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RESEARCH ARTICLE

CHARACTERIZATION OF ARBUSCULAR MYCORRHIZAL FUNGI ASSOCIATED TO THREE PROVENANCES OF PIGEON PEA (CAJANUS CAJAN L.) MILLSP. GROWN IN THREE SOILS FROM TWO DIFFERENT REGIONS IN SENEGAL

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Abstract

Pigeon pea (*Cajanus cajan* L.) Millsp., often referred to as "the meat of poor people," is a valuable underutilized agricultural resource in Africa, particularly in Senegal. This legume offers a diverse range of food and environmental benefits, making it a key element for food security and sustainability of agricultural systems, especially in tropical and subtropical regions. The aim of this study was to evaluate the density and diversity of arbuscular mycorrhizal fungi (AMF) associated to three provenances of pigeon pea cultivated in three soils from two regions in Senegal. Mycorrhizal parameters were determined in the three soil origins; and added to growth parameters after trapping culture with three pigeon pea provenances in these soils for 4 months. The results revealed significant differences among the soil origins and the presence or absence of the plant for AMF density and diversity, and mycorrhizal potential. Seven morphotypes of AMF belonging to five genera (*Rhizoglyphus*, *Scutellospora*, *Dendrospora*, *Gigaspora* and *Glomus*) were identified. Spore density was higher in Kaolack and lower in Kaffrine. The presence of pigeon pea plant increased spore density and mycorrhizal potential. Moreover, significant variations were expressed between the three provenances on the three soils. Spore density and root colonization were higher in Kaolack without cover with Fatick provenance. The best growth was observed with SL provenance in Kaolack without cover and with Kaffrine provenance in Kaffrine under cover. This study highlights the importance of taking into account the characteristics of soil sites for better exploitation of the agronomic and nutritional potentials of each pigeon pea provenance.

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Introduction:-

Out of approximately 50,000 edible plant species known in the world, only around 200 species, are utilized as food. Surprisingly, among those, only 15 crop plants provide 90% of the world's food energy intake. And, unfortunately, more than 50% of the calories consumed worldwide were provided by just three main crops which are wheat, rice and maize (UNSCN, 2020). While, it exists in this category, some neglected or underutilized plant species (NUS) which can constitute a good food supply alternative for diversification, contributing to food and nutrition security, and reduce the negative effects of climate change (Padulosi et al., 2019, Ndlovu et al., 2024). These unexploited plant species largely ignored in agricultural and food systems during a long period, have great potentials in food, nutrition, economy, environment and society. These traditional and orphan plant species that can help alleviate hidden hunger and malnutrition are now recognized as future smart crops by scientists and policymakers (Jairu et al., 2023).

Otherwise, Senegal, like low-income African countries, is affected by malnutrition in all its forms. One of the problems related to diet, namely undernutrition and malnutrition, affects the quality of life of many poor communities, especially women and children. Overall, 17.8% of women of reproductive age, have a chronic energy deficiency and 19% of children under 5 years of age suffer from stunted growth (Thiam et al., 2022; Faye et al., 2020). At the same time, there is a rapid transition from traditional diets based on local foods to a diet high in fat, salt and sugar, but low in fiber and essential micronutrients. However, a good diet not only meets the required level of calorie intake, but also the right balance of foods in the household food basket. Therefore, dietary patterns with a high diversity and variety of elements with a range of micro- and macronutrients are important to ensure nutritional security (Weerasekara et al., 2020). The exploration and exploitation of the potentials of these NUS crops will be a great opportunity and alternative to tackle these challenges.

Among these NUS crops, pigeon pea (*Cajanus cajan*, L.), caught our attention. This plant has enormous potential that is still underexploited in West African countries like Senegal, where this crop species is little known. Although it is the eighth most important legume crop in the world, the proportion of pigeon pea produced in West Africa remains low (Ohanenye et al., 2021). Despite its importance, especially in certain tropical and subtropical regions facing food and nutritional insecurity and abiotic stresses, little information on this crop in terms of genetic diversity, production and uses is available in Senegal. Indeed, pigeon pea is a drought-tolerant, multipurpose crop grown primarily for its edible seeds rich in dietary protein (Musokwa et al., 2021). Due to nitrogen fixation and improved soil fertility enabling mixed or intercropping, pigeon pea plays an important role in dryland farming systems, particularly chosen by smallholder farmers in many parts of the world (Namuyiga et al., 2022). Its ability to improve soil fertility is due to the association with symbiotic soil microorganisms such as nitrogen-fixing bacteria or rhizobia (Araujo et al., 2020; Bopape et al., 2023) and plant growth-promoting bacteria or Plant Growth-Promoting Rhizobacteria (PGPR) (Harshitha et al., 2020). This legume plant is also able to form symbiotic association with arbuscular mycorrhizal fungi to improve its growth and productivity (Dania & Fagbola, 2022). In fact, Arbuscular mycorrhizal fungi (AMF) might be considered as biostimulant and biocontrol agents of most terrestrial plants (Soudzilovskaia et al., 2020; Delaeter et al., 2024). The latter are particular soil microorganisms able to form symbiotic association with most terrestrial plants to improve plant growth and productivity through tolerance to numerous abiotic and biotic stresses (Wang et al., 2024a; Navarro & Morte, 2024). AMF can play a protector role against plant pathogens by improving photosynthesis and plant nutrition (Balestrini et al., 2020); and by modulating root microbiota through their extra-radical mycelium (Pérez-De-Luque et al., 2017). Recent studies had shown positive effects of AMF on pigeon pea growth and yield (Mathimaran et al., 2020; Yadahalli et al., 2022).

Little attention has been given to pigeon pea culture with its beneficial microbial environment and its agronomic potential in most African countries. In this study, we investigated to understand the relationship between pigeon pea and arbuscular mycorrhizal fungal community in their given environmental conditions. We hypothesized that, environmental conditions modulated by soil properties and geographical distance affected AMF density and diversity whatever the seed provenance.

Material and Methods:-

Study site description, soil sampling and physicochemical characterization:

In Kaolack, the study site is located in the community of Sing Sing. The geographical coordinates are 14°10'0" N and 16°7'60" W. Sing Sing has a hot and dry desert climate according to the Köppen-Geiger classification. The average temperature in June is around 29.8°C and the average rainfall is around 630 mm. At night, temperatures

drop to 24.6°C and during the day they can reach 37.2°C. In Kaffrine, the study site is located in the community of Sagna in the department of MalemHodar (14°05'12.5"N 15°23'00.0"W). The temperature is between 26°C and 39°C during the day. The annual rainfall is around 860 mm.

In Kaolack without cover, three sampling plots with 20 m of distance, were delineated in a field. In each plot, 4 points forming a 25 m square, completed by a fifth point in the center were considered for soil samples. The 5 soil samples taken at 0-20 cm depth were mixed to get a composite soil sample; giving a total of 3 composite soil samples. In Kaffrine, soils were sampled without cover and under pigeon pea plants. In Kaffrine without cover, the same approach as in Kaolack was adopted to get three composite soil samples. In Kaffrine under pigeon pea, soils were sampled in a farm field. Three plots containing plants were considered. In each plot, five plants with a distance of 5 m were chosen and soils were sampled in three points around the plant, then pooled. The soil samples of the five plants in each plot were mixed to get a total of three composite soils. The distance between plots was 10 m. All soil samples put in plastic bags were kept to the Laboratory for analyzes. It should be noted that we have not found any pigeon pea plants in Kaolack, which is why we do not have any undercover soil in this site. However, we have a prospect of introducing pigeon pea into the Kaolack site.

The physicochemical analyzes of soil were done at the Laboratory LAMA (Laboratoire des Moyens Analytiques, IRD, Dakar, Senegal). The soils were air dried at room temperature, ground to pass through a 2 mm sieve, and analyzed following the standard protocols. Soil organic carbon was estimated following the method of Bahadori & Tofghi (2017) by the Walkley-Black oxidation. Available P was determined using Olsen method (Battisti et al., 2021). Total N was determined using the Kjeldahl method (Hicks et al., 2022).

Evaluation of soil mycorrhizal potential from the different sites:

The most probable number (MPN) is a method that expresses the number of infective AMF propagules present in a soil and able to colonize roots of a plant. This procedure consists to mix the composite soil from each site with a sterilized sand in a dilution series of 0, 1/4, 1/16, 1/64, 1/256 and 1/1024 (Gagou et al., 2023). Each dilution was repeated five times and the five pots of each, containing 50 g of the diluted soil were used as substrate to grow seeds of maize plant (*Zea mays* L.), a highly mycorrhizal dependency plant. Plants were watered with demineralized water for 45 days. At the end of the assay, roots of all plants were extracted, cleaned and stained with Trypan blue as described in Ndoye et al. (2024). The roots were then cut into 1 cm segments and placed between a microscope slide in a glycerol drop and a cover slip for examination with a light microscope at a magnification of 100X. The MPN of AMF propagules that can colonize plant roots was calculated as follows: $\text{Log MPN} = x \log a - k$, where x represents mean of mycorrhized plants for all dilution ratios, a (factor of dilution) = 4, K= constant given by the table of Fisher & Yates (1970).

Determination of AMF spore density and diversity from field-collected soils:

AMF spore extraction was performed according to the wet sieving and decanting method as described in Ndoye et al. (2024). A quantity of 100 g of each dried-soil was placed in a 2L glass beaker, filled with tap water and agitated with a spoon. Then, the soil suspension was poured onto a series of 400-50 µm sieves, re-suspended in tap water and collected in 50 ml conical tubes. Centrifugation at 3000 g/min for 3 minutes was made after adding two solutions of sucrose at 20% and 60%. At the end, the supernatant was collected, passed through a 50 µm sieve, washed with tap water and placed in Petri dish (Sakha et al., 2024). AMF spore observation, description and identification were done under a dissecting microscope using the International Culture Collection of Arbuscular and Vesicular-Mycorrhizal Fungi for AM fungi description (<https://invam.wvu.edu/methods/spores/enumeration-of-spores>). The AMF spore density and abundance of each AMF species were expressed per 100 g of soil. Three replicates were made for each composite soil from each sampled site.

Characterization of AMF spore composition after trap culture:

A trapping culture method was performed to reveal the diversity of all AMF species in soil sites under pigeon pea plants. This method consisted of growing pigeon pea seeds in pots containing the sampled soils from each site. Three pigeon pea seed provenances (Saint Louis : SL, Fatik : FK and Kaffrine : KF) were used with two replicates per provenance in each soil site (Kaolack and Kaffrine). The seeds were pregerminated in agar Petri dish during 3 days before sowing. A total of 18 pots was displayed and watered until five days before the end of the experiment in order to stress the plants. This temporary period of water stress stimulates fungi adaptation to drastic conditions by favoring AMF spore production for its survival.

Assessment of plant growth and mycorrhization parameters after trap culture:

After 4 months of nethouse cultivation, the height and collar diameter of pigeon pea plants were measured, as well as the chlorophyll content of the leaves. The chlorophyll concentration was measured using the SPAD 502 Plus chlorophyll meter (Uddling et al., 2007).

The plant roots were also collected, rinsed with tap water, and kept cool to determine mycorrhization parameters (frequency and intensity of mycorrhization). For this, the plant roots were cleared with 10% KOH and stained with 0.05% Trypan blue solution as described by Ndeko et al. (2024a). Root fragments were then mounted between slides, coverslips in a glycerol drop and observed under a light microscope at x400. The frequency and intensity of mycorrhization were determined using the method of Trouvelot et al. (1986) as followed:

Frequency of mycorrhization (F%) = (number of mycorrhizal fragments / total number of observed fragments) x 100

Intensity of mycorrhization (M%) = $(95n_5 + 70n_4 + 30n_3 + 5n_2 + n_1) / N$

n_5, n_4, n_3, n_2 and n_1 respectively denoted the number of fragments of class 5, 4, 3, 2 and 1, and N was the number of fragments observed.

The soils were collected in plastic bags and stored in a cold room at 4°C for determination of AMF spore density and diversity, as previously described.

Statistical analysis:

Shapiro and Levene tests were performed on raw data to verify normality and homogeneity of variance. Normalized data were subjected to one- or two-way analysis of variance (ANOVA). Means of variables measured at the 5% probability level ($p \leq 0.05$) were compared using the Student Newman-Keuls test. Mycorrhization parameter values were transformed into arcsine square root of the value before statistical analysis. Tests and statistical analysis of the results were performed with XLSTAT software version 2010. Principal component analysis (PCA) method was performed for the relationship between plant and soil mycorrhizal parameters after trapping culture.

Results:-

Soil physicochemical properties:

The soil of Kaolack is richer in sand and clay than that of Kaffrine with 84% and 72.3% sand, and 7.9% and 4.5% clay, respectively (Table 1). However, the silt content is higher in Kaffrine (23.1%) than in the soil of Kaolack (7.31%). Contrary to the Nitrogen content, the organic Carbon and available Phosphorus contents are slightly higher in the Kaffrine soil.

The Magnesium, Calcium and Sodium contents are more important in Kaffrine and the Potassium content is practically not different between the two soils. The pH is slightly acid in Kaffrine soil (6.40) and slightly alkaline in Kaolack soil (7.83).

Table 1:- Physicochemical characteristics of soils without cover from Kaolack and Kaffrine

Soil components	Kaolack soil	Kaffrine soil
Clay (%)	7.90	4.50
Silt (%)	7.31	23.10
Sand (%)	84.40	72.30
Carbon (g/kg)	5.10	6.80
Available Phosphorus (mg/kg)	3.47	4.40
Total Nitrogen (g/kg)	0.43	0.30
Potassium (Cmol/kg)	4.60	4.50
Sodium (Cmol/kg)	0.35	1.30
Magnesium (Cmol/kg)	0.04	3.90

Calcium (Cmol/kg)	0.07	21.10
pH (H ₂ O)	7.83	6.40

Soil mycorrhizal potential infectivity:

The results on the most probable number of mycorrhizal propagules (MPN) in the soils from Kaolack and Kaffrine are shown in Figure 1. It indicated that this MPN was significantly higher in the kaffrine soil under cover (256 propagules/100g of soil), followed by that of the Kaolack soil without cover, and lower in the Kaffrine soil without cover (10 propagules/100g of soil).

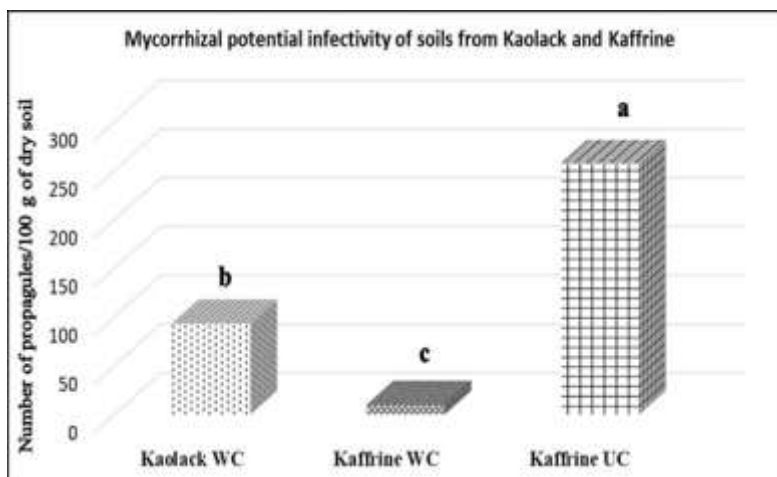


Figure 1:-Mycorrhizal potential infectivity (MPN) of soils from the two sites (Kaolack and Kaffrine)

WC: soil without cover, UC: soil under cover. Bars followed by the same letter were not significantly different according to the Student-Newman-Keuls test at the 5% threshold

AMF species diversity and spore density in soils from the study sites:

Results presented in Figure 2 revealed a total of 7 AMF morphotypes belonging to 5 genera (Racocetra, Dendiscutata, Scutellospora, Gigaspora and Glomus) and 2 families (Gigasporaceae and Glomeraceae) in the three soil origins.

The spore density of the AMF species (Table 2) was higher in the non-covered soil of Kaolack for 6 of the 7 species (Racocetra aff. gregaria, Dendiscutata sp., Scutellospora sp., Gigaspora aff. gigantea, Glomus sp1 and Glomus sp3). This AMF spore density was lower in the Kaffrine under-covered soil for 4 of these 7 species, namely Racocetra aff. gregaria, Scutellospora sp., Gigaspora aff. gigantea and Glomus sp1. It should be noted that the genus Glomus with the species Glomus sp3 was the most represented in the different soils.

Otherwise, the total AMF spore density was significantly higher in the Kaolack soil without cover (99 spores/100g of soil) than those in the Kaffrine soil under cover (49 spores/100g of soil) and Kaffrine soil without cover (45 spores/100g of soil), which were not significantly different between them.

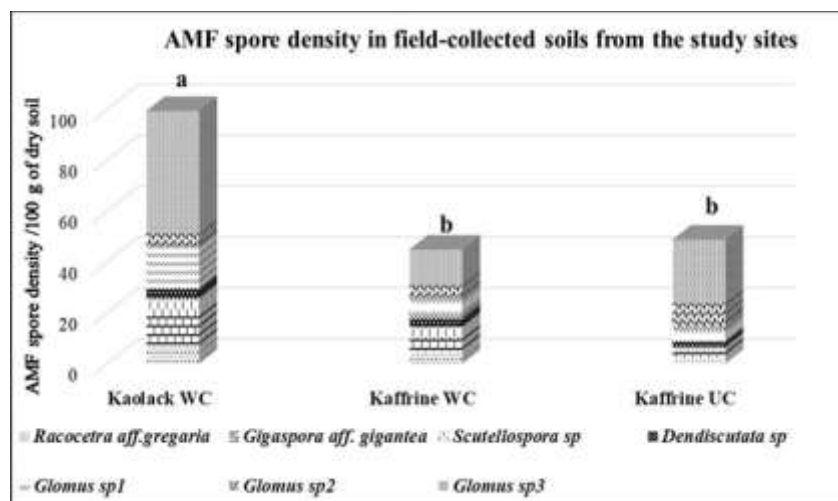


Figure 2:- AMF spore density in the three soil origins from the two study sites

WC: soil without cover, UC: soil under cover. Bars followed by the same letter were not significantly different according to the Student-Newman-Keuls test at the 5% threshold

Table 2:-Diversity of AMF species in soils from different origins

Families	Genera	AMF species	Kaolack WC	Kaffrine WC	Kaffrine UC
Gigasporaceae	Racocetra	Racocetraaff.gregaria	7 a	5 ab	2 b
	Gigaspora	Gigasporaaff. gigantea	12 a	6 b	3 b
	Scutellospora	Scutellosporasp	6 a	4 b	1 b
	Dendiscutata	Dendiscutatasp	4 a	3 a	3 a
Glomeraceae	Glomus	Glomus sp1	17 a	8 b	6 b
		Glomus sp2	5 b	5 b	9 a
		Glomus sp3	48 a	14 c	25 b
	Total number of AMF morphospecies		7	7	7
	Total AMF spore density		99a	45b	49b

WC: soil without cover, UC: soil under cover. In row, values followed by the same letter are not significantly different according to the Student-Newman-Keuls test at the 5% threshold

AMF spore composition after trap culture with three provenances of pigeon pea:

A total of 7 morphospecies of AMF (*Racocetraaff. gregaria*, *Dendiscutatasp*, *Scutellosporasp*, *Gigasporaaff. gigantea*, *Glomus sp1*, *Glomus sp2* and *Glomus sp3*) belonging to 5 genera

(*Racocetra*, *Dendiscutata*, *Scutellospora*, *Gigaspora* and *Glomus*) and 2 families (*Gigasporaceae* and *Glomeraceae*) were found in the Kaolack soil without cover with the 3 pigeon pea provenances (Table 3). These 7 AMF species were encountered with the 3 provenances in Kaolack without cover; with Saint Louis (SL) provenance in Kaffrine without cover and with Fatick (FK) provenance in Kaffrine under cover. Among these 7 morphospecies, 6 were recorded with KF (Kaffrine) and Fatick (FK) provenances in Kaffrine soil without cover, and with SL and KF provenances in Kaffrine soil under cover. In addition, *Glomus sp2* was not observed in the Kaffrine soil without cover, with the KF provenance.

For the SL pigeon pea provenance, the density of *Glomus sp3* under cover was dominant. They were followed by *Scutellosporasp* and *Dendiscutatasp* in Kaolack, *Racocetrasp* and *Gigaspora* in Kaffrine under cover; and finally, *Dendiscutatasp* and *Racocetrasp* in Kaffrine without cover.

For the FK pigeon pea provenance, spore density was higher with *Glomus* sp3 in Kaolack without cover and Kaffrine under cover; and with *Glomus* sp1 in Kaffrine without cover. They were followed by *Dendiscutatasp* and *Scutellosporasp* in Kaolack, *Gigasporasp* and *Dendiscutatasp* in Kaffrine under cover (UC); and finally, *Gigasporasp* and *Dendiscutatasp* in Kaffrine without cover (WC).

For the KF pigeon pea provenance, the density of *Glomus* sp3 and *Scutellosporasp* predominated in Kaolack, followed by those of *Glomus* sp3 and *Dendiscutatasp* in Kaffrine without cover; and finally, those of *Glomus* sp3 and *Glomus* sp1 in Kaffrine under cover.

Furthermore, the total density of AMF spores was significantly higher in Kaolack soil and lower in Kaffrine soil without cover, regardless of the pigeon pea provenance (Figure 3). This spore density was higher with FK provenance in Kaolack WC and Kaffrine UC. The lowest spore density was obtained with SL provenance in Kaolack WC, with SL and FK provenances in Kaffrine UC, and with KF provenance in Kaffrine WC.

Mycorrhizal root colonization of pigeon pea plants after trap culture :

The results on mycorrhization parameters of pigeon pea root plants from the 3 provenances grown in the three soils were reported in Table 4. They revealed that whatever the seed provenance, frequencies and intensities of mycorrhization were significantly higher in Kaolack soil without cover and Kaffrine soil under cover, compared to those in Kaffrine soil without cover. Furthermore, in Kaolack soil WC, the best values were recorded with KF and FK provenances, whereas in Kaffrine soil under cover, the frequencies and intensities of mycorrhization were higher in root plants of SL provenance and lower with those of FK provenance. In Kaffrine soil without cover, the provenance FK gave the greatest values of mycorrhizal parameters and the provenance KF the lowest values.

Table 3:- Density of AMF species in soils from the different origins cultivated with three pigeon pea provenances

Families	Genera	Species	Kaolack WC			Kaffrine WC			Kaffrine UC		
			SL	FK	KF	SL	FK	KF	SL	FK	KF
Gigasporaceae	Racocetra	Racocetraaff.gregar ia	2b	5a	2b	3ab	2b	1b	3ab	1b	2b
	Dendiscutata	Dendiscutatasp	8b	18a	6bc	5bc	5bc	2c	2c	2c	6bc
	Scutellospora	Scutellosporasp	8b	15a	17a	2c	0c	0c	0c	1c	0c
	Gigaspora	Gigasporaaff.gigant ea	2ab	5a	3ab	1b	5a	1b	4ab	4ab	2ab
Glomeraceae	Glomus	Glomus sp1	20a	20a	15bc	5c	7c	4c	5c	6c	4c
		Glomus sp2	3cd	7b	9a	3cd	1d	0d	2cd	2cd	4c
		Glomus sp3	15b	25a	17b	6cd	5d	7c	16b	19b	11c
Total number of morphospecies			7	7	7	7	6	6	6	7	6
Total spore density			58c	95a	68b	25d	25d	15e	32d	35d	29d

WC: soil without cover, UC: soil under cover, SL: pigeon pea provenance from Saint Louis, FK: pigeon pea provenance from Fatick, KF: pigeon pea provenance from Kaffrine. In row, values followed by the same letter are not significantly different according to the Student-Newman-Keuls test at the 5% threshold

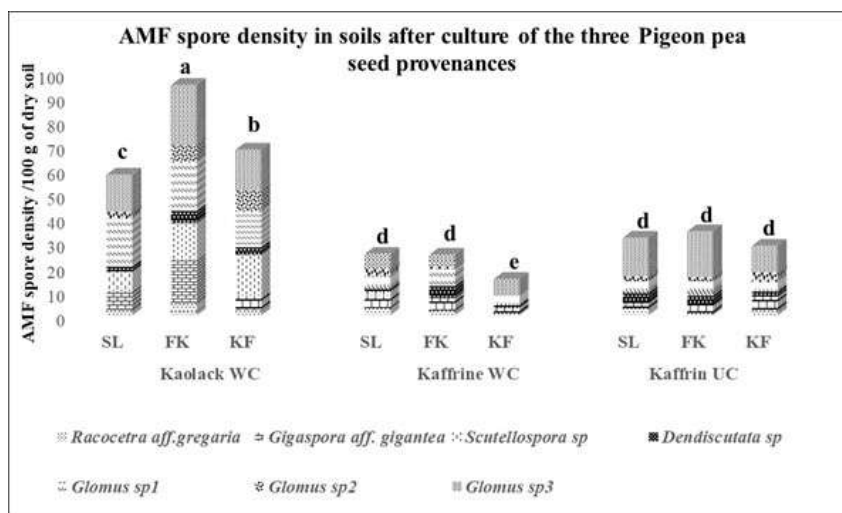


Figure 3:-AMF spore density in soils after culture of the three provenances of pigeon pea

WC: soil without cover, UC: soil under cover, SL: pigeon pea provenance from Saint Louis, FK: pigeon pea provenance from Fatick, KF: pigeon pea provenance from Kaffrine. Bars followed by the same letter were not significantly different according to the Student-Newman-Keuls test at the 5% threshold

Plant growth parameters after trap culture:

The results (Table 4) revealed that the height was higher in plants grown in Kaffrine soil under cover, followed by those in Kaolack soil without cover and significantly lower in those in Kaffrine soil without cover. However, no significant difference was noted between pigeon pea provenances regardless of the soil origin. Nevertheless, the height was higher in plants from SL provenance in Kaolack and Kaffrine soil without cover; and in those from FK provenance in Kaffrine soil under cover.

Statistical analysis showed a significant effect of the soil site on plant collar diameter with the highest values in Kaolack soil without cover and Kaffrine soil under cover. However, in the same soil, the significant effect of the provenance is less marked. Indeed, the provenance KF seemed to dominate in Kaffrine under cover with an average diameter of 3.2 mm.

Leaf chlorophyll content was significantly higher in plants from the 3 provenances grown in Kaolack soil without cover and Kaffrine soil under cover (between 37% and 50%); compared to those in Kaffrine soil without cover (around 22%). However, in the same soil, no significant effect of provenance on leaf chlorophyll content was noticed, even if it was higher with SL provenance in Kaolack WC and with FK provenance in Kaffrine UC.

Table 4:-Growth and mycorrhization parameters of pigeon pea plants in the different soils

Soil origins	Pigeon pea provenances	Height (cm)	Collar diameter (mm)	Chlorophyll content (%)	Frequency of mycorrhization (%)	Intensity of mycorrhization (%)
Kaolack	Saint Louis (SL)	36.50a	2.41b	46.34a	71.11a	19.82a
	WC					
	Fatick (FK)	28.00ab	2.14b	43.22a	85.70a	29.25a
	Kaffrine (KF)	27.30ab	2.44b	37.90a	86.10a	25.44a
Kaffrine	Saint Louis (SL)	26.80ab	1.50b	22.41b	33.87b	5.13b
	WC					
	Fatick (FK)	24.78b	1.67b	22.34b	37.23b	5.27b
	Kaffrine (KF)	22.84b	1.75b	22.18b	25.62b	3.04b

Kaffrine UC	Saint Louis (SL)	34.50ab	2.34b	43.71a	88.30a	35.50a
	Fatick (FK)	31.18ab	2.25b	49.80a	71.13a	22.61a
	Kaffrine (KF)	37.92a	3.19a	37.62a	82.24a	31.52a

WC: soil without cover, **UC:** soil under cover. In column, values followed by the same letter are not significantly different according to the Student-Newman-Keuls test at the 5% threshold

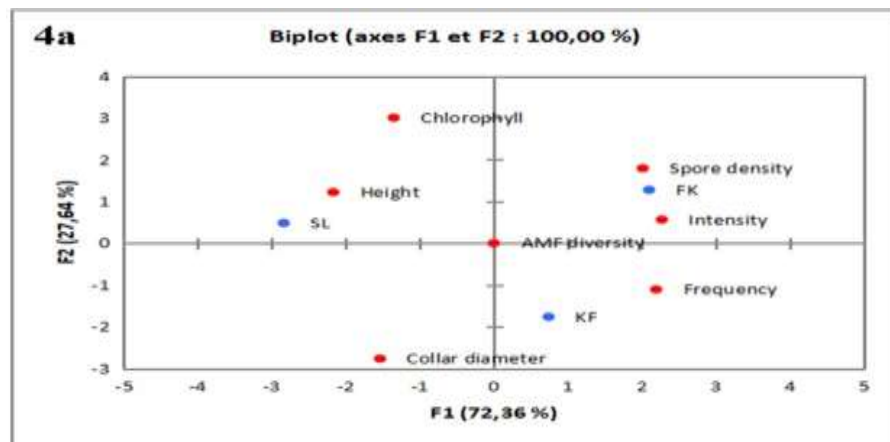
Coorelation between mycorrhizal and plant growth parameters after trapping under the three soils by principal component analyzis (PCA)

Whatever the soil origin, the PCA analyzes based on AMF mycorrhization and plant growth parameters (Figure 4) showed differences between pigeon pea provenances.

In the soil of Kaolack without cover (Figure 4a), the first two axes explained the totality of the variability. The first axis encompassed 72.36% of the total variation. This later is strongly and positively correlated to mycorrhizal parameters and strongly negatively correlated to plant height. The second axis expressed 27.64% of the total variation and is strongly positively related to plant collar diameter and leaf chlorophyll content. The PCA revealed a clear separation between the three provenances. The provenance Fatick at the upper right of the PCA had the greatest values of AMF spore density and root intensity of mycorrhization whereas the provenance Saint Louis at the upper left of the axes had the highest values of plant height and leaf chlorophyll content. The provenance Kaffrine with the greatest values of root frequency of mycorrhization was at the bottom right of the axes.

In the soil of Kaffrine without cover (Figure 4b), the first two axes of the PCA expressed 100% of the variability. A clear separation bewteen the three pigeon pea provenances, was also observed through the two axes. The axis F1 which explained 83.81% of the variability was strongly positively correlated to mycorrhizal and growth parameters except collar diameter which was strongly negatively correlated to the second axis. The first axis strongly discriminated the Saint Louis provenance in its right with the highest values of AMF diversity, plant height and leaf chlorophyll content; and the Kaffrine provenance in its left. The second axis showed a clear separation between Saint Louis provenance in the upper right and Fatick provenance in the bottom right with the greatest values of root colonization rates.

In the soil of Kaffrine under cover (Figure 4c), the total variance was explained by the first two axes. The first axis which encompassed 78.88% of the total variability was strongly positively correlated to AMF spore density and diversity and leaf chlorophyll content, and strongly negatively correlated to root mycorrhizal colonization and plant height. The second axis expressing 21.12% of the total variance was strongly negatively correlated to plant collar diameter. Along the first axis, the provenance Fatick in the upper right with the greatest values of AMF spore density, AMF diversity and leaf chlorophyll content was clear separated in one hand to the provenance Kaffrine in the upper left with the highest values of height and collar diameter. In the the other hand, they were distinguished to the provenance Saint Louis in the bottom left with the best values of root mycorrhizal colonization rates.



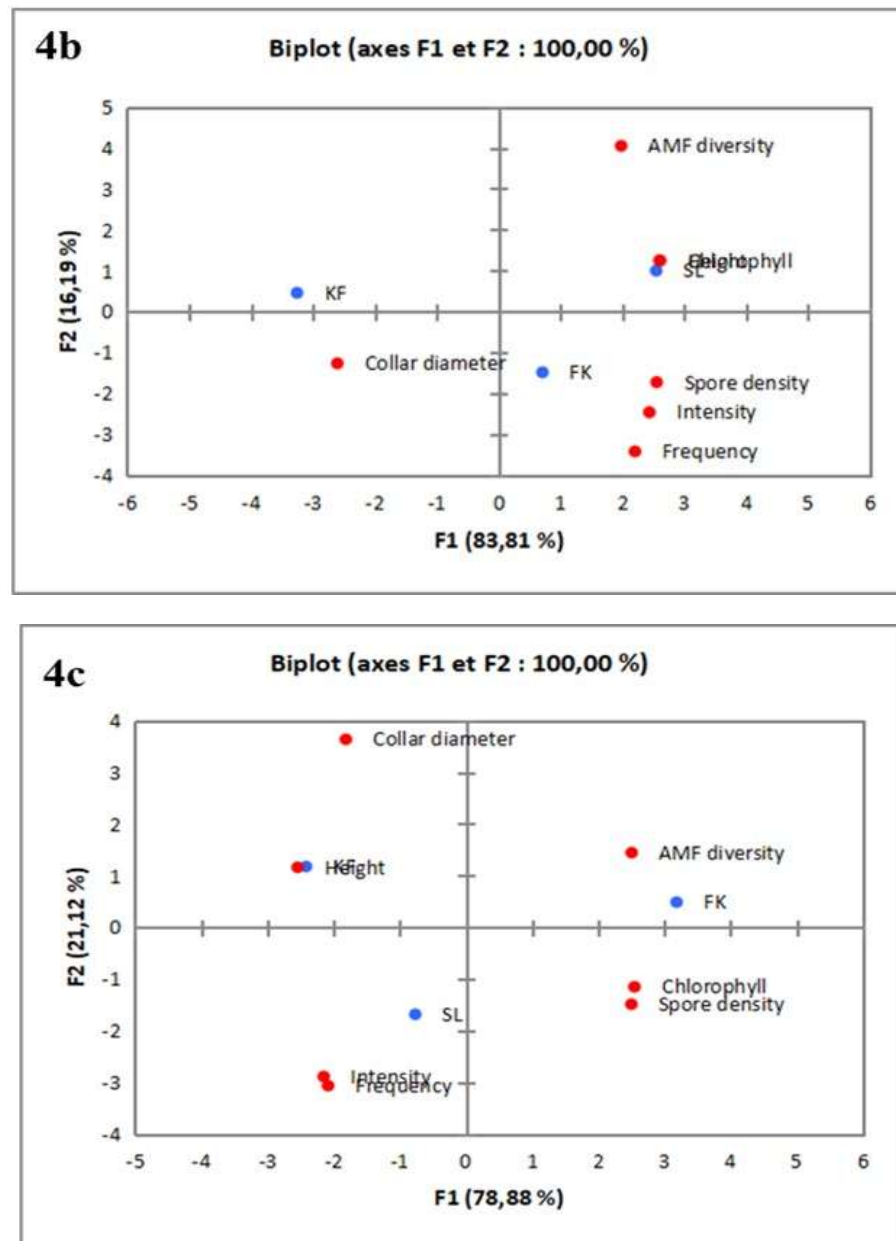


Figure 4:- PCA analyzes with Mycorrhizal and plant growth parameters after trapping under the three soils: Kaolack WC (4a), Kaffrine WC(4b) and Kaffrine UC (4c)

SL (provenance Saint Louis), FK (provenance Fatick), KF (provenance Kaffrine)

Discussion:-

To our knowledge, this study is the first one that aims to determine the diversity of AMF associated to different provenances of pigeon pea in different regions of Senegal.

We assessed key aspects of the AM fungal community, namely its density and diversity and also soil mycorrhizal infectivity of three soil origins ; as well as the AM colonization rate and growth of three pigeon pea provenances in these soils.

Soil mycorrhizal potential:

The results revealed that pigeon pea crop increased the soil mycorrhizal potential infectivity compared to the soils without cover. This was consistent with numerous studies on the positive effect of crops on soil fertility and soil-plant health (Tosi et al., 2023; Kabalan et al., 2024). Arruda et al. (2021) proved that cover crop might increase AMF inoculum potential for the succeeding crop in rotation or intercropping. Otherwise, in this present study, the soil mycorrhizal infectivity differs from one site to another. Similar results were obtained by Ech-Cheddadi et al. (2019) in the rhizosphere of sugarcane in the Gharb region of Morocco. Benelhadj et al. (2024) also found differences in mycorrhizal infectivity potential between three soils. Our results also emphasized that soil mycorrhizal potential is not only under the control of its AMF spore density but depends more on the quality of these spores and their capacity for infectivity. This fact was also underlined by Meddich et al. (2015).

AMF diversity and spore abundance in soil origins:

Based on morphological criteria, seven morphospecies belonging to five genera were recorded in this study. The same tendencies were revealed in different zones of Senegal, a Sahelian area, by several studies (Diop et al., 2021; Thiocone et al. 2023; Ndoye et al., 2024). Contrastingly, some studies have shown a high diversity of AMF in soils from tropical regions (Zhang et al., 2022; Wang et al., 2024a). It means that other factors might contribute to explain observed differences. For instance, Hazard et al. (2013) found that AMF community composition was strongly dependant on abiotic variables (soil type, rainfall, ...) and not on geographical distance or land use; whatever the plant species. Differences might also result from the AMF species preference to various host plants, abiotic factors and agroecological variations (Masebo et al., 2023).

From this work, it also emerged that the same AMF species were found in the three soils even if differences were observed in physicochemical characteristics. Similarly, no differences were found by Pereira et al. (2022) in AMF species richness between natural rainforests and adjacent sugarcane plantations. However, contrasting results showing the influence of physicochemical properties were recorded by Dilsiz et al. (2021) in some pastures of Turkey.

In addition, Gigasporaceae family was predominant whatever the soil origin. This was consistent with the study of Sakha et al. (2025) in tropical regions. This result was confirmed by those of Vieira and coworkers (2020) who found the predominance of Gigasporaceae family in sandy soils with low nutrient contents, organic matter and humidity. Moreover, Glomus was the dominant with three species and high spore densities in the different soil origins. It might be the result of its strong adaptability to stresses environments and its widest habitat (Noreen et al., 2023; Masebo et al., 2023). For instance, Yuwati et al. (2020) found the dominance of Glomus species in soils under the Galam trees at shallow peat of South Kalimantan (Indonesia).

Otherwise, it was demonstrated that AMF communities are mainly influenced by plant cover (Pérez-Redondo et al., 2025). We found a positive effect of pigeon pea plant on AMF spore density when compared to the non-cultivated soil. Similar results were highlighted by Egboka et al. (2022a) in one of their two locations in Nigeria with Cassava and Pineapple. In fact, the presence of vegetation cover is favorable for AMF reproduction (Birhane et al., 2020), as it depends on the host plant to complete its life cycle. This is also consistent with the findings of Masebo et al. (2023) showing greater AMF abundance in presence of plants cultivated in soil. In addition, soil carbon improvement might contribute to the increase in AMF spore abundance (Verbruggen et al., 2021). Otherwise, low soil fertility might induce AMF hyphae development in order to seek nutrients from host plants, thereby reducing their spore density (Rini et al., 2021).

Between the two locations, results revealed a significant difference in spore density in the two soils without cover as it was found by Pérez-Redondo et al. (2025). The highest AMF spore density was recorded in Kaffrine soil under pigeon pea plants compared to the soils without cover. It might be accredited to abiotic and biotic factors like soil physicochemical properties and/or climatic characteristics (Olguera et al., 2023). For instance, AMF spore density was higher in Kaolack soil with less available P content compared to Kaffrine soils. Noreen et al. (2023) also found an inverse correlation between AMF composition and P soil level. Else, soil from Kaolack which is rich in sand and clay contained the greatest number of AMF spores; whereas that of Kaffrine which is sandy-silt contained the highest amounts of available phosphorus and carbon. Differences in rainfall of the two locations (Kaolack and Kaffrine) might contribute to explaining the differences in spore density, Kaffrine receiving more rain than Kaolack. Otherwise, compared to numerous studies, low numbers of AMF spores were discovered in the soils from the different origins in this present work. Our results were in concordance with those of Egboka et al. (2022b) reporting

AMF spore densities between 39 to 71 spores 100 g^{-1} soil in three land use types and soil depths at two locations of southern Nigeria. Olguera et al. (2023) reported very low spore density (between 4 and 12 spores 100 g^{-1} soil) in different vegetation patches of a marginal Upland in Central Philippines. However, contradictories results were obtained by Belay et al. (2013) in soil from Acacia tree species in Ethiopia and Dare et al. (2012) in yam cropping systems in Nigeria with spore density that attended 1506 100 g^{-1} soil and 529 100 g^{-1} soil, respectively. The level of soil disturbance might also justify the differences in spore density between ecosystems.

AMF spore diversity and density after Pigeon pea trap culture:

In trap culture, AMF spore diversity varied slightly between pigeon pea provenances in Kaffrine soil whereas none difference was observed in Kaolack soil. This result might not be directly attributed to plant genotype; but rather to abiotic factors like soil physicochemical characteristics (Guzman et al., 2021). Similar results were observed by Musyoka et al. (2023) on Calotropis rhizosphere under two different locations in Kenya.

Our results showed variation in spore density between the three provenances regardless the site location. Plant seed genotype might contribute explaining this result. Compared to soil origins and whatever the location, spore density was decreased by trap culture in the present work. This might be explained by many factors such as culture conditions and age of plants in trap culture. This situation might be the result of the attitude of AMF in trap culture where they opte instead to germ and colonize roots, reducing their spore density in soil. This finding was not in agreement with the results of Husein et al. (2022) showing an increase in spore density after trapping with two plant species (Sorghum and Corn) under three different soil rhizospheres (Bambusa sp, Cichorium intybus and Pinus merkusii). AMF diversity was also decreased by trap culture in Kaffrine soils. Comparable results were reported by Tenzin et al. (2022) showing a loss of AMF diversity with the first Maize and Sorghum trapping culture. These authors demonstrated that under glasshouse conditions, one trap culture was not sufficient for AMF spore propagation. Bencherif et al. (2023) also denoted a loss of diversity after Alfalfa trap culture; and designated the root mycorrhizal rate as the best suitable criterion to evaluate the quality of AMF inoculum.

Pigeon pea root mycorrhizal colonization and growth after trap culture:

Regarding root colonization rate after trap culture, results revealed an influence of soil origin whatever the seed provenance. This was consistent with the results of Chafai et al. (2025) who observed differences in root mycorrhizal colonization rates of leek plants between four soil origins in Morocco after trapping. Abiotic factors such as physicochemical properties might justify these results (Masebo et al. 2023; Djotan et al., 2024).

In addition, in each soil origin, mycorrhizal colonization rates varied between seed provenances. However, site effect on mycorrhizal colonization was greater than that of seed provenance. This is in agreement with findings of Versuliené et al. (2024) on 10 wheat varieties cultivated under four pedoclimatic sites in Europe. It also aligned with the results of Xavier Martins and Rodrigues (2020) on different rice ecosystems and varieties. Our results confirmed these findings showing the strongest influence of site and climate on mycorrhizal colonization compared to that of genotypes (Meyer et al., 2021).

A positive effect of pigeon pea plant on root colonization was observed in Kaffrine site. It was assumed that vegetation has a greatest effect on soil biological properties such as colonization rate. It approved the results of Tyburska-Woś&Kieliszewska-Rokicka (2024) showing the influence of site on the root mycorrhizal frequency of Aesculus hippocastanum L.

Relationship between mycorrhization and plant growth parameters:

Our results revealed a positive correlation between root mycorrhizal parameters and AMF spore density and diversity in bulk soils of Kaolack and Kaffrine; whereas the latter was negative in Kaffrine under cover. This indicates that the presence of plant promotes the germination of AMF spores which will infect the roots improving their mycorrhization and reducing soil spore density. This result also underlined that, at least, one or more AMF species present in these soils were able to form a symbiotic association with pigeon pea root systems. A part of our findings was in agreement with those of Birhane et al. (2020) demonstrating a positive and significant correlation between root mycorrhizal colonization and spore density of woody species in a dry afro-montane forest in Northern Ethiopia. It was also reported by other researchers in different environments and host plants (Liang et al., 2021; Mohammed Ali et al., 2023). The study of Musyoka et al. (2023) also indicated no or negative relationship between mycorrhizal parameters. It was the case of Salim and coworkers (2020) who found that AMF root colonization had a negative relationship with the number of AMF spores in the soil.

Otherwise, the relationship between AMF spore density and diversity and plant growth was negative in the three soils except for chlorophyll content in Kaffrine soils and height in Kaffrine bulk soil. In contrast, some research highlighted the inverse relationship between AMF spore density and diversity and plant growth in tropical ecosystems (Wang et al., 2024b; Ndeko et al., 2024b). According to Holste et al. (2016), this situation might be the consequences of trees growing in low fertility soils combined with a proliferation of inefficient AMF. Our results were partially supported by Anandakumar and Kalaiselvi (2022) who found a positive correlation between AMF spore abundance and plant growth in *Vigna mungo* (L.) Hepper. Da Silva da Souza et al. (2023) also found the influence of AMF density on the growth of *Handroanthusserratifolius* seedlings and underlined the importance to determine the appropriate spore density in order to assess the symbiotic relationship between AMF and forest plants.

In addition, a positive correlation was noted between root mycorrhizal colonization and plant height in Kaffrine soils compared to Kaolack soil. Chen et al. (2022) reported the same tendency that might suggest the beneficial effect of AMF on plant growth in this context. In contrast, root mycorrhizal colonization was negatively correlated to plant height only in Kaolack bulk soil; to chlorophyll content in Kaolack bulk soil and Kaffrine under cover; and to collar diameter in Kaolack and Kaffrine bulk soil. The negative relationship between colonization and plant height was reported by Kilowasid et al. (2023) in rice. Many factors including environmental factors, AMF specificity and plant genotype and development stage might explain differences observed between locations. Negative relationship might be the result of the non-respect of the symbiosis pact between partners. In fact, symbiosis being an energy cost, the plant might allocate some carbohydrates to the fungus; and in return, if the AMF does not provide sufficient benefits, plant's resources can be depleted. Otherwise, competition for resources between the two partners might explained the negative correlation between AMF root colonization and plant growth.

Conclusion:-

This is the first report of the density and diversity of AMF associated to pigeon pea in different regions of Senegal. Interesting differences exist between the soil origins and between pigeon pea provenances in some times. It highlights a positive effect of this crop on soil mycorrhizal infectivity which can boost the native AMF propagation in these tropical soils. However, considering the very low level of mycorrhizal infectivity in these tropical soils, biofertilization by AMF would be very promising to improve soil fertility and productivity of crops such as pigeon pea. Trap culture might be a good method for AMF sporulation but its duration and/or its number might be modulated according to certain conditions.

Conflict of interest:-

The authors declare that there is no conflict of interest.

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References:-

1. Anandakumar S & Kalaiselvi T. 2022. Effect of *Glomus intraradices* spore abundance of the inoculum on percent mycorrhizal colonization and growth of *Vigna mungo* (L.) Hepper. *Plant Science Today*, 9(4): 829–836. <https://doi.org/10.14719/pst.1653>
2. Araujo J, Díaz-Alcántara CA, Urbano B & González-Andrés F. 2020. Inoculation with native *Bradyrhizobium* strains formulated with biochar as carrier improves the performance of pigeonpea (*Cajanus cajan* L.). *European Journal of Agronomy*, 113: 125985, ISSN 1161-0301. <https://doi.org/10.1016/j.eja.2019.125985>
3. Arruda B, Herrera WFB, Rojas-García JC, Turner C & Pavinato PS. 2021. Cover crop species and mycorrhizal colonization on soil phosphorus dynamics. *Rhizosphere*, 19: 100396. <https://doi.org/10.1016/j.rhisph.2021.100396>
4. Bahadori M & Tofighi H. 2017. Investigation of Soil Organic Carbon Recovery by the Walkley-Black Method Under Diverse Vegetation Systems. *Soil Science* 182(3): 101-106. <https://doi.org/10.1097/SS.0000000000000201>
5. Balestrini R, Brunetti C, Chitarra W & Nerva L. 2020. Photosynthetic traits and nitrogen uptake in crops: which is the role of arbuscular mycorrhizal fungi? *Plants (Basel)*, 9(9): 1105. <https://doi.org/10.3390/plants9091105>

6. Belay Z, Vestberg M & Assefa F. 2013. Diversity and abundance of arbuscular mycorrhizal fungi associated with acacia trees from different land use systems in Ethiopia. *African Journal of Microbiology Research*, 7(48): 5503-5515. <https://doi.org/10.5897/AJMR2013.6115>
7. Battisti M, Moretti B, Sacco D, Grignani C & Zavattaro L. 2021. Soil Olsen P response to different phosphorus fertilization strategies in long-term experiments in NW Italy. *Soil Use and Management* 38(1): 549-563. <https://doi.org/10.1111/sum.12701>
8. Bencherif K, Laruelle F, Tisserant B, Dalpé Y & Lounès-Hadj Sahraoui A. 2023. Engineering approach for production of arbuscular mycorrhizal inoculum adapted to saline soil management. *Stresses*, 3: 404-423. <https://doi.org/10.3390/stresses3020030>
9. Benelhadjelloul S, Nehal F, M'hamed I, Kadiri A & Bouchakour M. 2024. Assessment of mycorrhizal fungal diversity in legumes (*Medicago sativa*, *Medicago truncatula* and *Trifolium rubens*) from Algeria and their influence on soil physicochemical and microbiological properties. *Agricultural Science Digest*, 44(1): 06-13. <https://doi.org/10.18805/ag.DF-586>
10. Birhane E, Gebretsadik KF, Taye G, Aynekulu E, Rannestad MM & Norgrove L. 2020. Effects of forest composition and disturbance on arbuscular mycorrhizae spore density, arbuscular mycorrhizae root colonization and soil carbon stocks in a dry afro-montane forest in Northern Ethiopia. *Diversity*, 12(4): 133. <https://doi.org/10.3390/d12040133>
11. Bopape FL, Beukes CW, Katlego K, Hassen AI, Steenkamp ET & Gwata ET. 2023. Symbiotic performance and characterization of pigeonpea (*Cajanus cajan* L. Millsp.) rhizobia occurring in South African soils. *Agriculture*, 13(1): 30. <https://doi.org/10.3390/agriculture13010030>
12. Chafai W, El Yeznasni H & Khalid A. 2025. Isolation, multiplication, and characterization of arbuscular mycorrhizal fungi strains in soils of North-East Morocco: impact of soils and trap plants. *E3S Web Conference*, 632: 01013. <https://doi.org/10.1051/e3sconf/202563201013>
13. Chen Q, Deng X, Elzenga JTM et al. 2022. Effect of soil bacteriomes on mycorrhizal colonization by *Rhizophagus irregularis*—interactive effects on maize (*Zea mays* L.) growth under salt stress. *Biology & Fertility Soils*, 58: 515–525. <https://doi.org/10.1007/s00374-022-01636-x>
14. Da Silva da Sousa L, Correia TS, dos Santos dos Farias F, Santana MDS & Lara TS. 2023. Influence of arbuscular mycorrhizal fungi density on growth and metabolism of *Handroanthus serratifolius* (Vahl) S.O. Grose seedlings. *Physiologica Plantarum*, 1-14. <https://doi.org/10.1111/pp1.14067>
15. Dania SO & Fagbola O. 2022. Comparative Effects of soil nutrient status and texture on mycorrhiza – legume base cropping system in two soil types of Nigeria. *Journal of Current Opinion in Crop Science*, 3(1): 6-15. <https://doi.org/10.62773/jcocs.v3i1.149>
16. Dare MO, Abaidoo R, Fagbola O & Asiedu R. 2012. Diversity of arbuscular mycorrhizal fungi in soils of yam (*Dioscorea* spp.) cropping systems in four agroecologies of Nigeria. *Archives of Agronomy and Soil Science*, 59(4): 521–531. <https://doi.org/10.1080/03650340.2011.653682>
17. Delaeter M, Magnin-Robert M, Randoux B & Lounès-Hadj Sahraoui A. 2024. Arbuscular mycorrhizal fungi as biostimulant and biocontrol agents: a review. *Microorganisms*, 12(7): 1281. <https://doi.org/10.3390/microorganisms12071281>
18. Dilsiz A, Rezaee Danes Y, Sagu, Ç, Kaya Özdoğan D & Koyuncu N. 2021. Study on status of arbuscular mycorrhizal fungal populations in some pastures of Turkey. *International Journal of Agricultural Technology*, 17(6): 2473–2486. retrieved from <https://li04.tci-thaijo.org/index.php/IJAT/article/view/6414>
19. Diop I, Ndoye F, Diedhiou AG, Krasova-Wade T, et al. 2021. Diversity and spore density of arbuscular mycorrhizal fungi in the rhizosphere of Cowpea (*Vigna unguiculata* [L.] Walp.) cultivated in different soils in Senegal. *Journal of Animal & Plant Sciences* 48 (1): 8552-8565. <https://doi.org/10.35759/JANmPlSci.v48-1>
20. Djotan AKG, Matsushita N & Fukuda K. 2024. Within-Site Variations in soil physicochemical properties explained the spatiality and cohabitation of arbuscular mycorrhizal fungi in the roots of *Cryptomeria japonica*. *Microbial Ecology*, 87: 136. <https://doi.org/10.1007/s00248-024-02449-1>
21. Ech-Cheddadi S, Ouallal O, Fatine M, Moussa O et al. 2019. soil physicochemical properties and diversity of arbuscular mycorrhizal fungi in the rhizosphere of sugarcane in the Gharb Region of Morocco. *Plant cell biotechnology & molecular biology*, 20 (19&20): 828–845
22. Egboka NT, Leonard Chimaobi Agim LC, Michael Akaninyene Okon MA et al. 2022a. "population density of arbuscular mycorrhizal fungi and physico-chemical properties of soils as affected by cropping systems," *Journal Clean WAS (JCleanWAS)*, Zibeline International Publishing, 6(1) : 27-32. <https://doi.org/10.26480/jcleanwas.01.2022.27.32>

23. Egboka NT, Fagbola O, Nkwopara UN, Okoli NH, et al. 2022b. Density of arbuscular mycorrhizal fungi and nutrient status of soils in selected land use types and soil depths. *Sarhad Journal of Agriculture*, 38: 633-647. <https://dx.doi.org/10.17582/journal.sja/2022/38.2.633.647>
24. Faye MH, Dossou NI, Badiane A, Agne-Djigo A et al. 2020. Prevalence and associated factors of vitamin A deficiency among children and women in Senegal. *Journal of Nutrition Health & Food Science*, 8(2):1-11. <https://doi.org/10.15226/jnhfs.2020.001176>
25. Fisher RA & Yates F. 1970. *Statistical tables for biological agriculture and medical research*, 6th ed.; Hafner Publishing Company: Davien, CT, USA.
26. Gagou E, Chakroune K, Abbas M, Lamkani T. & Hakkou A. 2023. Evaluation of the mycorrhizal potential of date palm (*Phoenix dactylifera* L.) rhizosphere soils in the Figuig oasis (Southeastern Morocco). *Journal of Fungi*, 9(9): 931. <https://doi.org/10.3390/jof9090931>
27. Guzman A, Montes M, Hutchins L, DeLaCerde G et al. 2021. Crop diversity enriches arbuscular mycorrhizal fungal communities in an intensive agricultural landscape. *New Phytology*, 231: 447–459. <https://doi.org/10.1111/nph.17306>
28. Harshitha B, Goudar G, Krishnaraj PU & Koti RV. 2020. Characterization of plant growth promoting rhizobial isolates for pigeon pea (*Cajanus cajan* [L.] Mill sp). *International Journal of Current Microbiology and Applied Sciences*, 9(7): 3776-3788. <https://doi.org/10.20546/ijemas.2020.907.442>
29. Hazard C, Gosling P, van der Gast C et al. 2013. The role of local environment and geographical distance in determining community composition of arbuscular mycorrhizal fungi at the landscape scale. *ISME Journal*, 7: 498–508. <https://doi.org/10.1038/ismej.2012.127>
30. Hicks TD, Kuns CM, Raman C, Bates ZT, Nagarajan S. 2022. Simplified method for the determination of total Kjeldahl Nitrogen in waste water. *Environments*, 9(5) : 55. <https://doi.org/10.3390/environments9050055>
31. Holste EK, Holl KD, Zahawi RA & Kobe RK. 2016. Reduced aboveground tree growth associated with higher arbuscular mycorrhizal fungal diversity in tropical forest restoration. *Ecology & Evolution*, 6(20):7253-7262. <https://doi.org/10.1002/ece3.2487>
32. Husein M, Umamia N, Pertiwinigruma A, Rahmanb MM & Anantaa D. 2022. The Role of arbuscular mycorrhizal fungi density and diversity on the growth and biomass of corn and sorghum forage in trapping culture. *Tropical Animal Science Journal*, 45(1):37-43. <https://doi.org/10.5398/tasj.2022.45.1.37>
33. Jairu D, Quader SW, Pratiksha & Khalkho R. 2023. Neglected and Underutilized Species (NUS): The Smart Crop of the Future. *Food and Scientific Reports*, 4(12): 1-9. e-ISSN 2582-5437
34. Kabalan S, Kovács F, Papdi E, Tóth E, et al. 2024. Residues of symbiont cover crops improving corn growth and soil-dependent health parameters. *Agriculture*, 14(9): 1601. <https://doi.org/10.3390/agriculture14091601>
35. Kilowasid LMH, Manik Nevianti DS, Afa LO, Rakian TC et al. 2023. Effect of biochar on arbuscular mycorrhizae fungi, nutrient uptake and growth of local upland rice, Southeast Sulawesi, Indonesia. *IOP Conference Series: Earth Environmental Science*, 1162: 012006. <https://doi.org/10.1088/1755-1315/1162/1/012006>
36. Liang SM, Zheng FL, Fathi Abd Allah E, Muthuramalingam P et al. 2021. Spatial changes of arbuscular mycorrhizal fungi in peach and their correlation with soil properties. *Saudi Journal of Biological Science*, 28(11):6495-6499. <https://doi.org/10.1016/j.sjbs.2021.07.024>
37. Masebo N, Birhane E, Takele S, Belay Z et al. 2023. Diversity of arbuscular mycorrhizal fungi under different agroforestry practices in the drylands of Southern Ethiopia. *BMC Plant Biology*, 23: 634. <https://doi.org/10.1186/s12870-023-04645-6>
38. Mathimaran N, Jegan S, Thimmegowda MN, Prabavathy VR et al. 2020. Intercropping transplanted pigeon pea with finger millet: arbuscular mycorrhizal fungi and plant growth promoting rhizobacteria boost yield while reducing fertilizer input. *Frontiers in Sustainable Food Systems*, 4:88. <https://doi.org/10.3389/fsufs.2020.00088>
39. Meddich A, Hafidi M, Ait El mokhtar M & Boumezzough A. 2015. Caractérisation des paramètres physicochimiques et des potentialités mycorrhizogènes des sols salés de la palmeraie Nord-est de Marrakech. *Journal of Materials & Environmental Science*, 6(9): 2469-2475. ISSN: 2028-2508
40. Meyer E, Betancur-Agudelo M, Ventura BS et al. 2021. Mycorrhizal root colonization in maize fields is more affected by soil management and climate conditions than by plant genotype. *Archives of Microbiology*, 203: 4609–4618. <https://doi.org/10.1007/s00203-021-02429-w>
41. Mohammed Ali AKH, Adlan MAM & Musa LM. 2023. Occurrence and diversity of indigenous mycorrhizal fungi in different Sudan soils using sorghum [*Sorghum bicolor* (L.) Moench] as a host plant. *Chemical and Natural Resources Engineering Journal*, 7(1): 21–34. <https://doi.org/10.31436/cnrej.v7i1.75>
42. Musokwa M & Mafongoya P. 2021. Pigeonpea yield and water use efficiency: a savior under climate change-induced water stress. *Agronomy*, 11(1): 5. <https://doi.org/10.3390/agronomy11010005>

43. Musyoka DK, Kimiti JM, Muchugi AN, Jefwa JM, Sakha MA. 2023. Soil physic-chemical properties, arbuscular mycorrhizal fungi (amf) diversity and abundance in the rhizosphere of *Calotropis procera* in two wild locations of semi-arid Eastern Kenya. *International Journal of Current Microbiology & Applied Science*, 12(10): 181-201. <https://doi.org/10.20546/ijcmas.2023.1210.021>
44. Namuyiga DB, Stellmacher T, Borgemeister C & Groot JCJ. 2022. A Typology and preferences for pigeon pea in smallholder mixed farming systems in Uganda. *Agriculture*, 12(8): 1186. <https://doi.org/10.3390/agriculture12081186>
45. Navarro JM & Morte A. 2024. Arbuscular mycorrhizal fungi as biofertilizers to increase the plant quality of sour-orange seedlings. *Agronomy*, 14: 230. <https://doi.org/10.3390/agronomy14010230>
46. Ndeko AB, Diedhiou AG, Founoune-Mboup H, Chuma GB et al. 2024a. Site climate more than soil properties and topography shape the natural arbuscular mycorrhizal symbiosis in maize and spore density within rainfed maize (*Zea mays* L.) cropland in the eastern DR Congo. *PLOS ONE*, 19(12): e0312581. <https://doi.org/10.1371/journal.pone.0312581>
47. Ndeko AB, Diedhiou AG, Fall S, Diouf D et al. 2024b. Arbuscular mycorrhizal dependency and responsiveness of maize varieties from South-Kivu, eastern Democratic Republic of Congo. *Cereal Research Communications*, 52: 1873–1889. <https://doi.org/10.1007/s42976-024-00508-4>
48. Ndlovu M, Scheelbeek P, Ngidi M & Mabhaudhi T. 2024. Underutilized crops for diverse, resilient and healthy agri-food systems: a systematic review of sub-Saharan Africa. *Frontiers in Sustainable Food Systems*, 8: 1498402. <https://doi.org/10.3389/fsufs.2024.1498402>
49. Ndoye F, Diop I, Gueye MC, Sadio O. et al. 2024. Arbuscular Mycorrhizal Fungi and Soil Enzyme Activities in Different Fonio Millet (*Digitaria exilis* Stapf.) Agroecosystems in Senegal. *Journal of Pure and Applied Microbiology*, 19(18): 1866-1882. <https://doi.org/10.22207/JPAM.18.3.37>
50. Noreen S, Yaseen T, Iqbal J, Abbasi BA et al. 2023. Morphological and molecular characterizations of arbuscular mycorrhizal fungi and their influence on soil physicochemical properties and plant nutrition. *ACS Omega*, 8(36): 32468-32482. <https://doi.org/10.1021/acsomega.3c02489>
51. Ohanenye IC, Sun X, Sarteshnizi RA & Udenigwe CC. 2021. Germination alters the microstructure, in vitro protein digestibility, α -glucosidase and dipeptidyl peptidase-IV inhibitory activities of bioaccessible fraction of pigeon pea (*Cajanus cajan*) seeds. *Legume Science*, 3: e79. <https://doi.org/10.1002/leg3.79>
52. Olguera DT, Asio VB & Labides JLR. 2023. Soil physicochemical characteristics and spore density of indigenous arbuscular mycorrhizal fungi (amf) in different vegetation patches of a marginal upland in Central Philippines. *Mindanao Journal of Science and Technology*, 21(1): 46-72. <https://doi.org/10.61310/mjst.v21i2.1712>
53. Padulosi S, Phrang R & Rosado-May F. 2019. Supporting nutrition sensitive agriculture through neglected and underutilized species: Operational framework (p. 39). Rome, Italy: Bioversity International and IFAD.
54. Pérez-de-Luque A, Tille S, Johnson I et al. 2017. The interactive effects of arbuscular mycorrhiza and plant growth-promoting rhizobacteria synergistically enhance host plant defences against pathogens. *Scientific Reports*, 7: 16409. <https://doi.org/10.1038/s41598-017-16697-4>
55. Pérez-Redondo M, Jaizme-Vega M, González-Rodríguez ÁM et al. 2025. Arbuscular mycorrhizal density and propagation are driven by vegetation cover and plant phylogenetic diversity. *Plant Soil*, 1-15. <https://doi.org/10.1007/s11104-024-07127-2>
56. Pereira CMR, Lopez-García A, Maia LC, Frøslev TG et al. 2022. Arbuscular mycorrhizal fungal communities of pristine rainforests and adjacent sugarcane fields recruit from different species pools. *Soil Biology and Biochemistry*, 167 : 108585. <https://doi.org/10.1016/j.soilbio.2022.108585>
57. Rini MV, Yansyah MP & Arif MAS. 2021. The Application of Arbuscular Mycorrhizal Fungi Reduced the Required Dose of Compound Fertilizer for Oil Palm (*Elaeis Guineensis* Jacq.) in Nursery. *IOP Conferences Series: Earth and Environmental Science*, 1012: 012011. <https://doi.org/10.1088/1755-1315/1012/1/012011>
58. Sakha M, Gweyi-Onyango JP, Masso C & Baijukya FP. 2025. Diversity, characteristics, and abundance of native arbuscular mycorrhizal fungi in the semi-arid lands of Eastern Kenya. *Frontiers in Microbiology*, 16:1582476. <https://doi.org/10.3389/fmicb.2025.1582476>
59. Salim MA, Budi RSW, Setyaningsih L, Wahyudi II & Kirmi H. 2020. Root colonization by arbuscular mycorrhizal fungi (AMF) in various age classes of revegetation post-coal mine. *Biodiversitas*, 21 (10): 5013-5022. <https://doi.org/10.13057/biodiv/d211005>
60. Soudzilovskaia NA, Vaessen S, Barcelo M, He J et al. 2020. Fungal Root: global online database of plant mycorrhizal associations. *New Phytologist*, 227: 955–966. <https://doi.org/10.1111/nph.16569>
61. Tenzin UW, Noirungsee N, Runsaeng P, Noppradit P & Klinnawee L. 2022. Dry-season soil and co-cultivated host plants enhanced propagation of arbuscular mycorrhizal fungal spores from sand dune vegetation in trap culture. *Journal of Fungi*, 8: 1061. <https://doi.org/10.3390/jof8101061>

62. Thiam M, Diouf A, Coly OB, Kébé SD et al. 2022. Dietary intakes and nutritional status of mother-child (6-23 months old) pair targeted through the "organic residual products for biofortified foods for Africa project" in rural area in Senegal. *International Journal of Child Health and Nutrition*, 11(3): 119–130. <https://doi.org/10.6000/1929-4247.2022.11.03.1>
63. Thiocone KMO, Soumare A, Charahabil MM, Ndiaye L et al. 2023. Density and diversity of arbuscular mycorrhizal fungi in *Anacardium occidentale* L. plantations in Senegal. *African Journal of Microbiology Research*, 17(9): 221-229. <https://doi.org/10.5897/AJMR2023.9712>
64. Tosi M, Ogilvie CM, Spagnoletti FN, Fournier S et al. 2023. Cover crops modulate the response of arbuscular mycorrhizal fungi to water supply: a field study in corn. *Plants*, 12(5): 1015. <https://doi.org/10.3390/plants12051015>
65. Trouvelot A, Kough JL & Gianinazzi-Pearson V. 1986. Mesure du taux de mycorhization VA d'un système racinaire. Recherche de méthodes d'estimation ayant une signification fonctionnelle. In: *Physiological and Genetical Aspects of Mycorrhizae* (V Gianinazzi-Pearson, S Gianinazzi, eds), INRA, Paris, France, 217-221
66. Tyburska-Woś J & Kieliszewska-Rokicka B. 2024. Long-term monitoring studies of the mycorrhizal colonization of *Aesculus hippocastanum* L. roots and the vitality of soil microorganisms in urban and non-urban environments. *Forests*, 15(1): 84. <https://doi.org/10.3390/f15010084>
67. Uddling J, Gelang-Alfredsson J, Piikki K & Pleijel H. 2007. Evaluating the relationship between leaf chlorophyll concentration and SPAD-502 chlorophyll meter readings. *Photosynthesis Research*, 91(1): 3746. <https://doi.org/10.1007/s11120-006-9077-5>
68. UNSCN. 2020 – The COVID-19 pandemic is disrupting people's food environments: a resource list on Food Systems and Nutrition responses-UNSCN. [Unscn.org](https://www.unscn.org)
69. Verbruggen E, Struyf E & Vicca S. 2021. Can arbuscular mycorrhizal fungi speed up carbon sequestration by enhanced weathering? *Plants People Planet*, 3:445–453. <https://doi.org/10.1002/ppp3.10179>
70. Versulienė A, Hirte J, Ciulla F, Camenzind M et al. 2024. Wheat varieties show consistent differences in root colonization by mycorrhiza across a European pedoclimatic gradient. *European Journal of Soil Science*, 75(4): e13543. <https://doi.org/10.1111/ejss.13543>
71. Vieira LC, Silva DKAD, Escobar IEC, Silva JMD et al. 2020. Changes in an arbuscular mycorrhizal fungi community along an environmental gradient. *Plants*, 9:52. <https://doi.org/10.3390/plants9010052>
72. Wang Q, Liu M, Wang Z, Li J, Liu K & Huang D. 2024a. The role of arbuscular mycorrhizal symbiosis in plant abiotic stress. *Frontiers in Microbiology*, 14: 1323881. <https://doi.org/10.3389/fmicb.2023.1323881>
73. Wang J, Gao X, Wang J et al. 2024b. Host plants directly determine the α diversity of rhizosphere arbuscular mycorrhizal fungal communities in the National Tropical Fruit Tree Field Genebank. *Chemical & Biol Technologies in Agriculture*, 11: 20. <https://doi.org/10.1186/s40538-024-00540-w>
74. Weerasekara PC, Withanachchi CR, Ginigaddara GAS & Ploeger A. 2020. Understanding dietary diversity, dietary practices and changes in food patterns in marginalised societies in Sri Lanka. *Foods*, 9(11): 1659. <https://doi.org/10.3390/foods9111659>
75. Xavier Martins WF & Rodrigues BF. 2020. Identification of dominant arbuscular mycorrhizal fungi in different rice ecosystems. *Agricultural Research*, 9: 46–55. <https://doi.org/10.1007/s40003-019-00404-y>
76. Yadahalli GS, Doddamani BM & Vidyavathi GY. 2024. "Pigeonpea productivity and soil health as influenced by phosphorus levels and am fungi under different planting methods". *International Journal of Plant & Soil Science*, 36 (5): 929–937. <https://doi.org/10.9734/ijpss/2024/v36i54589>
77. Yuwati TW, Rahmi ANR, Hakim SS & Badruzaufari E. 2020. The Abundance of arbuscular mycorrhiza infective propagules under galam stand at shallow peat of South Kalimantan. *BIO Web of Conferences* 20, 03008 ICWEB 2019. <https://doi.org/10.1051/bioconf/20202003008>
78. Zhang M, Shi Z, Xu X & Wang X. 2022. Arbuscular mycorrhizal fungi associated with roots reveal high diversity levels at different elevations in tropical montane rainforests. *Diversity*, 14(8): 587. <https://doi.org/10.3390/d14080587>