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Characterization of Arbuscular Mycorrhizal Fungi associated with the green onion crop (*Allium fistulosum* L.) in Antioquia, Colombia

Caracterización de Hongos Micorrizicos Arbusculares asociados al cultivo de cebolla de rama (*Allium fistulosum* L.) en Antioquia, Colombia

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ABSTRACT

Onion crop (*Allium fistulosum* L.) has found an important niche in Colombian agriculture, with few studies of mycorrhizal symbiosis in productive zones. The objective of this study was to identify Arbuscular Mycorrhizal Fungi (AMF) and their relationship with agroecological zones in the onion production system. In the department of Antioquia, 23 rhizosphere soil samples were collected in lots with established onion crops for the determination of AMF, and GIS layers of the agroecological zones were applied to superimpose them. A total of 34 AMF morphospecies represented in the families Glomeraceae, Claroideoglomeraceae, Acaulosporaceae, Gigasporaceae, Ambisporaceae and Paraglomeraceae were found. The highest AMF importance value was presented by *Glomus trufemii*, followed by *Glomus tenerum, Claroideoglomus etunicatum, Glomus microcarpum* and *Glomus warcupii*. High affinity of the AMF Glomus sp., G. *warcupii*, *G. halonatum, G. badium, G. ambisporum, G. macrocarpum, G. glomerulatum*, *Gigaspora* sp., *Acaulospora* spp., *Rhizoglomus* sp. and *Rhizoglomus manihotis* with agricultural soils of high suitability classification for onion crops. It should be noted that the highest proportion of AMF morphospecies was associated with very low fertility soils with a greater representation of landscapes such as high plateaus, hills and hills, terraces and fan terraces. In conclusion, the importance of AMF diversity and their interaction in the green onion crop, as well as in the agroecological environment, especially in nutrient-deficient soils and erosion-prone landscapes, is highlighted.

Keywords: Diversity; ecosystems; soil; sustainability; symbiosis

RESUMEN

El cultivo de cebolla (*Allium fistulosum* L.) ha encontrado un nicho importante en la agricultura colombiana, con pocos estudios de la simbiosis micorrizíca en las zonas productivas. El objetivo del estudio fue identificar los Hongos Micorrizícos Arbusculares (HMA) y su relación con las zonas agroecológicas en el sistema productivo de cebolla. En el departamento de Antioquia, se realizó la recolección de 23 muestras de suelo rizosférico en lotes con cultivos establecidos de cebolla de rama, para la determinación de los AMF, adicionalmente se aplicó superposición de capas SIG de las zonas agroecológicas. Se encontró un total de 34 morfoespecies de HMA representado en las familias Glomeraceae, Claroideoglomeraceae, Acaulosporaceae, Gigasporaceae, Ambisporaceae y Paraglomeraceae. El mayor valor de importancia de los HMA lo presentó Glomus trufemii, seguido de *Glomus tenerum*, *Claroideoglomus etunicatum*, *Glomus microcarpum* y *Glomus warcupii*. Se presentó alta afinidad de los HMA *Glomus sp., G. warcupii, G. halonatum, G. badium*, *G. ambisporum, G. macrocarpum, G. glomerulatum, Gigaspora* sp., *Acaulospora* spp., *Rhizoglomus* sp. y *Rhizoglomus manihotis* con los suelos de vocación agrícola de clasificación de aptitud alta para el cultivo de cebolla. Se resalta que la mayor proporción de morfoespecies HMA se asociaron a los suelos de fertilidad muy baja con una mayor representación de paisajes como altiplanicies, lomas y colinas, terrazas y abanico terrazas. En conclusión, se resalta la importancia de la diversidad de los HMA y su interacción en el cultivo de cebolla de rama, así como en el entorno agroecológico, especialmente en suelos con deficiencias de nutrientes y propensos a la erosión.

Keywords: Diversidad; ecosistemas; simbiosis; sostenibilidad; suelos

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1. INTRODUCTION

The green onion crop (*Allium fistulosum* L.) dates back more than 4700 years and is native to the mountainous regions of Central Asia (Brewster, 2008). Due to its rapid growth, high productivity per unit area and the possibility of harvesting throughout the year in different regions of the country, the green onions crop has found an important niche in Colombian agriculture. Its low intensive care requirements and its adaptability to different types of soil and climatic conditions make it an attractive option for both small farmers and large producers.

According to the Observatory of Economic Complexity [OEC], (2024), by 2022, Colombia exported $2.13M in Onions, making it the 70th largest exporter of Onions in the world. In the same year, onions were the 441st most exported product in Colombia, with the main export destinations being the United States ($1.24M), Canada ($664k), United Kingdom ($71.1k), Curaçao ($56.9k), and Aruba ($43k).

The sustainability of onion crops has gained importance worldwide and this is where strategies in its management practices have become relevant. It is also important to understand the cropping system and its associated microbiota. Some root-colonizing fungi are found in the rhizosphere region; this symbiotic association between the fungi and the plant root is known as mycorrhiza. Recent reports in Colombia mention the use of onion plants as a trap crop for increasing inoculum of *Rhizoglomus irregulare* and *Acaulospora mellea* (Wilches-Ortiz et al., 2025), in Antioquia the study of (García Ávila et al., 2021) reported that soils in Girardota where onions are grown have the potential to allow the development of AMF (*Glomus* spp and *Acaulospora* spp), which was evidenced by comparison with a crude multisporic mycorrhizal inoculum.

The occurrence of arbuscular mycorrhizal fungal associations (AMF) in agricultural soils, especially in tropical and subtropical regions, is significantly high, representing 5-50% of all microbial symbioses in agricultural soils. There is limited knowledge of AMF diversity in onion crop in Colombia and its potential implications in the production system. Notably, AMF improves nutrient uptake, especially of phosphorus (Hayman & Mosse, 1971), and may allow reducing the amount of fertilizers applied by up to 50% of the total to be applied (Wilches Ortíz et al., 2022). In addition, AMF can protect the plant against biotic and abiotic stress and improve soil aggregation (Gosling et al., 2006; Wilches Ortíz et al., 2023). In this context, understanding the relationship between AMF and green onion crops is fundamental to optimizing sustainable agricultural practices and improving agricultural productivity. This natural assemblage may not only benefit plant health and yield but also contribute to the conservation of soil resources and promote more resilient and environmentally friendly agricultural systems. Therefore, the aim of the present study is to characterize the different AMF presents in soil.

2. MATERIALS AND METHODS

The study was conducted in the department of Antioquia (Figure 1), in four onion producing areas corresponding to the Valle de Aburrá (Girardota, Barbosa, Copacabana, San Jerónimo, San Antonio de Prado, Palmitas, San Cristóbal), Occidente (La Ceja, La Unión, Sonsón), Suroeste (Jericó, Jardín, Andes) and Norte (Giraldo, Santa Rosa). In these localities, farmers' plots with established green onions crops were selected, where rhizosphere soil samples were collected to determine the density of AMF spores per gram of soil, and to identify the main genera for each zone.

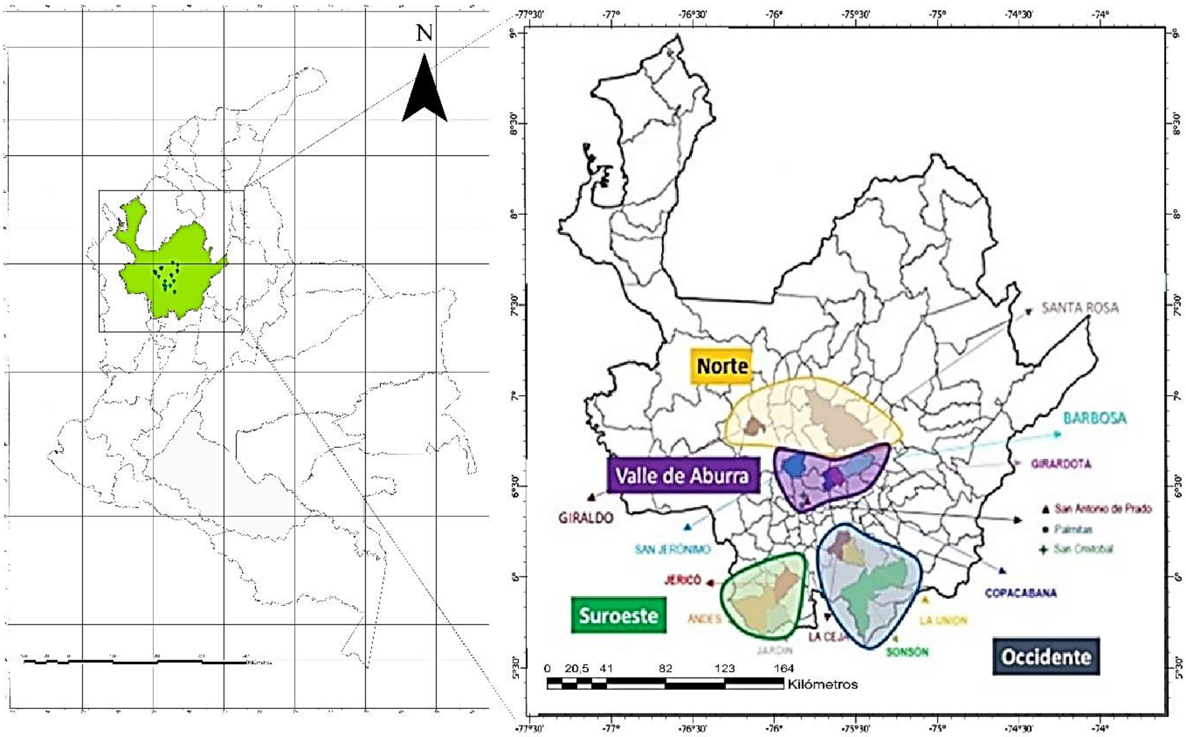

Figure 1. Location in Colombia of sampling in production areas of green onion (*Allium fistulosum* L.). Own source

Soil samples were collected from a total of 23 green onion production farms, in each of which between two and three samples were taken, depending on the heterogeneity of the crop in terms of topographic variability and soil type. Each sample consisted of five subsamples taken along a furrow. Georeferencing was performed on each sample for subsequent processing in ArcGIS PRO® to cross-reference the information from the AMF and the open access information from the agroecological zones of the Sistema de Información para la Planificación Rural Agropecuaria (SIPRA) and Agrology of the Instituto Geográfico Agustín Codazzi (IGAC).

Soil samples were processed in the agricultural microbiology laboratory of the Corporación Colombiana de Investigación Agropecuaria - Agrosavia. Spores were quantified and selected using the wet sieving and decantation methodology of (Gerdemann & Nicolson, 1963), from 10 g of soil and passed through a series of sieves (500µm No 35 with opening in inches 00197 and 38 µm No 400 with opening in inches 0015) in order to separate the organic matter, followed by centrifugation of the samples at 4000 x g for 5 minutes in 15 mL of 70% (m/v) sucrose solution, Afterwards, the interface formed was removed and deposited in a 38 µm sieve to remove remains of the sucrose solution and finally deposited in a Petri dish for observation and selection of spores in a Zeiss Stemi 508 stereoscopic microscope, then mounted on slides with polyvinyl alcohol-lactic acid glycerol (PVLG) or PVLG mixed with Melzer's reagent in a 1:1 (v/v) ratio (Brundrett et al., 1994) and allowed to dry for 24 hours at room temperature for microscopic observation. Identification of AMF spores to genus level was performed based on spore morphological characters such as: spore type formation (glomoid, acaulosporoid, entrophosporoid, gigasporoid and scutellosporoid), sporocarp formation, size and color; wall structure and staining, as well as germination characteristics. Classification keys from (Schenck & Pérez, 1990) and INVAM (International Culture Collection of Arbuscular and Vesicular-Arbuscular Mycorrhizal Fungi) were used as well as scientific publications that provide support in classification at genus and species level (Oehl et al., 2008; Redecker et al., 2013; Sieverding et al., 2015).

Additionally, the formulas described by (Wilches Ortiz et al., 2021) were used to establish the attributes of density, frequency, abundance and importance value (VI) of the AMF. The obtained agroecological variables of land use and vocation, landscape, climate, relief, lithology and soil fertility were correlated with the AMF found by means of a canonical correspondence analysis using the statistical software R® (The R Foundation, 2018) with the CCA (González et al., 2008) and ggplot2 (Wickham, 2009) libraries.

3. RESULTS

From the samples collected for each zone, the information was cross-checked to obtain the main agroecological characteristics detailed in Table 1. It should be noted that for the zoning of the zones, low fertility prevailed in the soils sampled.

Table 1. Agroecological characterization of the sample zones. SPCCC: Semi-intensive permanent cold climate crops. PP: Protection - production. CCTC: Cold climate intensive transient crops, PF: Protective forestry. Own source

ZONE	Municipality	Onion aptitude	Primary use	Vocation	Landscape	Weather	Topography	Lithology	Soil fertility
Norte	Santa Rosa	High aptitude	SPCCC	Agriculture	Highlands	Wet cold and very humid cold	Hillocks and hills	Igneous rocks	Very low
Norte	Giraldo	Medium aptitude	PP	Forestry	Mountain	Temperate humid to very humid	Backbones	Sedimentary rocks	Low to moderate
Occidente	La Ceja	Not suitable	PP	Forestry	Highlands	Wet cold and very humid cold	Hillocks and hills	Igneous rocks	Low to moderate
Occidente	La Unión	Medium aptitude	PP	Forestry	Montaña	Temperate humid to very humid	Filas y vigas	Igneous rocks	Low
Occidente	Sonsón	Not suitable	PP	Forestry	Highlands	Wet cold and very humid cold	Hillocks and hills	Igneous rocks	Very low
Suroeste	Jerico	Not suitable	SPCCC	Agriculture	Mountain	Cold wet to very humid	Rows and beams	Metamorphic rocks	Low to very low
Suroeste	Jardín	Not suitable	PF	Forestry	Mountain	Dry warm	Rows and beams	Igneous rocks	High
Suroeste	Andes	Medium aptitude	PF	Forestry	Mountain	Temperate humid to very humid	Backbones	Sedimentary rocks	Low to moderate

Valle de Aburrá	Medellín	High aptitude	PP	Forestry	Mountain	Cold wet to very humid	Rows and beamsv	Metamorphic rocks	Low to moderate
Valle de Aburrá	San Cristobal	No apta	SPCCC	Agriculture	Mountain	Cold wet to very humid	Rows and beams	Metamorphic rocks	Low to moderate
Valle de Aburrá	San Jerónimo	Medium aptitude	PP	Forestry	Mountain	Cold wet to very humid	Rows and beams	Metamorphic rocks	Low to moderate
Valle de Aburrá	Copacabana	High aptitude	PF	Forestry	Mountain	Temperate humid to very humid	Rows and beams	Metamorphic rocks	Low to moderate
Valle de Aburrá	Girardota	High aptitude	CCTC	Agriculture	Highlands	Wet cold and very humid cold	Terraces and terrace fans	Alluvial deposits	Low to moderate
Valle de Aburrá	Barbosa	Legal exclusion	PF	Forestry	Mountain	Temperate humid to very humid	Rows and beams	Igneous rocks	Low

The spore density of AMF found at the family level is illustrated in Figure 2, highlighting the relative density of the families Glomeraceae (67.7%), Claroideoglomeraceae (13.2%), Acaulosporaceae (12.1%), Gigasporaceae (2.8%) Ambisporaceae (2.0%), Paraglomeraceae (1.3%) and unidentified (0.8%).

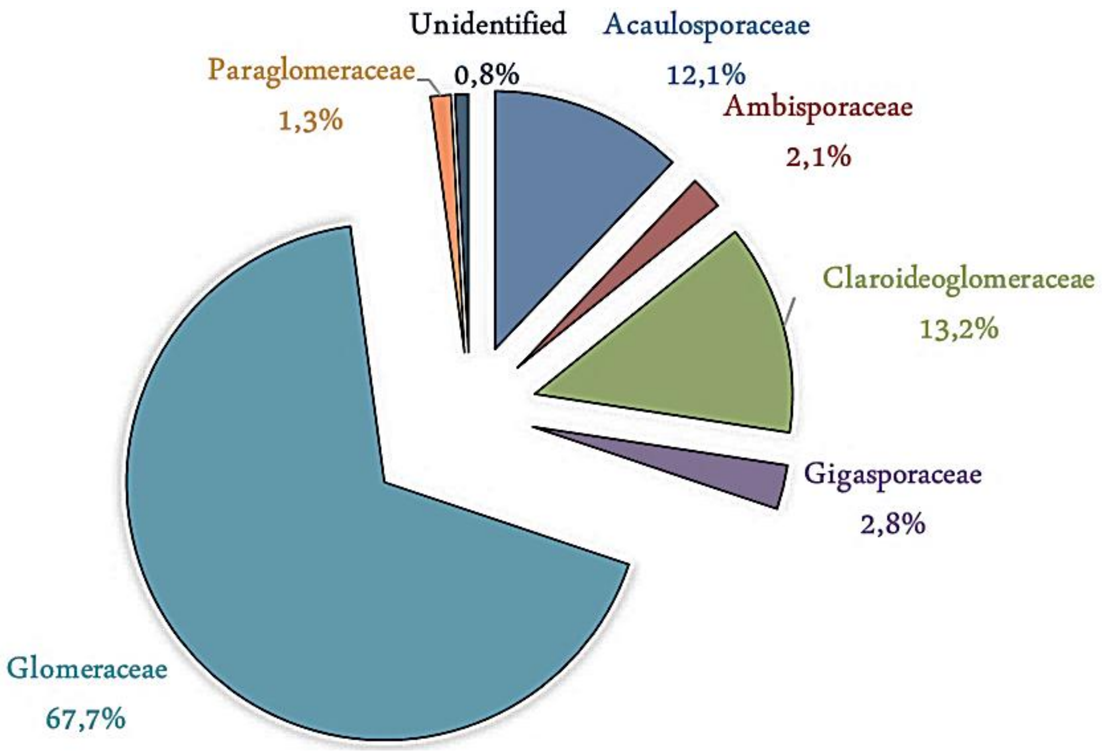

Figure 2. AMF spore relative density by family. Own source

The ecological importance of the AMF was determined by the Importance Value (IV), which is characterized by being dimensionless with a maximum accumulated value of 300 for all the species evaluated. In Figure 3, a radial graph shows the behavior of the individual AMF according to their IV, where the AMF with the greatest contribution are illustrated by a photograph of the spore: *Glomus trufemii* (30.3), *Glomus tenerum* (29.8) and *Claroideoglomus etunicatum* (28.6), consequently without photographs they are followed by *Glomus microcarpum* (28.4), *Glomus warcupii* (24.1), *Glomus macrocarpum* and *Glomus glomerulatum* among other AMF mentioned in the graph with their respective values being 6.3 the lowest IV found.

The canonical correspondence analysis (CCA) allows the analysis of the relationships between variables and is considered a useful and appropriate statistical tool to detect the relationship between the microbial community structure and environmental variables. Figure 4 illustrates this behavior and interaction of AMF and agroecological characterization of the sampled areas through the ACC with a total variance of 40.6%, confirming positive correlations for those variables that are closer to each other in the graph. Regarding the four sampled areas of Antioquia and their relationship with the AMF and agroecological attributes, these are mostly correlated if they are enclosed in circles within the graph. Thus, there was a high affinity of the AMF *Glomus* sp., *G. warcupii, G. halonatum, G. badium, G. ambisporum, G. macrocarpum, G. glomerulatum, Gigaspora* sp., *Acaulospora* spp., *Rhizoglomus* sp. and *Rhizoglomus manihotis* with agricultural soils classified as highly suitable for onion production. It is noteworthy that with an inertia percentage of 29.4%, the highest grouping of AMF morphospecies was obtained, associated with very low fertility soils with a greater representation of landscapes and reliefs such as high plateaus, hills and hills, terraces and terrace fans prone to erosion, highlighting the crucial role of AMF in improving the absorption of plant nutrients in these agroecosystems. The Norte zone showed the greatest interaction between AMF and agro-ecological characteristics compared to the other zones, followed by the Occidente, and lastly the Suroeste zone and Valle de Aburrá.

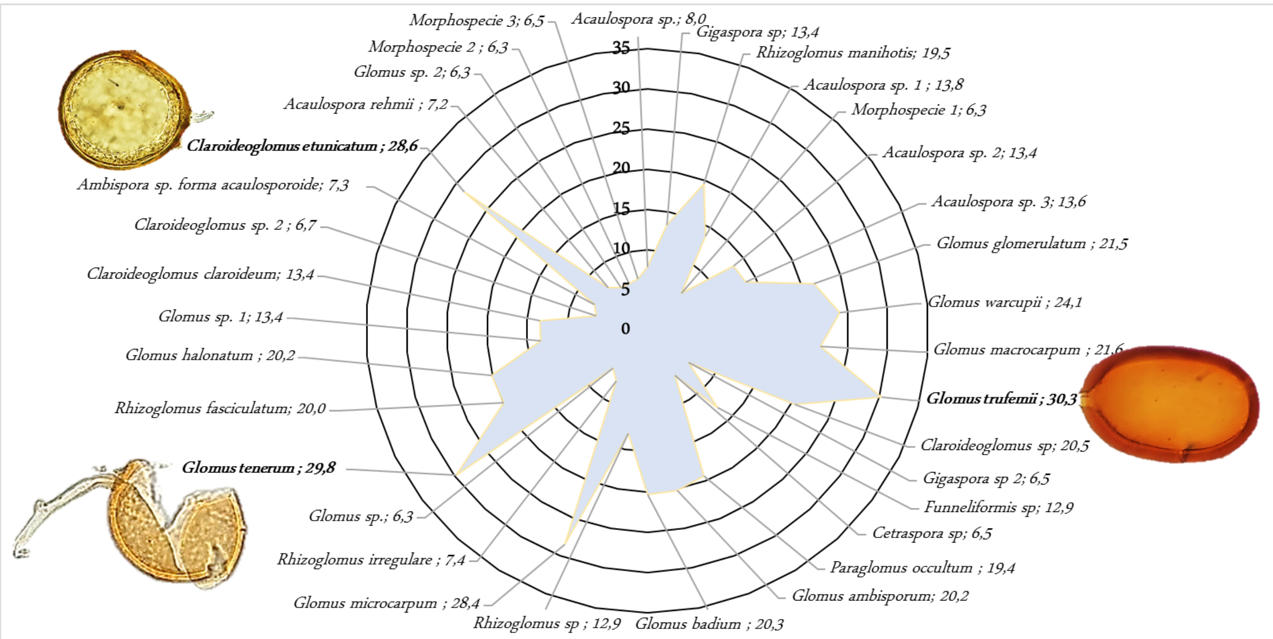

Figure 3. Radial plot of the importance value (IV) of AMF morphospecies. Own source

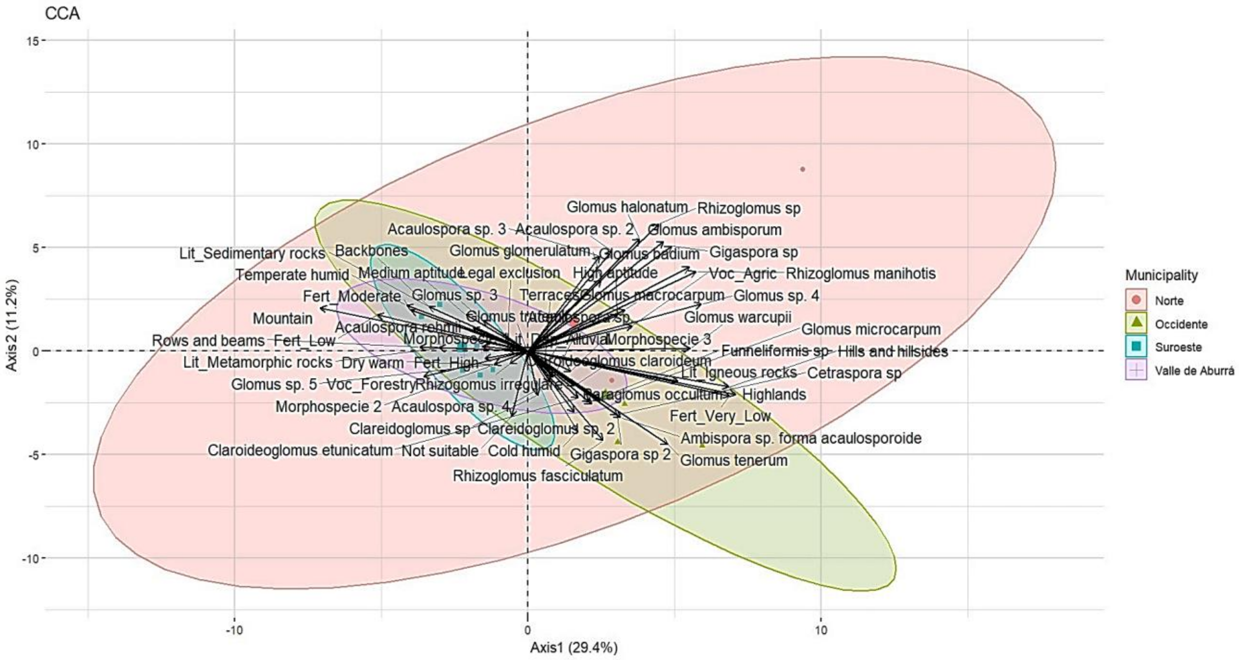

Figure 4. Canonical Correspondence Analysis of AMF morphospecies and their relationship with agroecological characteristics of the sampled areas. Own source.

4. DISCUSSION

Figure 2 highlights the Glomeraceae family (67.7%) with a high density of AMF spores compared to the other families. These results demonstrate the high presence of Glomeraceae in the different soils sampled, which coincides with the reports of (Gianinazzi et al., 2010), who report the high dominance of glomoid AMF in different agroecosystems.

According to Wilches Ortiz et al. (2021) the Importance Value (IV) is a parameter that estimates the contribution or ecological significance of each species in the community and its maximum value is 300, the closer a species is to this value, the greater its ecological importance and dominance over other species. Figure 3 shows the highest value of importance represented by *Glomus trufemii* followed by *Glomus tenerum*, *Claroideoglomus etunicatum*, *Glomus microcarpum* and *Glomus warcupii*. These results agree with the studies of Mathurin et al. (2022), where the diversity of fungal spores varies little from one agroecosystem to another with an outstanding dominance of spores of the *genus Glomus* extracted from soils.

In Figure 4, a high affinity of the AMF *Glomus* sp., *G. warcupii, G. halonatum, G. badium, G. ambisporum, G. macrocarpum, G. glomerulatum, Gigaspora* sp., *Acaulospora* spp., *Rhizoglomus* sp. and *Rhizoglomus manihotis* with agricultural vocation soils of high suitability classification for onion green crops was observed in the correspondence analysis. Coinciding with different studies associating these AMF with the onion crop and other agriculturally important horticultural production systems (Koshila Ravi et al., 2024; Papoui et al., 2022; Singh et al., 2021). The other AMF morphospecies were associated with very low fertility soils with a higher representation of landscapes and reliefs such as high plateaus, hills and hills, terraces and fan terraces predisposed to erosion, highlighting the crucial role of AMF in enhancing plant nutrient uptake in these agroecosystems, agreeing with Ebbisa (2023) who attribute enhanced nutrient uptake and stress resistance to AMF in agroecosystems with soil level deficits.

CONCLUSIONS

A wide diversity of AMF was found in the agroecological zones of the green onion crop in Antioquia, which allowed the identification of 23 morphospecies of AMF of the families Glomeraceae, Claroideoglomeraceae, Acaulosporaceae, Gigasporaceae, Ambisporaceae and Paraglomeraceae. It is noteworthy that the Glomeraceae family (67.7%) was the most abundant in all sampled areas. Part of the specific diversity of AMF was described with a significant value of importance for *Glomus trufemii* in the different zones, while other AMF species were more frequent in very low fertility soils and erosion-prone landscapes. The importance of the diversity of AMF and their interaction in the production system of green onion is highlighted, as well as in the agroecological environment, especially in soils that present fertility deficiencies and erosion-prone landscapes. It is recommended to continue with studies that delve into the variability of AMF in different seasons of the year and their possible impact on soil health and agricultural productivity.

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CONFLICT OF INTERESTS

There is no conflict of interest related to the subject matter of the work.

AUTHORS' CONTRIBUTION

Conceptualization, data curation, formal analysis, research, methodology, software, writing - original draft, writing - revision and editing: Wilches-Ortiz, W., Serralde-Ordoñez, D., Ramírez, L. & Castro-Gómez, Y.

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