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SOIL BETA-GLYCOSIDASE ENZYME ACTIVITY MONITORING FROM DIFFERENT PASTURELAND MANAGEMENT SYSTEMS

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Abstract
Organic carbon is essential for soil health, and conservation agriculture can minimize the negative effects of conventional practices on microbial communities and enzymatic activities, benefiting nutrient cycling and increasing soil organic matter. The enzyme beta-glucosidase (BG) stands out as a bioindicator of changes in the carbon cycle and is used to assess soil quality and sustainability. This study analyzed BG concentrations in the soil in different long-term pasture systems (14 years) at Embrapa Pecuária Sudeste, in São Carlos/SP, namely: crop-livestock (ILP), containing crops and pastures with cattle; intensive areas (INT), containing pastures with cattle; extensive pastures (EXT) with cattle, with indicative of some soil degradation; and native forest area (FLO). Higher BG values (average of 155.92 µg of p-nitrophenol/g of soil) were detected in ILP areas, indicating a higher concentration of enzymatic activity, followed by INT (average of 105.64 µg). ILP and INT have higher BG values than EXT areas, which have the lowest value of all the areas (63.17 µg). This concludes the importance of management in integrated areas, leading to better soil quality.

Keywords: enzymatic activity; beta-glucosidase, organic matter; soil health; UV-Visible light spectroscopy.

MONITORAMENTO DA ATIVIDADE DA ENZIMA BETA-GLICOSIDASE DO SOLO EM DIFERENTES SISTEMAS DE MANEJO DE PASTAGENS

Resumo
O carbono orgânico é essencial para a saúde do solo, e a agricultura de conservação pode minimizar os efeitos negativos das práticas convencionais sobre as comunidades microbianas e atividades enzimáticas, beneficiando a ciclagem de nutrientes e aumentando a matéria orgânica do solo. A enzima beta-glicosidase (BG) destaca-se como bioindicador de mudanças no ciclo do carbono e é utilizada para avaliar a qualidade e a

sustentabilidade do solo. Este estudo analisou as concentrações de BG no solo em diferentes sistemas de pastagem de longo prazo (14 anos) na Embrapa Pecuária Sudeste, em São Carlos/SP, sendo: lavoura-pecuária (ILP), contendo lavoura e pastagens com bovinos; áreas intensivas (INT), contendo pastagens com bovinos; pastagens extensivas (EXT) com bovinos, com indicação de degradação do solo; e área de floresta nativa (FLO). Foram detectados maiores valores de BG (média de 155, 92 µg de p-nitrofenol/g de solo) em áreas de ILP, indicando uma maior concentração de atividade enzimática e, seguidamente, INT (média de 105,64 µg). ILP e INT apresentam valores superiores de BG do que em áreas EXT, sendo estas as que têm menor valor entre todas as áreas (63,17 µg). Assim, conclui-se a importância do manejo em áreas integradas, propiciando uma maior qualidade do solo.

Palavras-chave: atividade enzimática; beta-glicosidase, matéria orgânica; saúde do solo; espectroscopia de luz UV-Visível.

1. Introduction

In the soil, organic matter (OM), rich in organic carbon, is a vital source of nutrients to maintain vitality and is an excellent indicator of soil quality. However, there is a decrease in carbon content in conventional soil management, compromising the productivity of the crop or production system, as well as the degradation of the soil microbial community, which plays an essential role in the decomposition of organic matter and nutrient cycling. However, soil conservation practices, such as the use of no-till farming and sustainable intensification, have shown benefits, such as increased soil OM and increased productivity of grains and animal production systems (Oliveira et al., 2024; Bento et al., 2025; Sá et al., 2025). Soil microorganisms produce enzymes that catalyze various biochemical reactions and are crucial in several biogeochemical cycles. Thus, when exposed to inadequate practices, soil disturbance and reduced OM, these microorganisms can suffer a drastic reduction in their activities, directly affecting the health and quality of the soil. In general, BG is responsible for degrading cellulose, one of the main components of plant organic matter, and for releasing glucose into the soil, acting in the carbon cycle. Due to its importance and its sensitivity as an indicator of changes in soil management, this study evaluated how the BG enzyme behaves, using UV-Visible light spectroscopy monitoring, in soils with animal production systems with pastures under different management and native forest, with the aim of assessing the quality and sustainability of the production system.

2. Materials and Methods

We analyzed 7 areas of different Crop-Livestock Integration (CLI) systems, intensive pasture (INT), extensive pasture (EXT) and native forest (FLO), in an area of native Cerradão (transition between Atlantic Forest and Cerrado) on the Canchim Farm - Embrapa Pecuária Sudeste, São Carlos/SP, with an experiment set up in 2011 and with soil samples collected in 2025, thus lasting 14 years. The samples were collected at a depth of 0-10 cm and dried in the open air, standardized to 2 mm. In each area, 6 points were collected and the analysis was carried out in triplicate for each point. This resulted in a total of 18 samples for each area analyzed, giving a total of 126 samples for this study. Beta-glucosidase (BG) activity was analyzed according to Tabatabai et al., 1994. This procedure is in line with the BioAS protocol (Mendes et al., 2021) and the descriptions available for grassland soils in Santos et al., 2022. To quantify BG activity, 4 mL of MUB universal buffer solution at pH 6.0 and 0.25 mL of p-nitrophenyl-β-D-glycopyranoside (PNG) 0.05 mol L-1 were added to one gram of soil as a substrate. The mixtures were

incubated at 37ºC for 1 hour. After the incubation time, the reaction was stopped by adding 1 mL of CaCl2 0.5 mol L-1. The control was differentiated by the addition of the MUB buffer solution to replace the substrate. Next, 4 mL of 0.1 mol L-1 tris hydroxymethyl aminomethane (THAM) buffer solution was added at pH 12.0 and the soil suspension was filtered through Whatman No. 2 filter paper. The intensity of the yellow color of the p-nitrophenol formed from the reaction was measured using a UV-Vis absorption spectrophotometer (UV-1601PC, Shimadzu) at 420 nm. Thus, the enzymatic activity was expressed as µg p-nitrophenol g-1 soil h-1 (µg of p-nitrophenol released per gram of dry soil) and estimated based on a standard curve prepared with known concentrations of p-nitrophenol. The data was statistically analyzed using descriptive measures, such as the arithmetic mean and standard deviation, in order to summarize the distribution of the results. Student's t-test was used to compare the two experimental groups, assuming a significance level of 5% ($p < 0.05$).

3. Results and Discussion

The results of BG enzyme activity in the soil revealed significant variations between the different animal production systems and native forest (FLO), reflecting the impact of management on the biological quality of the soil (figure 1).

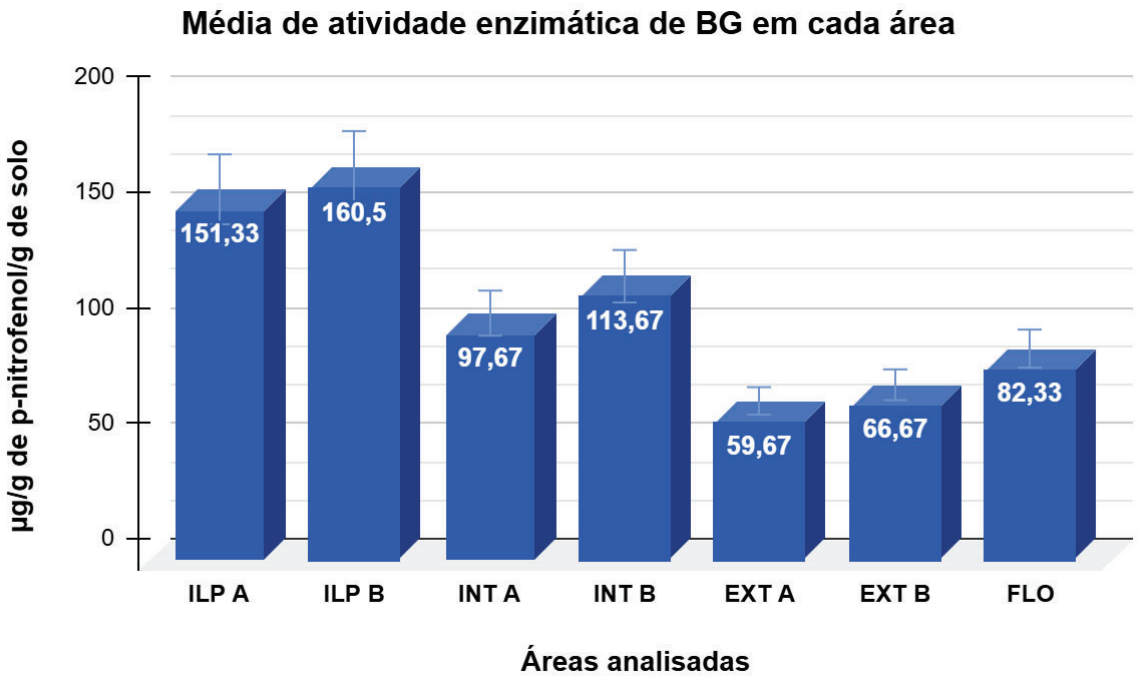

Figure 1: Average values of the enzymatic activity of the enzyme beta-glucosidase in each area analyzed (ILP - Integration of Crop and Livestock in areas A and B; INT - Intensive Pasture in areas A and B; EXT - Extensive Pasture in areas A and B and FLO - Forest) obtained by measuring absorbance at 420 nm with UV-Vis light spectroscopy.

The ILP area had the highest BG activity, with an average value of 155.92 µg p-nitrophenol/g soil of the two areas analyzed (A and B), indicating a soil with high microbial activity and carbon cycling. In contrast, the EXT area showed the lowest activity, with an average value of 63.17 µg p-nitrophenol/g soil, suggesting soil degradation and low organic matter renewal, which leads to low microbial activity. The INT pasture showed an average value of 105.64 µg of p-nitrophenol/g of soil, benefiting from more controlled

management practices, while FLO detected a value of 82.33 µg of p-nitrophenol/g of soil, reflecting a natural ecological balance of soil organic matter. The results on soil carbon stocks in the areas analyzed were partially published with soil samples collected in 2016. Bieluczyk et al., 2020, identified that the greatest growth in soil carbon stock, up to 40 cm, in the period from 2011 to 2016 occurred in the ILP system area, with a value of 1.96 ton C/ha.year, while the EXT pasture showed growth of 1.74 ton C/ha.year. Oliveira et al., 2024 measured carbon stocks at a depth of 1m, also for soil samples collected in 2016, and identified the following: ILP system - 103.52 ton C/ha and EXT pasture system - 99.08 ton C/ha. Thus, it can be seen that the ILP system had a higher rate of carbon accumulation in the soil than the EXT pasture, which was the lowest of all, coinciding with the BG values obtained. Thus, based on the data obtained in this work and other published data from the areas analyzed, the hypotheses to explain the differences are: in ILP management there was a significant increase in soil C stock (Oliveira et al., 2024; Bieluczyk et al., 2020) and a greater diversity of organic matter input with organic residues from pastures, agricultural crops and animal waste, which increased the amount of particulate organic matter (Bieluczyk et al., 2020); in the INT pasture, in general, there is a significant recent accumulation of organic matter due to the increase in biomass produced by the pasture as a result of soil fertilization, but with less diversity compared to the ILP system; in the EXT pasture area there is a lower C stock value in the soil (Oliveira et al., 2024 and Bieluczyk et al, 2020), with lower organic matter input and, consequently, low microbial activity, as well as some level of physical-chemical degradation of the soil; in the native forest (FLO), nutrient cycling tends to be efficient, although it may be slower and with lower enzymatic activity compared to intensive agricultural systems due to the presence of more recalcitrant organic material, reaching a higher value than EXT, but lower than INT and ILP. Despite this, microbial and enzymatic activity is sustained by the rich biodiversity of the soil and the adaptation of microorganisms to the natural environment. One of the few results available in the literature with pastures evaluated using the BioAS protocol is that of Santos et al., 2022, which was carried out with another long-term experiment at different levels of pasture intensification, also at Embrapa Pecuária Sudeste, with and without irrigation, and with different animal stocking rates, and with measurements of the soil's physicochemical properties, enzyme activities (BG and ARIL) and microbial biomass. In this experiment, higher BG values were also observed in pastures with dryland management, with soil fertilization and with moderate animal stocking, even when compared to the enzymatic activity for BG in native forest, while the reference area with extensive grazing showed the lowest level of BG. Therefore, the result presented here is consistent with that of Santos et al., 2022. This work is ongoing and will carry out further assessments, within the BioAS protocol, and with qualitative assessments of soil organic matter using different soil fractionation methods and spectroscopic techniques.

4. Conclusion

Beta-glucosidase is an enzyme that is sensitive to changes in land use and is an important bioindicator of soil health and functionality. The differences in the values obtained reinforce the importance of sustainable management practices that promote contributions of organic matter to maintain or improve the quality of edaphic ecosystems. Based on this work, we can see the importance of integrating soil management between crops and livestock in order to provide more organic matter, nutrients, microbial activity and, therefore, greater enzymatic activity, all of which contribute to an increase in soil carbon estimates, promoting an improvement in soil health and the edaphic system as a whole.

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