Journal of Positive School Psychology
2023, Vol. 7, No. 3, 947-953 http://journalppw.com

Presence Of Gigaspora Rosea In Rizosphere Of Pasture In Bothriochloa Pertusa (L) A. Camus

Alexander Pérez Cordero[1*], Donicer E. Montes Vergara[2] and Yelitza Aguas Mendoza[3]

[1] *Universidad de Sucre, Facultad de Ciencias Agropecuarias, Colombia *corresponding author: alexander.perez@unisucre.edu.co https://orcid.org/0000-0003-3989-1747*
[2] *Universidad de Sucre, Facultad de Ciencias Agropecuarias, Colombia donicer.montes@unisucre.edu.co https://orcid.org/0000-0002-2860-0505*
[3] *Universidad de Sucre, Facultad de Ingeniería, Colombia yelitza.aguas@unisucre.edu.co https://orcid.org/0000-0003-4880-4510*

ABSTRACT

The objective of this study was to isolate the diversity of arbuscular mycorrhizae associated with colosoana grass (Bothriochloa pertusa(L) A. Camus) from cattle farms in the municipalities of Corozal and Tolú, Department of Sucre, Colombia. The number of spores/100 g soil was determined. From the isolated morphotypes we proceeded to the identification of AM species, using taxonomic keys. The results of the analysis of variance for spore density/100 g soil showed a highly significant difference between the number of spores present in the soil. The number of spores/100 g soil and the percentage of root infection found on the cattle farms ranged from (900) 2600 - 4000 (7300) and (30) 41 - 65 (76) respectively. Thirty-one native morphotypes of arbuscular mycorrhizal fungi were isolated, distributed in two genera Glomus and Gigaspora, with Glomus species predominating. These soil microorganisms are an alternative for sustainable pasture recovery and production.

Keyword. Fungi, soil, diversity, pasture, sustainability.

1. INTRODUCTION

Cattle farming activity in the department of Sucre, Colombia, occupies 768,600 ha of pastures representing 13.7% of the livestock area of the Caribbean region; in the Sabanas de Sucre subregion there are 164,000 hectares of pastureland Bothriochloa pertusa, which corresponds to 21.3% of the total pasture area of the department (Pérez et al., 2015). In addition, dual purpose represents the main economic activity in the department of Sucre, where 94.9% of the total area destined to livestock activity is dedicated exclusively to cattle grazing (Gómez et al., 2010). The production of pasture and forage in Colombia is mainly for use in livestock, as a source of feed for livestock (Cardona and Peñas, 2012).

The colosuana or kikuyina grass (Bothriochloa pertusa (L). A. Camus), an introduced species, represents the largest sown area in the department of Sucre, with an approximate total sown area of 274.005 Hectares (Ha), distributed in 19 municipalities. Corozal has the largest area planted with this species in the region, representing 32,223 ha (Pérez et al., 2018).

The soils used for livestock farming in the department of Sucre show different degrees of compaction, erosion problems, low fertility levels, no fertilization, extensive grazing and degraded grasslands. Seasonal rainfall results in a shortage or total lack of fodder during the dry season. There is scientific evidence that arbuscular mycorrhizae play an important role in improving soil physico-chemical conditions; They stimulate the growth and nutritional quality of plant species, making them more tolerant to adverse abiotic and biotic conditions, improve production, nutritional quality and increase the tolerance of grass species to trampling, due to the supply of nutrients to the plant, which stimulate continuous regrowth and rapid recovery after defoliation by herbivorous animals (Pérez and Vertel 2010).

Arbuscular mycorrhizal symbiosis between plants and Glomeromycota, a phylum of early diverging Fungi; is widely hypothesized to have promoted the evolution of land plants during the middle Paleozoic. Arbuscular mycorrhizal fungi (AMF) perform vital functions in the phosphorus cycle that are fundamental to sustainable crop plant productivity. The unusual biological features of AMF ubiquist and abundant obligate biotrophs establishing an approximate association with 80% of vascular plants, have long fascinated evolutionary biologists. It has been suggested that the AMF maintain a stable assemblage of several different genomes during the life cycle, but this genomic organization has been questioned. In 2013, we have published the first genome of an AM fungus: Rhizophagus irregularis strain DAOM197198 (Glomerales). A unique haploid genome was identified in this strain with a repertoire of 28,232 genes. Glomeromycota are formed by distinct clades. Although AMF species have a very broad host spectrum, they differ by their fitness to plant genotypes and environment (Morin et al., 2019).

Based on the hypothesis that no studies of arbuscular mycorrhizal fungi associated with the species Bothriochloa pertusa (L) A. Camus have been carried out on the Colombian North Coast, for which reason this research will contribute and allow us to know the biodiversity of arbuscular mycorrhizal fungi associated with the species Colosoana grass in cattle farms, under the soil and climatic conditions of the municipality of Corozal.

2. MATERIALS AND METHODS

2.1 Sampling area. The present study was carried out in cattle farms, located in the municipality of Corozal, belonging to the department of Sucre. The municipality is located in the Northeast of the department of Sucre, at 8º 55" and 9º 19" North latitude, and between 75º 25" and 74º 42" East of the Greenwich meridian. It has an extension of 20.333 hectares (Has) approximately, it has 5100 Has with slopes between 0 and 3%, 5113.94 Has between 3 and 7%, 2528 Has between 12 and 25% and 7586.24 Has with more than 25%. The altitude ranges from 174 to 200 metres above sea level, average temperature of 28º C, average annual rainfall of 1105 mm and average annual relative humidity of 80%.

2.2 Sampling. Sampling was carried out in cattle farms established only with colosuana or kikuyina grass (Bothriochloa pertusa (L) A. Camus) in the municipality of Corozal. The samples were taken at a depth between 0-20 cm with the help of a PVC plastic tube of 3.8 cm in diameter and 25 cm in length, introducing, rotating and extracting the cylinder with the sample (soil and roots). In each farm between 15-20 samples were taken, these were homogenized per farm to form a sample with a weight of 2000 g (Pérez and Vertel, 2010).

2.3 Microbiological analysis. The microbiological analysis was carried out in the Microbiology laboratory of the University of Sucre in the city of Sincelejo, where the following activities were performed: sieving of soil samples, isolation of AMF spores, spore counting, separation of morphotypes, and identification of morphotypes at the genus level, following the methodologies proposed by (Invam, 2006; Pérez et al., 2012).

3. RESULTS AND DISCUSSION

Table 1 shows the values of the number of spores of arbuscular mycorrhizal fungi associated with colosoana grass roots in the municipality of Corozal. The zone with the highest spore presence was number 4, while zone 1 had the lowest density.

Table 1. Spore densities of arbuscular mycorrhizal fungi associated with colosoana grass on cattle farms.

Zones	Minimum, average and maximum value of spores/100 g of soils
1	(1300-) 1900 (-2500)
2	(2600-) 3500 (-4400)
3	(900-) 3550 (-6200)
4	(4000-) 5650 (-7300)

The ranges of spore density/100 g of soil found on cattle farms in the municipality of Corozal are higher than the values recorded by Picone (2000), who reports a range of 830 - 2600 spores/100 g of soil in a study of pastures in Nicaragua and Costa Rica.

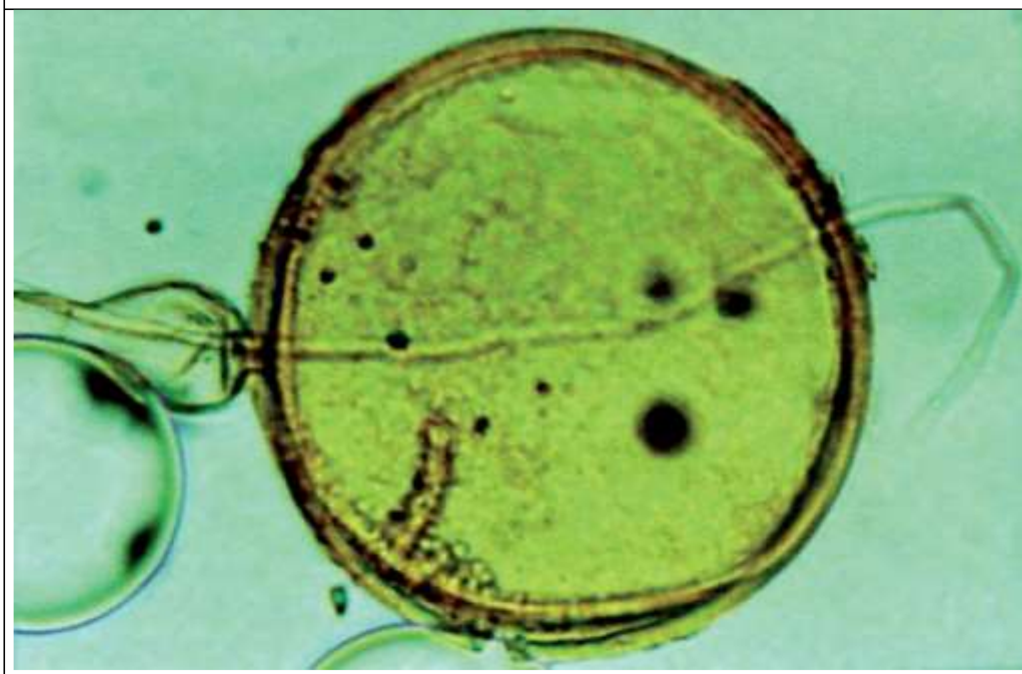
Of the isolated morphospecies, 96.9% correspond to species of the genus Glomus and 3.1% to the genus Gigaspora. The characteristics of the morphotypes show that there is a great diversity of species of the genus Glomus and these results are in agreement with those of Ahn-Heum et al., 2001 and Lugo and Cabello, 2002, who state that continuous grazing produces a decrease in the diversity of arbuscular mycorrhizae and increases the tolerance and adaptation of certain species to these conditions.

During the last 20 years, no agricultural practice has been carried out on the soils of these farms to improve the physical-chemical properties of the soil, mainly its aeration, nor has fertilization been carried out to make up for the nutrient extraction that the grass species has been doing over time. In spite of this series of soil and management limitations, the Bothriochloa pertusa (L) A. Camus grass species has some desirable characteristics for the livestock farmer, such as: rapid recovery after grazing., highly stoloniferous species, high seed production, resistance to trampling and good taste for livestock.

Given the conditions of livestock soils in the department of Sucre, and the possible strategies to implement in the sense that soil microbiological activity can be carried out by mutualistic associations between mycorrhizal fungi with other species of bacteria and fungi, so that they can synergistically contribute to the biological control of phytopathogens and stimulation of plant growth (Cano, 2011; King, 2011).

Bharadwaja et al. (2008) demonstrated that root colonization with HFMA can be increased in the presence of bacteria such as Pseudomonas, Stenotrophomonas and Arthrobacter, which can inhibit the growth of plant pathogens such as Erwinia carotovora, Phytophthora infestans and Verticillium dahliae. Also, in tropical ecosystems, phosphorus uptake occurs poorly on the soil surface, so plants and micro-organisms are responsible for immobilizing phosphorus. For example, saprophytic fungi are in charge of the non-soluble phase of phosphorus in organic matter, composed of nucleic acids, phospholipids and phosphoproteins; later on, this task is continued by slow-growing fungi (Peña-Venegas et al., 2006).

The main characteristics of the mycorrhizal fungus morphospecies identified as Gigaspora rosea Nicolson & Schenck, 1979 are listed in figure 1.

Morphospecies	**40X**
Gigaspora rosea Nicolson & Schenck, 1979.	
	
DESCRIPTION OF THE SPORE:	
Spore shape: ▪ Globosa	**Spore diameter:** ▪ 250 - 270 µm
Bulb colour: ▪ Hyaline	**Suspension bulb diameter:** ▪ 30 – 35 µm
Spore colour: ▪ Water: hyaline to pale yellow ▪ P V L: light yellow	**Cytoplasmic content:** ▪ Granular
Spore surface structure: ▪ Lisa	**Composition and wall type:** ▪ Single layer, laminated appearance.

Wall diameter: ▪ 5.0 – 5.5 µm	**Reaction with Melzer:** ▪ Negative
Hyphal connection diameter: ▪ 1 µm	**Hyphal colour:** ▪ Hyaline
Additional comments: ▪ No hyphal pore was observed.	**Taxonomic determination:** ▪ **Genus: Gigaspora** ▪ **specie:** Features similar to **Gigaspora rosea Nicolson & Schenck, 1979**.

Figure 1. Características morfológicas de la morfoespecie Gigaspora rosea Nicolson & Schenck, 1979. Fuente Pérez, 2020.

Species from the family of Gigasporaceae (Order Diversipsorales) form large hyphae and big spores (>200µm). According to plant hosts, Gigaspora spp can develop different types of intracellular structures, i.e. arbuscules and coils. Gigaspora rosea DAOM194757 can be easily cultivated on entire plant in pots and in vitro on root organ culture. Due to their ability to form long germinating hyphae, it was used to characterize the response to strigolactones (an early plant symbiotic signals) and the synthesis of lipo-chito-oligosaccharides (fungal symbiotic signals). G. rosea is mainly found in non-perturbed soil. Genome comparison with R. irregularis and other phylogenetically distinct species will give clues in deciphering shared and specific features involved in the symbiotic biology of AM fungi.

Arbuscular mycorrhizae-forming fungi are credited with increasing resistance to abiotic stresses such as drought and metal toxicity (Cardona et al., 2005), as they stimulate cell wall thickening and produce an increase in phenolic compounds, chlorophyll, arginine and citrulline (Peña-Venegas and Cardona, 2010). It is believed that at the genomic level, there are amino acid sequences that code for the expression of proteins homologous to metallothioneins, which allow fungi such as Gigaspora rosea to capture heavy metals (Guzmán-González and Farías-Larios, 2005).

4. CONCLUSIÓN

The total of isolated native morphospecies of arbuscular mycorrhiza-forming fungi in cattle farms in the municipality of Corozal, established with Bothriochloa pertusa (L) A. Camus grass, report two genera: Glomus and Gigospora, predominantly species of the genus Glomus. These results contribute significantly to the knowledge of the biodiversity of beneficial soil microorganisms. The cattle farms under study, despite not receiving adequate agronomic management, show high spore densities of arbuscular mycorrhizal fungi associated with Bothriochloa pertusa (L) A. Camus rhizosphere.

5. AUTHOR CONTRIBUTION.

Alexander Pérez Cordero: planning and execution of the experimental part. Donicer Montes V and Yelitza Aguas M, data analysis, conceptualization, writing - revision and editing. All authors have read and approved the manuscript.

6. CONFLICT OF INTEREST. The authors of the manuscript declare that there is no conflict of interest in the presentation and publication.

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