Vol.62: e19170737, 2019 http://dx.doi.org/10.1590/1678-4324-2019170737 ISSN 1678-4324 Online Edition



Article - Agriculture, Agribusiness and Biotechnology

Maize Productivity, Mycorrhizal Assessment, Chemical and Microbiological Soil Attributes Influenced by Maize-Forage Grasses Intercropping

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Received: 2017.11.17; Accepted: 2019.05.19.

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HIGHLIGHTS

- Maize-forage grasses promoted effects similar to monocropped maize under no-till system.
- Intercropping maize-tropical forage grasses represents an alternative for monocropped grains.

Maize-forages intercropping presented high rates of mycorrhizal colonization.

Abstract: Mycorrhizae are important components of agroecosystems and the diversification of crops stimulates the abundance of arbuscular mycorrhizal fungi and the participation of symbiosis in plant growth. This experiment examined mycorrhizal assessment, chemical and microbiological soil attributes in a maize-forage grasses intercropping compared to a maize-monocropping system. A complete randomized block design was used with crop systems installed under no-till management with three replicates, as follow: Maize (*Zea mays* L.); *Panicum maximum* Jacq. cv. Aruana; *Urochloa humidicola* (Rendle.) Schweickerdt.; Maize-*P. maximum* intercropping and *Maize-U. humidicola* intercropping. In 2015/2016 season, intercropping maize with *Panicum maximum* Jacq. cv. Aruana or *Urochloa humidicola* (Rendle.) Schweickerdt. promoted similar effects (Tukey test, p<0.05) to monocropped maize under no-till system on soil

chemical and biochemical parameters related to carbon cycling in the soil surface layer, as well as the dynamics of arbuscular mycorrhizal symbiosis in tropical soils, managed for a period of more than six years. Similar grain yield was verified among maize crop systems. This result indicates that intercropping maize-tropical forage grasses represents an alternative for monocropped grains, a very common practice that is used in intensive management, being able to guarantee equivalent productivities and to combine grain production with crop-livestock systems. As a result, intercropping promotes the diversification of the property's income source, adding environmental gains, such as more efficient land use by cultivated plants, keeping soil constantly cultivated, storing carbon and contributing to minimize the impact of climate change on agricultural systems and the sustainability of food production.

Keywords: Cropping systems; arbuscular mycorrhiza; tropical soils; grain crops.

INTRODUCTION

Maize (*Zea mays* L.) is one of the most important grain crops for humanity and Brazil is one of the largest producers worldwide. For 2017, the brazillian maize production is estimated at 88.014 million metric tons, with a harvested area at 16.630 million hectares [1]. For the country's economy, the gross revenue achieved by this crop should reach 171 billion USD [2] and represent about 21% of the international sales [3]. Successful maize production depends on the adequate application of agricultural amendments that will sustain the environment and promote agricultural production. According to the Green Revolution, these inputs are related to the use of highly soluble fertilizers, weed, insect and disease control, both applied in monocrop systems. Undoubtedly, these practices have expanded the supply of agricultural products but they also created negative consequences that have called this agricultural system into question. Nowadays, to ensure agriculture products supply the mind-set is shifting to prioritize for the way such increased production is accomplished [4]. In this sense, the use of technologies with less negative impacts as intercropping systems has been priorized.

Intercropping maize with different legumes seems to be the most common plant consortium employed in the world [5–8]. For Cerrado biome (Brazilian savanna), however, which accounts for 45% of maize production1 and 26% of pasture lands in Brazil [9], the consortium of maize with forage species has been of growing interest, mainly to anticipate the establishment of pastures and increase the soil cover in no-tillage management in areas destined to crop-livestock systems [10]. Intercropped maize with Panicum maximum increased soil microbial biomass nitrogen and microbial nitrogen quotient compared to maize in monoculture, resulting in higher nitrogen availability [11]. Indeed, a significant portion of the positive aspects provided by the intercropping seems to be directly related to the increased population of beneficial microorganisms.

Beneficial microorganisms may increase crop productivity due their abilities to produce plant growth regulators, to suppress disease and greater supply of nutrients to the plants [12,13]. Arbuscular mycorrhizal fungi (AMF) is a group of benefic microorganisms that represent a key link between plants and soil mineral nutrients, which can be considered a biotic soil components where, missing or impoverished, lead to a less efficient ecosystem functioning [14]. Fungal hyphae extend far beyond the roots, exploiting nutrients that are shared with the plant [15]. AMF may modulate the plant-water relations and determine physiological effects such as changes in stomatal conductance and transpiration, increasing the efficiency in water absorption by plants [16]. Recently, Bernardo et al. [13] reported that AMF association helps wheat roots reducing the osmotic stress and maintaining cellular integrity, mitigating the negative effects due to drought.

It is obvious that the soils from Cerrado biome are naturally poor in nutrients, the region presents a long dry season comprising 5-6 months (from April/May to September) and it is not uncommon the occurrence of dry spells during the rainy season [17]. Adoption of soil

and crop managements that favors the activity of microorganisms, specifically AMF, may thus favor crops due their action as environmental stress mitigators. It was therefore the objective of this work was to compare the influence of maize-forage grasses intercropping with maize-monocropping on the maize productivity, mycorrhizal assessment, chemical and microbiological soil attributes in a tipical Latosol from Cerrado bioma.

MATERIAL AND METHODS

Experimental site, design and management

The experimental area was located at the Fazenda Água Limpa, owned by the Universidade de Brasília, Brasília, Brasília, Brazil (15° 55' S; 47° 51' W; 1080 m). The regional climate is tropical and seasonal (Aw, Köeppen classification), with a dry season from April/May to September and a rainy season from October to March. Air temperature and average annual precipitation of the region is 18.0 to 28.5 °C and 1550 mm per year, respectively. The data were obtained from an automatic station model Campbell Sci. Inc., located in Fazenda Água Limpa. The soil was classified as a Dystrophic Red-Yellow Latosol, according to the Brazilian taxonomy system Embrapa [18], corresponding to a Typic Haplustox, according to the U.S. soil taxonomy system [19]. Previously, the soil was chemically characterized according to Embrapa [20] and presented the following results: 5.2 of pH CaCl₂; 0.5 mg dm⁻³ of P; 0.05 cmol_c dm⁻³ of K⁺; 0.2 cmol_c dm⁻³ of Al³⁺, 1.5 cmol_c dm⁻³ of Ca²⁺+Mg²⁺, 4.6 cmol_c dm⁻³ of H+Al³⁺ and 18.9 g dm⁻³ of total organic carbon. Particle size distribution was 525.0, 275.0 and 200.0 g kg⁻¹ of clay, silt and sand, respectively.

The maize-forage grasses intercropping experiment began in 2007 and have been conducted annually in the rainy season. A complete randomized block design was used with crop systems (treatments) installed under no-till management, as follow: 1 – Maize (*Zea mays*) monocropping (M); 2 - *Panicum maximum* monocropping (A); 3 - *Urochloa humidicola* monocropping (B); 4 - Maize-*P. maximum* intercropping (MA); 5 - Maize-*U. humidicola* intercropping (MB). Each treatment contained three replicate plots (8.0 m x 10.0 m). Next to the experimental plots, it was selected a reference area, with native Savanna vegetation.

Forages were sown manually, using 30.0 kg ha⁻¹ of seeds. Before maize sowing, forages and intercropped areas were cut with a grass cutter. The seeds were sown in between maize rows, after maize seeding. Biogene hybrid BG 7055 maize cultivar was sowing at 23 december 2015, planted at 7 plants m⁻¹ in rows of 0.5 m spacing. Lime rates were calculated to achieve a base saturation of 50% [21] and were manually applied uniformly on soil surface at beginning of the rainy season, according to soil chemical analysis [20] (Table 1). The experimental plots with maize received annualy 120.0 kg ha⁻¹ of N (20.0 kg ha⁻¹ of sowing application and 50.0 kg ha⁻¹ of 2 topdressing application, 15 and 30 days after maize emergence), 100.0 kg ha⁻¹ of P_2O_5 and 90.0 kg ha⁻¹ of K_2O . Treatments using forages alone received annualy 40.0 kg ha⁻¹ of N.

Table 1. Chemical characteristics of the soil before the season 2015/2016.

Management systems	pH (CaCl ₂)	TOC	Р	K⁺	Al ³⁺	H+AI	Ca ²⁺	Mg ²⁺	BS*
		g kg ⁻¹	mg	dm ⁻³		cmo	l _c dm ⁻³		%
M	5.1	36.5	3.7	50.7	00.3	3.8	3.6	0.9	47.6
MA	4.9	34.2	4.3	58.5	00.2	4.5	2.0	0.9	39.4
MB	5.0	35.4	11.6	62.4	00.2	4.0	2.4	1.0	45.9
Α	5.3	22.6	2.1	54.6	00.2	2.9	3.0	1.2	58.3
В	5.2	34.2	3.4	54.6	00.2	2.9	2.3	1.0	52.6

^{*}Base saturation values were calculated using the exchangeable bases and total acidity results at pH 7.0 (H +Al) [21]

Soil sampling and harvesting plants

Soil sampling and harvesting plants Bulk soil samples (0.1 m depth from soil surface) were taken during 2015/2016 crop season after 8 consecutive years of cultivation, forming one composite sample of each five subsamples from plot aiming processing chemical and microbial analyses. Roots samples (0.1 m depth from soil surface) were also taken from 5 plants in the central plot of each treatment, at same soil sample to evaluate mycorrhizal colonization. To determine maize productivity at maturity, plants from a 1 m-row segment of the 3 middle rows of each experimental plot (1.0 m²) were harvested for grain yield determination. The harvested plants were then sun-dried, manually thersshed and wheighed to calculate grain yield per hectare (ha).

Mycorrhizal assessment

The roots from maize and forage plants were washed free of soil and clarified as proposed by Phillips & Hayman [22]. Then, the clearfied roots were stained with Trypan blue and the percentage of root length colonization was estimated by the gridline intersection method [23]. The mean was calculated from three plot replications and considering 100 intersections. To evaluate AMF abundance, AMF spores were extracted from 50.0 g soil by routine wet sieving and decanting method according to Gerdemann & Nicolson [24]. Spores density was counted in Petri plate with concentric circles with the aid of stereomicroscope, and expressed as numbers of spores per 50 g soil⁻¹.

Microbiological soil attributes

Microbial biomass carbon (MBC) of soil samples was determined following the fumigation-extraction method [25], qMIC was obtained by the ratio between MBC and total organic carbon (TOC), easily extractable glomalin-related soil protein (EE-GRSP) was obtained through extraction in an autoclave, using 1.0 g of soil and 8.0 mL of sodium citrate at 20 mmol L⁻¹ (pH 7.4) at 121°C for 30 min. Subsequently a centrifugation (5000 g. 20 min) was performed and the supernatant was removed for protein quantification. Glomalin quantification (Bradford) was performed using bovine albumin-serum as standard [26]. Glomalin concentrations of both fractions were converted to mg g⁻¹ of soil considering the total volume of the supernatant and soil dry weight.

Chemical soil attributes

The soil samples (6 replicates for each system) were transported to the laboratory air dried sieved through a 2-mm screen and chemically analyzed according to [20]. Briefly, the pH was measured in water using a relationship of 1:2.5 (soil:solution); P and K⁺ were extracted by Mehlich–1 solution; Ca²⁺+Mg²⁺ and Al³⁺ were extracted with KCl 1 mol L⁻¹. Total organic carbon (TOC) was determined by the Walkley-Black procedures [27]. Mineral particle-size distribution was analyzed using the pipette method [20].

Reference area

In order to evaluate the modifications promoted by agricultural land use, a reference area with native Savanna vegetation was selected next to the plots and sampled in the same period of the experiment. This evaluation was used only for descriptive comparison between the Cerrado area and the areas under agricultural management. The following chemical and microbiological characteristics were observed in the reference area: 4.4 of pH H₂O, 3.0 mg dm⁻³ of P, 45.6 mg dm⁻³ of K⁺, 0.85 cmol_c dm⁻³ of Al³⁺, 0.3 cmol_c dm⁻³ of Ca²⁺+Mg²⁺, 0.15 cmol_c dm⁻³ of H+Al³⁺, 28.8 g dm⁻³ of TOC, 253.1 mg kg⁻¹ of MBC, 0.9 % of qMIC and 40 mg g⁻¹ of EE-GRSP. For mycorrhizal assessment, only spores 145 density of AMF was evaluated, which presented an AMF spore density of 151.0 spores 50 g⁻¹ soil.

Statistical analysis

The evaluation of intercropping systems effect on maize productivity and soil chemical and biological attributes, spore density and mycorrhizal colonization was conducted with a variance analysis using the statistical program Sisvar version 5.6 [28]. Variables in which a significant effect of the treatments was observed, and the averages compared by the Tukey test, considering the level of 5% of significance (p<0.05). The data of the recovered spores were transformed into log (X+1) and root colonization in arcsen (X/100)^{0.5} for this analysis. In order to evaluate the possible relationships between mycorrhizal assessment colonization and occurrence in the study areas, chemical and microbiological soil attributes, principal component analysis (PCA) was performed using the CANOCO program [29].

RESULTS AND DISCUSSION

Grain yield

Maize-*U. humidicola* (13857 kg ha⁻¹) and maize-*P. maximum* intercropping (12770 kg ha⁻¹) did not showed difference in maize yield comparing with maize in monocropping (15973 kg ha⁻¹). Despite this, maize productivity showed an absolute value of 19.9% higher in the monocropped system than the average of the intercropping. Maize was sensitive to competition in the beginning of the season [30] and may have been affected by the fast growth of the forage. Also, nitrogen may have its dynamics affected in intercropping systems [31] and in the present work, maize plants may have been negatively affected since a high forage sowing densities used (30 kg seeds ha⁻¹), probably resulting in great competition for nutrients by the roots of maize and grasses. Silva et al. [32] reported reduction of grain yield in maize-*U. brizantha* intercropping when forage density was higher than 8 kg ha⁻¹ of seeds.

Intercropping improve mitigation of environmental pollution, due to the higher carbon fixation by photosynthesis, besides allowing greater stability and productivity of grains and forage per unit area [33,34]. In tropical regions, such as Brazilian and African Savannas, intercropped tropical forages with grain crop rise as an efficient alternative to enhance maintenance of straw on the soil surface during winter-season, which is one of the mainly difficulties on no-till system, where cash crops alone often do not produce sufficient straw to adequately cover the soil throughout the year [35]. No grain yield difference between monocropped maize and intercropped maize-*Panicum* spp. has been reported by Almeida et al. [36], which agree with the present results and shows the viability of this kind of management in crop and livestock integration systems, as also described to other kind of tropical grasses [37].

Chemical and microbiological soil attributes

The values of pH, H+Al, Ca²⁺+Mg²⁺ and K⁺ showed similar averages for all treatments while exchangeable Al³⁺ and available P were higher in maize monocropping (Table 2). High exchangeable Al³⁺ in monocropped maize compared to the other management systems, maybe related with lime application and the presence of perennial forage species, which generally produce a stable soil structure and good porosity [38], which allows carbonate percolation through the soil profile. At intercropped treatments and sole forage areas, the lower available P (average of 22 mg dm⁻³) may be related with the higher nutrient uptake by forage grasses. Mendonça et al. [39] observed in maize-forage grasses intercropping using the genus *Urochloa* and *Megathyrsus* that the large amount of fine roots from the forage species and maize in the intercropping probably resulted in higher immobilization of P by plants, decreasing the levels of P.

Microbial biomass carbon, TOC, qMIC and EE-GRSP showed similar averages for all cropping systems (Table 3). Even so, MBC in maize-*P. maximum* intercropping (158.2 mg kg⁻¹ soil) was 25.7% and 33.8% higher than that observed in the maize-*U. humidicola* intercropping (125.9 mg kg⁻¹ soil) and maize monocropping (118.2 mg kg⁻¹ soil), respectively. Plant quality composition, litter deposition and root realeased exsudates are possible parameters that affect microbial biomass dynamics [17,40]. Coser et al. [11] found an

average MBC content (192.0 mg kg⁻¹) for all management systems in the same site during 2010/2011 season evaluation, with significative differences in *P. maximum* and maize-*P. maximum* intercropping (248.0 mg kg⁻¹ and 210.0 mg kg⁻¹. respectively). After 5 years, the MBC remains presenting very similar values (Table 3), reflecting that the stability of management system influence on soil microbial community.

Table 2. Soil chemical attributes in a Dystrophic Red-Yellow Latosol from Brazilian Savanna under different management systems. Averages followed by the same letter indicate no significance by Tukey test (p<.05).

	рН	Exchangeable Al ³⁺	H + Al ³⁺	Ca ²⁺ +Mg ²⁺	Available P	K⁺	
Management Systems		cmol _c dm ⁻³			mg dm ⁻³		
M	5.6 + 0.1 a	0.2 + 0.1 a	0.2 + 0.3 a	3.0 + 0.4 a	70.5 + 3.3 a	73.7 + 3.3 a	
MA	5.7 <u>+</u> 0.1 a	0.1 <u>+</u> 0.1 b	0.0 <u>+</u> 0.1 a	3.4 <u>+</u> 1.1 a	13.4 <u>+</u> 22.2 b	66.6 + 22.2a	
MB	5.6 <u>+</u> 0.2 a	0.1 <u>+</u> 0.1 b	0.0 <u>+</u> 0.1 a	3.7 <u>+</u> 0.7 a	22.9 <u>+</u> 23.2 b	68.6 <u>+</u> 23.2 a	
Α	5.8 <u>+</u> 0.1 a	0.1 <u>+</u> 0.1 b	0.0 <u>+</u> 0.1 a	3.4 <u>+</u> 0.3 a	15.5 <u>+</u> 15.4 b	49.6 <u>+</u> 15.4 a	
В	5.9 <u>+</u> 0.2 a	0.1 <u>+</u> 0.1 b	0.0 <u>+</u> 0.1 a	4.6 <u>+</u> 0.9 a	6.6 <u>+</u> 9.9 b	67.8 <u>+</u> 9.9 a	
Average	5.5 <u>+</u> 0.5	0.2 <u>+</u> 0.3	0.1 <u>+</u> 0.1	3.1 <u>+</u> 1.5	22.0 <u>+</u> 26.3	62.3 <u>+</u> 16.8	
CV (%)	2.4	14.6	212.1	23.0	66.6	24.7	

⁽M) Maize monocropping; (A) *Panicum maximum* monocropping; (B) *Urochloa humidicola* monocropping; (MA) Maize-*P. maximum* intercropping; (MB) Maize-*U. humidicola* intercropping; CV (%): coefficient of variation.

Total organic carbon ranged from 25.8 to 29.9 g kg⁻¹ (Table 3), with average of 27.7 g kg⁻¹, in agreement to the findings of [11]. Similar TOC among management systems resulted probably due the strong complexes formed between organic carbon and the oxides usually present in large quantities in Latosols from Cerrado. Thus microbial decomposition is minimized, decreasing oxidation of the organic matter [41]. Linking TOC of the same experimental area in the 2009/2010 growing season [42], the maize-forages intercropping promoted an average increase of 8.0% in the TOC in the period 2009/2010 (21.6 g kg⁻¹) to 2015/2016 (27.0 g kg⁻¹) in the 0-20 cm layer. Greater shoot and root biomass deposition from forage tropical grasses in the soil surface contribute to less soil erosion [43] and may be associated with TOC accumulation in the soil surface.

Table 3. Soil chemical and microbiological attributes in a Dystrophic Red-Yellow Latosol from Brazilian Savanna under different management systems. Averages followed by the same letter indicate no significance by Tukey test (p<0.05).

	MBC	TOC	qMIC	EE-GRSP
Management Systems	mg kg ⁻¹	g kg ⁻¹	-	mg kg ⁻¹
M	118.2 <u>+</u> 23.2 a	29.9 <u>+</u> 2.4 a	0.4 <u>+</u> 0.1 a	2.4 <u>+</u> 0.1 a
MA	158.3 <u>+</u> 36.4 a	27.4 <u>+</u> 1.0 a	0.8 <u>+</u> 0.4 a	2.3 <u>+</u> 0.2 a
MB	125.9 <u>+</u> 22.3 a	26.4 <u>+</u> 0.4 a	0.5 <u>+</u> 0.1 a	2.3 <u>+</u> 0.2 a
A	205.3 <u>+</u> 55.9 a	25.8 <u>+</u> 0.8 a	0.8 <u>+</u> 0.2 a	2.2 <u>+</u> 0.3 a
В	111.0 <u>+</u> 64.8 a	27.8 <u>+</u> 2.7 a	0.4 <u>+</u> 0.2 a	2.4 <u>+</u> 0.3 a
Average	172.4 <u>+</u> 76.3	27.7 <u>+</u> 2.1	0.6 <u>+</u> 0.3	2.4 <u>+</u> 0.7
CV (%)	32.2	6.9	31.3	8.4

⁽M) Maize monocropping; (A) *Panicum maximum* monocropping; (B) *Urochloa humidicola* monocropping; (MA) Maize-*P. maximum* intercropping; (MB) Maize-*U. humidicola* intercropping; CV (%): coefficient of variation.

Unlike the results for TOC, increases of 4% of soil organic carbon in the 0-20 cm layer were greater in intercrops than in monocrops, described for a 7 years field experiment, comparing rotational strip intercrop systems and ordinary crop rotations of maize/wheat [44]. This finding was based on a greater intercrop belowground productivity, compared to monocrop systems, suggesting that soil carbon storage potential of strip intercropping is

comparable to that of currently recommended agricultural systems to conserve organic matter in soil, like no-till management.

The microbial quotient (qMIC), which express soil microbial participation on TOC, ranged from 0.8% in P. maximum monocropping to 0.4% in the maize monocropping, but without statistical differences (Table 3). However, P. maximum monocropping and maize-P. maximum intercropping showed an average closer to the native Savanna vegetation (0.8% and 0.9% respectively), indicating that the soil's living component at this management systems was mantained. Unexpected deviations from this level should indicate that the management is changing and carbon is being released or accumulated [45]. Recognized by the relation with soil carbon content and aggregate stability [46], EE-GRSP was not influenced by the systems, showing an average of 2.36 g kg⁻¹ soil (Table 3), which is in agreement with other studies in agricultural annual crop and forage systems [47,48]. Fokom et al. [49] suggested the use of GRSP as an indicator of soil quality because the difference between crops and soils under native vegetation and other important relations as enzymes, carbon and nitrogen content. In the present study, the different systems showed average of EE-GRSP content lower than that observed in the reference area (2.6 and 4.0 g kg⁻¹ soil respectively), problably the due reduced primary productivity and litter deposition from native vegetation [50].

Microbiological soil attributes in the area with native vegetation showed MBC and TOC similar to others native Savanna regions [48], with qMIC nearby 0.9% and EE-GRSP of 4.0 g kg⁻¹ soil (Table 3). Land use change and the conversion of native vegetation areas into cropping systems reduce soil attributes as MBC, qMIC and EE-GRSP [48].

Mycorrhizal measurements

Arbuscular colonization was influenced by the managements, with the maize crops showing higher values than in the areas with forage grasses in monoculture (Fig. 1). Maize-P. maximun intercropping (51.8%) and maize-U. humidicola intercropping (48.1%), as well as maize monocropping (54.7%), were higher than P. maximum (24.3%) and U. humidicola (39.4%). However, U. humidicola presented similar mycorrhizal colonization to monocropped and intercropped maize. Host preference of AMF has been described for annual and perennial herbaceous species [51], especially related with cultivated species [52]. Higher root colonization on maize than verified in the present study, ranging from 60 to 80%, was observed by Garcá-Gonzáles et al. [53], influenced by barley (Hordeum vulgare L.. cv. Vanesa) and vetch (Vicia villosa L.. cv. Vereda) as cover crops during the fall/winter period. Maize and forage grasses, like that from the genus *Urochloa*, are described as high mycorrhizal dependency, with root colonization ranging from 51.0 to 75.0% and Panicum genus as average mycorrhizal dependency, ranging from 26.0 to 50.0% [54]. These grasses, mainly maize and *U. humidicola* are recognized as good mycorrhizal hosts because they have a spread, fine and fibrous roots, high photosynthetic capacity and high P uptake [55]. Intercropping systems are known as a management practice that support a more abundant and diverse AMF community compared to conventionally managed systems [56] and this knowledge may help in the choice of mixing mycotrophic crops, such as maize and cereals, which form common mycorrhizal networks and thus enhance exchange of nutrients and water, increasing global plant productivity [57]. Management systems modified AMF spore density, ranging from 427.0 to 584.0 AMF spores from 50 g⁻¹ of soil (Fig. 2). The higher AMF spores density was verified in monocropped maize, followed by intercropped with forage grasses (P. maximum and U. humidicola), P. maximum and U. humidicola, as 517.0, 584.0, 547.0, 541.0 and 427.0 spores 50 g⁻¹ soil, respectively. The reference area presented an AMF spore density of 151 spores 50 g⁻¹ soil.

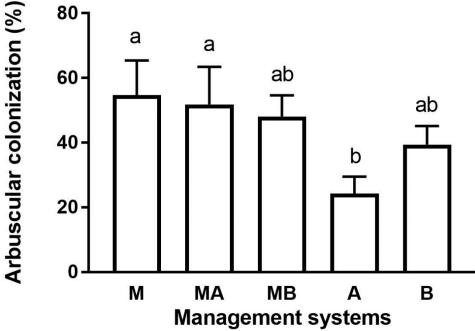


Figure 1. Arbuscular mycorrhizal colonization in several management systems: (M) Maize monocropping; (A) *Panicum maximum* monocropping; (B) *Urochloa humidicola* monocropping; (MA) Maize-*P. maximum* intercropping; (MB) Maize-*U. humidicola* intercropping. Bars represent the standard deviation and different letters the significance for the Tukey test (*p*<0.05).

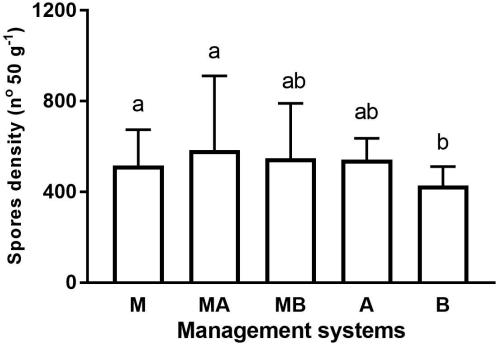


Figure 2. Arbuscular mycorrhizal fungi spore density (spore density) in several management systems: (M) Maize monocropping; (A) *Panicum maximum* monocropping; (B) *Urochloa humidicola* monocropping; (MA) Maize-*P. maximum* intercropping; (MB) Maize-*U. humidicola* intercropping. Bars represent the standard deviation and different letters the significance for the Tukey test (*p*<0.05).

Higher AMF spore density than verified in the present study was observed by García-Gonzáles et al. [53], on soil cultivated with maize and previously cultivated with barley (*Hordeum vulgare* L.. cv. Vanesa) and vetch (*Vicia villosa* L.. cv. Vereda) as cover crops during the fall/winter period. The authors associated high AMF spore density to the existence of a host for AMF during the intercrop period, which is consistent with the present study, where intercropped plots and forage plots were maintained during the year.

Increase or maintenance of AMF spore density in converted crop areas comparing with native vegetation soils were registered in Savanna and Amazon forest [58,59]. In maize monocropping and in the maize-forage intercropping areas, AMF spore density was almost 300% higher than Savana vegetation soil. No-tillage management system associated with mycotrophic crops (like maize and tropical forage grasses) are possible explanations for these observation [53].

Relations between soil attributes and arbuscular mycorrhiza influenced by crop management

In order to examine the relationship between soil chemical and microbiological attributes and the management systems adopted a principal components analysis was carried out (PCA) and the results are showed at the Figure 3.

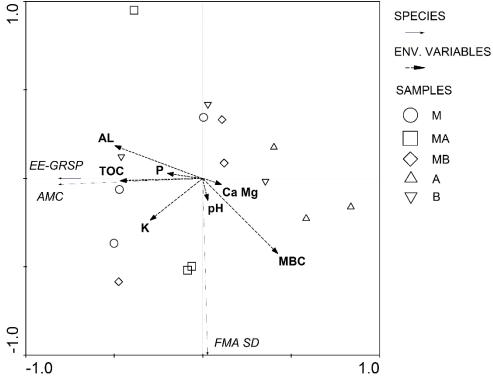


Figure 3. Principal component analysis of arbuscular mycorrhizal parameters (species: FMA SD - AMF spore density. AMC - arbuscular colonization. EE-GRSP – easily extractable-GRSP content) associated with several management systems: (M) Maize monocropping; (A) *Panicum maximum* monocropping; (B) *Urochloa humidicola* monocropping; (MA) Maize-*P. maximum* intercropping; (MB) Maize-*U. humidicola* intercropping. Enviromental variables: TOC. MBC. pH. Al³⁺. K⁺. Ca²⁺+Mg²⁺). available P.

In the PCA, environmental variables explained 61.7% of the variation observed in the study. About this total variation, 82.0% were explained by the 2 first principal components (axes), which is divided as 45.0% to principal component 1 (CP 1) and 37.0% to principal component 2 (CP 2). At the graphic representation, the CP 1 grouped the maize monocropping and maize-*P. maximum* intercropping (left side of the graphic) from the other systems. These managements showed higher arbuscular mycorrhizal colonization and EE-GRSP content, and were also associated with higher contents of COT, Al³⁺, K⁺ and in a minor proportion, available P. Tropical forage grasses and maize-*U. humidicola* reported lower content of EE-GRPS and arbuscular mycorrhizal colonization, related with higher MBC, like observed on PCA, positive correlation between GRPS and total organic carbon [60] and with arbuscular colonization [53], were described in other agricultural systems. Spore density of AMF presented next to CP 2 axe and the higher values of this variable were verified at maize-*P. maximum* intercropping, followed by maize monocropping and *P.*

maximum. Mycorrhizal symbioses allied with intercropping systems work together as tools of ecological intensification, managing soil resources use efficiency and also providing valuable ecosystems services, as nutrient and water cycling and carbon storage [61]. Select mycotrophic crops as annual grasses and forage grasses, like the present study, and combined with legume cover crops are useful strategies to enhance root from indigenous arbuscular mycorrhizal fungi through several soil types and conservative management systems, improving biologically based resource use in agricultural systems [61].

CONCLUSIONS

Intercropping maize and tropical forage grasses as *P. maximum* and *U. humidicola* resulted in similar grain yield to maize monocropping. Maize-forages intercropping presented high rates of mycorrhizal colonization and abundance of AMF spores, mainly related to TOC, K⁺, Al³⁺ and available P. Exchangeable Al³⁺ and available P were higher in maize monocropping than in the intercropping systems. Microbial biomass carbon, TOC, qMIC and EE-GRSP were not affected by the intercropping.

Conflicts of Interest: "The authors declare no conflict of interest." "The funders had no role in the design of the study; in the collection, analyses, or interpretation of data; in the writing of the manuscript, or in the decision to publish the results".

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