetration resistance, PR; hydraulic conductivity, K; soil organic carbon, SOC.

SOURCE: Author (2026).

Consistent with findings from the previous site, the multivariate analysis at Mellby also demonstrated a distinct separation between management strategies. Pillai's trace test (Table 16) confirmed that the presence of cover crops significantly altered soil attributes in the 0.00-0.20 m and 0.30-0.50 m layers.

Table 16 - Summary of Multivariate Analysis of Variance (MANOVA) using the Pillai test for soil management systems with and without cover crops at Mellby (0.00-0.20 m and 0.30-0.50 m depths).

Systems	With Cover Crops 0.00 - 0.20 m	With Cover Crops 0.30 - 0.50 m
Without cover crops 0.00 - 0.20 m	6.44×10^{-6} ***	
Without cover crops 0.30 - 0.50 m		4.07×10^{-7} ***

*** Significant at the 1% probability level according to Pillai's test; ns - not significant

SOURCE: Author (2026).

For Mellby in the 0.00-0.20 m layer, principal component analysis (PCA) explained 82.4% of the variance, with eigenvalues of 1.48 (PC1) and 0.61 (PC2) (Table 17). For PC1, the most significant contributions came from MaP, MIC, PAWC, PR and SOC. BD predominantly influenced PC2.

In the 0.30-0.50 m layer, PCA explained 76.7% of the variance, with eigenvalues of 0.20 (PC1) and 0.87 (PC4) (Table 6). For PC1, the most significant contributions came from BD, MaP, MIC, PAWC, K, and SOC. K predominantly influenced PC2.

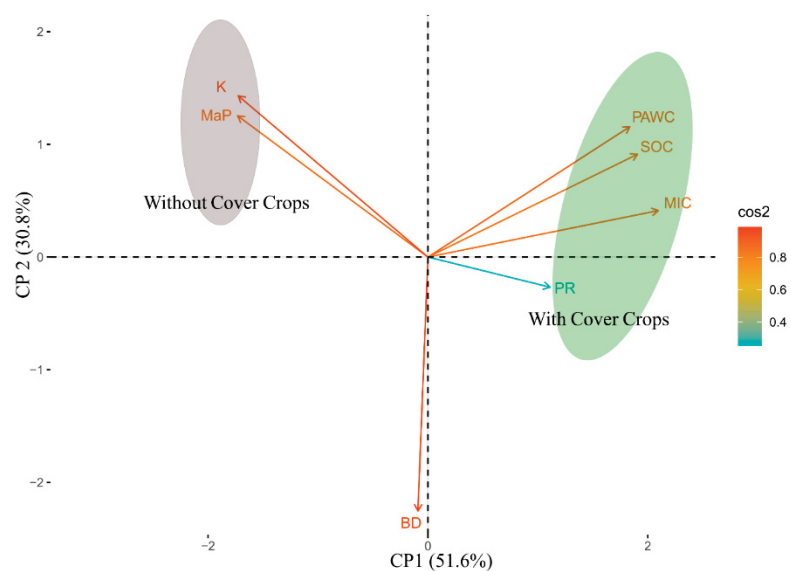
Table 17 - Summary of Eigenvalues, eigenvectors, and cumulative variance from Principal Component Analysis (PCA) of soil management systems with and without cover crops at Mellby (0.00-0.20 m and 0.30-0.50 m depths).

Soil attributes	CP 1		CP 2	
	Eigenvectors	Contribution (%)	Eigenvectors	Contribution (%)
0 - 0.20 m				
BD (Mg m ⁻³)	-0.04	0.04	-0.10	45.69
MaP (m ³ m ⁻³)	-0.76	16.08	0.30	14.13
MIC (m ³ m ⁻³)	0.92	23.60	-0.30	1.53
PAWC (m ³ m ⁻³)	0.80	18.12	-0.25	12.05
PR (MPa)	-0.76	15.99	-0.12	18.44
K (cm h ⁻¹)	0.48	6.62	0.85	0.64
SOC (g kg ⁻¹)	0.84	19.52	0.23	7.49
Eigenvalues	1.48		0.61	
Cumulative variance (%)	51.6		30.8	
0.30 - 0.50 m				
BD (Mg m ⁻³)	0.81	19.18	0.55	16.10
MaP (m ³ m ⁻³)	0.73	15.62	-0.18	1.71
MIC (m ³ m ⁻³)	-0.91	23.92	0.38	7.76
PAWC (m ³ m ⁻³)	-0.84	20.49	0.50	13.65
PR (MPa)	0.06	0.14	-0.86	39.35
K (cm h ⁻¹)	0.74	16.10	0.62	20.27
SOC (g kg ⁻¹)	-0.39	4.50	-0.14	1.11
Eigenvalues	0.20		0.87	
Cumulative variance (%)	49.6		27.1	

SOURCE: Author (2026).

At Mellby (0.00-0.20 m depth), the distinct clustering of confidence ellipses in the PCA biplots highlights the stratification of data based on management practices (Figure 14). The Without Cover Crops treatment was positively associated with hydraulic conductivity (K) and macroporosity (MaP). In contrast, the With Cover Crops system was defined by the association between microporosity (MIC), plant-available water capacity (PAWC), soil organic carbon (SOC), and penetration resistance (PR).

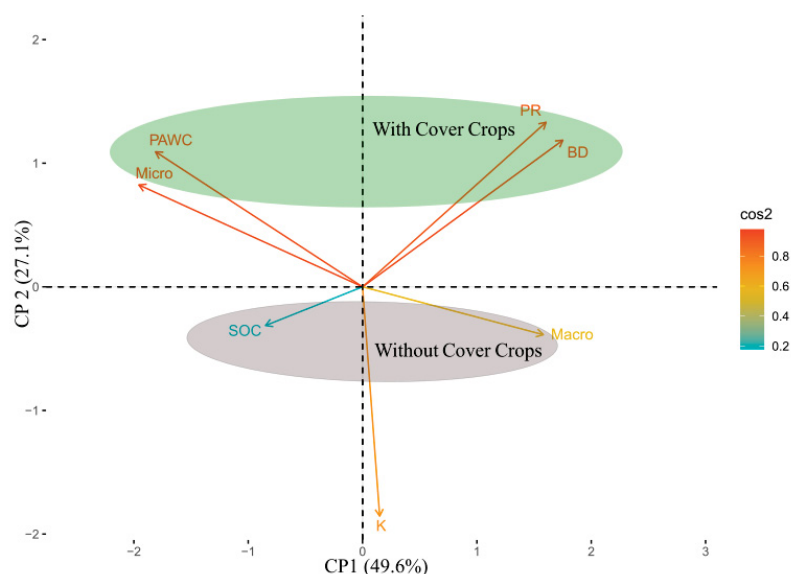
Figure 14 - Principal Component Analysis (PCA) of soil attributes for soil management systems with and without cover crops at Mellby (0.00-0.20 m depth). Ellipses represent the 95% Confidence interval. Soil organic carbon, SOC; hydraulic conductivity, K; penetration resistance, PR; plant-available water, PAWC; bulk density, BD; macroporosity, MaP; microporosity, MIC.



SOURCE: Author (2026).

In the subsurface layer (0.30-0.50 m), the PCA biplot reveals a clear divergence between treatments (Figure 15).

Figure 15 - Principal Component Analysis (PCA) of soil attributes for soil management systems with and without cover crops at Mellby (0.30-0.50 m depth). Ellipses represent the 95% Confidence interval. Soil organic carbon, SOC; hydraulic conductivity, K; penetration resistance, PR; plant-available water, PAWC; bulk density, BD; macroporosity, Macro; microporosity, MIC.



SOURCE: Author (2026).

The Without Cover Crops management was correlated with soil organic carbon (SOC) and macroporosity (Macro). Conversely, the With Cover Crops system was characterized by the interrelationship among hydraulic conductivity (K), bulk density (BD), penetration resistance (PR), and microporosity (MIC).

6.5 DISCUSSION

The multivariate approach (MANOVA) revealed a clear distinction between management systems, highlighting that the effect of cover crops in sandy soils is systemic rather than isolated to a single variable. At Fotegården, in the surface layer (0.00-0.20 m), the PCA results indicate that the With Cover Crops system favored the accumulation of soil organic carbon (SOC) and was associated with higher penetration resistance (PR). This pattern reflects the influence of *Lolium perenne*, which contributes to organic C inputs (Poeplau et al., 2015) while the absence of tillage during the cover crop cycle maintains a naturally firmer matrix compared to the frequent disturbance in the bare soil. Conversely, the association of the Without Cover Crops system with macroporosity (MaP) and hydraulic conductivity (K) indicates an unstable structure, likely facilitating rapid drainage but reducing the soil's capacity to filter nutrients, a typical behavior of degraded sandy soils under conventional tillage.

In the subsurface layer of Fotegården (0.30-0.50 m), the multivariate dynamics shifted, revealing the structural vulnerability of the bare soil. The Without Cover Crops system was associated with hydraulic conductivity (K) and penetration resistance (PR). This coexistence of high mechanical resistance (2.46 MPa) and rapid drainage (41.62 cm^{-1}) indicates a rigid, macropore-dominated matrix where water flows vertically with little interaction with the soil mass, typical of consolidated sandy soils (Lindén et al., 1999). Conversely, the With Cover Crops system was characterized by significantly lower PR (1.60 MPa) and moderated hydraulic conductivity.

Synthesizing the results for Fotegården, the multivariate segregation demonstrates that the With Cover Crops system favored the accumulation of soil organic carbon (SOC) and the maintenance of microporosity (MIC). This positive association suggests that the cover crop treatment significantly contributed to the soil's water storage capacity. As stated by Meurer et al. (2020), the interaction between organic carbon and the soil matrix is essential for creating and stabilizing capillary pores that retain water, preventing excessive drainage in coarse-textured soils.

At Mellby (0.00-0.20 m), the patterns mirrored those of the surface layer at Fotegården. The With Cover Crops system was strongly characterized by the relationship between microporosity (MIC), plant-available water capacity (PAWC), and SOC. This clustering indicates that the cover crop's primary physical role in this sandy loam soil is to enhance water retention and organic matter accumulation, counteracting the natural tendency of these soils to dry out rapidly. In contrast, the Without Cover Crops treatment was driven by K and MaP, suggesting that the absence of roots leave the soil pore system more open to rapid, potentially leaching-prone, vertical flow.

In the deeper layer of Mellby (0.30-0.50 m), the Without Cover Crops system was characterized by the association between soil organic carbon (SOC), macroporosity (MaP), and hydraulic conductivity (K). The presence of SOC in this system is likely dependent on the residual effects of historical cultivation and old crop residues rather than fresh biological inputs, resulting in a less resilient carbon pool. Furthermore, the correlation between MaP and K suggests that the Without Cover Crops treatment presents a more fragile soil structure. Conversely, the With Cover Crops system showed a complex interrelationship among bulk density (BD), penetration resistance (PR), and microporosity (MIC), indicating a biologically consolidated matrix where roots have increased heterogeneity and retention capacity.

Overall, the multivariate analyses across both sites demonstrate that *Lolium perenne* cover crops favored the accumulation of soil organic carbon (SOC) in these coarse-textured

environments. The recurrence of SOC vectors aligned with physical retention parameters (MIC/PAWC) in cover crop systems supports the findings of Romanoski et al. (2025) and Meurer et al. (2020) regarding the interdependence of carbon and pore functionality. While the bare soil systems favor macroposity and rapid flow (high K), which poses environmental risks for leaching (Svensson et al., 2026), the cover crop systems promote a more balanced structure that favors retention and controlled flux, essential for the resilience of sandy soils in temperate climates.

Although the univariate analysis did not show statistical differences for most attributes (Tables 12 and 15), evaluating these values against critical limits established in the literature is essential to understand the agronomic potential of these soils. Regarding soil aeration, the macroporosity (MaP) values in both Fotegården and Mellby, at all depths, were consistently superior to the critical limit of $0.10 \text{ m}^3 \text{ m}^{-3}$ (Tables 12 and 15). According to Grable & Siemer (1968), oxygen diffusion becomes restrictive to root growth below this threshold. Therefore, despite the sandy texture's susceptibility to consolidation, aeration is not a limiting factor for crop development in either management system.

Plant-Available Water Capacity (PAWC), however, presents a distinct scenario. The results indicate that Fotegården (at both 0.00-0.20 and 0.30-0.50 m depths) and Mellby in the surface layer (0.00-0.20 m) exhibit Plant-Available Water Capacity (PAWC) values around $0.20 \text{ m}^3 \text{ m}^{-3}$ (Tables 12 and 14). These values are considered ideal for promoting optimal root growth and development, falling within the "optimal" range proposed by Reynolds et al. (2009). However, at Mellby in the 0.30-0.50 m layer, PAWC values were considerably lower ($< 0.15 \text{ m}^3 \text{ m}^{-3}$), falling into a range considered "limited" or poor (Reynolds et al., 2009). This reduction in subsurface retention capacity highlights the vulnerability of the Mellby profile to drought stress if roots cannot access deeper water tables.

Analyzing the hydraulic conductivity (K), the values observed in the Without Cover Crops systems were generally higher, often exceeding 20 cm h^{-1} . According to the classification by Tobella et al. (2023) for coarse-textured soils, these values are categorized as "rapid" to "very rapid". While high permeability prevents surface runoff, in the context of nutrient management in Scandinavia, it exacerbates the risk of rapid leaching of nitrates and agrochemicals (Svensson et al., 2026). The reduction of K observed in the With Cover Crops systems (Tables 12 and 15), although statistically significant, maintains the conductivity within a "moderately rapid" range that is likely more favorable for nutrient residence time and uptake.

Soil bulk density (BD) values, which ranged between 1.35 and 1.55 Mg m^{-3} across sites, must be interpreted according to soil texture. For sandy loams and loamy sands, Pachepsky et

al. (2015) suggest that growth-limiting BD thresholds are generally > 1.60 or 1.65 Mg m^{-3} . Therefore, even though the multivariate analysis associated the cover crop systems with higher BD vectors (Figures 12 and 15), the absolute values observed (Tables 12 and 15) remain below the restrictive critical limits.

Finally, regarding soil organic carbon (SOC), the values observed (ranging from 12 to 25 g kg^{-1}) are typical for coarse-textured soils in temperate climates but are considered low for providing high structural stability resilience compared to clayey soils (Meurer et al., 2020). In sandy soils, the lack of physical protection by clay minerals makes SOC highly dynamic and vulnerable to mineralization (Yang et al., 2018). Although differences were not statistically significant in the univariate ANOVA, the consistent association of SOC with physical quality indicators (PAWC, MIC) in the PCA for cover crop systems underscores its crucial role. As noted by Poeplau et al. (2015) in Swedish soils, even small increments in SOC derived from *Lolium perenne* are vital for cementing sand particles, thus, any management that fails to maintain these levels (like the bare fallow) risks long-term physical degradation.

6.6 CONCLUSION

The results of this study confirmed the hypothesis that the long-term adoption of cover crops (*Lolium perenne*) promotes improvements in soil physical properties and organic carbon compared to bare soil. The With Cover Crops system presented an association between soil organic carbon (SOC) and microporosity, maintaining hydraulic conductivity within a balanced range ($31\text{-}38 \text{ cm h}^{-1}$) that favors water retention.

Conversely, the Without Cover Crops system exhibited excessive hydraulic conductivity values in the subsurface layers ($> 41 \text{ cm h}^{-1}$ at Fotegården and $> 66 \text{ cm h}^{-1}$ at Mellby). These findings emphasize the importance of biological management strategies to mitigate the risks of rapid vertical drainage and physical degradation in coarse-textured soils. In this context, the study reinforces that the integration of cover crops into agricultural landscapes promotes sustainability by balancing soil hydraulic functionality and carbon preservation

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7 GENERAL CONCLUSIONS

The results of this thesis confirm that sustainable agricultural intensification, driven by conservation management, is a key factor for restoring soil functionality, although specific physical mechanisms vary according to soil texture and climate.

From the subtropical clayey soils of Brazil to the temperate sandy soils of Sweden, the presence of living roots and the integration of components proved to be determining factors for soil quality. In Brazilian clayey Ferralsols, the challenge was to overcome physical constraints to flow; in this case, the introduction of crop rotations in Integrated Crop-Livestock Systems (ICLS) was crucial to increase hydraulic conductivity and air permeability. However, the study revealed an important functional trade-off: although increased macroporosity improves drainage and aeration, excessive aeration may compromise organic carbon stability.

On the other hand, in Swedish sandy soils, the challenge was retention; in this case, cover crops (*Lolium perenne*) acted to increase microporosity and carbon input, improving water retention capacity and mitigating leaching risks. Furthermore, the integrated assessment demonstrated that even when physical improvements are subtle (as in livestock-forestry systems), the systemic gain in ecosystem services, driven by carbon sequestration and timber provision, justifies the management complexity.

Therefore, it is concluded that the ideal soil structure is not a static standard, but a functional balance modulated by soil-plant dynamics. Integrated and conservation systems are successful because they allow the soil to self-organize: creating macropores for flow when necessary (clay) and micropores for retention when necessary (sand). Thus, this thesis provides robust evidence that fostering biological diversity is the most effective strategy to reconcile agricultural productivity with the urgent need for carbon sequestration and climate resilience.

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9 SOCIAL IMPACT OF RESEARCH

The social impact of this thesis is grounded in the translation of complex soil properties into applicable solutions for food sovereignty and climate resilience, aligning with the categories of practical, social, and political impact. The core of this contribution lies in the validation of conservation management practices as a tool for functional restoration, redefining the perception of soil beyond a mere productive support and consolidating it as a strategic asset in the provision of multifunctional benefits to society.

In the practical and economic sphere, the research demonstrates that both Integrated Crop-Livestock Systems (ICLS) and the use of cover crops not only mitigate physical constraints but also optimize the soil's pore functionality. In clayey soils, it was proven that the crop component improves flow properties by increasing saturated hydraulic conductivity and intrinsic air permeability. Concurrently, in sandy soils, the use of cover crops contributes to the increment of soil organic carbon, acting as a structural stabilizing agent. Together, these strategies offer the productive sector guidelines to reduce vulnerability to climate extremes, ensuring the stability of agricultural production in both tropical and temperate scenarios.

The social and environmental relevance of this thesis is consolidated through the application and analysis of the Ecosystem Services Index (ESI). The research revealed that complex systems, such as livestock-forestry, exhibit functional superiority (ESI: 0.71) compared to sole livestock systems (ESI: 0.52). More than just a metric, the ESI acts as an indicator of ecosystem efficiency, proving that biological diversification enables a fundamental balance: while the tree component enhances regulating services through carbon sequestration and wood provision, the soil's physical structure is maintained at adequate support levels. The discovery that pore connectivity explains up to 84% of the variation in soil organic carbon (SOC) provides an unprecedented scientific basis for carbon credit policies and environmental governance, allowing the ESI to be used as a parameter for monitoring the transition of rural properties to low-carbon models.

Consequently, this work contributes directly to the global Sustainable Development Goals, integrating SDG 2 (Zero Hunger and Sustainable Agriculture), SDG 13 (Climate Action), and SDG 15 (Life on Land). By unifying soil physics with carbon dynamics and the provision of ecosystem services, this thesis offers a scientific legacy that meets the pressing need to reconcile economic development with environmental preservation. The social impact generated herein resides in the provision of a protocol for maximizing the Ecosystem Services Index (ESI), capable of sustaining resilient communities and functional ecosystems for future

generations, reaffirming the role of academic research as a driver of social transformation and global climate resilience.

10 APPENDIX: TECHNICAL PRODUCTION – ORGANIZED EVENT

Product 8: Organized Event (Technical Product)

1. Event Characterization

Event Name: IV Brazilian Congress of Integrated Agricultural Production Systems (CBSIPA) and VIII SIPA SUL.

Type: National Congress and Field Day.

Year: 2025.

Location: Sector of Agricultural Sciences, Federal University of Paraná (UFPR).

Scope: National.

Target Audience: academics, researchers, farmers, and professionals in agricultural sciences.

2. Connection with Research (Adherence)

Research Line: NITA – Núcleo de Inovação Tecnológica em Agropecuária.

Linked Research Project: Relações entre o estoque de carbono e a permeabilidade ao ar e à água no solo sob diferentes sistemas de uso e manejo.

Purpose and Report: The event promoted the dissemination of technical-scientific knowledge on Integrated Agricultural Production Systems. The organization of the event allowed for the direct transfer of knowledge investigated in the doctoral thesis, such as pore functionality and soil organic carbon dynamics, to the technical and productive public through lectures and practical activities during the Field Day.

3. Impact and Applicability Analysis

Impact: Dissemination of crop-livestock-forestry integration protocols essential for carbon sequestration and the resilience of the agricultural ecosystem.

Applicability: High replicability of the management strategies discussed for optimizing field sustainability.

Complexity: High complexity, involving synergy between UFPR, organizing committees, and the productive sector.

Organizing Team

Permanent Faculty Member: Karina Maria Vieira Cavalieri Polizeli

Permanent Faculty Member: Leandro Bittencourt de Oliveira

Student (Doctorate): Vanessa Silva Romanoski

Supporting Documentation

Certificate 1: NITA Field Day – Integrated Crop-Livestock Systems (March 2024 – 16h).

Certificate 2: Field Day on Integrated Crop-Livestock Systems (September/October 2025 – 50h).

Certificate 3: IV CBSIPA and VIII SIPA SUL (October 2025 – 50h).

11 ANNEX – PROOF OF EXTENSION ACTIVITIES AND EVENT ORGANIZATION

Purpose: This appendix brings together the certificates for the organization of and participation in extension events linked to the Sector of Agricultural Sciences at UFPR. These activities served as the basis for technology transfer and the dissemination of scientific results obtained during the doctoral research on integrated crop-livestock systems (ICLS).

Certificate 1: NITA Field Day – Integrated Crop-Livestock Systems (March 2024 - 16h).

Certificate 2: Field Day on Integrated Crop-Livestock Systems (September/October 2025 - 50h).

Certificate 3: IV CBSIPA and VIII SIPA SUL (October 2025 - 50h).

Certificate 1: NITA Field Day – Integrated Crop-Livestock Systems (March 2024 - 16h).



MINISTÉRIO DA EDUCAÇÃO
UNIVERSIDADE FEDERAL DO PARANÁ
PRÓ-REITORIA DE EXTENSÃO E CULTURA
COORDENADORIA DE EXTENSÃO

CERTIFICADO

VANESSA SILVA ROMANOSKI participou como VOLUNTÁRIO(A) no(a) Evento de Extensão "DIA DE CAMPO: SISTEMAS INTEGRADOS DE PRODUÇÃO AGROPECUÁRIA (SIPA)" coordenado por LEANDRO BITTENCOURT DE OLIVEIRA, cumprindo 16h de atividades no período de 07/03/2024 a 08/03/2024, promovido pelo(a) Setor de Ciências Agrárias da Universidade Federal do Paraná.

06 de Abril de 2024

Profª. Drª. Mayara Elita Braz Carneiro
Pró-Reitor(a) de Extensão e Cultura

Profª. Drª. Mabel Karina Arantes Alves
Coordenador(a) de Extensão



Evento de Extensão

DIA DE CAMPO: SISTEMAS INTEGRADOS DE PRODUÇÃO AGROPECUÁRIA (SIPA)

Coordenador(a): LEANDRO BITTENCOURT DE OLIVEIRA

Data de Início: 08/03/2024

Data de Finalização: 08/03/2024

Atividades do membro:

- Organização : 8h
- Participante : 8h

<https://extensao.ufpr.br/publico/autenticacao.jsf> - Código para autenticação: 1BA32F5D99AC3D



Certificate 2: Field Day on Integrated Crop-Livestock Systems (September/October 2025 - 50h).



MINISTÉRIO DA EDUCAÇÃO
UNIVERSIDADE FEDERAL DO PARANÁ
PRÓ-REITORIA DE EXTENSÃO E CULTURA
COORDENADORIA DE EXTENSÃO

CERTIFICADO

VANESSA SILVA ROMANOSKI participou como ORGANIZADOR(A) no(a) Evento de Extensão "DIA DE CAMPO EM SISTEMAS INTEGRADOS DE PRODUÇÃO AGROPECUÁRIA" coordenado por LEANDRO BITTENCOURT DE OLIVEIRA, cumprindo 50h de atividades no período de 01/09/2025 a 10/10/2025, promovido pelo(a) Setor de Ciências Agrárias da Universidade Federal do Paraná.

10 de Novembro de 2025

Profª. Drª. Andrea Berriel Mercadante
Pró-Reitor(a) de Extensão e Cultura

Prof. Dr. Rodrigo Rosi Mengarelli
Coordenador(a) de Extensão



sigauFPR

Evento de Extensão

DIA DE CAMPO EM SISTEMAS INTEGRADOS DE PRODUÇÃO AGROPECUÁRIA
Coordenador(a): LEANDRO BITTENCOURT DE OLIVEIRA
Data de Início: 09/10/2025
Data de Finalização: 09/10/2025
Atividades do membro:
- Organização: 50h

<https://extensao.ufpr.br/public/autenticacao.jsf> - Código para autenticação: 89ADC11D2A0F78



sigauFPR

Certificate 3: IV CBSIPA and VIII SIPA SUL (October 2025 - 50h).

	<p>MINISTÉRIO DA EDUCAÇÃO UNIVERSIDADE FEDERAL DO PARANÁ PRÓ-REITORIA DE EXTENSÃO E CULTURA COORDENADORIA DE EXTENSÃO</p>
<h1>CERTIFICADO</h1>	
<p>VANESSA SILVA ROMANOSKI participou como ORGANIZADOR(A) no(a) Evento de Extensão "IV CBSIPA E VIII SIPA SUL" coordenado por LEANDRO BITTENCOURT DE OLIVEIRA, cumprindo 50h de atividades no período de 02/06/2025 a 10/10/2025, promovido pelo(a) Setor de Ciências Agrárias da Universidade Federal do Paraná.</p>	
<p>10 de Novembro de 2025</p>	
<p>Profª. Drª. Andrea Berriel Mercadante Pró-Reitor(a) de Extensão e Cultura</p>	<p>Prof. Dr. Rodrigo Rosi Mengarelli Coordenador(a) de Extensão</p>



<p>Evento de Extensão</p>	
<p>IV CBSIPA E VIII SIPA SUL Coordenador(a): LEANDRO BITTENCOURT DE OLIVEIRA Data de Início: 06/10/2025 Data de Finalização: 08/10/2025 Atividades do membro: - Organização: 50h</p>	
<p>https://extensao.ufpr.br/public/autenticacao.jsf - Código para autenticação: 3171E60A4D3E62</p>	



12 BIOGRAPHICAL SKETCH

Vanessa Silva Romanoski Itacarambi, daughter of Susana Marcia da Silva Romanoski and Edegar Paulo Romanoski. She holds a Bachelor's degree in Agronomy (2015-2019) from the Federal University of Western Bahia, Multidisciplinary Center of the Barra Campus (UFOB). She earned her Master's degree in Agronomy (Plant Production) from the Federal University of Paraná (2020-2022), focusing on the research line of Plant Production in Sustainable Agriculture, under the supervision of Professor Maristela Panobianco Vasconcello, PhD. She earned her PhD in Soil Science from the Graduate Program in Soil Science at the Federal University of Paraná (2022-2026), where she developed her thesis in the research area of Soil and Water Quality, Management, and Conservation, under the supervision of Professor Karina Maria Vieira Cavalieri Polizeli, PhD. During her doctoral studies, she completed an exchange program at the Swedish University of Agricultural Sciences (SLU), in Sweden.