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Bioindicadores de qualidade do solo em um sistema integrado de produção agropecuária

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BIOINDICADORES DE QUALIDADE DO SOLO EM UM SISTEMA INTEGRADO DE PRODUÇÃO AGROPECUÁRIA

Tese apresentada ao Curso de Pós-Graduação em Ciência do Solo, da Universidade Federal de Santa Maria (UFMS, RS), como requisito parcial para obtenção do título de **Doutor em Ciência do Solo.**

Orientador: Prof. Dr. Rodrigo Josemar Seminoti Jacques

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Santa Maria, RS
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DEDICATÓRIA

Aos meus pais Eduardo e Elvine e ao meu esposo Renan, dedico.

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"Tivesse a noite límpida milhões de estrelas,
mas equidistantes e de igual brilho, como se
admirariam o Cruzeiro do Sul, a Estrela
d'Alva, as Três Marias? A Diversidade é o
encanto da Vida."

(Leopoldo Magno Coutinho)

RESUMO

BIOINDICADORES DE QUALIDADE DO SOLO EM UM SISTEMA INTEGRADO DE PRODUÇÃO AGROPECUÁRIA

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ORIENTADOR: Rodrigo Josemar Seminoti Jacques

Os sistemas integrados de produção agropecuária (SIPAs) podem melhorar os atributos químicos, físicos e biológicos do solo. Porém, a intensificação do pastejo pode suplantar estes benefícios e causar graves prejuízos ao SIPA. A biota do solo é considerada uma boa bioindicadora ambiental e pode atestar sobre a qualidade do manejo deste sistema. O objetivo do estudo foi conhecer as implicações da intensificação do pastejo sobre a biota do solo em um SIPA de longa duração, verificar quais as condições edáficas mais influenciam os atributos biológicos, e avaliar se estes atributos podem ser utilizados como indicadores de qualidade ambiental do SIPA. O experimento é conduzido há 15 anos em uma área de 23 hectares com a sucessão *Glycine max* para produção de grãos no verão e *Avena strigosa* + *Lolium multiflorum* para o pastejo contínuo dos bovinos no inverno. Os tratamentos são constituídos pelas alturas de pastejo de 10, 20, 30 e 40 cm e por testemunhas sem pastejo. A amostragem foi realizada em quatro épocas entre 2014 a 2016, duas após o final da estação de pastejo e duas após a colheita da soja. Foram avaliados a respiração basal do solo, o conteúdo de carbono na biomassa microbiana (CBM), o quociente metabólico e a diversidade e abundância da meso e macrofauna epiedáfica, além das variáveis químicas, físicas e de cobertura vegetal para fins de correlação. No tratamento sem pastejo e nas maiores alturas da pastagem quantificaram-se a maior respiração microbiana, conteúdo de CBM, abundância e diversidade da fauna epiedáfica. Na menor altura de pastejo houve redução da cobertura vegetal e prejuízos aos atributos físicos do solo, o que resultou em menor teor de umidade e redução da atividade, abundância e diversidade dos organismos do solo. Estes resultados reforçam a necessidade de um manejo adequado da carga animal na pastagem para que não ocorra comprometimento à sustentabilidade dos SIPAs.

Palavras-chave: Microrganismo. Invertebrado do solo. Fauna edáfica. Bioindicador. Integração lavoura-pecuária. Sustentabilidade.

ABSTRACT

SOIL QUALITY BIOINDICATORS IN AN INTEGRATED CROP-LIVESTOCK SYSTEM

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Integrated crop-livestock systems (ICLS) can improve the chemical, physical and biological soil attributes. However, grazing intensification may outweigh these benefits and cause severe losses to the system. Soil biota is considered a good environmental bioindicator and can attest to the quality of the system management. This study aimed to know the implications of grazing intensification on soil biota in a long-term ICLS, to verify which soil conditions most influence biological attributes, and to evaluate if these attributes can be used as environmental quality indicators in ICLS. The experiment was started in 2001, on a 23 hectare area, with *Glycine max* in summer and *Avena strigosa*+*Lolium multiflorum* for continuous cattle grazing in winter. The treatments consisted of four sward heights (10, 20, 30, and 40 cm), plus an ungrazed area, as the control. Sampling was performed in four seasons between 2014-2016, two after the grazing season and two after soybean harvest. Soil basal respiration, microbial biomass carbon (MBC), microbial quotient, diversity and abundance of meso and macrofauna, as well as chemical, physical and vegetation cover variables were evaluated for correlation. The highest microbial respiration, MBC content, abundance and diversity of the soil fauna were quantified under moderate (20 and 30 cm) and light (40 cm) grazing intensities and under no grazing. At the high grazing intensity was a vegetal cover reduction and decreasing on the physical soil quality, which resulted in a lower soil moisture and activity, abundance and diversity reduction of soil organisms community. These results increase the need for an adequate management of pasture areas, without harming the ICLS sustainability.

Keywords: Microorganism. Soil invertebrate. Soil fauna. Bioindicator. Sustainability. Pasture areas.

SUMÁRIO

1 INTRODUÇÃO GERAL

1.1 SISTEMAS INTEGRADOS DE PRODUÇÃO AGROPECUÁRIA

Durante o último século a população mundial aumentou quatro vezes, fato possível devido à melhoria das técnicas agrícolas e industriais (TURMEL et al., 2015). Este crescimento populacional contínuo demanda um constante aumento da produção agropecuária, com manutenção da sustentabilidade dos sistemas agrícolas para as próximas gerações. A solução para essa questão está associada ao manejo sustentável do solo (LAL, 2007), com a intensificação do uso de áreas já utilizadas para a agricultura, através de alternativas que alcancem a sustentabilidade e aumentem a eficiência agropecuária (VILELA et al., 2008).

Neste sentido, os Sistemas de Integrados de Produção Agropecuária (SIPA) são apontados como uma alternativa agrícola sustentável para maximizar o uso do solo, com grande possibilidade de adoção pelos agricultores (CARVALHO et al., 2005). O SIPA utiliza-se das interações positivas entre planta e animal, o que resulta em melhorias ambientais e viabilidade econômica das propriedades rurais (CARVALHO et al., 2010).

Este sistema é planejado para explorar produtos e propriedades emergentes oriundos das interações entre solo, planta, animal e atmosfera (ANGHINONI, 2013), e baseia-se na diversificação, rotação, consorciação e/ou sucessão das atividades da agricultura e da pecuária dentro da propriedade rural (KLUTHCOUSKI et al., 1991). Este manejo, se aplicado de forma harmônica, traz benefícios para ambas as atividades, onde o solo é explorado durante o ano inteiro, o que favorece o aumento na oferta de grãos, de carne e de leite a um baixo custo (ALVARENGA e NOCE, 2005). Além dos benefícios econômicos, o SIPA é associado à melhoria dos atributos físicos e químicos do solo e redução de pragas e doenças (ASSMANN et al., 2015; CECAGNO et al., 2016; SILVA et al., 2014b; WESP et al., 2016).

O SIPA traz inúmeros benefícios ao solo, quando bem manejado. O solo atua como mediador dos vários processos aos quais o sistema é submetido, incorporando nutrientes e energia oriundos de uma diversidade vegetal, associados a novas vias de fluxo de nutrientes e água de origem animal (ANGHINONI, 2013). Como consequência tem-se a melhoria de seus aspectos físicos, químicos e biológicos, além do aumento de produtividade das culturas de interesse agrícola e das pastagens, e renda adicional com a venda dos animais resultando em melhoria da qualidade de vida do produtor (BERRY et al., 2003; VILELA et al., 2008). Aliado a isso está a facilidade do sistema se adaptar a qualquer tamanho de propriedade, desde que as características de solo não apresentem restrições.

O SIPA pode afetar direta e intensamente a biologia do solo através de vários processos. Destes, cabe citar a alteração na quantidade e qualidade dos resíduos aportados (SILVA et al., 2014a), o aporte diversificado de resíduos vegetais (SOUZA et al., 2010), as modificações na direção, magnitude e composição dos fluxos de nutrientes (ANGHINONI, 2013) e as alterações nos atributos físicos do solo (CECAGNO et al., 2016). Poucos foram os estudos realizados abordando as comunidades biológicas neste tipo de sistema. Para comunidades microbianas foi observada resposta rápida às mudanças no pastejo (LE ROUX et al., 2007), onde a intensidade de pastejo moderada pode apresentar aumentos tanto na atividade quanto na diversidade microbiológica, se comparadas a áreas de intenso ou nenhum pastejo (ZHOU et al., 2010). Para a fauna de invertebrados edáficos, o SIPA parece favorecer a manutenção da diversidade da fauna invertebrada (PORTILHO et al., 2011) e beneficiar um ambiente edáfico biologicamente mais ativo, se comparado aos sistemas convencionais (SILVA et al., 2011). Resultados na região subtropical do Brasil acerca da fauna e da microbiota do solo relacionados ao SIPA ainda são escassos.

1.2 COMUNIDADE DE ORGANISMOS EDÁFICOS COMO BIOINDICADORES DE QUALIDADE DO SOLO

A qualidade do solo afeta diretamente a produtividade e a sustentabilidade dos sistemas agrícolas. Pode-se representar a qualidade por um conjunto de parâmetros obtidos no solo, através de suas propriedades físicas, químicas e biológicas (LARSON e PIERCE, 1994), e a escolha dos parâmetros a serem utilizado depende do objetivo da avaliação a ser feita (GARDI et al., 2002). Os indicadores biológicos de qualidade do solo têm sido utilizados com frequência em diversos estudos para avaliar a qualidade em diferentes sistemas de plantio (BARTZ et al., 2013; CLUZEAU et al., 2012; FLOCH et al., 2011; VASCONCELLOS et al., 2013). As principais características exigidas para ser um bom indicador de qualidade do solo são apresentar estreita relação com funções do solo, sensibilidade e rápida resposta às mudanças no meio (DORAN e ZEISS, 2000). Pode-se considerar que a principal vantagem dos bioindicadores seja o fato de eles constituírem o atributo vivo do solo, apresentando, portanto, mais rápida resposta a variação no ecossistema se comparados com os atributos químicos ou físicos.

O ambiente edáfico representa um hábitat natural que comporta uma grande e diversa comunidade de seres vivos, que vão desde microrganismos até um grande grupo de espécies de macroinvertebrados. Estes organismos influenciam o meio no qual eles vivem, pois são responsáveis por funções ambientais importantes, atuando diretamente sobre a decomposição da matéria orgânica, a ciclagem de nutrientes, e a estruturação do solo (LAVELLE et al., 1993; LAVELLE e SPAIN, 2002). Da mesma forma, os organismos são influenciados pelo meio. A intensidade do uso do solo e o tipo e a qualidade da cobertura vegetal alteram de forma direta e significativa a biodiversidade edáfica (ROSA et al., 2015). A simplificação de hábitat gera mudanças no aporte de resíduos, na oferta de recursos, alterações na temperatura e umidade que podem levar a mudanças drásticas na composição das comunidades biológicas, levando ao seu declínio (BEDANO et al., 2016; LITTLE et al., 2013; PORTILHO et al., 2011).

Os microrganismos do solo, constituídos por bactérias, fungos, algas e protozoários, apresentam ativa participação nos processos de mineralização e imobilização temporária de nutrientes, ciclagem de nutrientes, fluxos de energia e transformação da matéria orgânica no solo (FERREIRA et al., 2011; KOTROCZÓ et al., 2014; SPOHN et al., 2016). Sua atividade é facilmente influenciada por fatores que alteram o solo, como manejo adotado, cobertura vegetal e tipo de fertilização (CARRERA et al., 2007; SHARKHUU et al., 2016), estágio do desenvolvimento de plantas e uso de pesticidas (FERREIRA et al., 2009; FERREIRA et al., 2008). Portanto, os microrganismos são considerados indicadores sensíveis da qualidade do solo (FERREIRA et al., 2011). Assim como as comunidades microbianas, os indicadores microbianos também se apresentam como boas alternativas de avaliação da qualidade do solo (LISBOA et al., 2012). Características como biomassa microbiana do solo (BMS), enzimas extracelulares e taxa de respiração basal do solo podem ser utilizadas como bioindicadoras de qualidade do solo, já que elas são ligadas intimamente com propriedades edáficas importantes, dentre as quais cabe citar os teores de matéria orgânica, a ciclagem de nutrientes e algumas propriedades físicas e químicas do solo (MOSCATELLI et al., 2012; VASCONCELLOS et al., 2013).

A comunidade de invertebrados do solo é diretamente relacionada à transformação, decomposição e liberação de nutrientes a partir de resíduos orgânicos, à estruturação do solo, a criação de bioporos e ao revolvimento do solo, incorporando matéria orgânica ao longo do perfil (BARTZ et al., 2014; ROVEDDER et al., 2009; STEFFEN et al., 2007). Apesar desta relevância ecológica, a atividade e diversidade dos organismos edáficos é sensível ao tipo de uso do solo adotado e pode ser prejudicada em sistemas mal manejados (BARETTA et al., 2014; BARTZ et al., 2014; ROSA et al., 2015). Assim, a abundância, a diversidade e a atividade da fauna edáfica podem fornecer indicativos úteis sobre a qualidade do solo (BARETTA et al., 2014; LAVELLE et al., 2006).

Várias são as opções de bioindicadores de qualidade do solo, porém estudos avaliando a comunidade biológica e a correlacionando com as variáveis obtidas em solos sob sistemas de SIPA são raramente efetuados. Sabe-se do potencial benéfico que o SIPA apresenta sobre os atributos químicos e físicos do solo, e da estreita ligação das comunidades edáficas com estes. Logo, torna-se necessária uma avaliação mais aprofundada do comportamento destas comunidades em solos submetidos ao SIPA.

2 HIPÓTESES E OBJETIVOS

2.1 HIPÓTESES

Em um sistema integrado de produção agropecuária quanto maior for a intensidade do pastejo, maior é a redução da atividade e da diversidade dos organismos do solo;

As áreas com maior intensidade de pastejo apresentam menor qualidade física do solo e menor cobertura vegetal, o que resulta em menor atividade e diversidade da biota edáfica;

Os atributos biológicos do solo são adequados indicadores de qualidade ambiental dos sistemas integrados de produção agropecuária.

2.2 OBJETIVOS

2.2.1 Objetivo Geral

Avaliar as comunidades de micro, meso e macrorganismos do solo após 15 anos de um SIPA manejado sob diferentes intensidades de pastejo e verificar se estes parâmetros biológicos podem ser utilizados como bioindicadores de qualidade neste sistema.

2.2.2 Objetivos Específicos

Determinar a biomassa e a atividade dos microrganismos do solo após 15 anos de pastejo com diferentes intensidades em um sistema integrado de produção agropecuária;

Avaliar a abundância e diversidade dos meso e macrorganismos epiedáficos em um SIPA de longa duração manejado sob diferentes intensidades de pastejo;

Realizar o levantamento das principais características físicas e químicas do solo e de cobertura vegetal após 15 anos de pastejo com diferentes intensidades em um sistema integrado de produção agropecuária;

Estabelecer relações entre os atributos físicos, químicos e de cobertura do solo com os atributos biológicos do solo.

Title

Effects of 15 years of intensive grazing on soil microbial biomass and activity in an integrated crop-livestock system*

Authors

Ângela Denise Hubert Neufeld, Hazael Soranzo de Almeida, Daiane Dalla Nora, Willian Braga dos Santos, Paulo César de Faccio Carvalho, Rodrigo Josemar Seminoti Jacques

Abstract

Integrated crop-livestock systems (ICLs) can improve soil chemical, physical and biological characteristics. However, intensive grazing may outweigh these benefits and cause serious damage to ICLs. Microorganisms are sensitive bioindicators and may indicate changes to soil quality. The aim of the present study was to investigate the effects of 15 years of intensive grazing in an integrated soybean-beef cattle system on soil microbial biomass and respiration, identifying which edaphic factors most affect these parameters. The experiment was conducted over a 15-year period in a 23-ha area, characterized by the crop succession *Glycine max* (summer grain production) and *Avena strigosa* + *Lolium multiflorum* (winter pasture). The treatments included four different grazing heights (10, 20, 30 and 40 cm) and one control without grazing. Four samplings were performed between 2014 and 2016, two following the grazing season and two following the soybean harvest. The following parameters were evaluated: soil basal respiration, microbial biomass carbon content (MBC) and metabolic quotient. The soil chemical and physical parameters and plant cover parameters were measured for correlation. Soil respiration and MBC were highest for the treatments without grazing and with higher grazing heights. Negative effects on physical soil characteristics were observed for

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the treatment with the lowest grazing height, resulting in decreased soil moisture and microbial biomass and activity.

Keywords: microorganism, metabolic quotient, bioindicator, integrated crop-livestock systems, sustainability.

1. Introduction

The introduction of animal grazing to crop production areas may improve soil physical, chemical and biological properties (Berry *et al.*, 2003; Vilela *et al.*, 2008). The presence of animals results in the incorporation of feces and urine into the soil, resulting in higher forage plant biomass, root activity and dry mass per growth season under grazing conditions. All this contributes to create a favorable environment for the growth and activity of soil microorganisms.

However, inadequate management of integrated crop-livestock systems (ICLs) due to an increased grazing load on pastures is often observed (Neves Neto *et al.*, 2013). Intensive grazing may outweigh the benefits of ICLs and have detrimental effects on soil. High grazing load has been reported to change the soil temperature and moisture regime (Klein et al., 2005), and intensive grazing was observed to result in decreased soil vegetation cover and organic carbon concentration as well as increased topsoil compaction (Kölbl et al., 2011). All these changes directly or indirectly affect microbial biomass and activity, and these parameters can therefore serve as bioindicators of an ICL’s ecosystem quality and balance.

Recent studies indicate that soil microbial communities may be affected by grazing. The introduction of grazing to croplands was observed to result in increased microbial activity and soil basal respiration rates in Mongolia (Sharkhuu *et al.*, 2016) and the United States (Adewopo *et al.*, 2015). In China, grasslands were observed to present a more stable C-CO_2 emission rate throughout the day than regenerated forests or farmlands (Liu *et al.*, 2016).

Although the ICL is increasingly acknowledged as an economic, social and environmental alternative to increase food production in several countries, few studies have focused on the long-term effects of intensive grazing on the dynamics of soil microbial communities, especially under subtropical soil and climate conditions. An experiment was performed to test the hypothesis that intensive grazing in a long-term ICL decreases soil microbial biomass and activity. The aim of the present study was to investigate the effects of 15 years of intensive grazing in an integrated soybean-beef cattle system on soil microbial biomass and respiration, identifying which edaphic factors most affect these parameters.

2. Materials and methods

2.1. Experimental site and conduction of the experiment

The study was performed as part of ongoing research conducted since 2001 by the Integrated Crop-Livestock Systems Research Group (Grupo de Pesquisa em Sistema Integrado de Produção Agropecuária) of the Federal University of Rio Grande do Sul (Universidade Federal do Rio Grande do Sul – UFRGS), in an area comprising approximately 23 ha, located in the municipality of São Miguel das Missões, Rio Grande do Sul, Brazil (29°03'10"S, 53°50'44"W). The soil is clayey (540 g kg^{-1} clay for a soil layer 0–20 cm deep) and classified as an Oxisol (Rhodic Hapludox - Soil Survey Staff, 1999). The climate is subtropical with hot and humid summers (type Cfa), according to the Köppen climate classification.

Before the experiment began, the area had been cultivated under no tillage since 1993, with black oat (*Avena strigosa* Schreb) in winter (only for soil cover) and soybean [*Glycine max* (L.) Merr.] in summer. From the beginning of the experiment (2001), until the present, the area has been continuously cultivated with soybean in summer (November to April) for grain production, under no tillage, and the cultivation of black oat + Italian ryegrass (*Lolium multiflorum* Lam.) was initiated in winter (May to October) for pasture, with four different grazing heights (treatments).

The treatments consisted of four grazing intensities: intense (10-cm grazing height), moderate (20- and 30-cm grazing heights), light (40-cm grazing height) and no grazing (control plots). A randomized block experimental design was used, with three replicates per treatment. The size of the plots with grazing varied from 0.9 to 3.6 ha. All plots had continuous grazing, a variable stocking rate and three tester animals and grazer animals as needed (200 kg average initial live weight), resulting in the different grazing heights tested.

Fertilization was based on the soil analysis and performed according to technical recommendations. In pastures, nitrogen fertilization was applied to obtain a yield between 4.0 and 7.0 t ha^{-1} of pasture dry matter. In soybean plantations, phosphorus and potassium fertilization was applied to obtain a yield of 4.0 t ha^{-1} (Assmann *et al.*, 2015; Schuster *et al.*, 2016).

2.2. Samplings and analyses

For the microbiological analyses, two samplings were performed immediately after the end of the grazing season (beginning of November 2014 and 2015) and two immediately after the soybean harvest (beginning of May 2015 and 2016). Five soil samples were collected from each plot, from the corners and center of a 36 x 60-m virtual quadrat placed in the center of the plot (coordinates obtained using a precision real time kinematic global positioning system). For all samplings, collections were always performed at the same points, from the 0–10 cm soil layer, using a soil-sampling auger. Soil samples were placed in plastic bags, stored in styrofoam boxes with ice and transported to the laboratory, where they were sieved and stored at 4°C for up to one week until analyzed.

Microbial biomass carbon (MBC) was determined using the fumigation-extraction method (De-Polli and Guerra, 1997), and carbon (C) was quantified using a total organic carbon analyzer (TOC-L Shimadzu). The soil basal respiration rate was determined by respirometry (Anderson and Domsch, 1978), consisting of soil incubation for 21 days in the dark at 26°C. The metabolic quotient for CO_2 (qCO_2) was calculated as the ratio between MBC and the accumulated respiration rate (Anderson and Domsch, 1993).

Samples for physical and chemical soil analyses were collected from the 0–10 cm soil layer in November 2014 and April 2015 at the same five points in each plot, as described above. The following soil chemical parameters were determined: total organic carbon (Walkley–Black); pH (water 1:1); P and K (Mehlich-1); Ca, Mg and Al (KCl 1 mol L^{-1}); V (% base saturation); and H+Al (Toledo *et al.*, 2012). The following soil physical parameters were determined: total porosity, macroporosity and microporosity, using a tension table (Embrapa, 1997); soil density, using the volumetric ring method (Blake and Hartge, 1986); and gravimetric moisture (Table 1).

Litter and plant shoot samples were collected from three points in each plot, on the same days as the soil collections, for microbiological analyses. Plant material was dried in a forced air ventilation oven at 65°C until constant weight was achieved. Rainfall and air temperature data were collected for two months for each sampling season, using a meteorological station situated in the experimental area (Figure 1).

2.3. Statistical analyses

An analysis of variance (ANOVA) was performed, followed by a Duncan test when significant differences were found, at $p \leq 0.05$, using the SASM-agri (version 3.2.4) software. Regression analyses were performed to analyze the relationship between MBC and the soil basal respiration rate, for the cycle following the grazing season and following soybean cultivation. The relationships between microbiological parameters and soil moisture, litter and plant biomass, for each sampling, were analyzed using the Pearson's correlation coefficient ($p \leq 0.05$), using the SigmaPlot (version 11.0) software.

3. Results

C-CO_2 production was higher for the treatments without grazing (control) and with higher grazing heights, for all samplings (Figure 2), indicating that intensive grazing decreased soil microbial activity. On average, C-CO_2 production was 16, 19, 26 and 30% lower for the treatment with the 10-cm grazing height than with the 20-, 30- or 40-cm grazing height or without grazing, respectively (Figure 2). The treatment with the 40-cm grazing height presented higher C-CO_2 production than the other grazing treatments, exhibiting values similar to those observed for the control treatment, for most seasons.

Similar to basal respiration, the MBC content also indicated detrimental effects of intense grazing on soil microorganism biomass. Except for the sampling performed in May 2015, MBC was highest for the treatments without grazing and with the highest grazing heights (Figure 2). On average, MBC was 2, 4, 9 and 18% lower for the treatment with the 10-cm grazing height than with the 20-, 30- or 40-cm grazing height or without grazing, respectively (Figure 2). A linear and positive correlation between MBC and basal respiration was observed for the samplings following grazing (R^2=0.88; Figure 3), but not for the samplings following the soybean harvest, for which a low coefficient of determination was observed (R^2=0.43). This was possibly due to the May 2015 sampling, which presented low basal respiration rates and high MBC.

The qCO_2 was low for all treatments and seasons, with a maximum value of 0.29 mg $C\text{-}CO_2$ kg^{-1} mg MBC kg^{-1} (Figure 2). The differences between treatments were therefore also very small, which contributed to the absence of an observable trend between treatments. The low qCO_2 values indicate that the microbial community in the studied ICL was stable, even under intense grazing.

Soil moisture was highest for the control treatment (without grazing) for all samplings, although not statistically significantly different for the first samplings, and tended to increase with increasing grazing height (Figure 4). This seemed to be due to a higher shoot and litter dry mass and better soil physical conditions, observed for the treatments without grazing and with the highest grazing heights. Soil density was lower and soil porosity was higher for these treatments (Table 1). The highest density and lowest porosity were observed for the 10-cm grazing height, which resulted in lower soil moisture (Figure 4).

For most samplings, the microbial basal respiration or MBC content were only significantly correlated with soil moisture and litter and shoot dry mass (Table 2). Basal respiration was positively and significantly correlated with shoot and litter dry mass for all samplings and with soil moisture for the May and November 2015 samplings. MBC was positively correlated with litter dry mass in November 2014 and with moisture, litter and shoot dry mass in November 2015.

4. Discussion

The present data were obtained from four samplings, performed over two years, as part of an experiment conducted over a 15-year period, with the same treatments, in a 23-ha area, supported by a very large soil, plant, animal and meteorological data set collected continuously during the duration of the experiment. This experiment therefore offers an excellent opportunity to study soil microbiology in an ICL and decreases the global knowledge gap on this subject.

Many farmers in southern Brazil currently use non-integrated crop production systems. In winter, 86% of the agricultural area is cultivated with cover plants for straw production, to be used in the no-tillage sowing of soybean in summer (Conab, 2011). This concentrates farmers' entire income on soybean, which is a great risk, considering weather and market instabilities. In turn, oat and Italian ryegrass winter pastures are highly productive; they can be used for animal grazing and still produce enough plant biomass for no-tillage sowing in the summer (Kichel and Miranda, 2006; Vilela *et al.*, 2008).

The introduction of cattle grazing in winter is an attempt to increase and stabilize farmers' incomes through the intensification of soil use (Berry *et al.*, 2003). However, whether introducing grazing can be detrimental to the soil is frequently questioned. Grazing may increase soil microbial activity and biomass, as it results in higher residue input and diversity, due to the incorporation of cattle urine and manure (Clegg, 2006; Lin *et al.*, 2009). In addition, forage plant root activity and growth are promoted by the defoliation caused by grazing, which results in an increased rhizodeposition turnover rate and fine root decomposition (Papatheodorou *et al.*, 2008; Hewins *et al.*, 2016). In turn, grazing may be detrimental to microbial activity because it increases topsoil compaction and decreases soil porosity, decreasing soil aeration and water infiltration, and removes part of the plant biomass, altering soil temperature and moisture (Jia *et al.*, 2006; Souto *et al.*, 2008).

The aim of the present study was to understand the effects of different grazing heights on soil microbial biomass and activity, with a focus on ICL sustainability. Soil microbial biomass and activity were chosen because they are highly sensitive bioindicators of environmental quality, and because high soil biological activity is a requirement for the sustainability of any production system. In biologically active soils, organic residue degradation, nutrient mineralization, soil organic matter formation, soil aggregation, biological control, plant hormone production and other processes occur in adequate levels, contributing to increased crop productivity and environmental conservation.

The present results indicate that intense grazing decreases soil microbial activity and biomass in an ICL. Overall, soil respiration and MBC were higher in treatments without grazing and with moderate to light grazing and were directly correlated to soil moisture and litter and shoot biomass. Soil moisture is one of the primary factors affecting soil microbial community composition and therefore soil respiration (Jia *et al.*, 2006; Chen *et al.*, 2015; Liu *et al.*, 2016). Previous studies have indicated that the seasonal variation of C-CO_2 production is predominantly governed by soil moisture and temperature (Risch and Frank, 2010).

Soil moisture was lower for the intense grazing treatment in all samplings, coinciding with lower plant cover, lower soil porosity and higher soil density, resulting in an environment with lower microbial activity and biomass. In addition, higher removal of shoot biomass results in decreased soil protection against high levels of solar radiation and consequently higher evaporation (Gong *et al.*, 2014).

The present results showed that the higher the grazing pressure is, the lower the soil plant and litter cover and the lower the basal respiration rate and MBC. A higher removal of shoot biomass, and consequent decrease in litter, results in lower soil protection against high levels of solar radiation and higher evaporation (Gong *et al.*, 2014), which causes the habitat conditions to be inadequate for microbial activity.

Overall, C-CO_2 fluxes were higher in areas without grazing or with moderate to light grazing. Peri et al. (2015) observed up to 30% higher microbial activity in areas under moderate grazing than under intense grazing. The exclusion of grazing also increases microbial activity (Prem *et al.*, 2014). However, C cycling was observed to slow following 10 years of grazing exclusion (Medina-Roldán *et al.*, 2012).

For all treatments, the lowest MBC values were observed for the first and last sampling periods, coinciding with the highest and lowest rainfall quantities, respectively. Soil O_2 supply may decrease in periods of high rainfall, which is directly reflected in the amount and activity of aerobic microorganisms (Souto *et al.*, 2008). In turn, low rainfall generates water stress and decreases soil diffusion, which is detrimental to the microbial community (Manzoni *et al.*, 2012; Sharkhuu *et al.*, 2016).

MBC typically corresponds to 2 to 4% of the total soil organic carbon stock (TOC) and is the organic matter fraction that is most sensitive to changes resulting from management (Gama-Rodrigues, 1999). In the present study, the contribution of MBC to TOC varied between samplings, averaging 2% for the first and fourth samplings, and 4% for the second and third samplings, indicating satisfactory microbial growth in the study area. The high MBC observed in the present study is in accordance with the MBC values between 0.9 and 1.8 g kg^{-1} observed in previous long-term grazing experiments (Prem *et al.*, 2014; Spohn *et al.*, 2016; Stevenson *et al.*, 2016). Except for the May 2016 sampling, no significant differences in MBC were observed between the moderate and light grazing treatment, indicating that the microbial communities were well adapted to the ICL. Pasture ecosystems with a high organic matter input rate promote the growth and activity of microbial communities, through the continuous inclusion of plant and animal residues, and promote root turnover, therefore increasing soil respiration rates (Peri *et al.*, 2015).

The qCO_2 was low for all treatments (lower than 0.3 mg C-CO_2 kg^{-1}), indicating a low level of physiological stress in the microbial community. A high qCO_2 indicates that adverse or stressful conditions are present in the microbial population and therefore that the soil organic carbon is being inadequately managed (Anderson and Domsch, 1993). Studies evaluating qCO_2 under several types of management reported 1.9 mg C-CO_2 kg^{-1} for pastures (Stevenson *et al.*, 2016) and 17.8 mg C-CO_2 kg^{-1} for soybean plantations (Zilli *et al.*, 2008). ICL characteristics, such as the frequent input of organic animal and plant residues, permanent soil cultivation and a long experimental duration, contribute to this stability.

Despite the overall low stress level of the microbial community observed for all treatments, decreased soil microbial biomass and activity were observed for lower grazing heights. The detriment to soil microorganisms caused by cattle grazing was minimized at the highest grazing heights. The taller living and dead plant biomass in these treatments led to better soil physical conditions, resulting in higher soil moisture and therefore higher microbial activity and biomass. These results emphasize the need for adequately adjusting the animal load on pastures, to not compromise ICL sustainability.

5. Conclusions

Soil microbial respiration and microbial biomass carbon content were higher without grazing and at the highest grazing heights.

Negative effects on the physical characteristics of soil were observed for the lowest grazing height, resulting in lower soil moisture content, microbial activity and biomass.

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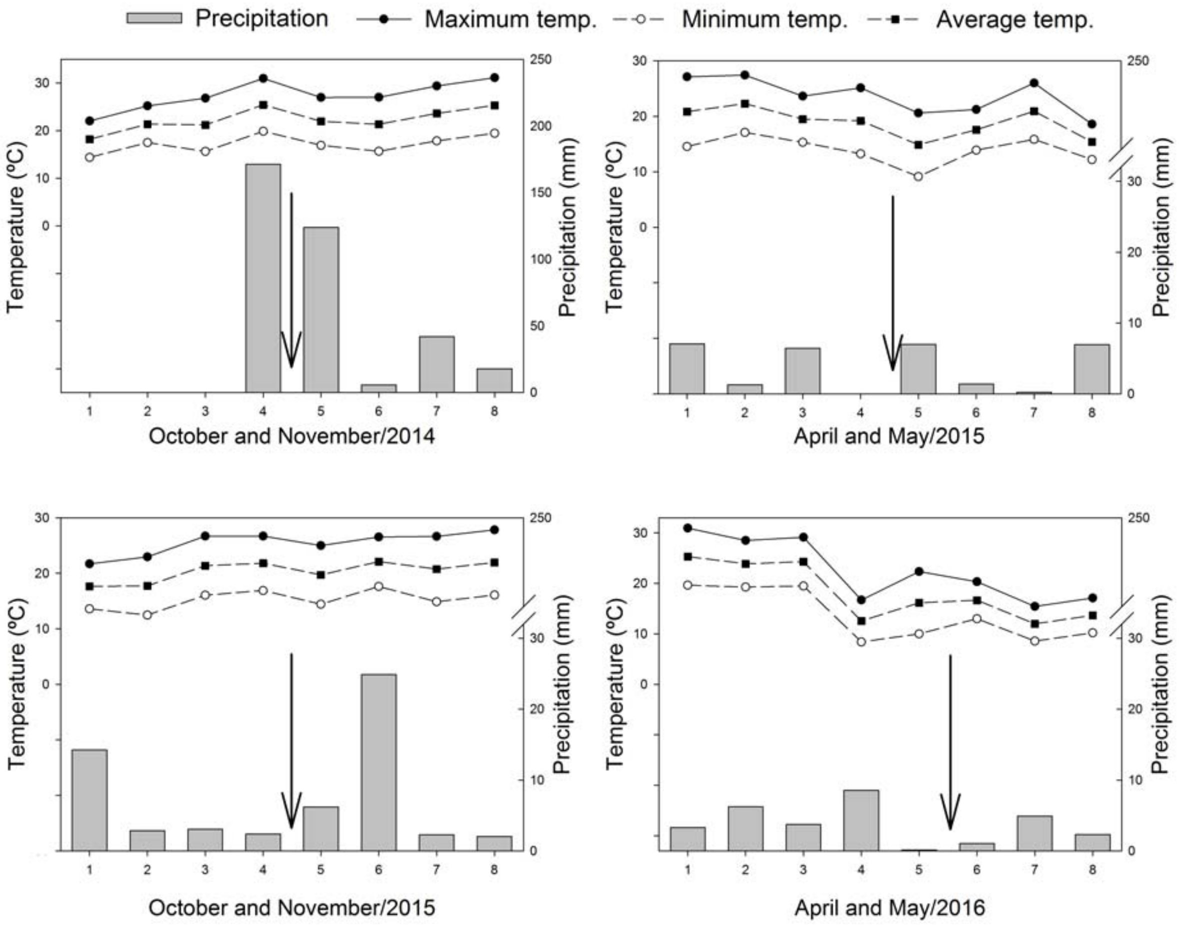

Figure 1: Rainfall and minimum, maximum and mean temperatures for all sampling periods. The arrow indicates the soil sampling date (11/05/2014; 04/29/2015; 11/01/2015; 05/05/2016).

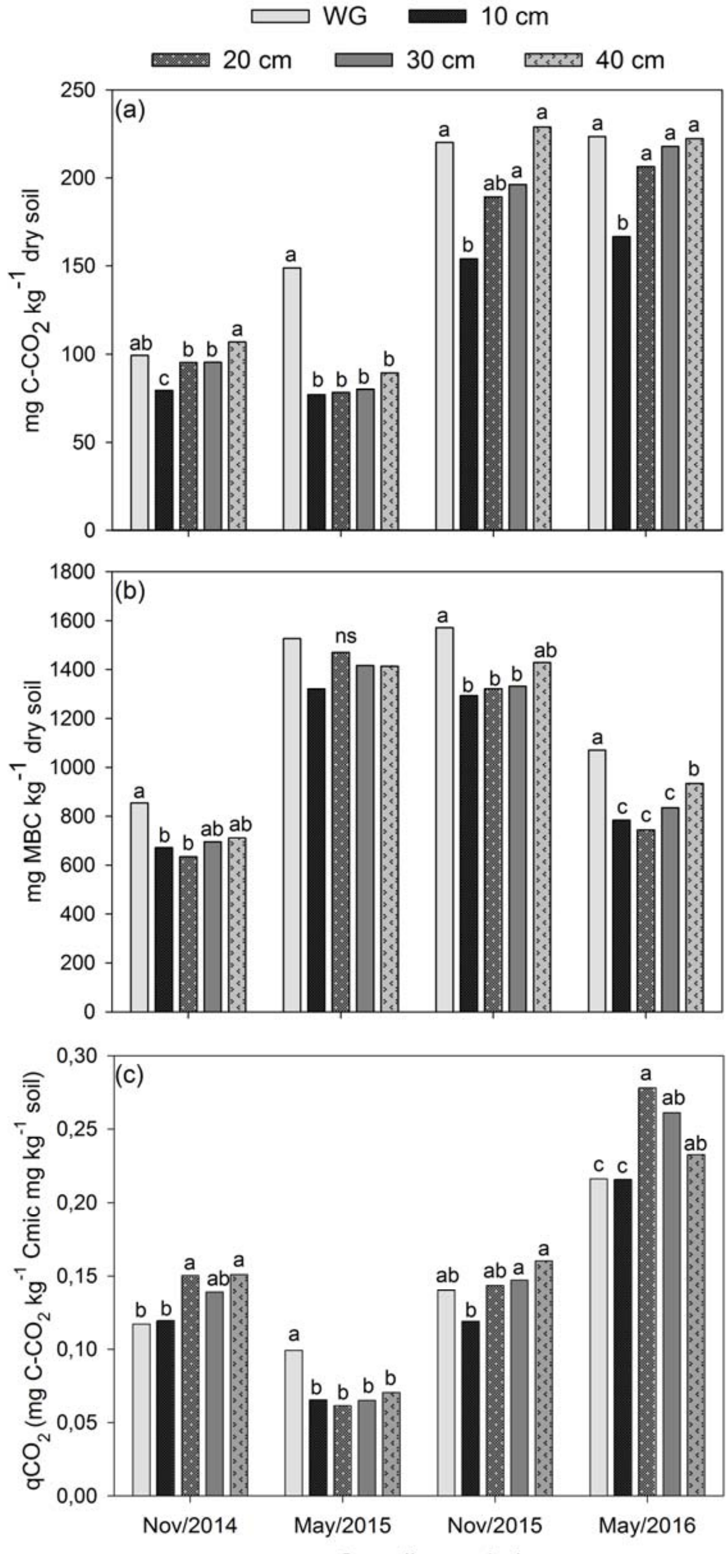

Figure 2: Basal respiration (a), microbial biomass carbon content (b) and metabolic quotient (c) at the 0–10 cm soil layer, following the cattle grazing season (Nov/2014

and Nov/2015) or soybean harvest (May/2015 and May/2016), for different treatments with different grazing heights (10, 20, 30 and 40 cm) or without grazing (WG), in a 15-year-old integrated soybean-beef cattle production system. Values are means of 15 replicates per treatment. Means followed by different letters were significantly different according to the Duncan test at p≤0.05.

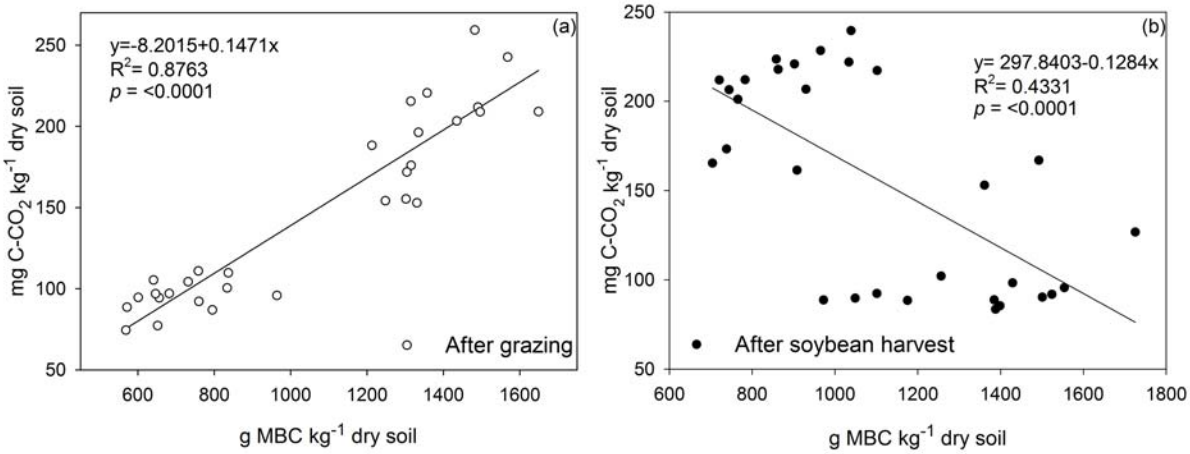

Figure 3: Linear regression between microbial biomass carbon and soil basal respiration rate for samplings performed following the cattle grazing season (a) or soybean harvest (b) in a 15-year-old integrated soybean-beef cattle production system with different grazing intensities (n=30).

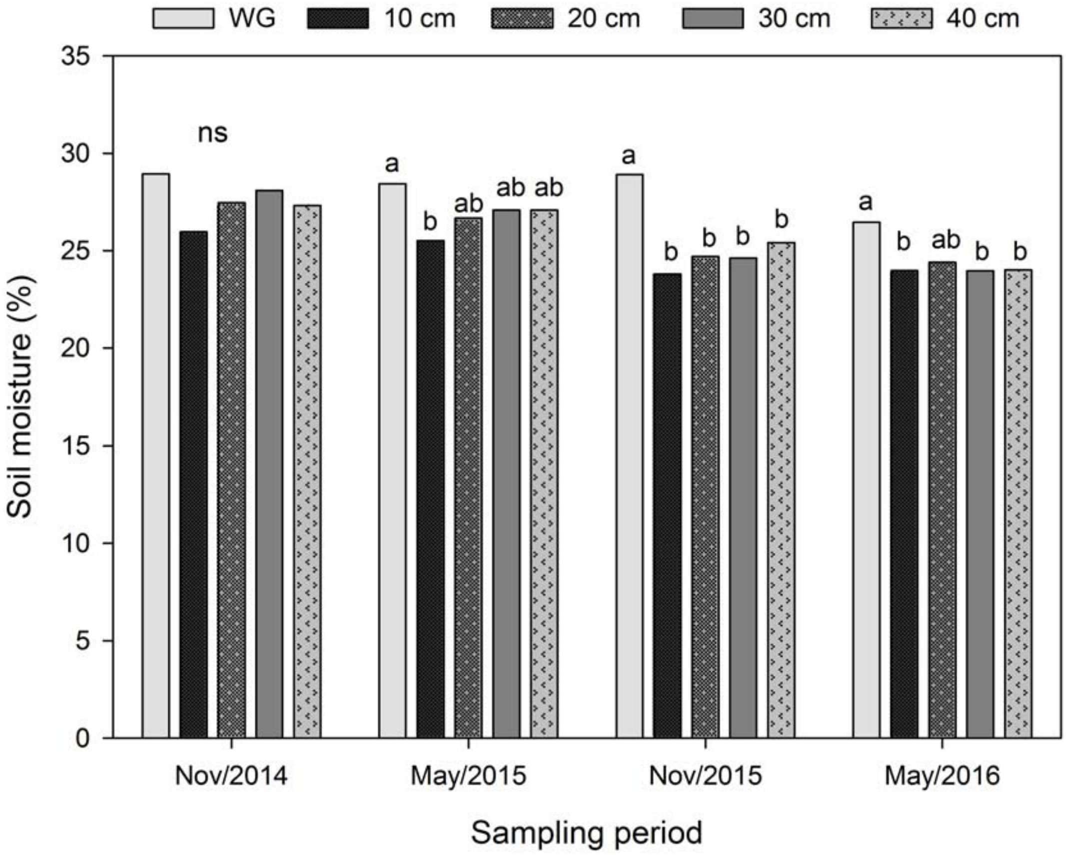

Figure 4: Gravimetric soil moisture at the 0–10 cm soil layer following the cattle grazing season (Nov/2014 and Nov/2015) or soybean harvest (May/2015 and May/2016), for different treatments with different grazing heights or without grazing (WG), in a 15-year-old integrated soybean-beef cattle production system. Values are means of 15 replicates per treatment. Means followed by different letters were significantly different according to the Duncan test at $p \leq 0.05$.

Table 1: Soil physical and chemical characteristics at the 0–10 cm soil layer and plant cover following the cattle grazing season (November) or soybean harvest (May), for different treatments with different grazing heights or without grazing (WG), in a 15-year-old integrated soybean-beef cattle production system. Values are means of 5 replicates per treatment.

Treat.	**SD[1]**	**Ma[2]**	**Mi[3]**	**pH**	**P**	**K**	**TOC[4]**	**V[5]**	**Al**	**Ca**	**Mg**	**H+Al**	**LDM[6]**		**SDM[7]**	
	g cm^{-3}	cm^3 cm^{-3}			mg dm^{-3}		g kg^{-1}	%		cmol$_c$ dm^{-3}			Mg ha^{-1}			
After grazing season													2014	2015	2014	2015
WG	1.26	0.13	0.42	4.3	13.1	219.3	-[8]	27.1	1.4	3.0	1.2	13.6	6.6	5.1	4.6	7.3
10 cm	1.40	0.09	0.40	4.8	9.0	187.4	-	48.5	0.8	4.1	1.7	6.9	1.3	1.0	1.8	0.9
20 cm	1.37	0.09	0.41	4.9	10.7	200.4	-	50.6	0.6	4.0	1.8	6.4	3.0	1.8	3.3	2.6
30 cm	1.33	0.12	0.42	5.0	9.2	163.4	-	52.5	0.6	4.5	1.9	6.4	3.1	2.1	3.4	3.3
40 cm	1.32	0.14	0.38	4.8	13.6	200.6	-	48.6	0.5	4.4	1.8	7.2	3.5	4.0	4.8	4.1
After soybean harvest													2015	2016	2015	2016
WG	1.34	0.09	0.45	4.3	19.5	222.6	23.1	36.4	1.3	3.9	1.5	11.2	9.2	8.5	0[9]	0
10 cm	1.37	0.09	0.41	4.6	9.7	136.3	22.2	44.6	1.1	4.3	2.0	8.5	4.1	4.3	0	0
20 cm	1.36	0.11	0.41	4.7	13.6	181.2	23.3	47.5	1.0	4.7	2.2	8.2	6.3	6.0	0	0
30 cm	1.36	0.09	0.43	4.8	9.4	148.3	22.8	49.0	0.7	4.6	2.1	7.5	6.6	6.0	0	0
40 cm	1.33	0.10	0.43	4.6	12.6	173.8	22.3	45.7	0.8	4.7	1.9	8.4	6.9	8.0	0	0

[1]Soil density; [2]Macroporosity; [3]Microporosity; [4]Total organic carbon; [5]Base saturation; [6]Litter dry mass; [7]Shoot dry mass; [8]Not sampled; [9]After the soybean harvest, no plants were growing in the area.

Table 2: Pearson correlation between soil basal respiration (C-CO_2) or microbial biomass carbon (MBC) and soil moisture, litter dry mass (LDM) and shoot dry mass (SDM), following the cattle grazing season (Nov/2014 and Nov/2015) or soybean harvest (May/2015 and May/2016), in a 15-year-old integrated soybean-beef cattle production system with different grazing heights (n=15).

Sampling period	C-CO_2			MBC		
	Soil moisture	LDM	SDM	Soil moisture	LDM	SDM
Nov/14	0.23	0.50*	0.77**	0.26	0.62**	0.35
May/15	0.63**	0.51*	-[1]	0.41	0.23	-
Nov/15	0.58*	0.70**	0.79***	0.75***	0.70**	0.77***
May/16	0.21	0.51*	-	0.47	0.45	-

[1] After the soybean harvest, no plants were growing in the area. *$p<0.05$; **$p<0.01$; ***$p<0.001$.

TITLE

Effect of long-term grazing intensification on epiedaphic faunal diversity in an integrated crop-livestock system**

AUTHORS

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ABSTRACT

Epiedaphic fauna is responsible for various key processes in the maintenance of soil and environmental quality. Intensification of grazing in integrated crop-livestock (ICL) systems may lead to a loss of diversity in soil organisms and compromise the sustainability of the system. This study aimed to identify the effects of 15 years of grazing intensification in an integrated soybean-beef cattle system on the diversity of epiedaphic fauna and to assess which edaphic conditions most affect the community of these organisms. The assessments were performed in an experiment conducted beginning in 2001 in a 23-ha area cropped with *Glycine max* for grain production in the summer and *Avena strigosa* + *Lolium multiflorum* for continuous cattle grazing in the winter. The treatments consisted of grass heights of 10, 20, 30, and 40 cm and controls without grazing. Sampling was performed in four collection periods from 2014 to 2016: two after grazing and two after soybean harvest. Epiedaphic faunal diversity was assessed using pitfall traps and chemical, physical, and soil cover variables for correlations. Grazing intensification for 15 years decreased the epiedaphic faunal abundance, richness, and diversity. The Collembola, Acari, Coleoptera, Araneae, Orthoptera, Dermaptera, and Hymenoptera groups were

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the most sensitive to the different grazing intensities. Vegetation cover and soil moisture were the factors that most affect the epiedaphic organisms in an integrated soybean-beef cattle system.

KEYWORDS: edaphic invertebrates; soil macrofauna; soil mesofauna; integrated crop-livestock system; soil physical quality.

1. INTRODUCTION

- Integrated crop-livestock (ICL) systems are based on the diversification, rotation, and combination and/or succession of crop and livestock production activities in farms (Kluthcouski et al. 1991). This management system, if applied correctly, brings benefits to both activities. The soil is exploited for nearly the entire year, which favors an increase in supplies of grain, meat, and/or milk and reduces production costs (Alvarenga and Noce 2005).
- ICL may improve the soil quality and resilience capacity, in addition to providing economic benefits (Salton et al. 2014). Studies have shown that this system can increase the carbon, nitrogen (Assmann et al. 2015), potassium, and phosphorus (Silva et al. 2014) cycling efficiency without damaging the soil physical quality when managed at moderate grazing intensity with grass heights of 20 to 30 cm (Cecagno et al. 2016). However, fewer research studies have focused on ICL biological attributes, particularly with long-term experimental protocols.
- Epiedaphic fauna consists of the invertebrates inhabiting the soil surface. The activities of these organisms improve agricultural productivity and environmental quality, resulting in the incorporation of soil surface residues, increased organic matter and plant nutrient availability, improved aggregation, biopore opening,

increased water infiltration and aeration, biological control, and other factors (Ferreira et al. 2007; Lavelle 1997; Rovedder et al. 2009). However, the management adopted in ICL may directly and intensely affect the epiedaphic fauna because these organisms are sensitive to soil chemical and physical changes and to changes in the vegetation and in the quantity and diversity of the residues deposited on the soil (Greenwood and McKenzie 2001; Souza et al. 2010).

- A problem commonly observed in ICL is the excessive stocking rate of grasslands, which results in reduced living plant and litter biomass availability (Kölbl et al. 2011), topsoil compaction and reduced water infiltration (Cecagno et al. 2016), and altered soil moisture and temperature regimes (Klein et al. 2005; Neves Neto et al. 2013), among other damage. Therefore, inadequate ICL management may offset its benefits and reduce soil biodiversity.

- Considering the importance of epiedaphic organisms to the sustainability of agro-ecosystems and the sensitivity of these organisms to management systems, this study tested the hypothesis that long-term grazing intensification in an ICL reduces the diversity of the main groups of soil invertebrates. Accordingly, we aimed to assess the effects of 15 years of grazing intensification in an integrated soybean-beef cattle production system on the diversity of the epiedaphic fauna and determine which edaphic conditions most affect the community of these organisms.

2. MATERIALS AND METHODS

2.1. Experimental site and procedures

- The study was performed as part of an experiment conducted beginning in 2001 in an area of approximately 23 ha by the Integrated Agricultural

Production System Research Group of the Federal University of Rio Grande do Sul (Universidade Federal do Rio Grande do Sul - UFRGS). The area is located in the municipality of São Miguel das Missões/ Rio Grande do Sul State/ Brazil (29°03'10" South, 53°50'44" West) and has a clayey soil (540 g kg^{-1} clay in the 0- to 20-cm layer) classified as Oxisol (Rhodic Hapludox - Soil Survey Staff 1999). The climate is subtropical, with a hot and humid summer (Cfa), according to the Köppen climate classification. Before 2001, the area was planted with black oat (*Avena strigosa* Schreb) in winters (only for soil cover) and soybean (*Glycine max* (L.) Merr.) in summers in a no-till crop production system. After the experiment had been established in the area, soybean planting was maintained for grain production in the summers (November to April), although the black oats + Italian ryegrass (*Lolium multiflorum* Lam.) were planted in the winters (May to October) for beef cattle grazing and remained thus until now.

- The grazing plots have variable sizes ranging from 0.9 to 3.6 ha. Continuous grazing was performed by three animals (of 200 kg initial mean live weight) per plot, with regulating animals being used to maintain the grass height at 10 cm (intense grazing), 20 cm (moderate grazing), 30 cm (moderate grazing), and 40 cm (light grazing). Plots without grazing were used as controls. The treatments were arranged in a completely randomized block design with three replicates.
- The fertilization, which followed the recommended norms and was based on the soil analysis values, consisted of grassland nitrogen fertilization and soybean phosphorus and potassium fertilization at doses for yields ranging from 4.0 and 7.0 t/ha of grassland dry matter and 4.0 t/ha of soybeans (CQFS RS/SC, 2004) (Assmann et al. 2015; Schuster et al. 2016).

2.2. Samplings and analyses

- Epiedaphic fauna was collected in four samplings: two conducted immediately after the cattle left the grassland (early November 2014 and 2015) and two immediately after the soybean harvest (early May 2015 and 2016). Nine pitfall traps were installed in each plot, totaling 27 per treatment. These traps were placed in the center of the plots located in the sampling grid. The grid consisted of three 30-m-long transects established 18 m apart from each other. Each transect had three collection points, totaling nine sample points per plot. The traps were treated with a 70% ethanol solution (v/v) and remained in the field for a 7-day period (11/05-11/11/2014; 03/29-04/6/2015; 11/1-11/7/2015; and 05/06-05/13/2016). After this period, the traps were transported to the laboratory for organism counts and identification to the Order level. The study was performed with the Authorization for Activities with Scientific Purposes (Autorização para Atividades com Finalidade Científica) number 4345-6 (Biodiversity Information and Authorization System (Sistema de Autorização e Informação em Biodiversidade - SISBIO)), issued by the Brazilian Ministry of the Environment (Ministério do Meio Ambiente do Brasil).
- In November 2014 and April 2015, soil samples were collected from the 0- to 10-cm layer in the same nine collection points of each plot, as described above, for physical and chemical analysis. The clay content (densimeter), total porosity, macroporosity, and microporosity according to a tension table (Embrapa 1997); soil density, using the volumetric ring method (Blake and Hartge 1986); and gravimetric moisture were measured to assess the soil physical traits. Total organic carbon (Walkley–Black); pH (water 1:1); P and K (Mehlich-1); Ca, Mg

and Al (KCl 1 mol L^{-1}); V (% base saturation); and H + Al (Toledo et al. 2012)were measured to assess the soil chemical traits (Table 1).

- Litter and plant shoot samples were collected from three points of the sampling grid. The plant material was dried in a conventional oven at 65°C to constant mass. Meteorological data on rainfall and air temperature were gathered for 2 months in each sampling period in a meteorological station installed in the experimental area (Figure 1).

2.3. Statistical analysis

- The abundance, richness, and diversity (Shannon) and the equitability (Pielou) indices of the epiedaphic organisms (Odum and Barrett 2007) were calculated using PAST software, version 2.17 (Palaeontological Statistics). Analysis of variance (ANOVA) was performed to compare the means of abundance and richness data, and the Duncan test (5% probability) was used when significant differences existed, using SASM-agri software (version 3.2.4). The frequency of each group was calculated in the different treatments. The organisms were grouped as "Others" when the means reached values of less than 1% in all treatments. Redundancy Analysis (RDA) was performed using the statistical software CANOCO 4.5 (ter Braak and Smilauer, 1998) to assess the effect of significant environmental (soil chemical, physical, and cover) variables on the epiedaphic community in the period after grazing and in one sampling period after the soybean harvest.

3. RESULTS

- A total of 65,672 individuals, divided into 20 taxonomic groups, were quantified from both samplings performed after grazing. The total number of individuals collected in both samplings performed after the soybean harvest was 54,274, belonging to 24 groups. Thus, the results showed greater taxon richness in the collections performed after the soybean harvest and a higher average number of individuals per treatment in the sample collections performed after grazing.

- Although this is a subtropical climate region (Cfa), with marked climatic variation between the four seasons, the conditions observed in the four sample collections were relatively stable (Figure 1). The average air temperature was only 3°C higher in collections performed after grazing than in collections performed after the soybean harvest. This condition was repeated in both sampling years. The rainfall was more variable, although the soil moisture showed no significant changes between the four collections (averaging 27.5, 26.9, 25.4, and 24.5% in the four collections; Table 1).
- The cattle grazing intensity affected the abundance of the epiedaphic organisms (Table 2). The collections performed after grazing showed that the higher the grazing intensity is, the lower the abundance of these organisms will be. In intense grazing, the abundance was approximately 40% lower than that of the treatment without grazing, which resulted in a significant difference. No significant differences occurred between the other treatments and the control, despite 20, 30, and 10% mean abundance reductions in treatments with the 20-, 30-, and 40-cm grazing heights, respectively. In the collections performed after the soybean harvest (Table 2), the number of organisms quantified in the treatment without grazing was 30% higher than the mean of the treatments with grazing, albeit without significant differences between treatments, given the wide variation in abundance between the years.
- Damage to the soil invertebrate diversity from the increased grazing intensity was also shown by the decreased group richness (Table 2). After the grazing

period, an increased richness was observed in light grazing, without significant differences from the treatments with moderate grazing and without grazing (Table 2). Intense grazing resulted in the lowest richness and significantly differed from light grazing. In collections performed after the soybean harvest, the highest richness was observed in the treatments without grazing and with light grazing (Table 2). Conversely, the treatment with the lowest diversity of epiedaphic organisms was grazing to a grass height of 20 cm, which showed no significant difference from the treatments at the 10- and 30-cm grass heights.

- The diversity indices indicate the same trend observed in abundance and richness (Table 2). The assessments performed after grazing showed an increased diversity of soil organisms in treatments without grazing and with grazing to a grass height of 30 and 40 cm. Damage to the soil ecological balance occurred in the treatments with grazing performed at higher intensities (10 and 20 cm), which resulted in lower diversity indices. The collections performed after soybean cultivation also showed an increased biodiversity of organisms in the treatment without grazing and decreased diversity in the grazing treatments.
- Acari, Araneae, Coleoptera, Collembola, Diptera, Hymenoptera, Hemiptera, Orthoptera, Dermaptera, and Larva (unidentified larvae) were the epiedaphic taxonomic groups most commonly found in the experimental area. These groups were mostly sensitive to changes caused by grazing intensification and showed changes in frequency with the grazing intensification. The least frequent organisms were grouped into “Others” and belonged to the Annelida, Chilopoda, Diplopoda, Isoptera, Blattodea, Thysanoptera, Lepidoptera, Neuroptera, Mollusca, Opilione, Scorpiones, Nematoda, and Odonata taxa.

- The mesofaunal members were the groups most commonly found in both assessment periods, and Collembola and Acari accounted for more than 60% of the organisms collected in nearly all the treatments (Figure 2). In the collection performed after grazing, Collembola maintained nearly the same frequency in all treatments, whereas the mites had a higher frequency in grazing to a height of 20 cm. However, in the collection performed after the soybean harvest, the frequency of mites tended to increase, and the frequency of Collembola tended to decrease with the grazing intensity.

- Coleoptera were the most commonly found epiedaphic macrofaunal organisms (Figure 2). The frequency of this group decreased at the lowest grass heights, in collections performed after both grazing and the soybean harvest. On average, the frequency of Coleoptera was 18% in the control and the lower grazing intensities and 10% with grazing to grass heights of 10 and 20 cm. Only the Hymenoptera group showed an increase in frequency with the grazing intensity in the collection performed after cattle grazing. This is most likely associated with a more disturbed habitat because these organisms may indicate environmental imbalances. Similar to the Coleoptera group, the frequency of organisms of the Araneae, Orthoptera, and Dermaptera orders decreased with grazing to the lowest grass heights, thus indicating that grazing intensification affects several groups of edaphic fauna.

- The comparison between collection periods shows that soybean cropping decreased the macrofauna frequency and increased the mesofauna frequency, mainly due to the increase in mites (Figure 2). On average, macroorganisms accounted for 36% of the organisms collected after cattle grazing, whereas this percentage decreased to 23% in the collection performed after the soybean harvest. Various non-edaphic, adult individuals of the Diptera order were captured in the four collections. However, their presence was associated with attraction to the trap ethanol solution. Therefore, these data were disregarded.

- The matrix of the soil chemical and cover variables explained 22% of the variation in the epiedaphic fauna data (*p*=0.002), whereas the matrix of soil physical variables explained 10% of this variation (*p*=0.01) in the redundancy analysis of the collection performed after grazing. The exclusive percentage effect of each significant variable was 10% for the shoot dry matter, 6% for the pH, 3% for the calcium, 6% for the moisture, and 3% for the soil density. Conversely, the matrix of the soil chemical and cover variables only explained 5% of the variation in the data for the epiedaphic fauna (*p*=0.05), and the matrix of the soil physical variables explained 36% of this variation (*p*=0.001) in the collection performed after the soybean harvest. In this case, an exclusive percentage effect of each significant variable was 5% for pH, 31% for moisture, and 6% for microporosity.

4. DISCUSSION

- Grazing intensification for 15 years significantly decreased the epiedaphic faunal diversity. In general, the control (without grazing) and the low-intensity grazing treatments had the highest abundance and richness and the best diversity indices. Low soil invertebrate diversity was associated with high-intensity grazing treatments. Grasslands managed sustainably have microhabitats with improved edaphic conditions, increased plant biomass cover (Gill 2007), improved physical structure (Conte et al. 2007), increased residue input (Bayer et al. 2009), and increased soil organic matter (Rosenzweig et al. 2016). Furthermore, the vegetation height in the grazing areas and the invertebrate abundance are positively correlated (*p*=0.009) because a more structured and complex vegetation cover results in better food and habitat conditions (Hoste-Danyłow et al. 2010). Conversely, grazing managed intensively may lead to a decreased abundance in organisms from key edaphic arthropod orders due to habitat simplification (Little et al. 2013; Swengel 2001).

- The frequency of the edaphic mesofaunal organisms (Collembola and Acari) increased in both assessment periods. These organisms are the most abundant and widespread arthropods in most soils (Bedano et al. 2011). Their activity improves soil quality and plant yield because they are highly active in soil organic matter decomposition, nutrient cycling, and biological pest control (Moreira et al. 2010). In both sampling periods, the mites were frequent at the 20-cm grass height. This result may indicate edaphic mite preference for areas with high bovine manure availability and good soil plant cover. These conditions are found at the 20-cm grass height. The plots with the lowest grass heights are also the smallest and therefore have a higher input of manure because the number of animals is the same in all plots. According to Silva et al. (2014), in this same experimental protocol, the manure dry mass production in a grazing season was 669, 478, 366, and 213 kg ha^{-1} for the 10-, 20-, 30-, and 40-cm grass height treatments, respectively.
- Collembola was present at a higher frequency than mites were in the period after grazing in all treatments. This relation indicates a slightly disturbed environment (Mateos 1992). However, the collection performed after the soybean harvest showed that the plots with an increased frequency of mites also showed a decreased frequency of Collembola, which also has been reported in other studies (Rieff et al. 2014; Rieff et al. 2016). This behavior may

be associated with predation because some groups of mites are microarthropod predators (Mejía-Recamier et al. 2013).

- The Coleoptera order is abundant in most Brazilian soils and is extremely important for the soil chemical and physical quality (Correia and Oliveira 2005; Pompeo et al. 2016; Portilho et al. 2011). The results from the present study show the importance of an adequate grassland management for ICL sustainability. Coleoptera were more commonly found at low grazing intensities and in sample collections performed after grazing, regardless of the sampling period. All results show that most living or dead plant cover provides a more suitable habitat for the survival of these organisms (Pompeo et al. 2016; Rosa et al. 2015). These effects may be direct, through increased food and shelter availability, or indirect, through improved chemical and physical conditions, as outlined in Table 1.

- Organisms of the Araneae, Dermaptera, and Orthoptera Orders were also adversely affected by the increase in grazing intensity. Spiders are the most diverse and abundant arthropod predators in grazing ecosystems (Horváth et al. 2009). Therefore, they have a key role in community structure and natural pest control (Sunderland and Samu 2000). The abundance and diversity of these organisms are directly associated with the vegetation vertical structure and height (Bell et al. 2001; Harris et al. 2003; Horváth et al. 2009). Some Dermaptera groups are important for crop production because they contribute to biological pest control (Buzzi 2013). Conversely, Orthoptera are important in grazing areas because they account for most of the biomass of the edaphic invertebrates in those systems (Little et al. 2013).

- Hymenoptera (ants) are one of the bioindicators of disturbed environments most studied in the literature (Pereira et al. 2007; Rocha et al. 2015). A study conducted in the Brazilian Midwest showed that ICL may favor the ant community, depending on the management adopted, and they may be used as bioindicators in this type of soil management (Crepaldi et al. 2014). In the collection performed after grazing, the frequency of these organisms was highest in the treatment with 10 cm of grass height, which is the treatment with the most disturbed edaphic environment.
- Redundancy analysis indicated that soil moisture was a key factor in the composition of the community of epiedaphic organisms (Figure 3). Soil moisture was higher in treatments without grazing and with moderate and light grazing in both times. This performance may result from the improved edaphic conditions observed in these treatments due to increased soil compaction and vegetation and litter mass in the area. Intense shoot biomass extraction, as observed in intense grazing, converts the soil into an environment less protected from the high levels of solar radiation and therefore with increased evaporation (Gong et al. 2014).
- Increased soil density and decreased porosity, as observed in intense grazing, limit the activity of these gallery-builder organisms and the abundance of those requiring porous soil space to survive (Moço 2006). Furthermore, the area fails to provide quality habitats, resources, and shelter to support the high diversity of edaphic organisms, given its decreased soil cover (Bedano et al. 2016). The damage caused by 15 years of intense grazing (10 cm) to the soil physical quality severely affected the epiedaphic fauna. Such losses were not even

offset by the bovine manure input per area in intense grazing, which was three times higher (669 kg ha^{-1}) than that in light grazing (213 kg ha^{-1}).

- The pH and calcium content were the chemical factors that most affected the epiedaphic fauna. The pH adversely affected the epiedaphic fauna in both sample collection times. This characteristic is important to establish specific groups, including ants (Jacquemin et al. 2012), and is usually inversely correlated with the soil abundance of the organisms (Harada and Bandeira 1994; Nowak 2001). In the sample collection performed after grazing, the calcium content positively affected the distribution of the edaphic organisms, which may be attributed to the importance of this nutrient for the physiology of some groups of invertebrates because calcium is associated with mechanisms of osmotic regulation and ecdysis (Rosa et al. 2015). Soil physical, chemical, and cover attributes, changed by the different grazing intensities, significantly affected the epiedaphic organisms, which was also observed in other studies assessing different management systems (Bartz et al. 2014; Souza et al. 2016)

5. CONCLUSIONS

- Grazing intensification for 15 years reduces the epiedaphic faunal abundance, richness, and diversity in an integrated soybean-beef cattle production system.
- The Collembola, Acari, Coleoptera, Araneae, Orthoptera, Dermaptera, and Hymenoptera groups are the most sensitive to the different grazing intensities.
- Plant cover and soil moisture are the factors that most affect the epiedaphic organisms in an integrated soybean-beef cattle production system.

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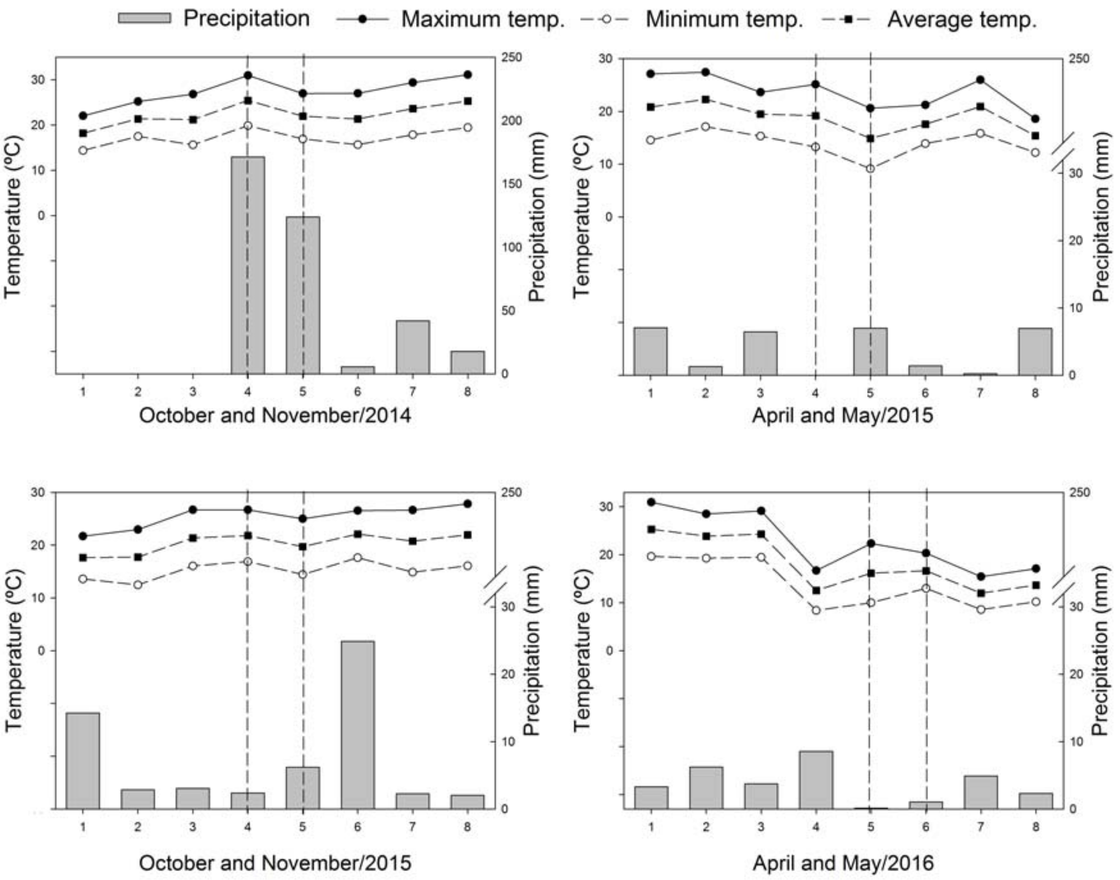

Figure 1: Precipitation and minimum, mean, and maximum air temperatures in the collection periods. The period between dotted lines indicates the days on which the traps remained in the field.

Table 1: Physical and chemical attributes of the 0- to 10-cm soil and plant cover layer in the treatments with different grass heights or without grazing (WG), in collections performed after cattle grazing and soybean harvest, in a 15-year integrated soybean‑beef cattle production system. The data are expressed as the means of five replicates per treatment.

	Physical variables			**Chemical variables**											**Plant cover variables**			
Treatment	**SD[1]**	**Ma[2]**	**Mi[3]**	**Moisture**		**pH**	**P**	**K**	**TOC[4]**	**V[5]**	**Al**	**Ca**	**Mg**	**H+Al**	**LDM[6]**		**SDM[7]**	
	g cm^{-3}	cm^3 cm^{-3}		%			mg dm^{-3}		g kg^{-1}	%		$cmol_c$ dm^{-3}			Mg ha^{-1}			
				2014	2015			After grazing							2014	2015	2014	2015
WG	1.26	0.13	0.42	28.9	28.9	4.3	13.1	219.3	-[8]	27.1	1.4	3	1.2	13.6	6.6	5.1	4.6	7.3
10 cm	1.40	0.09	0.40	25.9	23.7	4.8	9.0	187.4	-	48.5	0.8	4.1	1.7	6.9	1.3	1.0	1.8	0.9
20 cm	1.37	0.09	0.41	27.4	24.6	4.9	10.7	200.4	-	50.6	0.6	4	1.8	6.4	3.0	1.8	3.3	2.6
30 cm	1.33	0.12	0.42	28.0	24.6	5.0	9.2	163.4	-	52.5	0.6	4.5	1.9	6.4	3.1	2.1	3.4	3.3
40 cm	1.32	0.14	0.38	27.3	25.4	4.8	13.6	200.6	-	48.6	0.5	4.4	1.8	7.2	3.5	4.0	4.8	4.1
				2015	2016			After soybean harvest							2015	2016	2015	2016
WG	1.34	0.09	0.45	28.4	26.4	4.3	19.5	222.6	23.1	36.4	1.3	3.9	1.5	11.2	9.2	8.5	0[9]	0
10 cm	1.37	0.09	0.41	25.5	23.9	4.6	9.7	136.3	22.2	44.6	1.1	4.3	2.0	8.5	4.1	4.3	0	0
20 cm	1.36	0.11	0.41	26.6	24.4	4.7	13.6	181.2	23.3	47.5	1.0	4.7	2.2	8.2	6.3	6.0	0	0
30 cm	1.36	0.09	0.43	27.0	23.9	4.8	9.4	148.3	22.8	49.0	0.7	4.6	2.1	7.5	6.6	6.0	0	0
40 cm	1.33	0.10	0.43	27.0	24.0	4.6	12.6	173.8	22.3	45.7	0.8	4.7	1.9	8.4	6.9	8.0	0	0

[1] Soil density; [2] Macroporosity; [3] Microporosity; [4] Total organic carbon; [5] Base saturation; [6] Litter dry mass; [7] Shoot dry matter; [8] Data not shown; [9] No growing plants remained after the soybean harvest.

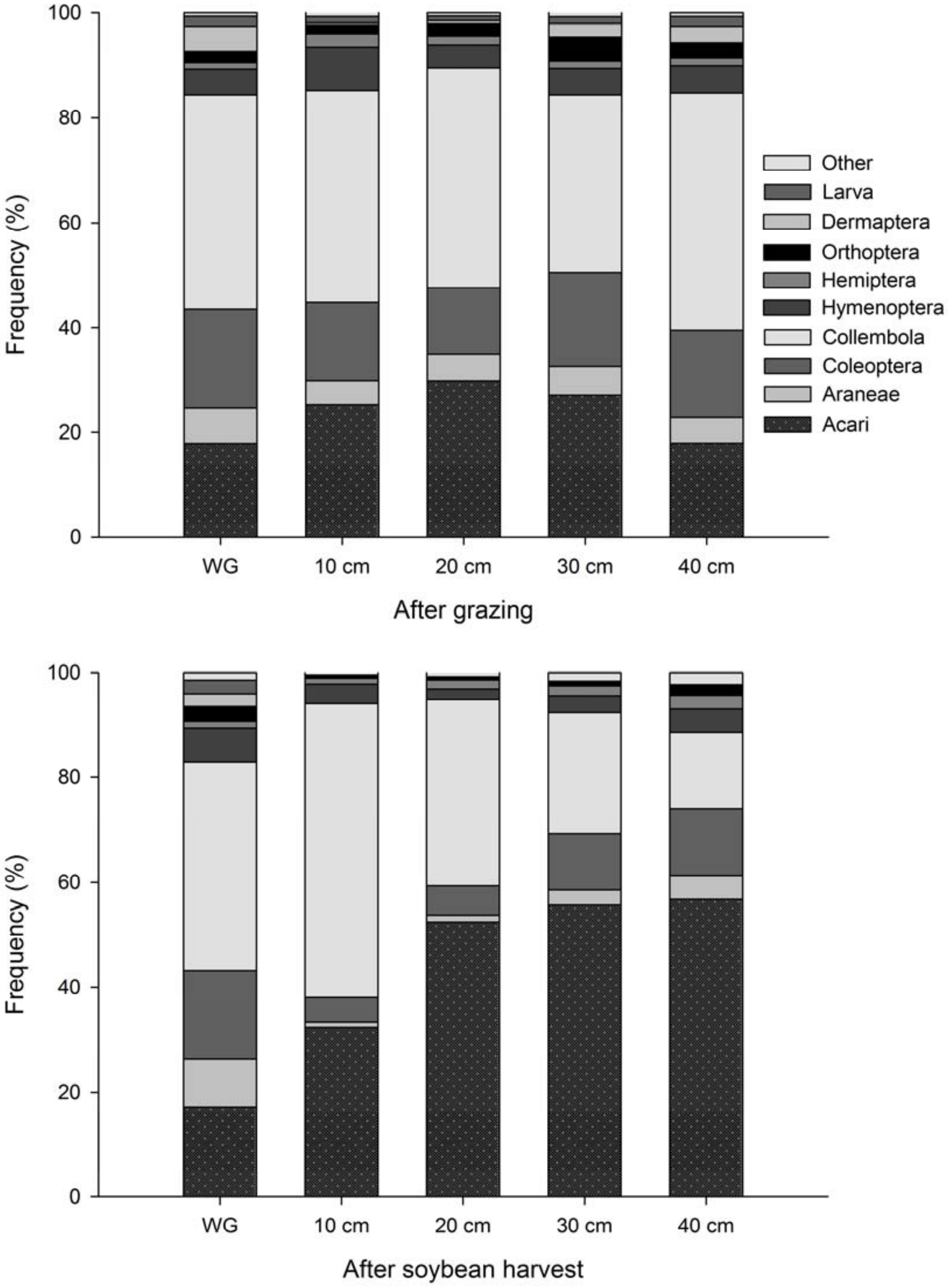

Figure 2: Relative frequency of the epiedaphic fauna groups after cattle grazing (November 2014 and 2015) and after the soybean harvest (May 2015 and 2016) in the treatments with different grass heights or without grazing (WG) in a 15-year integrated soybean‑beef cattle production system. The data are expressed as the means of 27 replicates per treatment.

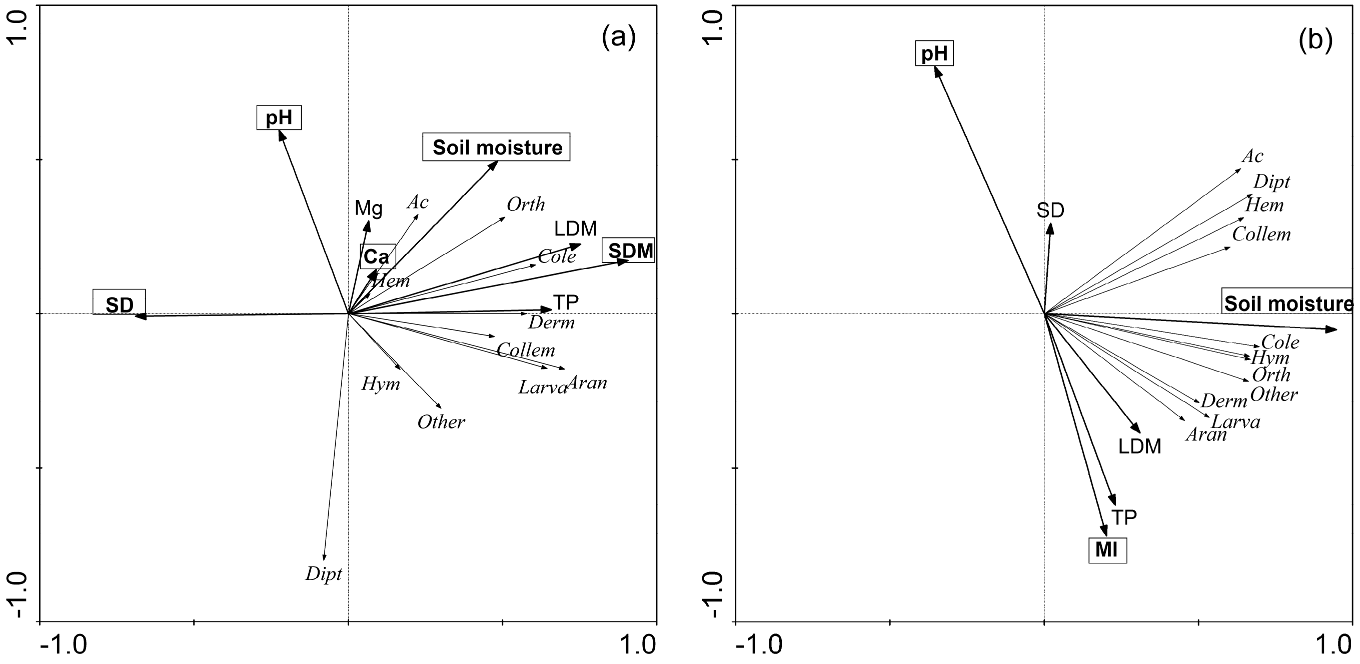

Figure 3: Redundancy analysis (RDA) after cattle grazing (November 2014 and 2015; a) and after the soybean harvest (May 2015 and 2016; b) in a 15-year integrated soybean-beef cattle production system managed with grass heights of 10, 20, 30, and 40 cm or without grazing. The variables with significant responses are highlighted with a box. Ac (Acari); Dipt (Diptera); Hem (Hemiptera); Collem (Collembola); Cole (Coleoptera); Hym (Hymenoptera); Orth (Orthoptera); Derm (Dermaptera); Aran (Araneae); Larva (unidentified larvae); Others (sum of the less frequent groups); pH; SD (soil density); TP (total soil porosity); MI (soil microporosity); TOC (total organic carbon); Ca (calcium); Mg (magnesium); LDM (litter dry mass); and SDM (shoot dry mass). The data refer to 27 replicates per treatment.

Table 2: Abundance, richness, and diversity indices of epiedaphic fauna collected after cattle grazing and soybean harvest, in the treatments with different grass heights or without grazing (WG), in a 15-year integrated soybean‑beef cattle production system. The data are expressed as the means of 27 replicates per treatment.

Variable/ Index	Treatments				
	WG	10 cm	20 cm	30 cm	40 cm
			After grazing		
Abundance	361a	224b	283ab	256ab	324ab
Richness	12ab	11b	12ab	12ab	13a
Shannon	1.83	1.69	1.69	1.85	1.82
Pielou	0.73	0.69	0.67	0.74	0.71
			After soybean		
Abundance	173ns	156	146	93	89
Richness	12a	10ab	9b	10ab	12a
Shannon	1.67	1.43	1.38	1.42	1.38
Pielou	0.68	0.62	0.63	0.62	0.56

5 DISCUSSÃO GERAL

Os dados obtidos neste estudo resultaram de quatro amostragens realizadas ao longo de dois anos, em um experimento manejado com sistema integrado de produção agropecuária conduzido há 15 anos sob diferentes pressões de pastejo, em uma área de 23 hectares. Este trabalho é suportado por um conjunto muito grande de dados de solo, planta, animal e meteorológicos coletados ininterruptamente neste período. Constitui-se, portanto, numa excelente oportunidade para o estudo da biota do solo neste tipo de sistema, visando reduzir a grande carência de informações sobre o tema a nível mundial.

O modo produtivo atualmente utilizado por grande parte dos agricultores no sul do Brasil é um sistema puramente agrícola. No inverno, 86% área agrícola é cultivada somente com plantas de cobertura para a produção de palha para o plantio direto da soja no verão (CONAB, 2011). Este modo produtivo concentra na soja toda a renda do agricultor, o que é um grande risco, tendo em vista as instabilidades meteorológicas e mercadológicas. Por outro lado, as pastagens de invernos de aveia e azevém são altamente produtivas e podem ser utilizadas para o pastejo animal e ainda produzirem biomassa vegetal suficiente para o plantio direto do verão (KICHEL e MIRANDA, 2006; VILELA et al., 2008).

Frente a este contexto, a introdução do pastejo bovino no inverno representa a busca por aumento e estabilidade de renda aos agricultores, com intensificação uso do solo (BERRY et al., 2003). Porém frequentemente questiona-se se a introdução do pastejo pode prejudicar o solo. Para os organismos do solo, o pastejo pode aumentar sua diversidade, atividade e biomassa, pois há introdução de maior quantidade e diversidade de resíduos através da urina e esterco incorporados pelos animais (CLEGG, 2006; LIN et al., 2009). Além disso, a atividade e o crescimento radicular das forrageiras são incentivados pela desfolhação provocada pelo pastejo, o que gera um incremento na taxa de rotatividade de rizodeposições e decomposição de raízes finas (HEWINS et al., 2016; PAPATHEODOROU et al., 2008). Por outro lado, o pastejo pode prejudicar a atividade biológica, pois promove compactação da camada superficial do solo, reduz a porosidade e por consequência a aeração e a infiltração de água, retira parte da biomassa vegetal, o que altera a temperatura e a umidade do solo (JIA et al., 2006; SOUTO et al., 2008).

Preocupados com a sustentabilidade dos SIPAs, buscou-se neste trabalho conhecer as implicações das diferentes alturas de pastejo dos bovinos sobre a comunidade de organismos edáficos. Foram avaliadas a atividade e a biomassa microbiana através da taxa de respiração basal do solo e do carbono da biomassa microbiana, e a diversidade de invertebrados epiedáficos. Estes parâmetros foram escolhidos por serem bioindicadores de alta sensibilidade da qualidade do ambiente e porque uma alta atividade biológica do solo é requisito para a sustentabilidade de qualquer sistema produtivo (BARETTA et al., 2014; CLUZEAU et al., 2012; PONGE et al., 2013). Nos solos biologicamente ativos, os processos de degradação dos resíduos orgânicos, mineralização dos nutrientes, formação da matéria orgânica do solo, agregação do solo, controle biológico, produção de fitohormônios, etc., ocorrem em uma magnitude satisfatória, contribuindo para a aumentar a produtividade e preservar o ambiente(BARTZ et al., 2014; SHARKHUU et al., 2016).

Para este trabalho, tanto os parâmetros microbiológicos quanto a diversidade da comunidade epiedáfica responderam aos tratamentos aplicados. Dos atributos do solo, a umidade foi determinante para a composição e atividade biológica no solo, assim como a cobertura vegetal. Quanto maior a pressão de pastejo aplicada, menor foi a cobertura de biomassa vegetal, a serapilheira e a umidade do solo. Nestes tratamentos de pastejo mais intenso foram observadas a menor taxa de respiração basal e o menor teor de carbono na biomassa da comunidade microbiana, assim como uma menor diversidade da comunidade epiedáfica. O pastejo manejado de forma intensiva pode levar a um decréscimo da abundância e atividade da biota do solo devido a simplificação de hábitat (LITTLE et al., 2013; PERI et al., 2015; SWENGEL, 2001), o que pode acarretar em prejuízos importantes para a qualidade do solo.

As avaliações microbiológicas mostraram que tanto a taxa de respiração basal do solo, quanto o conteúdo de carbono da biomassa responderam às intensidades de pastejo e aos distúrbios ocasionados por ela. Quanto maior foi a pressão de pastejo aplicada, menor foi a atividade microbiana. Desta forma, estes atributos podem ser utilizados como bioindicadores de qualidade do solo, assim como observado em outros estudos avaliando diversos indicadores microbiológicos em outros sistemas agrícolas (FERREIRA et al., 2011; LISBOA et al., 2012; MOESKOPS et al., 2012; MOSCATELLI et al., 2012; VASCONCELLOS et al., 2013). Da mesma forma alguns grupos integrantes da fauna epiedáfica do solo também podem ser utilizados como bioindicadores da qualidade do SIPA (BARETTA et al., 2014). Os ácaros, colêmbolos, coleópteros, aranhas e ortópteras se mostraram sensíveis às pressões de pastejo aplicadas, sendo assim, adequados ao uso como bioindicadores neste sistema.

Nas maiores alturas de pastagem os prejuízos causados pelo pastejo bovino aos organismos do solo são minimizados. A maior biomassa vegetal viva ou morta nestes tratamentos proporciona melhores condições físicas do solo, que resulta em maior umidade do solo e por consequência maior atividade, biomassa e biodiversidade dos organismos. Estes resultados reforçam a necessidade de um ajuste adequado da carga animal na pastagem para que não ocorra comprometimento à sustentabilidade dos SIPAs.

6 CONCLUSÃO GERAL

A partir dos resultados obtidos nos dois estudos conduzidos em um SIPA de longa duração pode-se concluir que:

Há maior respiração microbiana e maior conteúdo de carbono na biomassa microbiana do solo no tratamento sem pastejo e quando o pastejo ocorre nas maiores alturas da pastagem;

Na menor altura da pastagem há prejuízos aos atributos físicos do solo, o que resulta em menor teor de umidade, e redução da atividade e da biomassa dos microrganismos;

A intensificação do pastejo reduz a abundância e a diversidade da fauna epiedáfica;

A perda da qualidade física do solo sob pastejo intenso resulta em menor teor de umidade e este fator é o que mais contribui para a redução da abundância e da riqueza dos meso e macrorganismos;

Os atributos biológicos do solo avaliados no presente estudo podem ser utilizados como bioindicadores de qualidade do solo em um SIPA de longa duração.

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