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Morphological Diversity of Arbuscular Mycorrhizal Fungi and Mycorrhization of Okra (*Abelmoschus esculentus* L.) Cultivars in the Peri-Urban Area of Abidjan (Côte d'Ivoire)

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Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

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Original Research Article

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ABSTRACT

In Côte d'Ivoire, okra (*Abelmoschus esculentus* L.) plays a key role in market-gardening systems due to its nutritional and economic importance. However, the productivity of this crop remains limited by soil degradation and excessive use of chemical inputs. From a sustainable agriculture perspective, the use of arbuscular mycorrhizal fungi (AMF) represents a promising alternative. This study evaluated the morphological diversity of AMF communities and the mycorrhization abilities of seven okra cultivars to identify those showing the highest affinity for this beneficial symbiosis. The experiment was conducted on an experimental plot in the district of Abidjan. Soil samples were analyzed by wet-sieving and morphological observation to characterize AMF communities, while stained root samples from the different cultivars were examined to determine mycorrhizal colonization frequency and intensity. The results revealed an average density of 4.48 ± 0.24 spores/g of soill, with a predominance of spores measuring 90 µm (52.30%) and 45 µm (40.11%). Ten morphotypes were identified belonging to the genera *Glomus*, *Gigaspora*, *Acaulospora*, and *Scutellospora* with a clear dominance of *Glomus*. Analysis of the roots showed high variability in mycorrhization depending on the cultivar. Colonization frequencies reached 100% in Noura F1 and Djonan F1, while the highest intensity (56.12%) was recorded in Noura F1, followed by Kopê F1. Conversely, the Divo cultivar had the lowest values. These results open up prospects for selecting more mycorrhizogenic cultivars and the promotion of AMF as a sustainable biotechnology for okra cultivation.

Keywords: Okra, Abelmoschus esculentus; Arbuscular mycorrhizal fungi; AMF; microbial diversity; cultivars; sustainable agriculture.

1. INTRODUCTION

"Agriculture in Côte d'Ivoire, constitutes the main driver of socio-economic development, accounting for nearly one-quarter of the Gross Domestic Product (GDP) and providing livelihoods for a large proportion of the rural population" (Ducroquet et al*.,* 2017). Within this sector, vegetable production plays a strategic position, both for food security and income generation. Vegetable crops such as okra (*Abelmoschus esculentus* L.), tomato (*Solanum lycopersicum* L.), and eggplant (*Solanum melongena* L.) not only serve as staple foods for households but also contribute significantly to nutritional balance and the fight against malnutrition. "This contribution is particularly important in urban and peri-urban areas, where demand for fresh vegetables is steadily increasing" (Bancal & Tano, 2019; Toure et al*.,* 2015). "Okra, an annual species of the Malvaceae family, is among the most widely cultivated and consumed vegetables worldwide" (FAOSTAT, 2023). Global production is estimated at about 300,000 tons, and the crop holds a privileged place in diets and local markets across West Africa (Nana, 2005). Beyond its economic importance, okra is highly valued for its nutritional properties, being rich in dietary fiber, proteins, vitamins (A, C, B6, K, folic acid), and essential minerals. "These attributes contribute to maintaining physiological functions and preventing various deficiencies" (Nzikou et al*.,* 2006; Petropoulos et al*.,* 2018; Agregán et al*.,* 2022). However, "okra productivity remains constrained by multiple biotic and abiotic stresses, compounded by an increasing reliance on chemical inputs. While the use of mineral fertilizers and synthetic pesticides often leads to short-term yield gains" (Jochems-Tanguay, 2014), "their negative impacts on soil fertility, biodiversity, water quality, and human health are well documented" (Griffon, 2010; Roussy et al*.,* 2015). "In this context, the transition toward more sustainable agricultural practices has become a necessity to reconcile productivity, food security, and ecosystem preservation" (Bezu et al*.,* 2014; Gebru et al*.,* 2021). "Among innovative approaches, the exploitation of arbuscular mycorrhizal fungi (AMF) appears particularly promising. These root symbionts play a key role in agroecosystems by enhancing phosphorus and other nutrient uptake, improving plant tolerance to abiotic stresses, and stimulating

resistance against certain soilborne pathogens" (Wang & Qiu, 2005; Diouf et al., 2009 ; Da Silva et al., 2017). "Beyond their positive effects on growth and yield, AMF also contribute to product quality and the resilience of farming systems" (Lounès-Hadj, 2013). "Their integration into crop management practices thus represents a credible alternative to chemical inputs, paving the way for ecologically intensive agriculture" (Haro et al., 2012). In this perspective, the present study aims to assess the mycorrhizal responsiveness of different okra cultivars grown in southern Côte d'Ivoire. Specifically, it seeks (i) to characterize the diversity of AMF present in the soils of the experimental plot and (ii) to quantify the susceptibility of the tested cultivars to mycorrhizal symbiosis. The goal is to determine the extent to which harnessing this biotechnology could contribute to the sustainable improvement of okra productivity in Ivorian vegetable farming systems.

2. PRESENTATION OF THE STUDY AREA

The study area is the district of Abidjan. Located in southern Côte d'Ivoire, between latitudes 4°10' and 5°30' North and longitudes 3°45' and 4°10' West. It comprises ten municipalities and three sub-prefectures: Bingerville, Songon, and Anyama. With a total area of 2,119 km², including 566 m² of lagoon, and an estimated population of approximately 5,616,633 inhabitants according to the 2021 General Population and Housing Census (INS, 2021), the district of Abidjan is bounded to the north by the contact line between the bedrock and the sedimentary basin, to the south by the Ébrié lagoon, to the west by the Agnéby and Niéké rivers, and to the east by the Mé river and the Aghien and Potou lagoons. The experiment was conducted at Félix HOUPHOUËT-BOIGNY University, on the experimental plot (Fig. 1) of the Plant Physiology and Pathology Teaching and Research Unit (UPR PPV).

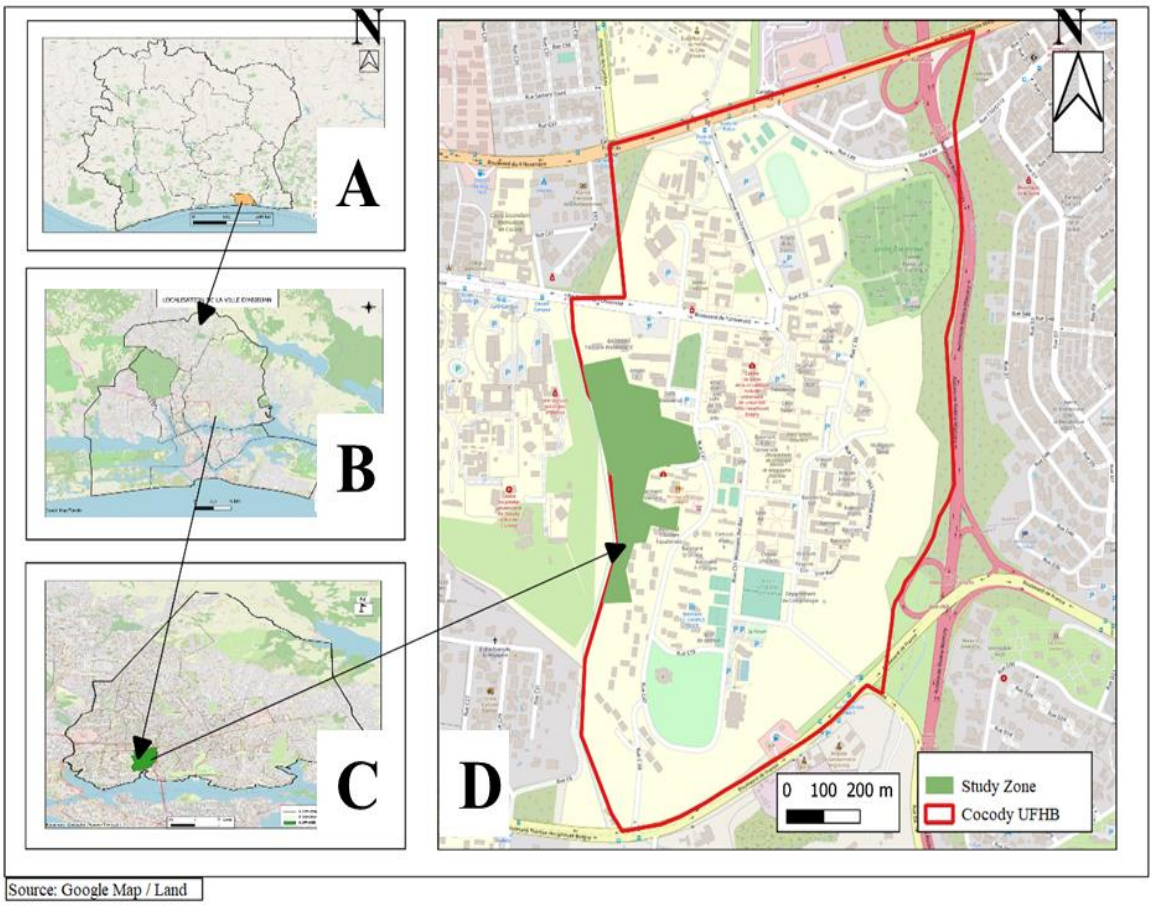


Fig. 1. Map showing the study area
(Source: Tuo et al., 2023)

3. MATERIALS AND METHODS

3.1 Materials

3.1.1 Plant material

The plant material used in this study consisted of the roots of seven (7) okra cultivars (*Abelmoschus esculentus* L., Moench), namely: Noura F1, Teriman, Djonan F1, Clemson spineless, Divo, Kopê F1, and Local. These genotypes were selected based on several key agronomic and socioeconomic criteria, including their wide availability throughout Côte d'Ivoire throughout the year, their relatively short phenological cycle, which makes them a good early variety, and their obvious appeal to producers in terms of yield and to consumers in terms of their organoleptic and nutritional qualities.

3.1.2 Soil material

The soil material used in this study consisted of soil samples taken from the experimental site of the Plant Physiology and Pathology Teaching and Research Unit (UPR-PPV) at Félix HOUPHOUËT-BOIGNY University (Cocody, Côte d'Ivoire), representative of local soil and climate conditions and used as an analytical support for experimental investigations.

3.1.3 Chemical material

The chemical materials used in the laboratory analyses included various reference reagents such as glycerin, potassium hydroxide (KOH), hydrochloric acid (HCl), a dye based on fountain pen ink blue, sucrose ($C_{12}H_{22}O_{11}$), and hydrogen peroxide (H_2O_2). These substances were used as solvents, bases, acids, colorimetric agents, energy sources, and oxidants, respectively, depending on the methodological requirements of the experiments.

3.2 Methods

3.2.1 Characterization of the community of native arbuscular mycorrhizal fungi at the experimental site

3.2.1.1 Soil sampling

At the experimental site, soil samples were taken using an auger from the topsoil layer between 0 and 20 cm deep, following a diagonal pattern. Five (5) soil samples were taken from five points (Fig. 2). The collected samples were then homogenized to form a representative composite sample, which was transported to the laboratory of the Plant Physiology and Pathology Teaching and Research Unit for various analyses, including the extraction and analysis of CMA spores.

3.2.1.2 Extraction of Arbuscular Mycorrhizal Fungal (AMF) spores

The extraction of AMF spores was carried out following the wet sieving technique described by Gerdeman and Nicolson (1963). The collected soil was first air-dried in the laboratory (25 °C, 24 h) and then sieved through a 2-mm mesh sieve to remove the coarse particles. A 20 g soil subsample was then placed in a beaker containing 500 ml of tap water. After vigorous stirring, the suspension was allowed to settle for 10 min. The supernatant was subsequently passed through a series of sieves with decreasing mesh sizes (710 µm, 200 µm, 90 µm, and 45 µm). The residue retained on the 710 µm sieve, consisting mainly of coarse debris, was discarded. The residues retained on the 200 µm, 90 µm, and 45 µm sieves were collected separately and transferred into Eppendorf tubes containing a 70% sucrose solution. These suspensions were centrifuged at 2500 rpm for 7 min. The resulting supernatants were then re-filtered through the same series of sieves (710 µm, 200 µm, 90 µm, and 45 µm). Finally, the residues were thoroughly rinsed with sterile distilled water and recovered in sterile Petri dishes (90 mm in diameter). Five (5) independent extractions were performed from the composite sample, thus constituting five replicates for each sieve fraction (200 µm, 90 µm, and 45 µm).

3.2.1.3 Spore counting

The isolated spores were counted using a binocular magnifying glass (Zeiss) in the laboratory of the Plant Physiology and Pathology Teaching and Research Unit at UFHB (Ivory Coast). The parameters evaluated included: (i) spore density at the experimental site, (ii) proportion of spores according to color, and (iii) proportion according to size. These last two criteria made it possible to distinguish and classify the different morphotypes of the MA fungal spores. The CMA spore density was expressed as the number of spores per gram of dry soil.

Fig. 2. Soil sampling before establishing the okra field

3.2.1.4 Identification of arbuscular mycorrhizal fungi (AMF)

The spores were identified based on their morphological characteristics (Schenck and Perez-Collins, 1990). First, a few representative specimens of each morphotype were mounted between a slide and cover slip in the presence of glycerin and a mixture of glycerol and Melzer's reagent. Observations were made at the laboratory of the African Center of Excellence on Climate Change, Biodiversity, and Sustainable Agriculture (CEA-CCBAD) using an AX10 microscope equipped with a camera. This microscope was used to capture images using the Zeiss ZEN 3.1 (blue edition) application. The species were identified and described using morphological reference keys, based in particular on the descriptions available in the International Collection of Arbuscular and Vesicular-Arbuscular Mycorrhizal Fungi (INVAM) database.

3.2.2 Field trial

3.2.2.1 Land preparation and sowing

The experimental plot, previously under fallow, was first cleared using a machete and a hoe (daba) and then manually plowed to incorporate plant residues into the soil (Figs. 3A and 3B). The plot layout was subsequently carried out using wooden stakes and a cord, in accordance with the planned sowing density (Fig. 3C). Sowing was performed immediately after rainfall, at a rate of three (3) to four (4) seeds per hill, at a depth of 2 to 3 cm. One week after sowing, non-germinated hills were resown. Two (2) weeks after sowing, thinning was carried out to retain only one vigorous plant per hill (Fig. 3D). In the event of rainfall shortage, the plot was irrigated every two (2) days in the morning with tap water.

3.2.2.2 Experimental design

The trial was conducted using a randomized complete block design (Fisher's block) with three (3) replications (3 blocks). Each block was subdivided into seven (7) subplots corresponding to the different okra cultivars tested (treatments). The main factor studied was the cultivar, with seven (7) treatments: Noura F1, Divo, Djonan F1, Clemson Spineless, Kopê F1, Teriman, and a local cultivar. Each subplot was assigned to one (1) cultivar, represented by two rows of 10 plants each, for 20 plants per cultivar (Fig. 4). Each replication contained 140 okra plants, giving 420 plants for the entire trial. The spacing between the hills was 0.6 m, and the spacing between the rows was also 0.6 m, resulting in a planting density of 27,776 plants per hectare for each cultivar. The total experimental area covered approximately 300 m² (Fig. 4). The experimental unit was represented by a single okra plant.

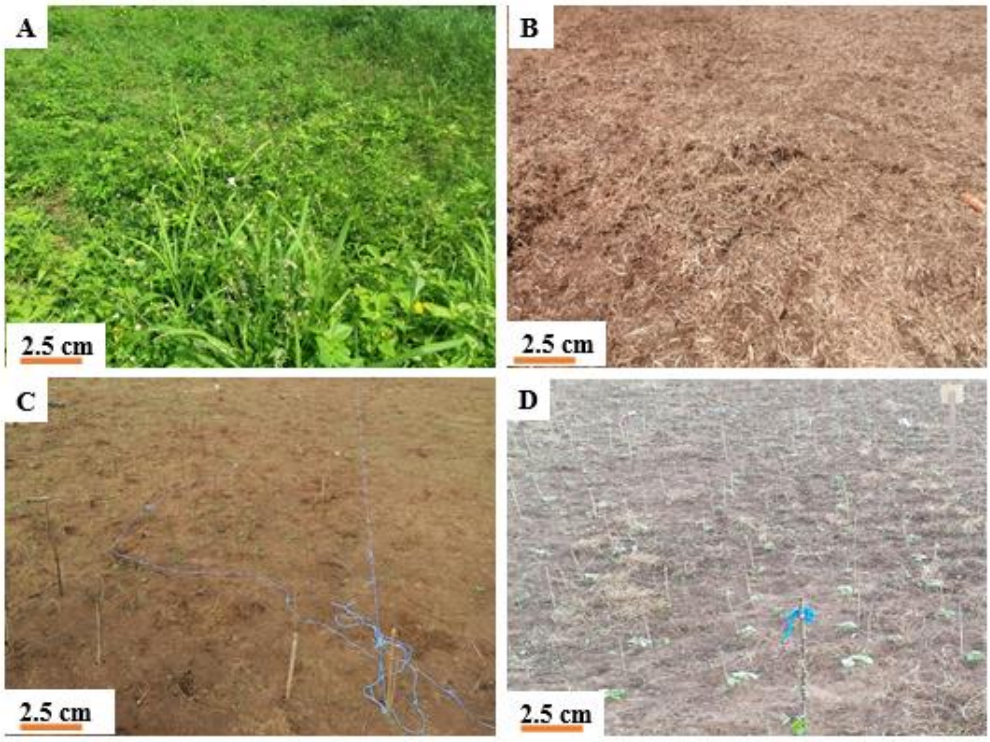


Fig. 3. Setting up the trial on the experimental plot

A: Fallow plot; B: Cleared and plowed plot; C: Staking of the plot; and D: Okra seedlings transplanted onto the plot.

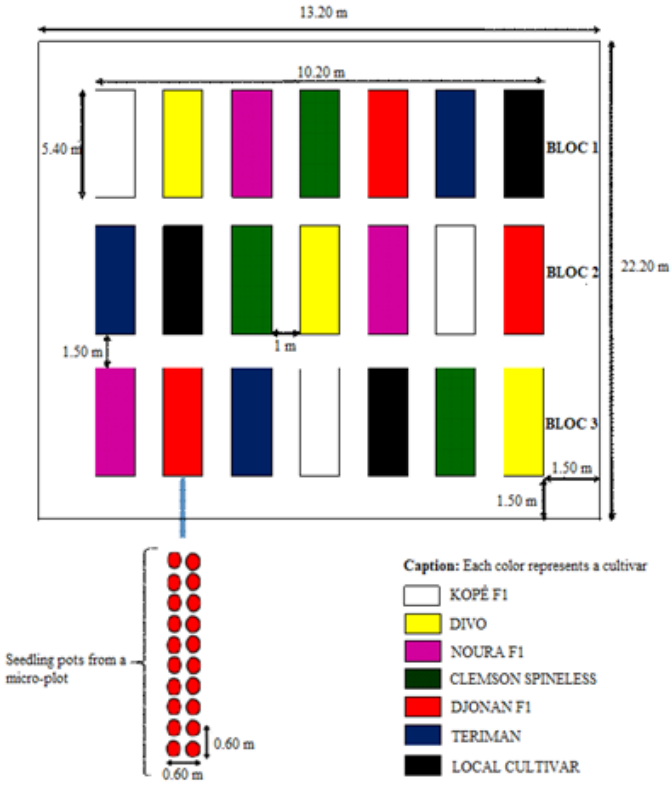


Fig. 4. Fisher block experimental setup

3.2.2.3 Monitoring and maintenance of the trial

To maintain the experimental plot, four manual weedings were carried out using a hoe when the micro-plots were overgrown with weeds. For fertilization of the okra plants, 240 kg/ha of NPK (12-22-22 + $2SO_3$ + 1MgO + 5CaO) fertilizer was applied in two installments (on the 14th and 21st days), followed by 240 kg/ha of urea (46% N) in a single application as soon as the first flowers appeared. As for pest management, five (5) insecticide treatments were carried out on the plot. Deltamethrin at a dose of 75 ml in 15 L of water was sprayed using a backpack sprayer.

3.2.3 Determination of the mycorrhization vevel in different Okra cultivars

3.2.3.1 Root sampling

Root sampling was carried out on the same day for all cultivars, at the end of the fruiting stage of the plants, on the experimental plot of the UPR of Plant Physiology and Pathology, whose soil has a sandy-loam texture (Tuo et al*.,* 2023). The root system of five plants per cultivar and per block was collected using a hoe. Thus, 15 plants of each cultivar were sampled. Subsequently, the root system of the collected plants was separated from the aerial part using pruning shears and washed with tap water. The obtained root samples were kept in water for approximately one minute to separate the fine soil particles from the coarser ones and then stored in a refrigerator before further analyses.

3.2.3.2 Root staining

Fine roots were cut into fragments using sterile scissors in a Petri dish and transferred into test tubes. In each tube, 2 ml of a 10% KOH solution was added to clarify the roots. The tubes were then placed in a water bath maintained at 90 °C for 10 min. After cooling, two drops of hydrogen peroxide were added to each tube, followed by a new 2-min incubation in the water bath. The roots were then thoroughly rinsed with distilled water, immersed in 2 ml of 1% HCl solution, and placed again in the water bath at 90 °C for 1 min. After discarding the HCl solution and cooling, the roots were covered with an ink-blue solution until fully immersed (Vierheilig et al*.,* 1998). The tubes were returned to the water bath at 90 °C for 15 min to fix the staining. Finally, the roots were rinsed with distilled water to remove excess dye.

3.2.3.3 Quantification of mycorrhizal colonization

Quantification of mycorrhizal colonization was performed on stained root samples mounted between the slide and coverslip. For each cultivar, approximately 30 stained root fragments were arranged in parallel, with 10 (10) fragments per slide. Observations were carried out under a light microscope, focusing on the presence of characteristic fungal structures such as spores, hyphae, arbuscule, and vesicles. Colonization was assessed following the method of Truvelot et al*.* (1986), which is based on scoring and the calculation of quantitative parameters. The parameters considered were :

- Frequency of mycorrhizal colonization (F, %)
- Intensity of colonization in the root cortex (M, %)
- Intensity of mycorrhization in the colonized fragments (m, %),
- Arbuscular intensity within the colonized portion (a, %),
- Arbuscular intensity within the whole root system (A, %).

This method enabled the evaluation of (i) root colonization by mycorrhizal fungi for each fragment observed by assigning a score from 0 to 5 :

- n0: Number of fragments scored 0
- n1: Number of fragments scored 1
- n2: Number of fragments scored 2
- n3: Number of fragments scored 3
- n4: Number of fragments scored 4
- n5: Number of fragments scored 5 and (ii) the abundance of arbuscule per fragment, ranging from A0 (no arbuscules), A1 (few arbuscules), A2 (presence of arbuscules) to A3 (abundant arbuscules). This also allowed the calculation of the colonization parameters.

- **Frequency of mycorrhizal colonization**

The frequency, denoted as F and expressed as a percentage, corresponds to the proportion of colonized root fragments relative to the total number of fragments examined. This parameter reflects the importance of fungal penetration points in the colonization of the root system (Trouvelot et al*.,* 1986) :

$$F\ (\%) = \frac{\mathrm{n1} + \mathrm{n2} + \mathrm{n3} + \mathrm{n4} + \mathrm{n5}}{\mathrm{n0} + \mathrm{n1} + \mathrm{n2} + \mathrm{n3} + \mathrm{n4} + \mathrm{n5}} \times 100$$

- **Intensity of root cortex colonization**

The intensity of root colonization was obtained by calculating the proportion of the root cortex colonized in relation to the entire root system. It shows the extent of colonization of the root system (Trouvelot et al*.,* 1986) :

$$M\ (\%) = \frac{95\mathrm{n5} + 70\mathrm{n4} + 30\mathrm{n3} + 5\mathrm{n2} + \mathrm{n1}}{2\mathrm{n0} + \mathrm{n1} + \mathrm{n2} + \mathrm{n3} + \mathrm{n4} + \mathrm{n5}}$$

- **Intensity of mycorrhization of mycorrhizal fragments**

The intensity of mycorrhization is the product of the intensity of mycorrhization (M) expressed as a percentage relative to the frequency of mycorrhization (F) (Trouvelot et al*.,* 1986) :

$$m\ (\%) = \frac{\mathrm{M}}{\mathrm{F}} \times 100$$

- **Arbuscular intensity of the mycorrhizal part :**

$$a\ (\%) = \frac{100\mathrm{mA3} + 50\mathrm{mA2} + 10\mathrm{mA1}}{100}$$

With mA3 = ((95n5A3 + 70n4A3 + 30n3A3 + 5n2A3 + n1A3)/number of mycorrhizal fragments) x 100/m, the same applies to A2 and A1.

- **Arbuscular intensity in the root system**

This parameter indicates the ratio of the product of the arbuscular intensity of the mycorrhizal part (a %) and the colonization intensity of the root cortex, expressed as a percentage (Trouvelot et al*.,* 1986) :

$$A\ (\%) = \frac{\mathrm{M}}{100} \times a\ \%$$

3.2.4 Statistical analyses

The collected data were entered and organized using Microsoft Excel 2016 (Microsoft Corporation, Redmond, WA, USA), which was also used to create graphical representations. Descriptive and inferential statistical analyses were performed using STATISTICA software, version 12.5 (StatSoft Inc., Tulsa, OK, USA). To normalize the distribution of variables expressed as percentages, they were first transformed using the arcsin√x function (Snedecor & Cochran, 1989). When the conditions for applying analysis of variance (one-way ANOVA) were not met, the data were subjected to nonparametric tests, notably the Kruskal-Wallis test (Kruskal & Wallis, 1952), followed by the Steel-Dwass-Critchlow-Fligner post hoc test (Critchlow & Fligner, 1991) for multiple comparisons. The statistical significance threshold was set at $p < 0.05$.

4. RESULTS AND DISCUSSION

4.1 Results

4.1.1 Native mycorrhizal communities in the experimental plot

4.1.1.1 Spore density of arbuscular mycorrhizal fungi and their proportion by diameter

Stereomicroscopic observation revealed the presence of arbuscular mycorrhizal fungi (AMF) in the experimental plot before the establishment of the trial, with a density of 4.48 ± 0.24 spores per gram of soil. Fig. 5 illustrates the distribution of AMF spores according to their diameter. Analysis of variance indicated a significant variation in the proportions depending on the spore size ($p < 0.05$). The 90 µm and 45 µm diameter classes were the most represented, accounting for 52.30% and 40.11% of the total spores, respectively.

4.1.1.2 Spore proportion by color

Observation of the spores isolated from the soil of the experimental plot allowed the distinction of five (5) color classes: black, yellow-orange, hyaline, brown, and yellow (Fig. 6). Statistical analysis showed a significant variation in spore proportions according to color ($p < 0.05$). Multiple comparison tests revealed that yellow-orange spores were the most abundant, whereas hyaline spores represented the least frequent group in the experimental soil.

4.1.1.3 Number of arbuscular mycorrhizal fungal morphotypes in the soil

The combined analysis of morphological traits, notably spore color and diameter, allowed the identification of 10 morphotypes of arbuscular mycorrhizal fungi (Table 1). Among these, yellow-orange spores of 90 µm diameter and black spores of 45 µm diameter were the most abundant, representing 21.15% and 21.57% of the total, respectively. In contrast, hyaline spores with a diameter of 200 µm were the least represented, with a proportion of 0.87%. Based

on their morphological features, the isolated spores were attributed to the genera Glomus (morphotypes 1a, 2a, 3b, 4b, 6, and 8), Gigaspora (morphotypes 2b, 3a, and 4a), Acaulospora (morphotype 2b), and Scutellospora (morphotype 10).

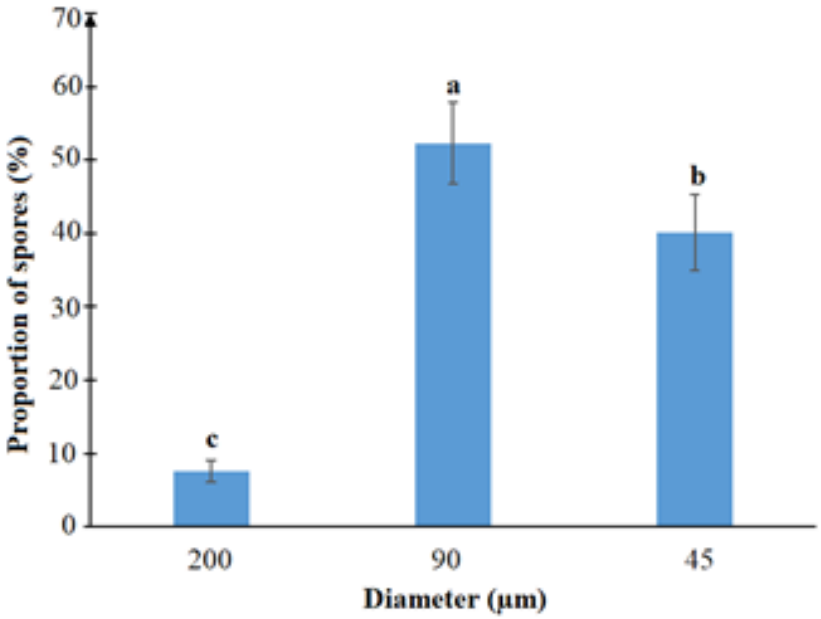


Fig. 5. Proportions of arbuscular mycorrhizal fungal spores in the experimental plot according to the diameter

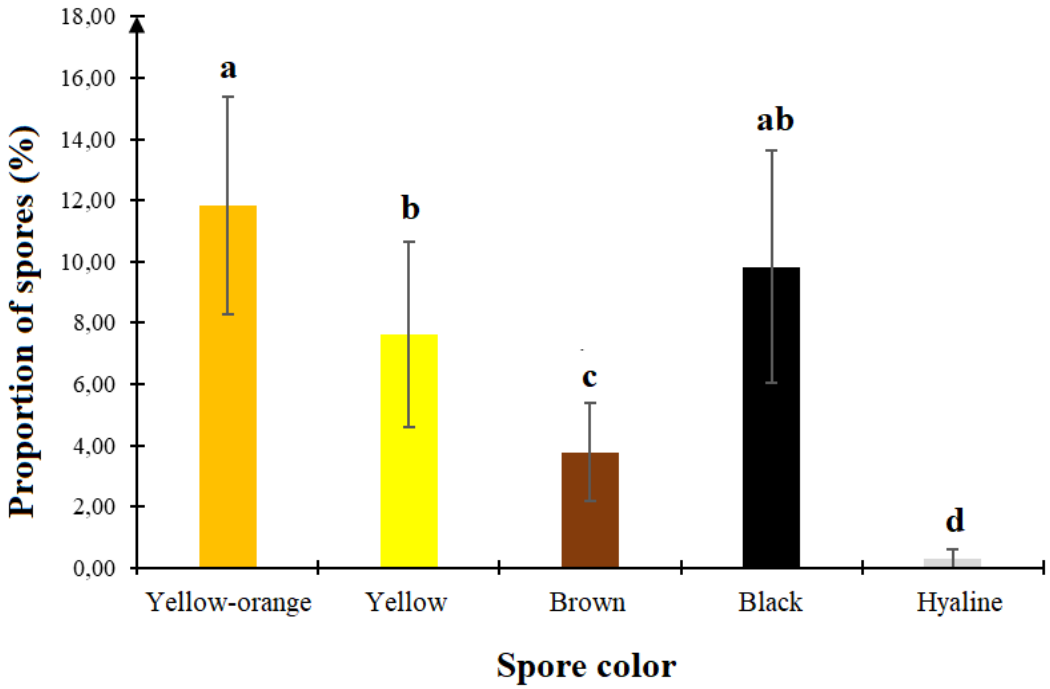
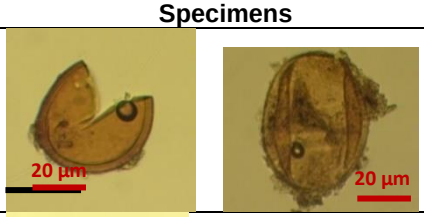
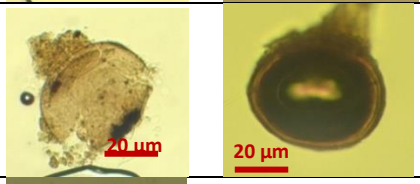
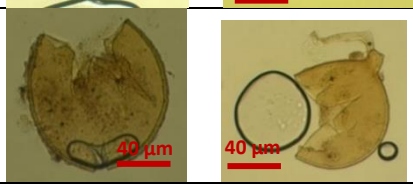
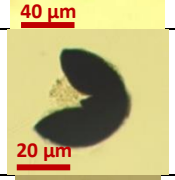
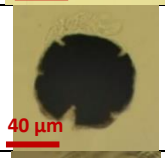
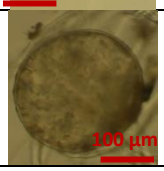


Fig. 6. Proportions of arbuscular mycorrhizal fungal spores according to color)

Table 1. Morphotypes of mycorrhizal fungi

N	Color	Diameter	Proportion (%)	Specimens
1	Yellow-orange	45 µm	21.15	
2		90 µm	14.34	
3	Yellow	45 µm	5.55	
4		90 µm	14.63	
5		200 µm	2.63	Nothing
6	Brown	45 µm	5.02	
7		90 µm	6.28	
8	Black	45 µm	21.57	
9		90 µm	7.92	
10	Hyaline	200 µm	0.87	

4.1.2 Degree of mycorrhizal colonization in the Okra cultivars studied

Microscopic examination of the stained root fragments revealed the presence of characteristic structures of arbuscular mycorrhizal fungi (AMF) in all the cultivars analyzed. Specifically, arbuscules, intraradical hyphae, and spores were observed within the cortical tissues of the roots of all the tested cultivars (Fig. 7).

4.1.2.1 Frequency and intensity of mycorrhizal colonization

The analysis showed a significant difference among the okra cultivars only with respect to the frequency and intensity of mycorrhizal colonization (Table 2). Regarding the colonization frequency, 100% of the observed roots of the cultivars Djonan F1 and Noura F1 exhibited signs of mycorrhizal symbiosis. Conversely, the lowest frequency was recorded in the cultivar Divo. Regarding colonization intensity, the values ranged from 10.57% to 56.12%. Post hoc testing allowed the grouping of okra cultivars into two main clusters. The first group, consisting of the cultivars Kopê F1 and Noura F1, exhibited the highest intensity values. The second group, comprising the local cultivar and Divo, displayed the lowest colonization intensities. For the intensity of colonization in the mycorrhizal fragments, the values ranged between 14.51% and 50.78%.

4.1.2.2 Arbuscular proportions in the different cultivars

Table 2 presents the proportion of arbuscules observed in the mycorrhizal fragments and the arbuscular intensity at the root system level. Statistical analyses revealed that both variables were significantly influenced by the cultivar. The proportion of arbuscules in the colonized fragments varied considerably among cultivars: the lowest values were recorded in Djonan F1 (0.29%) and Clemson Spineless (0.50%), whereas the highest value was obtained in Teriman (45.38%). Concerning the arbuscular intensity at the root system scale, the values ranged from 0.08% to 11.61%. The post hoc test also separated the cultivars into two main groups: Teriman and Noura F1, which exhibited high values, and Djonan F1 and Clemson Spineless, which showed low values.

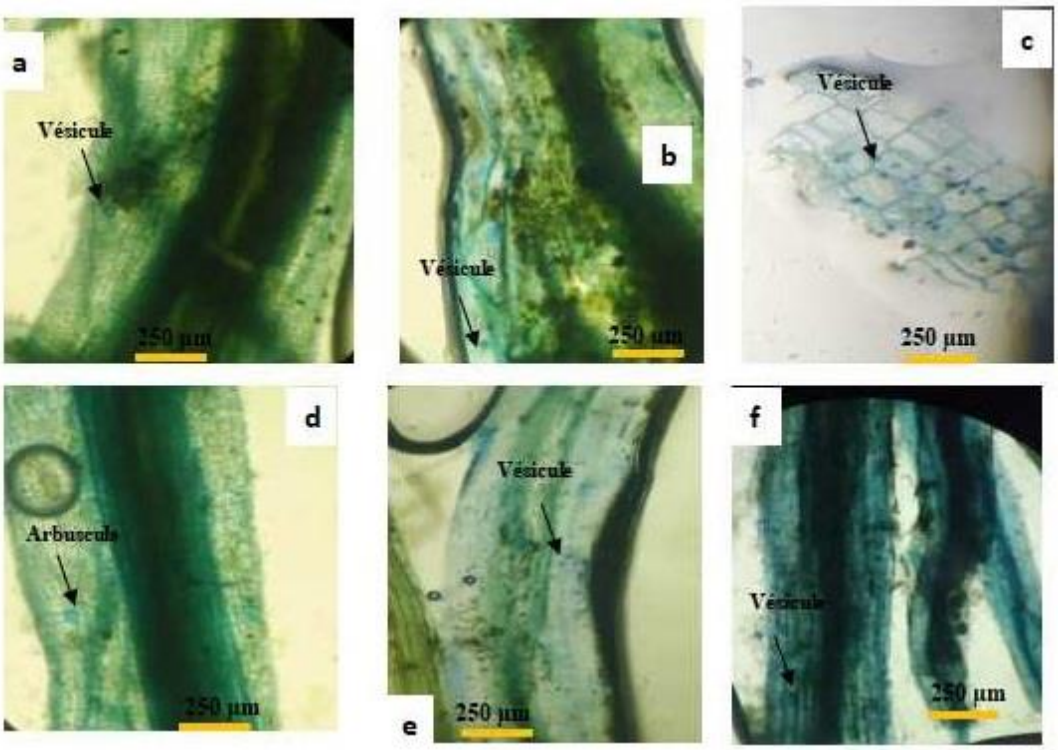


Fig. 7. Mycorrhizal structures in the root sections of different Okra cultivars
a: Teriman F1, b: Djonan F1, c: Clemson spineless, d: Noura F1, e: Kopê F1, f: Divo

Table *2*. Root mycorrhization index according to the cultivars used

Cultivars	F (%)	M (%)	m (%)	a (%)	A (%)
Teriman	87.78 ± 6.19 **a**	23.31 ± 2.79 **ab**	26.42 ± 1.85 **b**	45.38 ± 10.06 **a**	11.10 ± 3.32 **a**
Djonan F1	100.00 ± 0.00 **a**	23.07 ± 4.75 **ab**	23.07 ± 4.75 **b**	0.29 ± 0.29 **b**	0.08 ± 0.08 **b**
Local	77.27 ± 5.32 **a**	11.06 ± 0.45 **b**	14.51 ± 1.53 **c**	39.40 ± 11.74 **a**	4.46 ± 1.44 **ab**
Kopê F1	93.33 ± 6.67 **a**	45.70 ± 7.84 **a**	50.78 ± 12,85 **a**	11.43 ± 4.84 **ab**	5.85 ± 2.96 **ab**
Clemson spineless	97.22 ± 2.78 **a**	38.21 ± 13.10 **ab**	39.15 ± 12.88 **ab**	0.50 ± 0.50 **b**	0.10 ± 0.10 **b**
Divo	55.95 ± 5.95 **b**	10.57 ± 2.37 **b**	20.48 ± 3.57 **b**	21.05 ± 12.77 **ab**	3.19 ± 1.19 **ab**
Noura F1	100.00 ± 0.00 **a**	56.12 ± 7.74 **a**	56.12 ± 7.74 **a**	17.75 ± 2.37 **ab**	11.61 ± 2.84 **a**
Moyennes	**87.71 ± 3.91**	**29.72 ± 4.21**	**56.12 ± 7.74**	**19.39 ± 4.41**	**5.20 ± 1.20**

Values in the same column followed by the same letter are not significantly different at the 5% level (Newman–Keuls test).

F % (frequency of mycorrhization), M % (intensity of mycorrhization in the root cortex), m % (intensity of mycorrhization in colonized root fragments), a % (arbuscular intensity within the colonized portion), and A % (arbuscular intensity in the whole root system)

4.2 Discussion

This study aimed to promote the sustainable production of okra (*Abelmoschus esculentus* L.) through the exploitation of the benefits associated with mycorrhizal symbiosis. The results confirmed the presence of arbuscular mycorrhizal fungi (AMF) in the experimental plot of the Teaching and Research Unit in Plant Physiology and Pathology (UPR-PPV) at Félix Houphouët-Boigny University, the site where the varietal trial was implemented. The study highlighted the morphological diversity of spores from native mycorrhizal communities in the soil and the degree of root colonization of the different okra cultivars tested. The mean spore density recorded (448 spores per 100 g of soil) was relatively high. This could be explained by the absence of mechanical disturbances in the plot before the trial, creating an environment favorable to spore proliferation. It is worth noting that the experimental site had been under fallow conditions for more than 12 years. These values exceed those reported by Jamilou et al*.* (2023), who observed a mean spore density of 110 to 345 spores per 100 g of soil, but they are consistent with the findings of Droh et al*.* (2022) in Côte d'Ivoire on maize cultivation, where high spore density was also observed in minimally disturbed soils. Analysis of spore distribution by size revealed a predominance of spores ranging between 45 and 90 µm (52.30% and 40.11%, respectively). This result can be explained by the relationship between the spore abundance and the mesh diameter of the sieves used. These observations are consistent with the findings of Koffi et al*.* (2021), who reported the dominance of small- and medium-sized spores across all agroecological zones studied in Benin for maize (*Zea mays* L.). Morphological examination of the spores revealed five distinct colorations: black, orange-yellow, hyaline, brown, and yellow, with the orange-yellow spores being significantly more abundant. This colorimetric diversity suggests the co-existence of several AMF morphotypes at the experimental site, in agreement with the observations of Blaszkowski and Czerniawska (2006), who also identified notable morphological variability. From a taxonomic perspective, 10 morphotypes were identified, belonging mainly to the genera *Glomus, Gigaspora, Acaulospora*, and *Scutellospora*. The marked dominance of the genus *Glomus* is consistent with the findings of Abbas (2014), who emphasized its prevalence in tropical soils, and aligns with the conclusions of Droh et al*.* (2023), who showed that the Glomeraceae were widely represented in the Bouaflé and Niellé regions of Côte d'Ivoire. These results differ from those of Jamilou et al*.* (2023), who identified *Scutellospora*, *Gigaspora*, and *Glomus* in tomato cultivation. Nevertheless, they confirmed that *Glomus* represents a ubiquitous and ecologically dominant group in the cultivated soils of West Africa. Microscopic examination of the roots revealed the presence of mycorrhizal structures (hyphae, spores, and arbuscules) in the cultivarsvars studied, confirming their ability to establish mycorrhizal symbiosis, as previously reported by Ndoye et al*.* (2013). Mycorrhization parameters indicated colonization frequencies close to 100% for some cultivars (notably Djonan F1 and Noura F1), with intensities ranging from 10.57% to 56.12%. Although variable across genotypes, this level of colonization remained higher than that observed by Koffi et al*.* (2021) and Bossou et al*.* (2019), who reported mean colonization frequencies of 80% with intensities below 2.47% in the areas studied. The arbuscular colonization intensity of

the root fragments ranged from 14.51% to 50.78%, reflectthe heterogeneity among the cultivarsvars. The highest values were recorded in Teriman and Noura F1, while Djonan F1 and Clemson Spineless showed lower levels. This result reflects the specificity of the plant–AMF interactions, as previously emphasized by Duponnois et al*.* (2013). However, the arbuscule colonization observed here was low (< 5%), in contrast to the study by Koffi et al*.* (2021), who reported arbuscule percentages exceeding 56% in Acacia mangium. These discrepancies illustrate the decisive influence of plant material and edaphic conditions on the establishment of symbiosis. Finally, although Hetrick et al*.* (1992) noted that plant growth is not systematically correlated with the degree of colonization by AMF, the presence of arbuscules, hyphae, and vesicles within the root cortex clearly indicates the presence of vesicular-arbuscular endomycorrhizal fungi (VAMF) in the experimental site. These VAMFsays contribute to the minerall uptake and physiological resilience of okra cultivars. Overall, these results are consistent with recent studies conducted in Côte d'Ivoire (Droh et al*.,* 2022; Droh et al*.,* 2023) and underscore the importance of further exploiting AMF diversity in tropical vegetable production systems to reduce dependence on chemical inputs and promote sustainable agriculture.

5. CONCLUSION

This study revealed the diversity and structure of arbuscular mycorrhizal fungal (AMF) communities associated with okra cultivation in the soils of southern Côte d'Ivoire. The average spore density recorded (4.48 ± 0.24 spores/g of soil) and the predominance of small- to medium-sized spores indicated a significant mycorrhizal potential, dominated by the genera *Glomus*, *Gigaspora*, *Acaulospora*, and *Scutellospora*, with a marked predominance of *Glomus*. The results also confirmed a pronounced inter-varietal variability in response to mycorrhization, reflected in significant differences in the frequency and intensity of root colonization, with superior performances observed in the cultivars Kopê F1 and Noura F1. These findings highlight that mycorrhizal responsiveness represents a discriminating trait for varietal improvement and the development of sustainable cropping practices. However, the low arbuscular intensity recorded (< 5%) suggests that the functional efficiency of the symbioses remains limited, likely due to the complex interactions between soil characteristics, agricultural practices, and cultivar–AMF specificity. Overall, these results underscore the need for integrated strategies combining the selection of mycotrophic varieties, rational input management, and the valorization of mycorrhizal inocula to enhance the efficiency of symbioses and, consequently, the productivity and sustainability of vegetable production systems.

DISCLAIMER (ARTIFICIAL INTELLIGENCE)

Author(s) hereby declare that NO generative AI technologies such as Large Language Models (ChatGPT, COPILOT, etc.) and text-to-image generators have been used during the writing or editing of this manuscript.

COMPETING INTERESTS

Authors have declared that they have no known competing financial interests or non-financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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