Impact of the *Brachiaria* hybrids on both soil health and carbon stock on livestock production

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Abstract: Pastures occupy 30% of Earth and 80% of the entire agricultural area of the planet. To ensure food to the world and contribute to the quality of the environment, pasture-based animal production systems will also have to undergo through a more intense evolution. The intensification of tropical grasslands is an important strategy of land utilization in developing countries, contributing to increase production and minimize environmental impact through the best management practices. In this sense, the use of *Brachiaria* hybrids represents an excellent option, since combining the best traits of different *Brachiaria* species, with higher nutritive value, forage, and seed yield. Here we have evaluated six *Brachiaria* hybrids' effects on both soil health and carbon stock. We observed that in all *Brachiaria* genotypes the mean carbon stock varied significantly in at least two soil depth categories. In general, carbon stock tends to get smaller as soil depth increases. Enzyme activity analysis showed there were no significant differences in the mean enzyme activity except in hybrid GP 3660 for β -glucosidase enzyme. Therefore, the adoption of *Brachiaria* hybrids might also help farmers to produce in an environmentally friendly manner, due to the potential benefits of *Brachiaria* to soil life enzyme activity and carbon mitigation.

Introduction

In the last 30 years agriculture has evolved dramatically, especially in large crops such as corn and soybeans. Since 1961 global area with those crops increased by 150% but production increased by 1200%. To ensure food to the world and contribute to the quality of the environment, pasture-based animal production systems will also have to undergo through a more intense evolution. However, the speed of the adoption of new management practices that increase production efficiency in grazing systems is nowhere near what we have seen in agriculture recently. In this scenario, the intensification of tropical grasslands is an important strategy for land utilization in developing countries. This means increasing production and minimizing environmental impact through the best management practices. In this sense, the use of *Brachiaria* hybrids represents an excellent option, since combine the best traits of different *Brachiaria* species, with higher nutritive value, forage, and seed yield. Here we present the effect of six *Brachiaria* hybrids on both soil health and carbon stock and discuss the potential impact of these hybrids on nutrient cycling and carbon sequestration in the soil.

Methods

Experimental design

The *Brachiaria* hybrids trial was established in 2018 in randomized block design (RBD) with three replications (4m X 6.4m each). A total of six *Brachiaria* hybrids were tested: GP 0423; GP 1435; GP 1467, GP 3025; GP

3207; GP 3660 (Figure 2). The experimental field is highly homogeneous and has historically been subjected to the same management. The commercial hybrids were planted with a spacing between the lines of 20 cm, 8 seeds per meter linear and density of 400.000 seeds per cultivated hectare.

Soil sample analysis

Briefly, 1kg of 0-20 cm depth soil were collected and used for chemical, physical and biological analysis according to Júnior et. al., (2012). Biological analyses of soil enzymatic activity were performed according to Tabatabai (1994). Enzymes evaluated as biological indicators of soil quality were: arylsulfatase, β -glucosidase, acid phosphatase and N-acetyl- β -D-glucosaminidase (NAG). Carbon concentration from 0 to 90 cm depth was determined by the combustion method (Rayment, 1992).

Data analysis

A two-way mixed ANOVA, combined with one-way and one-way repeated measures ANOVAS, was conducted to evaluate whether there were significant differences in the mean carbon stock (t/ha) along soil depth and plant genotypes. We also conducted one-way ANOVA to assess significant differences in the mean enzyme activity between genotypes considering Arylsulfatase, β -glucosidase, NAGase and Acid phosphatase. The assumptions of normality of residuals, homogeneity of variances and covariances of between-subject factor (genotype), and sphericity were checked with the Shapiro-Wilk, Levene's, Box's M-test and Mauchly's test respectively. The Greenhouse-Geisser sphericity correction was applied to the within-subject factor (soil depth) violating the sphericity assumption. The significance level α =0.05 was used for all analyses. In order to identify the differences, we conducted multiple comparison tests using Tukey's test with p-values adjusted for the number of comparisons. Statistical analyses were performed in R v.3.6.3 software (R Core Team, 2020).

Results

Carbon stock

A two-way mixed ANOVA was conducted to evaluate whether there is an interaction between soil depth categories and *Brachiaria* hybrid genotypes on the mean carbon stock (Figure 1). The Shapiro-Wilk test showed that the model residuals are normally distributed (p-value = 0.07). Levene's and Box's M-tests were also non-significant (p-value = 0.56, and 0.07, respectively) then the variances and covariances were considered homogeneous between the genotypes. Greenhouse-Geisser correction was applied because the sphericity assumption was violated (p-value < 0.001). The results indicated a two-way interaction between soil depth and *Brachiaria* hybrid genotypes on the mean carbon stock (F(13.98,33.55)=5.35, p-value < 0.001). Therefore, the mean carbon stock along different soil depths depends on the *Brachiaria* genotype (Figure 1).

One-way ANOVA (with Bonferroni correction) was performed to assess the effects of *Brachiaria* genotypes on the mean carbon stock at each level of soil depth. The black dots in Figure 1B indicate genotypes with significantly different mean carbon stocks in each soil depth category. All but two (10-20 cm and 40-60 cm) soil depth levels display differences in mean carbon stock between at least two genotypes. We highlight GP 3660 at soil depth levels 0-10 cm, GP 1435 at 20-30 and 30-40 cm, and GP 3025 at 60-80 cm soil depth levels, with carbon stocks of 13.47 \pm 0.535, 9.77 \pm 2.387, 7.26 \pm 0.464 and 12.45 \pm 2.086, respectively, which are significantly higher than the mean carbon stocks of the remaining genotypes in the corresponding soil depth level.

Additionally, we observed that in all genotypes the mean carbon stock varied significantly in at least two soil depth categories (Figure 1C). In general, carbon stock tends to get smaller as soil depth increases. The exceptions

are the GP 1467 e GP 3025 genotypes which accumulate significantly more carbon in deeper soil depths, especially on 40-60cm (12.27 ± 3.503) and 60-80cm (12.45 ± 2.086), respectively. In addition, GP 3660 genotype mean carbon stock (13.47 ± 0.535) is significantly higher in the most superficial soil depth category (0-10cm, see Figure 1C, coloured symbols).



Figure 1. Mean carbon stock (t/ha) along soil depth levels and genotype. A) Mean carbon stock overall visualization; B) Carbon stock comparison between soil depth levels in each genotype. C) Carbon stock comparison between genotypes along each soil depth level. Colored symbols: adjusted p-values for pairwise comparisons between soil depth levels for each genotype. Black symbols: adjusted p-values for comparison between genotypes in each soil depth level. *: p-value ≤ 0.05 ; **: p-value ≤ 0.01 ; ***: p-value ≤ 0.001 ; ***: p-value ≤ 0.001 .

Enzyme activity

We have also evaluated if there is an effect of *Brachiaria* genotypes on the mean activity of Arylsulfatase, β -glucosidase, NAGase, and Acid phosphatase enzymes (Figure 2). The results have revealed there were no significant differences in the mean enzyme activity (nMol pNP x g⁻¹ x h⁻¹) of Arylsulfatase (F(5,12)=1.59, p-value=0.235, Figure 2A), NAGase (F(5,12)=1.15, p-value=0.388, Figure 2C), and Acid phosphatase (F(5,12)=1.63, p-value=0.277, Figure 2D) between *Brachiaria* genotypes. For β -glucosidase, however, the mean carbon stock was significantly lower (F(5,12)=3.98, p-value=0.023, see Figure 2B, black symbols) for GP 3660 (187.09±44.987) in comparison with two other genotypes, GP3207 (252.25±37.923) and GP 3025 (257.8±54.513).



Figure 2. Mean enzyme activity (nMol pNP x g^{-1} x h^{-1}) of different plant genotypes. **A)** Arylsulfatase; **B)** β -glucosidase; **C)** NAGase; **D)** Acid phosphatase. Black symbols: adjusted p-values for Tukey multiple comparison tests between genotypes. ***:** p-value ≤ 0.05 ; ****:** p-value ≤ 0.001 ; *****:** p-value ≤ 0.001 ; *****:** p-value ≤ 0.001 .

Conclusions

The adoption of *Brachiaria* hybrids might also help farmers to produce in an environmentally friendly manner, due to the potential benefits of *Brachiaria* to soil life enzyme activity and carbon mitigation. The positive impacts of the *Brachiaria* hybrids are even higher when combined with other species. Diversification of pastures and the incorporation of key functional plant groups (legumes, for instance) generally improve nutrient cycling and often lead to increased carbon sequestration in the soil (Gaviriaia-Uribe et al, 2020). Papalotla Group claims based on internal data that its *Brachiaria* hybrids tested here (Cayman – 0423 and Camello – 3025) showed higher forage yield, especially under environmental stress conditions as severe drought (Camello) or poorly drained soil (Cayman) (not published data). Camello, for instance, have demonstrated high carbon accumulation in deeper soil depth (Figure 1), suggesting the potential positive impact of its root system against drought to carbon fixation. Higher forage production combined with better nutritive value led to not just higher meat and dairy production, but better digestibility, higher carbon stocks in the soil and lower methane emission (Ruden-Restrepo et al., 2018). Furthermore, improved *Brachiaria* hybrids can also benefit crops as corn and soybean by avoiding nematodes multiplication (not published data), recycling nutrients from deeper layers in the soil and adding organic matter to the agriculture system.

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