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Growth, root colonization and yield attribute responses of five groundnut (*Arachis hypogaea* L.) varieties toward arbuscular mycorrhizal fungal inoculation in a Senegalese agricultural soil

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The aim of this study was to isolate the most specific and effective arbuscular mycorrhizal fungi (AMF) for groundnut and to determine the degree of variability in the response of groundnut varieties to inoculation. The seeds of five varieties: 55-437, Fleur 11, Sunu Gaal, Amoul Morom, and Essamaay were inoculated individually with five AMF (*Funneliformis mosseae*, *Rhizophagus aggregatus* and *Rhizophagus fasciculatus*, the indigenous isolates, and *Rhizophagus irregularis* and *Gigaspora rosea*, isolated from Canada). Plants were grown under greenhouse conditions in a mixture of non-sterile sandy soil and sterilized soil at 120°C for 20 min (1:1, v/v). The results obtained in terms of root AMF colonization and nodule formation showed a positive effect of AMF inoculation in all varieties. Furthermore, we showed that inoculation efficacy did not depend on the origin of the inoculated AMF and no clear relationship was found between the fact that the varieties used were traditional or modern. However, our data indicated that Amoul Morom, Essamay, and 55-437 were more responsive to AMF inoculation, showing the greatest increase in plant growth, leaf chlorophyll content, and yield parameters. The results therefore confirm the functional variation among the inoculated AMF, which is crucial for establishing potential formulations of AMF inoculants to improve groundnut productivity. According to this study, further selection of compatible AMF partners would be useful to improve inoculation success with Fleur 11 and Sunu Gaal.

Key words: Peanut (*Arachis hypogaea*), bioferlilizers, arbuscular mycorrhizal fungi (AMF), symbiotic performance, plant growth, yield parameters.

INTRODUCTION

Soil microbial communities are involved in several functions in agroecosystems, such as nutrient availability, pathogen control, and resilience to abiotic stresses (Aguégué et al., 2023; Lu et al., 2023; Sharma et al., 2023). Arbuscular mycorrhizal fungi (AMF) are among these important soil-dwelling microorganisms and can have a strong influence on plant growth and productivity. They form mutualistic associations with over 80% of all vascular plants, affecting plant fitness and competitive interactions (Johnson et al., 1997; Aguégué et al., 2023). They are well known for assisting host plants with phosphorus uptake (Smith and Read, 2008; Lu et al., 2023), but can also provide other benefits such as protection from pathogens (Cardoso and Kuyper, 2006; Sharma et al., 2023), assistance with the uptake of other nutrients such as nitrogen and copper, and improved water relations (Smith and Read, 2008; Sene et al., 2010; Lu et al., 2023; Sharma et al., 2023). AMF hyphae also play a role in the formation and structural stability of soil aggregates (Miller and Jastrow, 2000; Zhang et al., 2023) and contribute to the composition of plant community structures (van der Heijden et al., 2015; Chen et al., 2023). In return, AMF receive photosynthetic products from the host plant (Smith and Read, 2008; Lu et al., 2023).

Soil microorganisms are now being promoted as smart fertilizers for a new green revolution in the 21st century (Sene et al., 2010; Fortin et al., 2015; Lesueur et al., 2016; Mohanty and Swain, 2018; Rocha et al., 2019; Sene et al., 2021, 2023). Microbial inoculants offer low-cost alternatives to expensive mineral fertilizers and provide a means to maintain or improve soil fertility (Hart et al., 2015; Itelima et al., 2018; Begum et al., 2019). A large body of scientific evidence demonstrates not only improved crop yield and resistance to environmental stress in AMF crops, but also improvements in many food quality attributes, such as increased levels of desirable antioxidants, vitamins and minerals (Sene et al., 2010; Calvo et al., 2014; Fortin et al., 2015; Hart et al., 2015; Rocha et al., 2019).

Groundnut, also known as peanut (*Arachis hypogaea* L.), is an important grain legume grown in the tropics and subtropics, including sub-Saharan Africa. It is an important source of oil and protein and also contains vitamin B as well. Groundnut is consumed worldwide for human and animal feeding (Noba et al., 2014). In Senegal, groundnut has been a cash crop for more than a century, contributing to 60% of the country's agricultural gross domestic product (GDP) and about 80% of its export earnings (Sene et al., 2010; Noba et al., 2014). It is the most important oil-producing crop, and the four oil factories established in the country formed the backbone of the national industrial fabric. After a long period of decline, groundnut yields have increased in the last five years. However, the factors that determine these increases, that is, soil fertility, have steadily deteriorated, with a reduction in fallow land and low levels of fertilizer use (Sene et al., 2010). Various agricultural practices, including the use of chemical fertilizers, have been adopted to increase yields and alleviate food shortages. However, the high cost of chemical fertilizer and the need for sustainable alternative sources have increased the strategic importance of microbial inoculation. The study was undertaken to isolate the most specific and effective AMF inoculants for five modern and traditional Senegalese groundnut varieties and to use elite strains as inoculants. Our hypothesis was that the response of groundnut to AMF inoculation would vary between varieties and that this variability would differ between modern and traditional varieties.

MATERIALS AND METHODS

Plant

Five local groundnut (*A. hypogaea* L.) varieties obtained from the Centre National de Recherche Agronomique (CNRA) in Bambey, Senegal, were used in this experiment. These varieties were selected on the basis of the taste desired by the local population and their characteristics are shown in Table 1.

Arbuscular mycorrhizal fungal materials

The arbuscular mycorrhizal fungal (AMF) inoculants used in this study are from the collection of the Laboratoire Commun de Microbiologie (LCM) IRD/ISRA/UCAD, Dakar, Senegal. Three of them (*Funneliformis mosseae*, *Rhizophagus aggregatus* and *Rhizophagus fasciculatus*) are indigenous and isolated from Senegalese soils (Table 2). In this experiment, they were tested against two exotic AMF inoculants (*Rhizophagus irregularis* and *Gigaspora rosea*).

Greenhouse experimental design

The experiment was set up in the greenhouse (Bel Air experimental station, 14°44'N, 17°30'W in Dakar) using a non-sterile soil from Sangalkam, 30 km east of Dakar, mixed with sterilized soil at 120°C for 20 min (1:1, v/v). This soil has a pH of 6.5 with 58.15, 32.8 and 3.6% of sand, loam and clay, respectively and contains 0.06% total N, 0.54% total C, 39 mg P kg^{-1} total P, 4.8 mg P kg^{-1} available P. It was sieved (< 1 mm), homogenized and used to fill up the pots. Seeds of selected groundnut varieties (listed in Table 1) were first surface sterilized (to avoid seed-borne diseases) with 5% sodium hypochlorite (NaOCl) for 5 min, 70% ethanol for 3 min and thoroughly rinsed with sterile distilled water. The seeds were then placed on Petri dishes containing moist filter paper for germination under sterile conditions and kept in the dark at 25°C. The germinated seeds were manually transplanted to a depth of 2-3 cm into 1.5 L plastic pots disinfected with a solution containing 1.81% of calcium hypochlorite and filled with the soil substrate. Two germinated seeds were planted in each pot. The plants were dismantled on the 3rd day after planting and before inoculation to one plant per pot. The pots were arranged in randomized blocks, with a single inoculation and five replications. The pots were placed at 10 and 40 cm within and between the rows for the varieties Fleur 11, 55-437 and Sunu Gaal. The distance between the pots was 10

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Table 1. Characteristics of the groundnut varieties used in the study.

Variety	Type	Growth habit	Growth cycle (days)	Registration in Senegal
Fleur 11	Spanish	Erect	90	Traditional, since 1955
55-437	Spanish	Erect	90	Traditional, since 1993
Sunu Gaal	Spanish	Erect	95	New, since 2017
Essamay	Virginia	Semi-erect	105	New, since 2017
Amoul Morom	Virginia	Semi-erect	120	New, since 2017

Table 2. Characteristics of the arbuscular mycorrhizal fungal (AMF) strains used in the study.

AMF strains	Origin of AMF strain isolation
Funneliformis mosseae (formerly *Glomus mosseae*)	Diokoul -Senegal
Rhizophagus aggregatus (formerly *Glomus aggregatum*) R13	Djignaki - Senegal
Rhizophagus fasciculatus (formly *Glomus fasciculatum*) R10	Kabrousse - Senegal
Rhizophagus irregularis (formerly *Glomus intraradices*)	Exotic, Canada
Gigaspora rosea	Exotic, Canada

and 60 cm for the varieties Amoul Morom and Essamay. The plants were grown for 65 days under greenhouse conditions (temperature of 27-35°C, relative humidity of 70-80% and 12 h of light) and were watered every two days with tap water without added nutrients.

Inoculant preparation and inoculation

The greenhouse experiment was composed of six treatments: three with application of indigenous AMF inoculants compared with two exotic AMF, and a negative control without inoculation for each variety. The AMF inoculants were propagated on *Zea mays* L for 12 weeks under greenhouse conditions on sterilized substrate (soil and sandy 1:1 v/v). For AMF inoculation, 10 g of the substrate containing an average of 40 spores g^{-1} soil and root fragments with 85% of colonized roots length, were placed adjacent to roots of seedlings. Treatments without AMF inoculants received 10 g of autoclaved inoculants in order to avoid differences in soil nutrient content linked to the addition of AMF inoculants.

Collection of growth and yield variables

Data on growth variables (plant height and number of branches) and leaf chlorophyll content for each variety were collected at flowering [30 days after planting (DAP)], pod filling (45 DAP) and pod maturity periods (60 DAP). Plant height (cm) was measured with a ruler from the base of the stem to the apex, while the number of branches was counted manually. Leaf chlorophyll content was quantified at 30, 45 and 65 DAP using a SPAD-502Plus chlorophyll meter (Konica-Minolta). At harvest, whole groundnut plants were uprooted. The soil adhering to the roots was removed under running tap water and nodules were picked and counted. The pods were manually stripped from the plants to record the yield components. For each variety, above-ground and root biomass, root colonization (intensity and frequency of AMF in the roots) and the yield attributes (number of pods per plant, pod weight) were determined. Above-ground and root biomass, nodule weight and the yield attributes were determined by weighing sample parts after over-drying to constant weight at 65°C.

Root arbuscular mycorrhizal fungal colonization

The roots obtained at 65 DAP were washed properly and used to examine the level of AMF colonization. Randomly selected lateral roots, which are more likely to form mycorrhizae, were collected, cleared in KOH [10% (w/v)] at 80°C for 30 min and stained with trypan blue (0.05% (w/v) in 0.8% acetic acid solution) at 80°C for 35 min (adapted from Phillips and Hayman, 1970). Roots were cut into 1-2 cm pieces and placed on slides for microscopic observation (x250). A total of 100 root pieces were taken randomly from each sample. AMF colonization was quantified according to the method of Mcgonigle et al. (1990).

Data analyses

All data were tested for normality and homogeneity using the Shapiro-Wilk and Levene tests, respectively. Data for plant growth and yield parameters were statistically analyzed using univariate analysis with one-way analysis of variance (ANOVA) using the R software v3.4.4 (R Core Team, 2020). AMF colonization data for each treatment and plot were square-root transformed and subjected to a two-way ANOVA followed by Tukey's multiple means tests to analyze how the response variable varied between treatments. Significantly different means were separated using the Tukey (HSD) test at the 5% probability threshold. Means and standard errors are presented throughout and $P < 0.05$ is considered significant.

RESULTS

Mycorrhizal root colonization and root nodulation

Groundnut plants of all varieties were naturally colonized by autochthonous AMF as shown for the control root plants. However, root AMF colonization levels at 65 DAP were generally low in the uninoculated plants, ranging from 9.96 ± 1.84 to 13.06 ± 2.3%. Furthermore, the

Table 3. Root arbuscular mycorrhizal fungal (AMF) colonization and nodulation (number of nodules) of five groundnut varieties (*A. hypogaea*) under single inoculation with AMF 65 days after planting.

Treatments	Groundnut varieties									
	Amoul Morom		Essamaye		55-437		Fleur11		Sunu Gaal	
	IRC (%)	NNum	IRC (%)	NNum	IRC (%)	NNum	IRC (%)	NNum	IRC (%)	NNum
F. mosseae	33.1 ± 2.52^{a}	59.3 ± 4.72^{a}	38.4 ± 4.03^{b}	44.5 ± 4.12^{b}	24.55 ± 3.05^{b}	33.0 ± 4.94^{bc}	18.73 ± 1.16^{b}	68.0 ± 10.7^{ab}	23.46 ± 2.28^{a}	43.8 ± 11.4^{b}
R. aggregatus	27.01 ± 2.47^{b}	34.0 ± 4.82^{bc}	24.6 ± 2.55^{c}	72.8 ± 6.02^{a}	42.73 ± 3.07^{a}	27.8 ± 5.56^{c}	28.6 ± 1.86^{a}	80.3 ± 16.29^{a}	26.95 ± 2.40^{a}	57.8 ± 11.7^{ab}
R. fasciculatus	18.06 ± 2.63^{c}	41.3 ± 4.11^{b}	27.9 ± 2.84^{c}	34.3 ± 6.40^{bc}	21.0 ± 2.66^{b}	37.7 ± 4.62^{bc}	26.31 ± 2.98^{a}	60.8 ± 11.3^{abc}	27.39 ± 2.11^{a}	41.8 ± 9.54^{b}
G. rosea	21.14 ± 1.98^{c}	64.3 ± 11.0^{a}	23.8 ± 2.36^{c}	66.7 ± 5.49^{a}	24.3 ± 1.05^{b}	42.3 ± 5.71^{b}	19.43 ± 1.15^{b}	72.0 ± 12.2^{ab}	24.09 ± 2.34^{a}	74.0 ± 7.55^{a}
R. irregularis	38.4 ± 3.78^{a}	60.0 ± 11.3^{a}	62.1 ± 5.61^{a}	48.0 ± 10.8^{b}	44.6 ± 4.65^{a}	61.8 ± 10.5^{a}	29.59 ± 2.35^{a}	64.8 ± 8.85^{ab}	27.73 ± 2.60^{a}	43.3 ± 8.3^{b}
Control	9.96 ± 1.84^{d}	26.7 ± 3.06^{c}	13.06 ± 2.3^{d}	29.3 ± 2.22^{c}	12.13 ± 1.25^{c}	14.0 ± 3.73^{d}	11.74 ± 1.96^{c}	43.5 ± 7.23^{c}	13.05 ± 2.17^{b}	14.3 ± 3.8^{c}

NNum: Nodule number; IRC: intensity of root AMF colonization; Values (mean ± standard error) are an average of five replications; means ± standard error within the same column followed by the same superscript letters are not statistically different at the 5% probability according to Tukey test.

results showed that there was a significant difference ($p < 0.05$) in AMF colonization (ranging from 18.06 ± 2.63% to 62.1 ± 5.61%) between the inoculated and non-inoculated plants (Table 3), irrespective of the groundnut variety. Interestingly, the highest root AMF colonization occurred with the exotic strain *R. irregularis*, especially in the modern groundnut varieties. In addition, the indigenous AMF *R. aggregatus* also showed high root AMF colonization (42.73 ± 3.07%) in the traditional variety 55-437, whereas the root colonization was still low in the modern varieties Amoul Morom, Essamay and Sunu Gaal. Furthermore, the variety Essamay showed an overall higher AMF root colonization rate than the other varieties, irrespective of the AMF inoculated. In contrast, *R. fasciculatus* and *G. rosea* showed relatively low root AMF colonization compared to the other AMF inoculants (Table 3).

The results showed that there was a significant difference ($p < 0.05$) in the number of nodules between the inoculated and uninoculated treatments, except for *R. fasciculatus* when inoculated on Essamay and Fleur 11 and for *R. aggregatus* when inoculated on Amoul Morom (Table 3). In this case, the inoculated plants showed a higher number of nodules, including those inoculated with *G. rosea*, although such an increase was not clear for root AMF colonization. Irrespective of the groundnut variety, plants in the *F. mosseae*, *G. rosea* and *R. irregularis* treatments were more nodulated than the uninoculated plants (Table 3), indicating that these AMF inoculants had a high capacity to increase root nodule occupancy. The native AMF *R. aggregatus* also showed high nodulation on plants of the varieties Essamay and Fleur 11.

Leaf chlorophyll content

For the six varieties, leaf chlorophyll content at 30, 45 and 65 DAP after inoculation with AMF ranged from 29.8 ± 6.84 to 40.5 ± 2.72 (Table S1), 31.9 ± 3.23 to 44.0 ± 2.40 (Table S2), and 33.9 ± 2.86 to 46.7 ± 2.07 (Table 4), respectively, and was higher for groundnut variety Amoul Morom followed by the variety Sunu Gaal. At 30 and 45 DAP, the data showed no significant difference between the inoculated and non-inoculated plants, irrespective of the variety (Tables S1 and S2). However, leaf chlorophyll content increased significantly at 65 DAP for both Amoul Morom and Sunu Gaal when plants were inoculated with all native AMF and *G. rosea* for the former and only native AMF for the latter (Table 4).

Growth response of groundnut varieties to AMF inoculation

Plant growth parameters affected by AMF inoculation with the indigenous and exotic inoculants are shown in Table 5. Overall, the results showed that Amoul Morom, 55-437 and Essamay were more responsive to AMF inoculation in terms of plant height and collar diameter. Inoculation with 80 and 50% of our AMF collection showed the ability to improve plant height and collar diameter in Amoul Morom and 55-437, respectively. Interestingly, the plant height at 65 DAP showed a significant difference ($p < 0.05$) with the highest height and collar diameter observed on the *R. aggregatus* and *G. rosea* inoculation treatments. Only *R. irregularis* showed

Table 4. Leaf chlorophyll content at 65 days after planting in response to groundnut varieties (*A. hypogaea*) single inoculation with AMF strains.

Treatments	Groundnut varieties				
	Amoul Morom	Essamaye	55-437	Fleur11	Sunu Gaal
F. mosseae	45.3 ± 2.54^{a}	39.6 ± 1.17^{a}	36.0 ± 2.70^{a}	38.0 ± 1.52^{b}	40.2 ± 3.32^{a}
R. aggregatus	44.8 ± 2.23^{a}	36.6 ± 4.86^{ab}	38.1 ± 1.29^{a}	38.1 ± 1.82^{b}	40.6 ± 2.10^{a}
R. fasciculatus	45.5 ± 2.73^{a}	39.2 ± 1.67^{a}	37.6 ± 0.95^{a}	43.0 ± 1.71^{a}	42.3 ± 2.05^{a}
G. rosea	46.7 ± 2.07^{a}	35.4 ± 3.08^{ab}	36.0 ± 1.94^{a}	38.0 ± 1.59^{b}	39.4 ± 3.71^{ab}
R. irregularis	42.4 ± 3.24^{ab}	35.3 ± 1.69^{b}	39.2 ± 1.45^{a}	40.2 ± 2.67^{ab}	39.4 ± 3.47^{ab}
Control	41.3 ± 1.01^{b}	34.3 ± 1.80^{b}	36.0 ± 2.55^{a}	37.7 ± 1.30^{b}	33.9 ± 2.86^{b}

Values (mean ± standard error) are an average of five replications; means ± standard error within the same column followed by the same superscript letters are not statistically different at the 5% probability according to Tukey test.

Table 5. Growth (plant height and collar diameter) response of groundnut varieties (*A. hypogaea*) to single inoculation with AMF 65 days after planting.

Treatments	Groundnut varieties									
	Amoul Morom		Essamaye		55-437		Fleur11		Sunu Gaal	
	Height (cm)	CD (mm)	Height (cm)	CD (mm)	Height (cm)	CD (mm)	Height (cm)	CD (mm)	Height (cm)	CD (mm)
F. mosseae	14.3 ± 1.04^{ab}	5.45 ± 0.63^{a}	16.8 ± 1.19^{ab}	5.24 ± 1.03^{a}	21.4 ± 2.50^{ab}	4.32 ± 0.32^{ab}	18.5 ± 2.67^{a}	5.03 ± 0.79^{a}	23.0 ± 2.16^{a}	5.28 ± 0.59^{a}
R. aggregatus	15.7 ± 1.15^{a}	6.68 ± 0.32^{a}	18.8 ± 1.55^{ab}	5.12 ± 0.26^{a}	22.1 ± 1.89^{a}	4.76 ± 0.37^{a}	21.6 ± 2.50^{a}	4.52 ± 0.77^{a}	23.3 ± 1.71^{a}	4.33 ± 1.00^{a}
R. fasciculatus	16.0 ± 1.41^{a}	5.65 ± 0.95^{a}	16.6 ± 1.11^{ab}	5.24 ± 1.03^{a}	17.8 ± 1.26^{b}	4.73 ± 0.58^{ab}	19.4 ± 1.60^{a}	4.69 ± 0.31^{a}	24.6 ± 1.49^{a}	4.71 ± 0.39^{a}
G. rosea	15.9 ± 1.03^{a}	6.01 ± 0.28^{a}	18.4 ± 2.25^{ab}	5.08 ± 0.50^{a}	24.4 ± 2.45^{a}	4.85 ± 0.78^{a}	21.8 ± 2.22^{a}	4.56 ± 0.45^{a}	23.2 ± 1.58^{a}	4.57 ± 0.29^{a}
R. irregularis	16.0 ± 1.50^{a}	5.32 ± 0.18^{a}	19.5 ± 1.87^{a}	5.15 ± 0.26^{a}	21.0 ± 2.24^{ab}	4.65 ± 0.04^{ab}	19.4 ± 1.18^{a}	4.65 ± 0.61^{a}	23.1 ± 1.65^{a}	4.75 ± 0.42^{a}
Control	13.5 ± 0.71^{b}	5.18 ± 0.85^{a}	15.6 ± 1.38^{b}	5.04 ± 0.32^{a}	16.7 ± 1.76^{b}	3.41 ± 0.68^{b}	19.6 ± 2.14^{a}	4.41 ± 0.45^{a}	22.7 ± 2.08^{a}	4.26 ± 0.47^{a}

CD: Collar diameter; Values (mean ± standard error) are an average of five replications; means ± standard error within the same column followed by the same superscript letters are not statistically different at the 5% probability according to Tukey test.

a significant increase in plant height with Essamay (19.5 ± 1.87 cm plant^{-1}) compared to the uninoculated treatment (15.6 ± 1.38 cm plant^{-1}). However, no significant difference was found in the varieties Fleur 11 and Sunu Gaal (two genetically close varieties, Faye I. personal communication) when comparing the growth parameters of inoculated and uninoculated plants, but inoculated plants performed better than uninoculated plants for all groundnut varieties (Table 5).

Groundnut dry matter and yield parameters

The varieties Amoul Morom and Essamay responded better in terms of biomass production. For these varieties, all inoculated AMFs increased both shoot dry weight (SDW) and root dry weight (RDW). However, the differences observed were only significant for SDW. Furthermore, we found no significant difference in biomass production between plants inoculated with *R. irregularis* and the uninoculated plants, regardless of the variety used. The RDW was increased in three AMF treatments (*R. aggregatus*, *R. fasciculatus* and *G. rosea*), but the SDW was increased only when *G. rosea* was inoculated. Only the SDW was increased in the *R. fasciculatus* and *F. mosseae* treatments for the varieties Fleur 11 and Sunu Gaal, respectively. Both varieties were less responsive to AMF inoculation (Table 6).

Yield characteristics were improved in 40% of the treatments for each of the varieties Amoul Morom and 55-437. However, an improvement in

Table 6. Biomass production (above-ground and root biomass) of groundnut varieties at harvest.

Treatments	Groundnut varieties									
	Amoul Morom		Essamaye		55-437		Fleur11		Sunu Gaal	
	SDW (g)	RDW (g)	SDW (g)	RDW (g)	SDW (g)	RDW (g)	SDW (g)	RDW (g)	SDW (g)	RDW (g)
F. mosseae	3.27 ± 0.53[a]	1.31 ± 0.24[a]	2.82 ± 0.75[a]	1.13 ± 0.12[ab]	1.52 ± 0.40[ab]	0.51 ± 0.07[ab]	1.72 ± 0.18[b]	0.73 ± 0.16[ab]	3.38 ± 0.45[a]	0.88 ± 0.17[a]
R. aggregatus	2.72 ± 0.30[ab]	1.05 ± 0.15[ab]	2.83 ± 0.31[a]	1.33 ± 0.24[a]	1.58 ± 0.48[ab]	0.67 ± 0.07[a]	1.78 ± 0.17[b]	0.77 ± 0.23[ab]	2.54 ± 0.21[ab]	0.81 ± 0.14[ab]
R. fasciculatus	2.37 ± 0.31[b]	1.11 ± 0.15[ab]	2.76 ± 0.49[a]	1.32 ± 0.14[a]	1.53 ± 0.22[ab]	0.75 ± 0.36[a]	2.54 ± 0.14[a]	0.87 ± 0.11[a]	2.62 ± 0.65[ab]	0.75 ± 0.11[ab]
G. rosea	2.73 ± 0.52[ab]	1.27 ± 0.22[a]	2.88 ± 0.54[a]	1.06 ± 0.16[ab]	1.95 ± 0.73[a]	0.68 ± 0.21[a]	2.17 ± 0.62[ab]	0.86 ± 0.16[ab]	2.28 ± 0.34[ab]	0.68 ± 0.11[ab]
R. irregularis	2.22 ± 0.12[bc]	0.92 ± 0.11[ab]	2.58 ± 0.39[ab]	1.07 ± 0.18[ab]	1.42 ± 0.28[ab]	0.48 ± 0.11[ab]	1.66 ± 0.22[b]	0.71 ± 0.10[ab]	2.51 ± 0.82[ab]	0.62 ± 0.01[ab]
Control	1.52 ± 0.27[c]	0.68 ± 0.32[b]	1.56 ± 0.29[b]	0.82 ± 0.11[b]	0.60 ± 0.11[b]	0.16 ± 0.09[b]	1.65 ± 0.21[b]	0.52 ± 0.12[b]	1.96 ± 0.46[b]	0.59 ± 0.04[b]

SDW: Shoot dry weight; RDW: root dry weight; Values (mean ± standard error) are an average of five replications; means ± standard error within the same column followed by the same superscript letters are not statistically different at the 5% probability according to Tukey test.

yield attributes was observed in 20% of the treatments for the varieties Sunu Gaal and Essamay, whereas none of the inoculated AMFs showed a significant improvement in yield parameters for the variety Fleur 11. This suggests that the response of groundnut inoculation in terms of yield attributes is variety dependent, but not related to the fact that the varieties are traditional or modern. Among theinoculated AMF strains, only *G. rosea* showed a significantly better agronomic performance for variety Sunu Gaal and no significant difference was observed for variety Fleur 11. However, Amoul Morom showed a significant yield improvement when plants were inoculated with *F. mosseae* or *R. aggregatus*. Improved pod number and weight were also observed in variety 55-437 inoculated with *R. aggregatus*, while *R. irregularis* improved pod number. Only inoculation with *G. rosea* showed an improvement in pod number for varieties Essamaye and Sunu Gaal, but no significant difference was found for variety Fleur 11. Essamaye had a higher yield than the other four varieties, with a maximum of 1.38 ± 0.22 g plant^{-1} in the *G. rosea* treatment (Table 7).

DISCUSSION

Legume crops are closely associated with symbiotic microbial communities that influence plant traits related to plant growth and yield (Cardoso and Kuyper, 2006; Calvo et al., 2014; Lesueur et al., 2016; Begum et al., 2019; Xiang et al., 2022; Aguégué et al., 2023). In the present study, five traditional and modern groundnut varieties were tested for requirements with or without indigenous and exotic arbuscular mycorrhizal fungi (AMF) inoculants. The efficacy of the AMF inoculants was assessed in terms of their ability to increase root AMF colonization, plant growth, leaf chlorophyll content and yield parameters. The results confirm the functional variation among the inoculated AMF, which is crucial in establishing potential formulations of AMF inoculants to enhance groundnut productivity. The efficacy of inoculated AMF was specifically dependent on the groundnut genotype used, with the varieties Amoul Morom, 55-437 and Essamay being more responsive to AMF inoculation than the varieties Fleur 11 and Sunu Gaal.

High root colonization ability is an important requirement for the selection of AMF inoculants in crop production (Calvo et al., 2014; Hart et al., 2015; Aguégué et al., 2023). The fact that groundnut is a root-hairless crop (Nambiar et al., 1983; Wissuwa and Ae, 2001) suggests that its dependence on AMF for nutrient uptake would be high, highlighting the importance of AMF fertilizers in groundnut. In this study, as predicted, the AMF inoculants tested appeared to be infective even in the presence of native AMF. In the inoculated treatments, there was a significant increase in the rate of root AMF colonization of all groundnut varieties compared to the control plants. Evidence of increased root AMF colonization by mycorrhizal inoculation has been reported previously (Cely et al., 2016; Thioub et al., 2019; Adeyemi et al., 2021; Sene et al., 2021, 2023) and our results are consistent with such previous findings. The inoculated strains may compete with indigenous AMF for colonization sites and spread rapidly within the host roots.

Furthermore, the results showed that root AMF colonization varied greatly depending on the groundnut variety used. Specifically, inoculation

Table 7. Yield attributes (number of pods per plant, pod weight) of groundnut varieties at harvest.

Treatments	Groundnut varieties									
	Amoul Morom		Essamaye		55-437		Fleur11		Sunu Gaal	
	No. of pods	Wt. of Pods (g)	No. of pods	Wt. of Pods (g)	No. of pods	Wt. of Pods (g)	No. of pods	Wt. of Pods (g)	No. of pods	Wt. of Pods (g)
F. mosseae	5.00 ± 1.63^{a}	0.27 ± 0.03^{a}	6.00 ± 3.83^{ab}	0.56 ± 0.12^{b}	4.25 ± 1.50^{ab}	0.44 ± 0.15^{ab}	4.75 ± 0.96^{a}	0.59 ± 0.04^{a}	4.00 ± 1.41^{b}	0.49 ± 0.14^{a}
R. aggregatus	3.25 ± 1.50^{ab}	0.15 ± 0.02^{b}	5.75 ± 2.87^{ab}	0.52 ± 0.14^{b}	7.25 ± 1.71^{a}	0.69 ± 0.07^{a}	4.75 ± 1.26^{a}	0.58 ± 0.11^{a}	5.75 ± 0.96^{ab}	0.46 ± 0.07^{a}
R. fasciculatus	3.00 ± 0.00^{ab}	0.10 ± 0.01^{bc}	6.75 ± 1.50^{ab}	1.11 ± 0.20^{a}	4.33 ± 2.31^{ab}	0.55 ± 0.24^{ab}	5.25 ± 2.22^{a}	0.53 ± 0.15^{a}	4.25 ± 1.26^{b}	0.50 ± 0.15^{a}
G. rosea	3.00 ± 1.00^{ab}	0.10 ± 0.01^{bc}	10.0 ± 1.41^{a}	1.38 ± 0.22^{a}	4.00 ± 2.00^{ab}	0.37 ± 0.11^{ab}	6.50 ± 2.38^{a}	0.75 ± 0.25^{a}	7.50 ± 1.73^{a}	0.65 ± 0.09^{a}
R. irregularis	2.50 ± 0.58^{b}	0.10 ± 0.02^{bc}	4.25 ± 2.06^{b}	0.51 ± 0.04^{b}	7.25 ± 1.71^{a}	0.56 ± 0.20^{ab}	5.00 ± 1.15^{a}	0.56 ± 0.02^{a}	3.33 ± 1.15^{b}	0.50 ± 0.09^{a}
Control	2.50 ± 0.58^{b}	0.08 ± 0.01^{c}	3.75 ± 0.50^{b}	0.44 ± 0.13^{b}	3.00 ± 0.00^{b}	0.35 ± 0.03^{b}	4.75 ± 0.96^{a}	0.55 ± 0.10^{a}	4.00 ± 0.00^{b}	0.50 ± 0.21^{a}

No. of pods (number of pods per plant); Wt. of Pods (weight of pods per plant); Values (mean ± standard error) are an average of five replications; means ± standard error within the same column followed by the same superscript letters are not statistically different at the 5% probability according to Tukey test.

with *R. irregularis* resulted in the highest root AMF colonization in the varieties Amoul Morom, Essamay and 55-437. The increased root AMF colonization with *R. irregularis* is consistent with the report of Köhl et al. (2016). These authors reported that the *R. irregularis* has a broad niche with the ability to successfully compete with native AMF, and thus can successfully colonize root plants in a wide range of soils. In the case study of the present study, such high root AMF colonization was not observed in the varieties Fleur 11 and Sunu Gaal. In addition, *R. aggregatus* also showed a higher colonization of variety 55-437 than the other varieties. This indicates a discrepancy in the ability of AMF inoculants to compete and colonize the groundnut varieties, and supports a report by Jie et al. (2013) on soybean (*Glycine max* L.). To date, there is no convincing evidence of AMF host specificity, but host preference and selectivity have been widely reported (Torrecillas et al., 2012; Bender et al., 2016; Köhl et al., 2016), and variability amongst different AMF species in root AMF colonization has been investigated in several studies (Wagg et al., 2015; Berruti et al., 2017; Thioub et al., 2019).

It has been previously reported that inoculation causes a change in the root system morphology in groundnut through lateral root development (Yano et al., 1996; Gutjahr and Paszkowski, 2013). Such changes in the root system are generally considered to have a large uptake capacity (Smith and Read, 2008; Fortin et al., 2015; Aguégué et al., 2023). Although root length was not assessed in the present study, there is sufficient evidence that AMF inoculation had a positive effect on this parameter, as root dry weight (RDW) increased significantly in almost all inoculated treatments. This could lead to an increase in the volume of root tissue that can be colonized by AMF or rhizobia. Therefore, a very clear difference in nodule formation was observed between the inoculated and uninoculated plants. In this case, the inoculated plants showed a significantly higher number of nodules, regardless of the groundnut variety. In contrast, no such increase in root AMF colonization was observed in any of the varieties.

Leaf chlorophyll content was generally higher in inoculated than in uninoculated plants, irrespective of the variety used. The AMF association has been reported to affect the host plants in terms of stomatal movement and leaf photosynthesis. This has been shown to increase chlorophyll content and the rate of transpiration and photosynthesis (Sheng et al., 2008; Ye et al., 2022). On the other hand, the highest leaf chlorophyll content could be due to the highest nodule formation in the inoculated plants, suggesting a potential synergistic effect between inoculated AMF and indigenous rhizobia and thus the basic function of rhizobia in N_2 fixation. The efficiency of N_2 fixation in groundnut may result in the accumulation of nitrogen in plant tissues, which in turn reflects the synthesis of chlorophyll.

The results of this study also showed that different varieties responded differently to the AMF applied in terms of plant growth and yield parameters. Significantly higher plant growth and yield parameters for Amoul Morom, Essamay and 55-437 varieties were reported with AMF inoculation. This could be attributed to a higher responsiveness of these groundnut varieties to the inoculated AMF strains. The inoculation of efficient and compatible AMF may help to establish symbioses earlier than the indigenous AMF populations, resulting in increased plant growth benefits. Indeed, studies using P^{32}-labelled phosphate have clearly shown that P is translocated from the soil to the root by the AMF mycelium (Qin et al., 2022), and perhaps the efficiency differs between plant genotypes. In contrast, the genetically closely related varieties Fleur 11 and Sunu Gaal were less responsive to the AMF inoculation in the case study of this present study. This suggests functional differences between AMF inoculants and is consistent with a number of recent studies reporting differences in host genotypes in response to AMF inoculation (Calvo-Polanco et al., 2016; Duc et al., 2017; Bazghaleh et al., 2018; Frew, 2020). Furthermore, the increased nodule numbers in the *R. irregularis* and *G. rosea* treatments did not translate into plant growth or yield parameters in the Fleur 11 and Sunu Gaal varieties. This was not expected and indicates the need for further selection of highly efficient and appropriate AMF inoculants for successful inoculation of Fleur 11 and Sunu Gaal. Similar negative or neutral effects after AMF inoculation were observed by Chotangui et al. (2022) on two groundnut varieties in the Western Highlands of Cameroon. However, the potential of inoculated AMF in our case study may be underestimated as the confined space of the pots does not allow for maximum root development.

Conclusion

The demand for microbial inoculants is increasing, driven by the need for sustainable and environmentally friendly agricultural practices and safer and healthier food. To select the best arbuscular mycorrhizal fungi (AMF) inoculants for five traditional and modern Senegalese groundnut varieties, we hypothesized that the response of groundnut to indigenous and exotic AMF inoculation is cultivar dependent and that there is a different degree of variability between traditional and modern cultivars. The results of this study showed that the AMF inoculants tested promoted increases in various parameters analyzed. In particular, inoculation efficacy did not depend on the origin (exotic or indigenous) of the inoculated AMF and no clear relationship was found between the fact that the varieties used were traditional or modern. However, the response to AMF inoculation differed between varieties, demonstrating the differential feedback between groundnut genotypes and AMF partners. Groundnut varieties such as Essamay, Amoul Morom and 55-437 responded better than the closely related genotypes Fleur 11 and Sunu Gaal. These results confirm the functional variation among inoculated AMF, which is crucial for establishing potential formulations of AMF inoculants to improve groundnut productivity. According to this study, further selection of compatible AMF partners would be useful to improve inoculation success with these latter varieties.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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Table S1. Leaf chlorophyll content at 30 days after planting in response to groundnut varieties inoculation with arbuscular mycorrhizal fungi

	Groundnut varieties				
Treatments	**Amoul morom**	**Essamaay**	**55-437**	**Fleur 11**	**Sunu Gaal**
F. mosseae	41.1 ± 3.79[a]	35.8 ± 4.37[a]	34.2 ± 4.92[a]	34.8 ± 1.31[a]	37.9 ± 3.34[a]
R. aggregatus	41.5 ± 2.72[a]	31.6 ± 4.70[a]	31.3 ± 4.63[a]	37.0 ± 2.61[a]	39.0 ± 2.76[a]
R. fasciculatus	38.5 ± 2.55[a]	32.3 ± 3.34[a]	33.8 ± 4.10[a]	39.5 ± 5.97[a]	36.0 ± 5.81[a]
G. rosea	38.9 ± 1.40[a]	33.5 ± 6.06[a]	35.0 ± 3.29[a]	35.3 ± 1.10[a]	35.8 ± 6.59[a]
R. irregularis	38.9 ± 3.28[a]	34.5 ± 1.66[a]	31.6 ± 1.82[a]	37.3 ± 3.46[a]	40.0 ± 1.86[a]
Control	37.8 ± 1.68[a]	29.8 ± 6.84[a]	30.5 ± 3.33[a]	34.7 ± 2.00[a]	33.2 ± 5.45[a]

In columns, means with identical superscript letters are statistically equivalent at the 5% probability level.

Table S2. Leaf chlorophyll content at 45 days after planting in response to groundnut varieties inoculation with arbuscular mycorrhizal fungi

	Groundnut varieties				
Treatments	**Amoul morom**	**Essamaay**	**55-437**	**Fleur 11**	**Sunu Gaal**
F. mosseae	43.2 ± 2.40[a]	32.5 ± 3.64[a]	35.2 ± 1.73[a]	35.0 ± 1.73[a]	36.2 ± 5.19[a]
R. aggregatus	44.0 ± 2.40[a]	37.5 ± 3.24[a]	35.7 ± 1.90[a]	35.7 ± 1.58[a]	36.8 ± 2.17[a]
R. fasciculatus	43.2 ± 1.11[a]	32.7 ± 5.12[a]	35.9 ± 2.30[a]	36.3 ± 0.67[a]	37.8 ± 2.43[a]
G. rosea	43.1 ± 3.12[a]	36.0 ± 3.40[a]	34.4 ± 3.16[a]	36.4 ± 1.54[a]	37.0 ± 1.68[a]
R. irregularis	41.8 ± 2.06[a]	32.1 ± 1.44[a]	36.4 ± 2.44[a]	34.4 ± 2.28[a]	37.8 ± 3.73[a]
Control	40.8 ± 2.33[a]	31.9 ± 3.23[a]	33.4 ± 3.40[a]	33.0 ± 1.16[a]	33.0 ± 1.95[a]

In columns, means with identical superscript letters are statistically equivalent at the 5% probability level.