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Efficiency of Arbuscular Mycorrhizal Fungi in the Development of Cowpea

Roberta de Souza Santos[1], Juliana Gomes Barreto Souza Leite[1], Elismar Pereira de Oliveira[2], Divino Levi Miguel[2], Joice Andrade Bonfim[3], Patrícia Lopes Leal[1*]

[1] Federal University of Bahia, Multidisciplinary Institute of Health, Vitória da Conquista, BA, Brazil.

[2] State University of Bahia, Department of Agricultural and Soil Engineering, Vitória da Conquista, BA, Brazil.

[3] Federal Institute of Education, Science and Technology of Bahia, Guanambi, BA, Brazil.

ABSTRACT: Cowpea is an important and strategic agricultural crop for Northeast of Brazil, once it combines nutritional value, climate adaptability and soil quality improvements. The objective of this study was to evaluate the efficiency of arbuscular mycorrhizal fungi (AMF) communities on the growth and mineral nutrition of cowpea. The experiment was conducted in a completely randomized design (CRD), with three replications e envolveu tratamentos com inoculação de FMA referências; Inoculação de Comunidades de FMA oriundas de diferentes sistemas de uso do solo; Ausência de inoculação de FMA. After 80 days of cowpea cultivation, were determined plant growth and nutrition parameters, mycorrhizal colonization of the roots and indentifications of AMF species. Results showed that the AMF community from coffee intercropped with grevílea had results close to the RT, with a dry mass value (DMAP+RDM) of 4.31 g and phosphorus content of 7.60 g/kg-1. High species richness of AMF from the study areas was recovered after 80 days of cowpea cultivation.

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Corresponding Author: Patrícia Lopes Leal

KEYWORDS: Agricultural management; Agroecosystem; Glomeromycota.

INTRODUCTION

Cowpea (*Vigna unguiculata* L. Walp) is a crucial subsistence crop in Brazil, with approximately 90% of its one million hectares cultivated in the Northeast region (Bezerra et al., 2010). Despite its hardiness and tolerance to various environmental stressors, cowpea productivity remains low, averaging 300-400 kg ha^{-1} (Freire Filho et al., 2011). This low yield is often attributed to phosphorus deficiency prevalent in tropical soils, exacerbated by phosphorus fixation, which limits plant uptake and necessitates phosphate fertilization (Faquim & Andrade, 2004).

To address this challenge, the use of biofertilizers, particularly AMF (Phylum Glomeromycota), has emerged as a promising strategy for enhancing soil fertility and cowpea yield (Silva, et al., 2018). AMF are ubiquitous in soil biota, forming symbiotic relationships with the roots of over 80% of terrestrial plants (Redecker et al., 2013). Notably, species like Funneliformis mosseae, *Rhizophagus clarus*, and *Scutellospora heterogama* have demonstrated significant improvements in phosphorus uptake, growth, and grain yield in cowpea (Silva et al., 2009; Cruz et al., 2017).

While AMF are not host-specific, their effects on plants can vary significantly, exhibiting functional specificity at both the species and isolate levels (Pouyu-Rojas et al., 2006). Furthermore, AMF community composition, including spore diversity and abundance, is influenced by ecosystem characteristics and soil management practices (Silva et al., 2009; Silva et al, 2018). Therefore, it is crucial to evaluate the effectiveness of AMF communities and isolates derived from specific edaphoclimatic conditions.

Despite this, studies on mycorrhizal efficiency tend to use AMF species from microbial culture collections without considering their origin or adaptation to local conditions (Pagano et al., 2008; Oliveira et al., 2015;). This limitation is particularly evident in research on traditional crops like cowpea in Northeastern Brazil, where the efficacy of native AMF communities and isolates remains pool understood (Silva et al., 2009; Cruz et al., 2017). This study represents a screening of communities of efficient AMF for inoculation of cowpea crops, according to the specific soil conditions of a region where this legume is typically cultivated. therefore, AMF communities isolated from soils under different land use systemswere evaluated for mycorrhizal colonization and efficiency in cowpea development.

MATERIAL AND METHODS

Inoculum soil sampling

This study was conducted in Vitória da Conquista, Southwest Bahia, Brazil (14°52′52″ S, 40°34′46″ W; 847 m altitude). The region has a high-altitude tropical climate, characterized by a dry winter season and hot, humid summers, according to the Köppen classification. Average annual precipitation is 758 mm, with temperatures ranging from 16 to 26 °C and an average relative humidity of 78.4% (Figure 1).

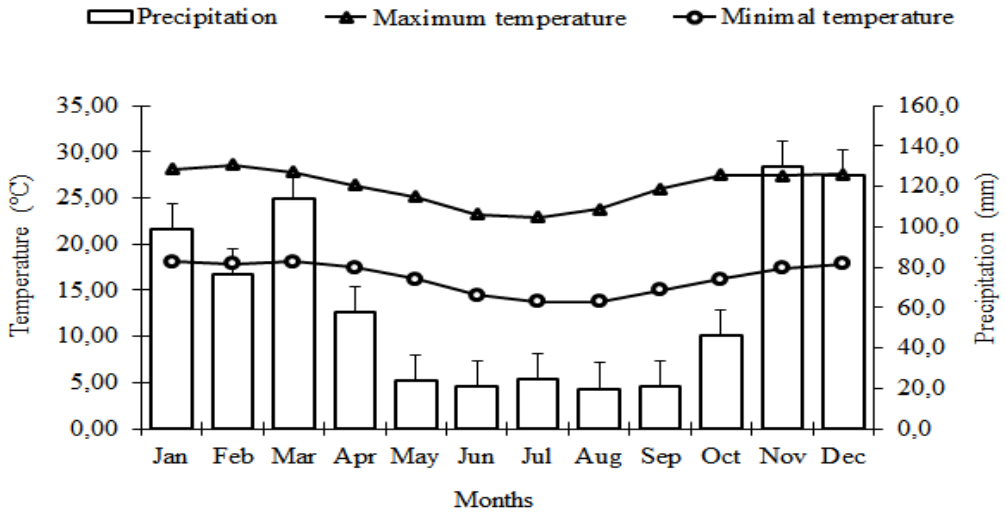

Figure 1 . Annual averages of precipitation, maximum and minimum temperatures (2018) in study areas localized at Vidigal farm, in the municipality of Barra do Choça, State of Bahia, Brazil according National Institute of Meteorology of Brazil.

Soil samples were collected from different land use systems: coffee plantation intercropped with grevillea (CG), full sun coffee plantation with 2.5 x 0.5 m spacing (CS1), full sun coffee plantation with 1.70 x 0.70 m spacing (CS2), pasture (PS), and native forest (NF). Pasture and native forest were included as comparative systems representing degraded and conserved areas, respectively, both adjacent to the coffee plantations (Figure 2 and Table 1).

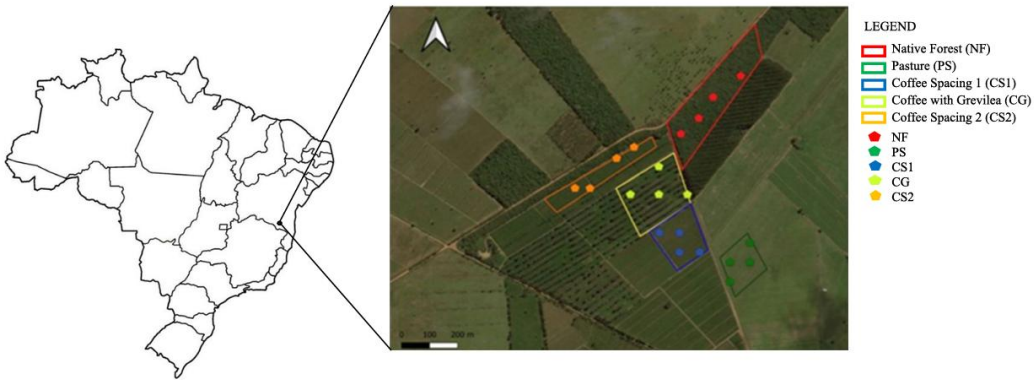

Figure 2. Study areas and sampling location at Vidigal farm in the municipality of Barra do Choça/BA, Brazil. NF: native forest (Mata de Cipó); PS: pasture cultivated with Brachiaria brizantha; CS1: cof fee in full sun and spacing of 2.50 × 0.50 m; CG: coffee combined with Grevilea; CS2: coffee in full sun and spacing of 70 × 0.70 m.

Table 1. Characteristics of the study sites in farm Vidigal: composition, plant area (ha) and spacing, and planting time.

Sampling units	Composition	Area (ha) and spacing	Planting Time
CG	Catuaí coffee crop (IAC 144) intercropped with *Grevillea robusta* in order to protect coffee plants from strong winds and minimize strong sunlight	12 hectares of planted area with spacing of 2.0 x 0.50 m between coffee plants and 20.0 x 4.0 m between grevillea plants	The grevilleas were planted 20 years ago and the coffee culture was implemented 10 years later.
CS1	Catuaí coffee grown in full sun	4 hectares of planted area with spacing of 2.50 x 0.50 m.	10 years
CS2	Catuaí coffee grown in full sun	5 hectares of planted area with spacing of 1.70 x 0.70 m.	12 years
PS	Cultivation of *Brachiaria brizantha* used to raise beef cattle.	30 hectares of planted area	10 years
NF	Native vegetation of Montana seasonal semideciduous forest. Composed of medium-sized woody plants (10 to 20 m), with markedly deciduous characteristics, presence of lianas, and Fabaceae dominance.	Fragment consisting of 10 hectares	20 years

CG: Coffee combined with grevillea; CS1: Coffee in full sun with 2.50 x 0.50 m spacing; CS2: Coffee in full sun with 1.70 x 0.70 m spacing; PS: Pasture cultivated with *Brachiaria brizantha*; NF: Native Forest (Mata de Cipó).

Within each area, a 25 m transect was established with four equidistant sampling points. At each point, three soil samples were collected at a 20 cm depth using an auger and then homogenized, yielding four composite samples per area. Part of the samples were used as inoculum soil in cowpea and another part was intended for physicochemical characterization (Table 2)

Table 2. Chemical characteristics of the soil, at a depth of 0-20 cm, under three coffee management systems, a pasture area and a native forest area.

Field areas	pH	P	K^+	Ca^{2+}	Mg^{2+}	Al^{3+}	H^+	S.B	t	T	V	m
		mg/dm^3	cmol$_c$/dm3								%	
CG	4,60 b	9,0 a	0,13 a	3,20 a	1,12 b	1,02 b	8,82 b	4,45 b	5,47 b	14,20 a	33,50 b	19,50 b
CS1	4,67 b	4,5 b	0,20 a	2,75 a	0,95 b	1,15 b	9,57 b	3,50 b	4,65 b	14,22 a	24,75 b	24,75 b
CS2	4,84 b	2,7 b	0,15 a	2,75 a	0,97 b	0,92 b	9,10 b	3,35 b	5,15 b	14,25 a	31,25 b	19,50 b
PS	5,82 a	2,7 b	0,14 a	3,65 a	2,77 a	0,12 c	5,95 b	6,60 a	6,72 a	12,67 a	52,00 a	2,00 c
NF	4,42 b	1,0 b	0,08 a	0,67 b	0,67 b	2,82 a	14,12 a	1,45 c	4,27 b	18,40 a	8,00 c	66,00 a
CV%	5,32	32,68	34,38	26,61	33,65	42,24	25,67	31,32	16,29	17,59	29,09	34,32

Means followed by the same letter in the column, do not differ statistically by the Tukey test, at 5% probability. Average values obtained from four repetitions in each collection unit. Original means K+ were transformed into X^0.5 to meet the assumptions of the analysis of variance. CG: Coffee combined with grevillea; CS1: Coffee in full sun with 2.50 x 0.50 m spacing; CS2: Coffee in full sun with 1.70 x 0.70 m spacing; PS: Pasture cultivated with *Brachiaria brizantha*; NF: Native Forest (Mata de Cipó). S.B: Sum of bases; t: effective CEC; T: CEC in pH 7; V: Base saturation; m: Aluminum saturation.

Cultivation and conduct of the experiment

Cowpea seeds were pre-treated (disinfested and with dormancy broken) and sown in 3 kg pots containing sterilized sand under greenhouse conditions. After germination, three seedlings per pot were maintained and grown for 80 days. The experiment followed a completely randomized design (CRD) with three replications and included the following treatments: Reference treatment (RT): Inoculation with *Claroideoglomus etunicatum* and *Rhizophagus clarus* (100 spores of each species per pot); Non-inoculated control (NI): No AMF inoculation; Native AMF communities: Inoculation with AMF communities from the five land use systems (CG, CS1, CS2, PS, and NF).

Inoculation with native AMF communities involved adding 100 g of inoculum soil, layered between portions of sterile sand in each pot. For the non-inoculated control, 100 g of sterilized inoculum soil was added to maintain similar soil conditions across treatments.

Growth and nutrition parameters

Eighty days after planting, cowpea seedlings were evaluated for height, and the aerial parts were harvested at the collar height. Stems and leaves were collected and placed in paper bags. Roots were carefully washed, and a 5 g subsample was stored in glass containers with 50% alcohol for subsequent mycorrhizal colonization analysis. The remaining roots, stems, and leaves were dried separately in a forced-air circulation oven at 65 °C for 72 hours. Dry matter was determined for the aerial parts (DMAP) and roots (RDM), and total dry matter was calculated (DMAP + RDM). Dried aerial parts were ground and analyzed for nitrogen (N) and phosphorus (P) content according to Malavolta et al., 1997).

Mycorrhizal colonization was assessed using the slide count method. Roots were stained with methyl blue and examined under a microscope to determine the percentage of colonization (Giovannetti & Mosse, 1980).

Extraction, identification and counting AMF spores

AMF spores were extracted from 50 g of each soil sample using the wet sieving technique (Gerdemann & Nicolson, 1963). Spores were separated by morphotype and mounted on slides for microscopic examination. Spore identification was performed at the genus and species levels based on phenotypic characteristics. Taxonomic identification was conducted using the comparative descriptions provided by the International Culture Collection of (Vesicular) Arbuscular Mycorrhizal Fungi (INVAM; http://invam.ku.edu) and Redecker et al., 2013.

Statistical analysis

All data were submitted to the normality and homogeneity test and then performed the analysis of variance and test of means compared by the Scott-Knott test at 5%, using the SISVAR statistical program.

RESULTS AND DISCUSSION

Composition of AMF communities after cowpea cultivation

A diverse community of 37 AMF morphotypes, including 27 identified species, was recovered after 80 days of cowpea cultivation. (Table 3). This richness surpasses the 23 AMF species recovered by Leal et al. (2009) from trap cultures using various host plants, including cowpea, and inoculum soils from different Amazonian land use systems.

Acaulosporaceae and Glomeraceae were the dominant families, aligning with common findings in cowpea rhizospheres (Silva et al., 2018). Species richness varied significantly among land-use systems, with the highest richness observed in NF (n = 23) and CS1 (n = 21). No significant differences in richness were found among the other treatments: CS2 (n = 12), CG (n = 10), and PS (n = 7) (Table 3).

AMF sporulation did not differ significantly among treatments. Average spore abundance was 187 in CG, 193 in CS1, 211 in CS2, 187 in NF, and 98 in PS. *A. scrobiculata* exhibited the highest spore density (n = 435), followed by *G. fuegianum* (n = 407), *A. mellea* (n = 316), *Glomus* sp.3 (n = 301), and *G. ambisporum* (n = 283). All other species had a total density of fewer than 200 spores per 50 mL of soil (Table 3). The reference AMF treatment (*R. clarus* and *C. etunicatum*) had a mean sporulation of 124 spores, showing no significant difference from the other treatments.

Table 3. Species of arbuscular mycorrhizal fungi (AMF) isolated from study areas and used as inoculum in the efficiency of the cultivation of cowpea.

AMF Families/Species	Number of spores (50 mL of soil)					TS[2]
	Study areas[1]					
	CG	CS1	CS2	PS	NF	
Acaulosporaceae						
Acaulospora delicata (C. Walker, C.M. Pfeiff. &Bloss)		12	48		4	64
Acaulospora denticulata (*Sieverd. & S. Toro, Angewandte Botanik*)		95			29	124
Acaulospora foveata (Trappe & Janos)	14	2				16
Acaulospora kentinensis (C.G. Wu & Y.S. Liu ex Kaonongbua, J.B. Morton & Bever)	114					114
Acaulospora mellea (Spain & N.C. Schenck)	36	76	37		167	316
Acaulospora reducta (Oehl, B.T. Goto & C.M.R. Pereira)					23	23
Acaulospora scrobiculata (Trappe)	35	52	348	2	5	442
Acaulospora spinosa (C. Walker & Trappe)		1				1
Acaulospora tuberculata (Janos & Trappe)		14				14
Ambisporaceae						
Ambispora sp.					8	8
Archaeosporaceae						
Archaeospora trappei (R.N. Ames & Linderman) J.B. Morton & D. Redecker			2		22	24
Claroideoglomeraceae						
Claroideoglomus etunicatum (W.N. Becker &Gerd.) C. Walker &Schuessler		61			32	93
Dentiscutataceae						
Dentiscutata biornata (Spain, Sieverd. & S. Toro) Sieverd., F.A. Souza & Oehl		1			10	11
Gigasporaceae						
Gisgaspora albida (N.C. Schenck & G.S. Sm)		3		28		31
Gigaspora rosea (T.H. Nicolson & N.C. Schenck)		3	25	32		60
Gigaspora sp.				138		138
Scutellospora calospora (T.H. Nicolson & Gerd.) C. Walker & F.E. Sanders					1	1
Scutellospora cerradensis (Spain & J. Miranda)						
Glomeraceae					2	2
Glomus ambisporum (G.S. Sm. & N.C. Schenck)	251	32				283
Glomus fuegianum (Trappe & Gerd)		52			22	74
Glomus geosporum (T.H. Nicolson & Gerd.) C. Walker	129	128	65	245	153	720
Glomus sp.	63		17		50	130
Glomus sp.1		36	89		66	191

Glomus sp.2		90	21			111
Glomus sp.3		65	157		107	329
Glomus sp.4		19				19
Glomus sp.5					8	8
Rhizophagus fasciculatus (C. Walker & A. Schüßler)		7				7
Rhizoglomus fasciculatum (Sieverd., G.A. Silva & Oehl)	82		32		15	129
Rhizoglomus intraradices (N.C. Schenck & G.S. Sm.) Sieverd., G.A. Silva & Oehl	10					10
Septoglomus constrictum (Trappe) Sieverd., G.A. Silva & Oehl					2	2
Septoglomus sp.	26					26
Intraornatosporaceae						
Intraornatospora intraornata (B.T. Goto & Oehl) B.T. Goto, Oehl & G.A. Silva			5		16	21
Racocetraceae						
Racocetra fulgida (Koske & C. Walker) Oehl, F.A. Souza & Sieverd					2	2
Racocetra intraornata (B.T. Goto & Oehl)		15		14	2	31
Racocetra sp.				38		38
Racocetra verrucosa (Koske & C. Walker) Oehl, F.A. Souza & Sieverd		11			2	13
Species Richness	10 b	21 a	12 b	7 b	23 a	
Total spores per area	760 a	775 a	846 a	497 a	748 a	

[1] NF: Native forest (Mata de Cipó); PS: pasture cultivated with *Brachiaria brizantha*; CG: Coffee intercropped with grevillea; CS1: Full ground coffee, spacing of 2.50 X 0.50 m; CS2: Full ground coffee, spacing of 1.70 x 0.70 m.
[2] Total spore density of each identified morphotype.

Biometric characteristics

No significant differences were observed among treatments for plant height (Table 2). However, distinct effects were found for average dry matter of shoots (DMAP), roots (RDM), and total dry matter (DMAP + RDM) (Table 4).

Among the native AMF communities, the CG inoculum resulted in the highest DMAP (3.19 g), RDM (1.11 g), and total dry matter (4.31 g). These values were most similar to those of the reference AMF treatment, which produced the highest averages overall: DMAP (5.59 g), RDM (1.96 g), and total dry matter (7.55 g) (Table 4).

Table 4. Height, dry matter of aerial part (APDM), roots (RDM) and total dry matter (APDM + RDM) of cowpea plants (*Vigna unguiculada* L.), after 80 days, under different treatments involving AMF inoculation.

Treatments*	DMAP (g)	RDM (g)	DMAP+RDM (g)	Height (cm)
CG	3.19 b	1.11 b	4.31 b	17.62 a
CS1	2.56 b	0.84 c	3.40 b	17.45 a
CS2	2.91 b	1.04 b	3.95 b	18.10 a
PS	2.45 b	0.83 c	3.28 b	17.60 a
NF	1.86 c	0.90 c	2.76 c	16.82 a
RT	5.59 a	1.96 a	7.55 a	19.00 a
NI	1.80 c	1.03 b	2.83 c	16.23 a
CV%	18.62	11.59	12.92	8.53

*See Table 3 for identification of AMF communities present in treatments. CG: Coffee intercropped with grevillea; CS1: coffee in full sun with a spacing of 2.50 x 0.50 m; CS2: coffee in full sun with a spacing of 1.70 x 0.70 m; PS: pasture cultivated with *Brachiaria brizantha*; NF: native forest; RT: AMF references (*R. clarus* and *C. etunicatun*); NI: Not inoculated. Means followed by equal letters in the columns do not differ by the Scott-Knott test at 5%.

Table 5. Contents of leaf phosphorus (P), leaf nitrogen (N) and percentage of root mycorrhizal colonization in cowpea (*Vigna unguiculada* L.) after 80 days, under different treatments involving AMF inoculation.

Treatments*	Leaf Phosphorus	Leaf Nitrogen	Colonization
	(g/kg^{-1})		(%)
CG	7.60 a	7.79 c	85.16 a
CS1	5.38 b	15.70 b	69.47 b
CS2	7.59 a	8.49 c	72.18 b
PS	7.61 a	12.74 b	77.68 a
NF	4.13 c	26.75 a	80.63 a
RT	4.19 c	23.68 a	85.90 a
NI	4.47 c	27.07 a	0 c
CV (%)	4.86	5.64	9.11

See Table 3 for identification of AMF communities present in the treatments. CG: Coffee intercropped with grevillea; CS1: coffee in full sun with a spacing of 2.50 x 0.50 m; CS2: coffee in full sun with a spacing of 1.70 x 0.70 m; PS: pasture cultivated with *Brachiaria brizantha*; NF: native forest; RT: AMF references (*R. clarus* and *C. etunicatun*); NI: Not inoculated. Averages followed by equal letters in the columns do not differ from each other by the Scott-Knotta 5% test.

R. clarus and *C. etunicatum* are well-known for their ability to promote mycorrhizal colonization, plant growth, and nutrient uptake in cowpea (Andrade et al., 2009; Cruz et al., 2017). Reference AMF species generally outperform native species because they do not compete with existing soil microorganisms, leading to a more efficient symbiosis with the plant.

Nutrient content and mycorrhizal association

The CG, PS, and CS2 treatments resulted in higher phosphorus (P) content in the cowpea shoots ($p < 0.05$), with an average of 7.6 g kg^{-1} (Table 5). While that NF, RT, and NI treatments had the lowest leaf P content (average of 4.26 g kg^{-1}) and did not differ significantly from each other (Table 5).

Notably, leaf P content across all treatments exceeded the values reported by Silva et al. (2018), who observed a range of 0.78 to 1.52 g kg^{-1} in cowpea across 23 treatments with different AMF species. This difference underscores the importance of P availability in the soil for nutrient uptake and translocation by AMF, particularly in P-deficient soils (Cardoso et al., 2010).

The CG inoculum contained ten AMF species: *A. foveata*, *A. kentinensis*, *A. mellea*, *A. scrobiculata*, *G. ambisporum*, *G. geosporum*, *Glomus sp.*, *R. fasciculatum*, *R. intraradices*, and *Septoglomus* sp. Of these, only *A. foveata* and *R. intraradices* have been previously reported as efficient in promoting cowpea development (Silva et al., 2009; Nascimento et al., 2024). This suggests that other species within the CG inoculum may also contribute to P uptake and cowpea growth.

Nitrogen (N) content in cowpea leaves varied significantly among treatments (Table 5). The highest leaf N content was observed in RT (23.68 g kg^{-1}), NF (26.75 g kg^{-1}), and NI (27.07 g kg^{-1}), with no significant differences among these treatments. The lowest N content was found in CG (7.79 g kg^{-1}) and CS2 (8.49 g kg^{-1}) (Table 3). The high leaf nitrogen (N) content in the non-inoculated control suggests that AMF may not significantly enhance N uptake in cowpea. This is likely due to the high mobility of N in the soil, which allows for efficient plant uptake without AMF assistance (Cardoso et al., 2010).

However, in treatments with lower leaf N content, co-inoculation with AMF and nitrogen-fixing bacteria could be a strategy to improve N uptake (Silva et al., 2009). This approach capitalizes on the synergistic relationship between mycorrhization and nodulation, optimizing N acquisition, as the benefits of AMF extend beyond P nutrition (Arumugam et al., 2010).

All treatments, except the non-inoculated control, exhibited high mycorrhizal colonization (69.47-85.90%) (Table 5). This suggests that cowpea is highly mycotrophic and can be effectively colonized by a diverse range of AMF, potentially leading to improved plant growth (Silva et al., 2018).

CONCLUSIONS

1. The AMF community from coffee plantations intercropped with grevillea (CG) was particularly effective in promoting cowpea growth and phosphorus uptake.

2. Agroforestry systems can serve as valuable sources of beneficial AMF inocula.
3. High mycorrhizal colonization across most treatments highlights the crucial role of AMF in cowpea development.
4. This study advances our understanding of native AMF communities and their potential for sustainable agriculture in the study region.

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REFERENCES

1. Andrade, M. M. M., Stamford, N. P., Sousa, C. A., Silveira, A. C. G. A., Freitas, A. D. S., & Santos, C. E. R. S. (2009). Mineral fertilization and rock biofertilizer with Bradyrhizobium and mycorrhizal fungi on cowpea. *Revista Brasileira de Ciências Agrárias*, *4*(3), 289-292. https://doi.org/10.5039/agraria.v4i3a10
2. Arumugam, R., Rajasekaran, S., & Nagarajan, S. M. (2010). Response of Arbuscular mycorrhizal fungi and Rhizobium inoculation on growth and chlorophyll content of Vigna unguiculata (L) Walp Var. Pusa 151. *Journal of Applied Sciences and Environmental Management*, *14*(4), 113–115. https://doi.org/10.4314/jasem.v14i4.63282
3. Bezerra, A. K. P., Lacerda, C. F., Hernadez, F. F. F., Silva, F. B., & Gheyi, H. R. (2010). Cowpea/corn crop rotation utilizing waters of different salinities. *Ciência Rural*, *40*(5), 1075-1082. https://doi.org/10.1590/S0103-84782010000500012
4. Cardoso, E. J. B. N., Cardoso, I. M., Nogueira, M. A., Maluche, B. C. R. D., & De Paula, A. M. (2010). Micorrizas arbusculares na aquisição de nutrientes pelas plantas. In J. O. Siqueira, F. A. De Souza, E. J. B. N. Cardoso, & S. M. Tsai (Eds.), *Micorrizas: 30 anos de pesquisas no Brasil* (Vol. 1, pp. 153-215). Editora UFLA.
5. Cruz, E. C., Sobreira, A. C., Barros, D. L., & Gomide, P. H. O. (2017). Doses de fósforo e fungos micorrízicos arbusculares no crescimento e produção do feijão-caupi em Roraima. *Boletim do Museu Integrado de Roraima*, *11*(1), 21-28. https://doi.org/10.24979/bolmirr.v11i01.792
6. Faquim, V., & Andrade, A. T. (2004). *Nutrição mineral e diagnose do estado nutricional de hortaliças* (lato sensu specialization work). Universidade Federal de Lavras, Lavras (MG). https://www.docsity.com/pt/nutricao-mineral-de-hortalicas/5435641/
7. Freire Filho, F. R., Ribeiro, V. Q., Rocha, M. de M., Silva, K. J. D., Nogueira, M. S. R., & Rodrigues, E. V. (2011). *Feijão-caupi no Brasil: produção, melhoramento genético, avanços e desafios*. Embrapa Meio-Norte. https://www.alice.cnptia.embrapa.br/bitstream/doc/916831/1/feijaocaupi.pdf
8. Gerdemann, J. W., & Nicolson, T. H. (1963). Spores of mycorrhizal endogone species extracted from soil by wet sieving and decanting. *Transactions of the British Mycological Society*, *46*(2), 235-244. https://doi.org/10.1016/S0007-1536(63)80079-0
9. Giovannetti, M., & Mosse, B. (1980). An evaluation of techniques for measuring VA mycorrhizal infection in roots. *New Phytologist*, *84*(3), 489-500. https://doi.org/10.1111/j.1469-8137.1980.tb04556.x
10. Leal, P. L., Stürmer, S. L., & Siqueira, J. O. (2009). Occurrence and diversity of arbuscular mycorrhizal fungi in trap cultures from soils under different land use systems in the Amazon, Brazil. *Brazilian Journal of Microbiology*, *40*, 111-121. https://doi.org/10.1590/S1517-83822009000100019
11. Malavolta, E., Vitti, G. C., & Oliveira, S. A. (1997). *Avaliação do estado nutricional das plantas: princípios e aplicações*. Ed. Potafos.
12. Nascimento, I. de O., Silva, B. S. de, Almeida, B. S., Cunha, W. L., Sobrinho, N. de A., de Oliveira, J. D., Nobre, C. P., & Rodrigues, A. A. C. (2024). Eficiência simbiótica de fungos micorrízicos arbusculares do cerrado maranhense associados à cultura do feijão-caupi (Vigna unguiculata (L.) WALP.). *Cuadernos De Educación Y Desarrollo*, *16*(1), 1774–1791. https://doi.org/10.55905/cuadv16n1-092
13. Oliveira, J. R. G., Silva, E. M., Rios, T. T., Melo, N. F., & Yano-Melo, A. M. (2015). Response of an endangered tree species from Caatinga to mycorrhization and phosphorus fertilization. *Acta Botanica Brasilica*, *29*(1), 94-102. https://doi.org/10.1590/0102-33062014abb3420
14. Pagano, M. C., Cabello, M. N., Bellote, A. F., Só, N. M., & Scotti, M. R. (2008). Intercropping system of tropical leguminous species and Eucalyptus camaldulensis, inoculated with rhizobia and/or mycorrhizal fungi in semiarid Brazil. *Agroforest Syst*, *74*, 231–242. https://doi.org/10.1007/s10457-008-9177-7
15. Pouyu-Rojas, E., Siqueira, J. O., & Santos, J. G. D. (2006). Compatibilidade simbiótica de fungos micorrízicos arbusculares com espécies arbóreas tropicais. *Revista Brasileira de Ciência do Solo*, *30*, 413-424. https://doi.org/10.1590/S0100-06832006000300003

16. Redecker, D., Schüßler, A., Stockinger, H., Stürmer, S., Morton, J., & Walker, C. (2013). An evidence-based consensus for the classification of 80 arbuscular mycorrhizal fungi (Glomeromycota). *Mycorrhiza, 23*(7), 515-531. https://doi.org/10.1007/s00572-013-0486-y
17. Silva, G. A., Siqueira, J. O., & Sturmer, S. L. (2009). Effectiveness of arbuscular mycorrhiza fungal isolated from soils under different land use systems in the Alto Solimões river region in the Amazon. *Acta Amazônica, 39*(3), 477-488. https://doi.org/10.1590/S0044-59672009000300001
18. Silva, G. A., Siqueira, J. O., Sturmer, S. L., & Moreira, M. S. (2018). Effectiveness of Arbuscular Mycorrhizal Fungal Isolates from the Land Uses of Amazon Region in Symbiosis with Cowpea. *Anais da Academia Brasileira de Ciências, 90*(1), 357-371. https://doi.org/10.1590/0001-3765201820160189