



Morphological and Structural Characterization of Rhizospheric Endomycorrhiza Communities Associated with Rice Grown in the Sahelian Zone (Chad)

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Authors' contributions

This work was carried out in collaboration among all authors. Author YN designed the study, performed the statistical analysis, wrote the protocol and wrote the first draft of the manuscript. Authors STT and AN managed the statistical analyses of the data. Author RT managed the literature searches. All authors read and approved the final manuscript.

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ABSTRACT

In Chad, rice is grown for its starchy seeds or caryopses, but is also for consumption in the form of seeds pasta with soup. However, little or no work has been conducted in Chad to assess the composition of endomycorrhiza community inhabiting the plant rice rhizospheres. Hence, the main objective of this study was to investigate on native endomycorrhiza that are efficient to establish a symbiotic relationship with two rice varieties in the Sahelian zone in Chad. Two rice varieties were grown in a pot experiment on composite soils samples from Laï, Kelo, Kolobo and N'Djamena for 3 months. Parameters such as mycorrhizal frequency, intensity, specific density and richness were determined following to the standard methods. After spore extraction, species description and

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characterization were obtained through the informations provided by the International Vesicular Mycorrhizal fungi collection (INVAM): <http://invam.caf.wv.edu/fungi/taxonomy/species ID.htm>. The highest specific endomycorrhizal density (97.3%) and richness (11%) were registered respectively at kelo, Lai and Kolobo. The endomycorrhizal frequency and intensity were respectively between 4.33-7.33% and 0.8-2.9% for the two rice varieties. Eleven endomycorrhiza species belonging to six genera were identified from different soil samples. These include *Septoglomus* (*S. constrictum*, *S. deserticola*); *Rhizophagus* (*R. aggregatus*, *R. fasciculatus*, *R. intraradices*); *Acaulospora* (*A. lacunosa*, *A. rugosa*, *A. trappei*); *Claroideoglomus lamellosum*; *Glomus pansihalos* and *Diversispora epigae*. *S. constrictum* was the most dominant and frequent species found in all the soil sampling sites, while *R. aggregatus* was the less frequently encountered specimen. The lowest macorrhizal richness and frequency obtained is a proof that cultivation of rice dependent on endomycorrhiza in this zone. The multi-native endomycorrhiza spores identified are the active principle to be included in the bioinoculants production in order to increase and improve the production of rice in the south of Chad.

Keywords: Chad; endomycorrhiza species; mycorrhizal intensity; mycorrhizal frequency; rice varieties; specific richness; specific density.

1. INTRODUCTION

During the process of improving the living conditions of populations, Chad is undertaking several development projects, including the one concerning the improvement of rice production in the Tandjile, Mayo Kebbi and the two Logones zones, with the aim of consolidating the food security [1]. Due to the low soil fertility, rice is usually grown in the aforementioned zones with the addition of chemical fertilizers to balance the mineral balance and allow the plants to express their potentials, although this practice clearly causes serious food and environmental risks of pollution [2].

Sustainable agronomic research has shown that food production can be better conducted with technologies based on biological processes [3]. Among these techniques, organic farming with the application of biofertilizers has been highly recommended [4-9]. In fact, endomycorrhiza fungi are one group of the soil microorganisms, which are generally involved in the improvement of plant growth, as well as in their protection against plant pests and diseases [10]. They also reduce environmental contamination by their ability to capture soil nutrients, causing a decrease in the use of chemical fertilizers while ensuring good crop yields [11]. Currently, it is estimated that 90% of cultivated plants are associated to mycorrhiza [12].

In Chad, rice is grown for its starchy seeds or caryopses, but is also consumed in the form of seeds pasta with soup. Although some studies have been carried out in Cameroon on the diversity of mycorrhiza associated to maize and cowpea [13,14], little or no work has been

conducted in Chad to assess the composition of endomycorrhiza community associated to rice. The main objective of this study was to investigate on endomycorrhiza that are able to establish a symbiotic relationship with two rice varieties in the Sahelian zone in Chad. In short, the endomycorrhiza spores diversity and distribution within the rhizosphere of rice are to be evaluated, as a step forward to the production of elite endomycorrhizal biofertilizers to boost the rice production in Chad.

2. MATERIALS AND METHODS

2.1 Description of Study Sites and the Experimental Set Up

Soils were sampled in the Tandjile regions comprising Kelo (N 9°18'50.7"), Lai (N 9°23'24"), Mayo-Kebbi (Kolobo): (N 9°54'18.2") and the city of N'Djamena (N 9°54'18.6"), as shown on Fig. 1 Soils were collected within the 0-20 cm depth horizon in the five selected sites. In each site, soils were collected at 5 different locations along a transect at a rate of 10 kg per location, for a total of 50 kg soil per site that was stored in 2 kg plastic pots.

Fig. 2 summarizes the experimental design in a random bloc A, B, C, D representing the soil sampling sites, while at each site M (Maditolngar) and T (Tox 728-1) indicate the two rice varieties repeated each in twelve 2 kg plastic pots.

2.2 Determination of the Physico-chemical Properties of Soils

Soils were analysed at the Chadian Institute for Agronomic Research for Development (ITRAD)

in N'djamena. The parameters analysed were: Organic carbon (CO), organic matter (OM), hydrogen potential (pH), total nitrogen (N), available phosphorus (P) and available potassium (K). Soil parameters were analysed by the PALINTEST Kit using a 5000 Direct Water Reading Proof Spectrophotometer that determines the soil physico-chemical properties.

When choosing a test, the instrument automatically selects the parameters required for accurate analysis, including wave length and reaction time. After the completion of the test, additional optional tests are available and the results can be converted into different units, depending on the studied parameter.

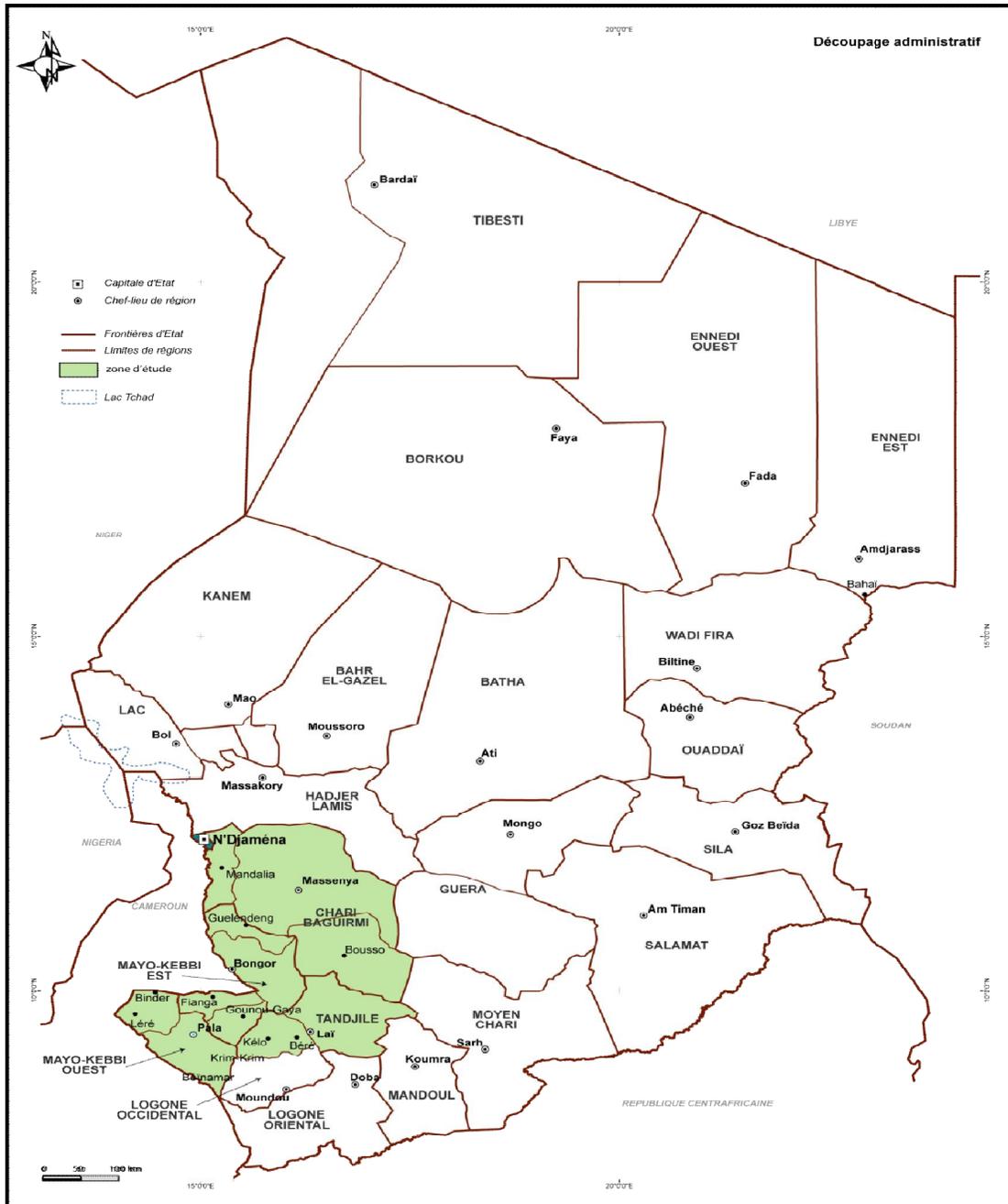


Fig. 1. Map of Chad showing the soil sampling study sites (in green)

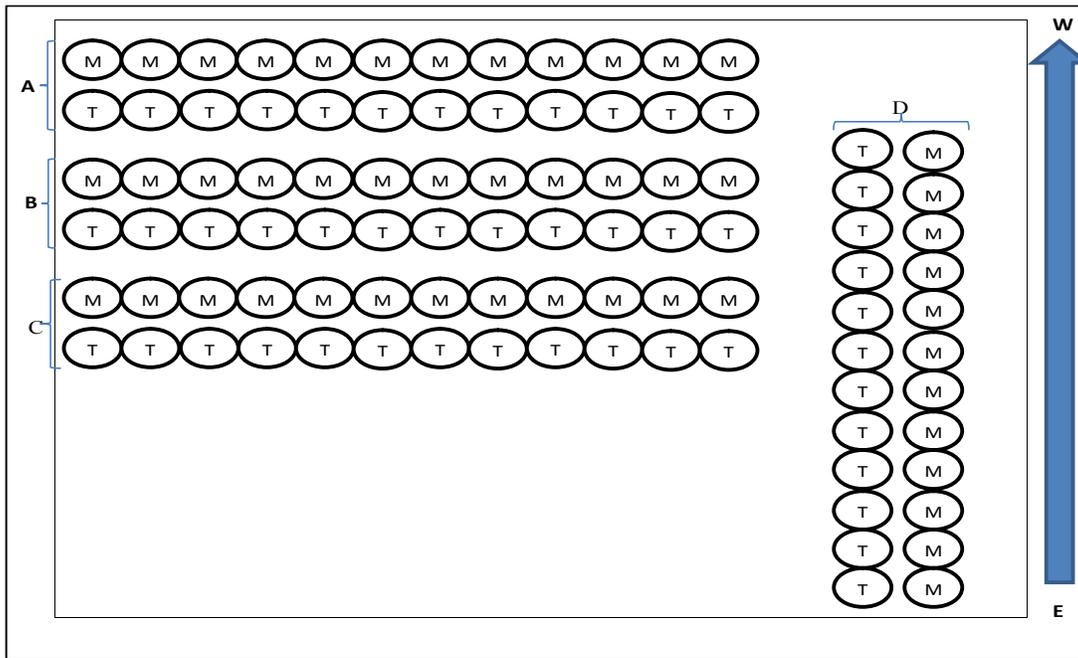


Fig. 2. Experimental layout of rice varieties in different soil types

T: TOX 728-1; M: Madjitolngar; A: Laï soil (Tandjilé); B: N'Djaména soil (Chari Baguirmi); C: Kolobo soil (Mayo Kebbi); D: Kelo soil (Tandjilé)

2.3 Trapping of Endomycorrhizal Spores from Soil Samples

Endomycorrhizal spores inhabiting the rhizosphere of rice plant were trapped according to the method described [15] and modified as follows: Local rice varieties Tox728-1 and Madjitolngar seeds originated from the Malla Agricultural Research station (Tandjile) were sown in twelve different pots, each containing 1 kg of composite soil per site. Pots placed out of the contact with soil were watered with natural rainfall for three months (July to September 2016). Plants roots at maturity and rhizospheric soils were sampled and preserved in the refrigerator at 4°C for laboratory analysis.

2.4 Determination of the Degree of Mycorrhization in Infested Roots

Fine harvested rice roots were thinned through the method to assess endomycorrhizal infestation structures [16]. Youngest roots were cut into 1-2 cm length, before they were successfully washed, inserted into a test tube containing 10% potassium hydroxide, and heated in a water bath at 90°C for 30 minutes to clear the roots. Potash was discarded then, the solution was filtered through a sieve before neutralization by rinsing with acidified water.

Neutralized roots were depth into cotton blue under a water bath for 15 minutes, filtered again through a sieve and rinsed with distilled water. Some roots were mounted in water for direct observations, while other were mounted in glycerine for later observations. The estimation of mycorrhization parameters was evaluated as previously described [17].

2.5 Evaluation of the Mycorrhizal Frequency and Intensity in Rice Plant Roots

The frequency of mycorrhization F (%) was expressed as the percentage of colonization of the rice plant roots by arbuscular fungi as follow:

$$F (\%) = 100(N-N_0)/N$$

With N : The number of root fragments observed, and N_0 the number of non-mycorrhizal fragments.

The intensity of mycorrhization was referred to as the degree of roots colonized by arbuscular fungi and expressed as:

$$M\% = (95n_5 + 70n_4 + 30n_3 + 5n_2 + n_1)/N,$$

where n_5, n_4, n_3, n_2 and n_1 are the number of root fragments noted number 5, 4, 3, 2 and 1.

The proposed scoring system is based on an overall assessment of each of the 30 fragments, which is assigned a grade of between 0 and 5 corresponding to the estimate of the proportion of cortex colonized by the mycorrhizal symbiont.

2.6 Extraction of Endomycorrhiza Spores

Endomycorrhizal spores were extracted using the wet method [18] with slight modifications. It consists essentially of suspending 50 g soil sample in 1L water, then after mechanical agitation, passing the solution through a series of sieves ranged 50, 100, 200, 300, 400 μm to separate organic matter from other soil particles. The density gradient was created by centrifugation to obtain a suspension of particles with size and density similar to that of endomycorrhiza [19]. All spores were collected after filtration through a 25 μm sieve.

2.7 Morphological and Structural Characterization of Endomycorrhizal Spores in Rice Root

During the identification process, extracted endomycorrhiza spores were grouped into morphotypes based on their size, shape and color. A slide (76 x 26 mm) carrying two groups of spores was used. At the right end of the slide was deposited a few drops of solvent Polyvinyl-Lactic Acid-Glycerol (PVGL) that will allow the observation of the external morphology of spores. At the left end of the slide, the spores are deposited in the PVLG + Melzer reagent useful in the study of membrane layers [20]. A space was left at one end of the slide that will bear the name of the specimen.

The determination of morphotypes at the level of genus was assessed based on the classifications criteria [21]. The original descriptions of species, as well as the descriptions provided on the website of the International Vesicular Mycorrhizal fungi collection [22]: <http://invam.caf.wv.edu/fungi/taxonomy/speciesID.htm> were used as the reference during the identification process. Morphological characters of spores were compared with those of standard specimens and the reference strains. Parameters used to characterize endomycorrhizal spores were evaluated based on the formula proposed [17].

2.8 Analysis of Endomycorrhiza Diversity in Soil Samples

Specific Richness measures the diversity directly based on the total number of species in a specific site. This method depends on the sample size and does not consider the relative abundance of different species. Its ecological value is therefore limited. As an ecological concept, abundance is another important component of diversity defined as heterogeneity, thus, the equitability of the distribution of individuals among species. Biological indices provide a better expression of diversity in a given ecosystem.

The species richness refers to the total number of different morphotypes recorded in a 100 g soil sample, and was expressed by:

R (%): $N/100$ g, where N is the number of different specimens.

The specific density indicates the number of spores recovered in 100 g soil sample, and was express as:

D (%): $N / 100$ g, where N is the number of spores.

The diversity of endomycorrhizal species in all the sites was calculated using Shannon-Weaver diversity index (H) [23].

The Shannon index is given by the formula below:

$-H = -\sum p_i \ln p_i$ Where $p_i = S/N$, S is the total number of individuals of one species, N is the total number of all individuals in the sample and $\ln =$ logarithm to base e. The proportion of species relative to total number of species (p_i) was calculated, and multiplied by natural logarithm of this proportion ($\ln p_i$). The results were summed across the species, and multiplied by -1.

2.9 Data Analysis

The data has been statistically analyzed using the "stratigraphics 5.0" program that performs the Analysis of Variance (ANOVA). The averages of results from different regions (Adamawa, North and Far North) were separated using the Less Significant Difference (LSD) at the indicated

probabilities. The microsoft excel program was used to plot graphs.

The relationships between the different parameters were determined by the Pearson correlation coefficient using the "Stratigraphics 5.0" program.

3. RESULTS AND DISCUSSION

3.1 Physico-chemical Properties of Soils

The physico-chemical properties of soils were different from one soil sampling site to another (Table 1). Soil pH values failed between 4.73 and 5.54, indicating that they are acid. The organic matter content was between 0.149 and 0.170%, whereas the organic carbon varied from 0.086 to 0.098%. All these values were lower than 1%, demonstrating a weak restitution of organic matter to soil. The elevated soil nitrogen (0.14-1.89 mg/l), phosphorus (0.19-1.82) and potassium (0.5-8.1 mg/l) concentrations were probably attributed to intensive uses of chemical fertilizers by farmers in the sites. The characteristics of such soils were qualified as soils with low degree of fertility [24].

3.2 Mycorrhization Frequencies of Rice Varieties within and between Localities

The mycorrhization frequencies values registered in localities varied from 4.66% at Kolobo, 5.33% at Kelo, 7.33% at Laï, to 4.33% at N'Djamena (Fig. 3). Apart from the Kolobo site with 5.33% mycorrhization frequency, the TOX728-1 variety showed frequencies lower than that of Magitongar in all the other sites. These observed values are lower than those reported in Maroc where mycorrhization frequencies in *Tetraclines articulata* roots from 7 sites was comprised

between 86-100% [25]. Similarly, 30-70% mycorrhization frequency was obtained in *Olea europa* roots between 5 different studied sites [16]. In contrast, plants of the Poaceae family have been reported to present less than 10% mycorrhization frequencies [26]. On the overall, the mycorrhization frequency differed from one rice variety to another, the Magitolngar variety showing more affinity to endomycorrhiza than the TOX728-1 variety. This was justified by the fact that one fungi species can behave differently during the infestation process of plants belonging to the same taxon [27].

3.3 Mycorrhization Intensities of Rice Varieties within and between Localities

As far as the Madjitolngar variety is concerned, the mycorrhization intensity was significantly ($p = 0.0021$) more elevated at Laï (2.9%) than at other localities where it was almost similar with 1.8% at Kolobo, 2.09% at Kelo, and 1.8% N'Djamena (Fig. 4). Although a similar mycorrhization intensity of 1.1% was reported in lignous species in Niger [28], these values were lower than between 35-55% pointed out in Maroc within *Citrus* rhizosphere [29]. For the Tox variety, the mycorrhization intensity was significantly ($p = 0.0043$) higher at N'Djamena (2.37%) than Kelo (0.8%), Kolobo (1.53%), and Laï (1.46%). More elevated mycorrhization intensities of 21% have been reported within *Ceratonia siliqua* rhizosphere [30], or 70% within *Saccharum officinarum* rhizosphere in Maroc [31]. The fact that the mycorrhization intensity of the rice variety Magitongar was better than that of TOX728-1 variety can indicate that Madjitolngar might possess a higher endomycorrhizal dependency than the TOX728-1 variety. The genetic pool of endomycorrhiza render them able to better associate to some plant species than others [32].

Table 1. Variation of soil properties from sampling sites

Soil parameters	Laï	Kolobo	Kelo	N'Djamena	P-value
Carbone (%)	0.094a	0.097a	0.098a	0.086a	0.99
OM (%)	0.162a	0.167a	0.170a	0.149a	0.99
pH	4.85a	4.73a	5.03a	5.54a	0.99
Total nitrogen (%)	1.89b	0.14a	1.19a	0.21a	0.01
Phosphorus (mg/l)	0.19a	0.34a	0.22a	1.82b	0.04
Potassium (mg/l)	1.8a	0.5a	2.8a	2.5a	0.30
C/N ratio	0.049a	0.692a	0.082a	0.409a	0.15

OM: Organic Matter; Ddf= 3. For each soil parameter in a raw, values affected with the same letter are no significantly different between the sampling sites at the indicated level of probability

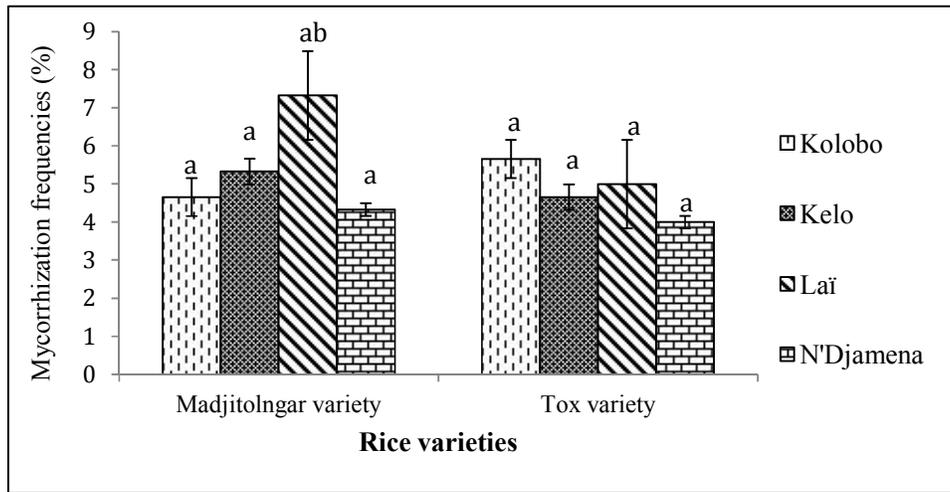


Fig. 3. Variation of mycorrhization frequencies within the sites and between the two rice varieties

Magitolngar variety $F = 2.32$; $p = 0.15$; $Ddl: 3$; $LSD: 95\%$; *Variety Tox*: $F = 0.87$; $p = 0.49$; $Ddl: 3$; $LSD: 95\%$. For each soil rice variety, bars indicating the mycorrhization frequencies affected with the same letter are no significantly different between the sampling sites at the indicated level of probability

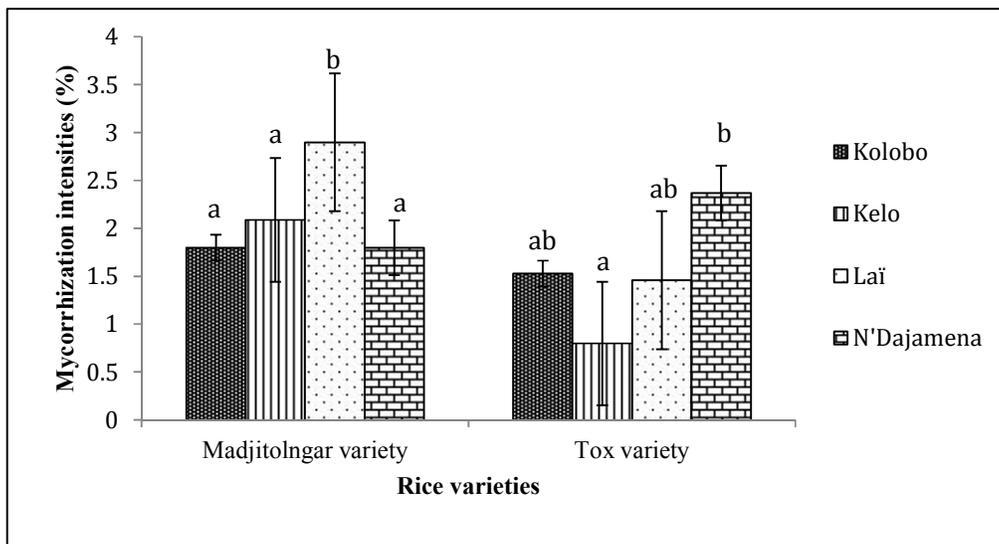


Fig. 4. Variation in the mycorrhization intensities of rice varieties grown in different localities in Chad

Magitolngar variety: $F = 12.70$; $P = 0.002$; $LSD: 95\%$; $Ddl: 3$; *Tox variety*: $F = 10.05$; $p = 0.004$; $Ddl: 3$; $LSD: 95\%$. For each soil rice variety, bars indicating the mycorrhization intensities affected with the same letter are no significantly different between the sampling sites at the indicated level of probability

3.4 Differences in the Specific Densities of Spores between Rice Varieties at Different Sites

The number of endomycorrhiza spores in 100 g of soil varied between localities and between the Madjitolngar and TOX728-1 (Fig. 5) rice varieties. For Madjitolngar rice variety, the

specific density of spores was significantly ($p < 0.0001$) greater Kelo (78%) and Lai (72%) compared to those of Kolobo (64%) and N'Djamena (17%). These values are not far from 59% reported in the rhizosphere of *Zea mays* grown in Northern Cameroon [13]. As for the TOX728-1 variety, the specific density of spores found at Kelo (97.3%) was significantly ($p <$

0.0001) and consistently more elevated than those observed at Laï (94%), Kolobo (77%) and N'Djamena (13%). Globally, for the two rice varieties, the specific density of spore was more important at Kelo than at other localities, based on the low content of its soil in nitrogen (0.14%), phosphorus (0.34%) and potassium (0.5%). These findings are in line with high specific density of spores in low phosphorus soil content reported in Benin [10].

3.5 Differences in the Specific Richness of Spores between Rice Varieties at Different Sites

The specific richness of spores observed for the rice variety Madjitolngar was significantly greater ($p = 0.0033$) at Laï (11%) and Kolobo (10%) than at Kelo (5%) and N'Djamena (4%). As far as the rice variety TOX728-1 is concerned, the specific richness of spore was significantly higher ($p = 0.0033$) at Kolobo (11%) and Kelo (10%) than at N'Djamena (7%) and Laï (6%) (Fig. 6). These values are closed to those obtained within *Zea mays* rhizosphere (6%) in Northern Cameroon [14], compared to 0-6% range reported in the roots of *Ammophila arenaria* growing in maritime dunes of Bornholm-Danemark [33]. Conversely, the specific richness of spores in a low land cropping system was reported to be between 30-60% in Tunisia [26]. The lowest specific richness of spores was seen as the result of the effect of cultural practices, such as labour and crop rotation, that exert a selection pressure on soil

mycorrhizal community. This selection was translated as the modification of the population structure of endomycorrhiza [34], a direct cultural system being favorable to mycorrhization [35]. In short, the specific richness of spores TOX728-1 rice variety was greater than that of the Magitolngar variety in all the sites except at Lai, indicating that TOX728-1 variety has a high and wide affinity to endomycorrhiza compared to Madjitolngar variety.

3.6 Morphological and Structural Characteristics of Isolated Endomycorrhiza Spores

The morphological and structural analysis of isolated endomycorrhizal spores revealed the presence of eleven species grouped into six genera (Fig. 7). The authors of the fungal names are those presented at the URL web page <http://www.indexfungorum.org/Authors Of Fungal Names.htm>. Three species were of the genus *Rhizosphagus*, three of the genus *Acaulospora*, two of the genus *Septoglomus*, whereas each of the genera *Glomus*, *Diversispora* and *Claroideoglomus* was represented only by one species. In a related study conducted in Northern Cameroon, which also belongs to the Sahelian zone, six species were reported in *Zea mays* rhizosphere, groups in four different genera [14]. It is convenient to state that common endomycorrhiza found in the two plant rhizospheres were *R. intraradices* and *D. epigae*, and thus are considered as promiscuous to the

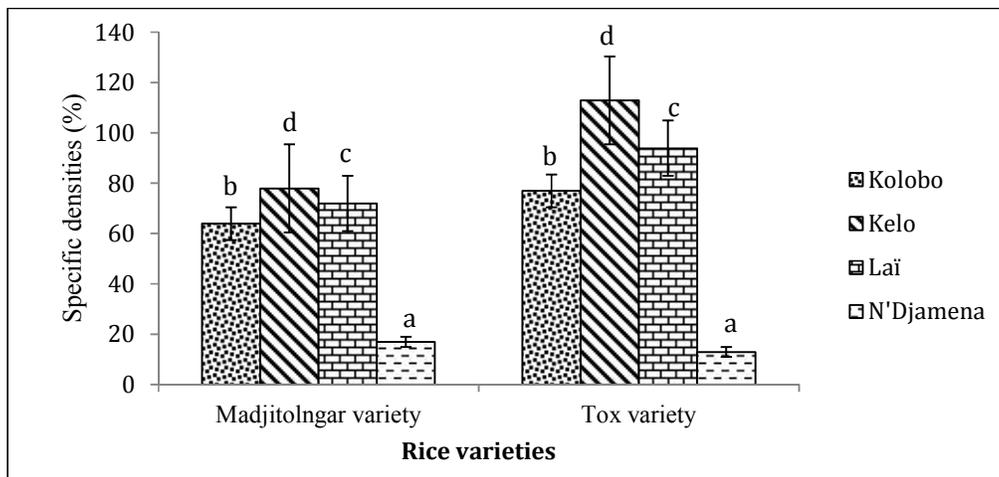


Fig. 5. Variation in the specific densities of spores in rice varieties grown in different localities in Chad

Magitolgar: $F = 243.45$; $p < 0.0001$; $Ddl: 3$; $LSD: 95\%$; *Tox*: $F = 648.66$; $p < 0.0001$; $Ddl= 3$; $LSD: 95\%$. For each soil rice variety, bars indicating the specific densities affected with the same letter are no significantly different between the sampling sites at the indicated level of probability

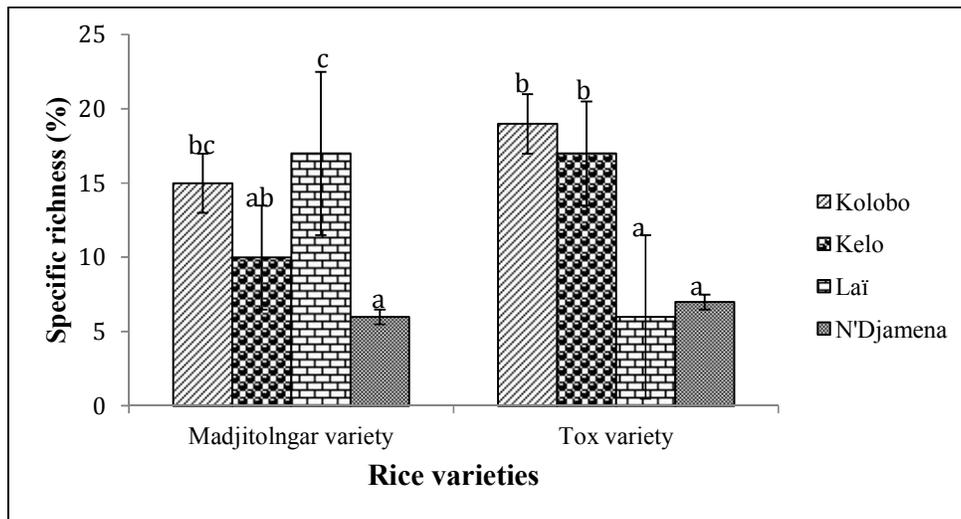


Fig. 6. Differences in specific richness of rice varieties between localities

Magitolngar: $F = 11.00$; $p = 0.0033$; $Ddl = 3$; $LSD = 95\%$; Tox: $F = 12.50$; $p = 0.0022$; $Ddl = 3$; $LSD = 95\%$. For each soil rice variety, bars indicating the specific richness affected with the same letter are no significantly different between the sampling sites at the indicated level of probability

two plants. Structurally, some species were protected by three membrane layers (*D. epigae*, *G. pansihalos*, *R. aggregatus*, *S. deserticola*, *A. lacunosa*, *S. constrictum*), other by two membrane layers (*R. intraradices*, *A. rugosa*, *Acaulospora trapei*, *R. fasciculatus*, *C. lamellosum*), while in addition some harboured a suspensor hyphae (*S. constrictum*, *A. trapei*, *G. pansihalos*, *R. intraradices*).

3.7 Distribution of Endomycorrhizal Spores within the Rhizosphere of Two Rice Varieties at Different Localities

The distribution of endomycorrhizal spores within the rhizosphere of two rice varieties at different sampling sites is illustrated in Table 2. *R. aggregatus* appeared as the most uncommon species, since it was only encountered within the rhizosphere of Tox variety at Kolobo, Kelo and N'Djamena. In contrast, *S. constrictum* was present in all the localities and was frequently encountered. These observations are in agreement with reported results [35], which pointed out that arbuscular mycorrhiza belonging to the genus of *S. constrictum* were the most represented in the fungi community within a plot submitted to direct cultural system for a long period. Therefore, *S. constrictum* seems to be ubiquitous, since it appears to be adapted to a diverse pedoclimatic conditions represented here by soil sites. The highest specific diversity was observed at Laï for Magitolngar rice rhizosphere

($H' = 2.24$), opposite to the lowest specific diversity observed at N'Djamena in rhizosphere of the same rice variety ($H' = 1.21$).

3.8 Correlation between Some Studied Parameters of Rice Plants

Table 3 reveals significant positive and significant correlations were observed between endomycorrhizal richness and clay content ($r = 0.726$, $p < 0.044$) on one hand, the mycorrhization frequency and mycorrhizal intensity ($r = 0.712$, $p < 0.033$) on the other.

These findings are an indication that the specific richness of endomycorrhizal spores increases with the clay content and support the findings that substrates such as clay are important for the multiplication of *Glomus intraradices*, *G. mosseae* and *G. verruculosum* in *Zea mays* rhizosphere [44]. In contrast, there were negative correlations between sand and clay ($r = -0.932$, $p < 0.0004$) on one hand, species richness and sand content ($r = -0.795$, $p = 0.001$) on the other. The decrease in sand content with specific richness may indicate the intensive cultural practices applied in the studied soil sites that negatively affect the community of endomycorrhiza in cultivated soils. Our results also line with the reversal linear relationship between the lower endomycorrhizal density and the higher phosphorus content in crop rhizosphere [9].

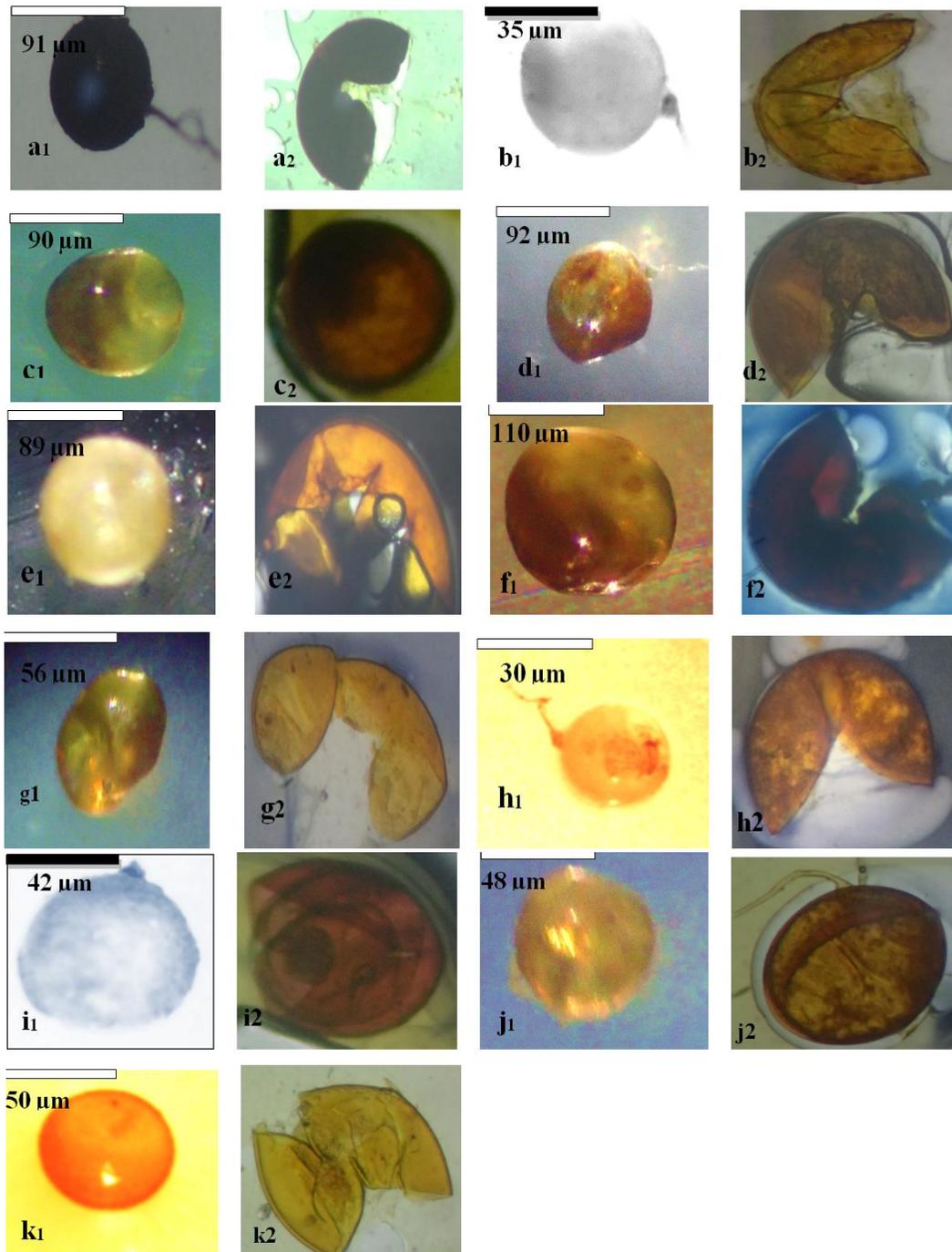


Fig. 7. Morphological and structural diversity of isolated endomycorrhiza spores from sampled soils

For each specimen, on the left are intact spores, while on the right are spores showing wall layers 2-3 after intensive staining in Melzer's reagent. They are recognised based on the comparison with the descriptions first provided by different authors; A. *Septoglomus constrictum* [36]; B. *Acaulospora trapei* [37]; C. *Rhizophagus fasciculatus* [38]; D. *Acaulospora lacunosa* [37]; E. *Claroideoglomus lamellosum* [39]; F. *Septoglomus deserticola* [36]; G. *Rhizophagus aggregatus* [40]; H. *Glomus pansihalos* [41]; I. *Acaulospora rugosa* [37]; J. *Rhizophagus intraradices* [42]; K. *Diversispora epigae* [43]; two wall layer spores (a, c, d, f, g, h, k); three wall layer spores (b, e, i, j)

Table 2. Distribution of spores in different soil sampling sites

Spore diversity	Kolobo Tox	Kolobo Madjitolngar	Kelo Tox	Kelo Madjitolngar	Laï Tox	Laï Madjitolngar	N'djamena Tox	N'djamena Madjitolngar
<i>A. lacunosa</i>	+	-	+	/	+	-	/	/
<i>A. rugosa</i>	+	-	+	/	-	++	++	/
<i>A. trappei</i>	++	+	/	/	/	+	/	+
<i>C. lamellosum</i>	-	++	+	++	+	+	/	++
<i>D. epigae</i>	++	-	+	++	+	+	/	++
<i>G. pansihalos</i>	+	+	+	++	++	+	/	/
<i>R. aggregatus</i>	+	/	+	/	/	-	+	/
<i>R. fasciculatus</i>	/	+	+	/	++	+	/	/
<i>R. intraradices</i>	+	-	-	++	-	+	+	/
<i>S.constrictum</i>	+++	++	++	+	+	+	+++	*
<i>S.deserticola</i>	++	++	++	/	-	+	++	/
H'	2.09	2.01	2.11	1.57	1.90	2.24	1.76	1.21

/ : Absent; - : [0-5]; + : [6-15]; ++ : [16-30]; +++ : [31-50]; *: [51- ∞]; H': Shannon diversity index;

Table 3. Correlation between some soil physico-chemical soil properties and endomycorrhization parameters (%)

	P	pH	Clay	Sand	F	I
pH	0.262 0.679ns					
Clay	0.354 0.352 ns	-0.434 0.341ns				
Sand	-0.264 0.535 ns	0.027 0.825ns	-0.932 0.0004**			
F	-0.238 0.464 ns	-0.023 0.875ns	-0.494 0.101ns	0.578 0.187ns		
I	0.076 0.807 ns	0.458 0.210ns	-0.3280 0.389ns	0.334 0.306ns	0.712 0.033**	
R	-0.003 0.931ns	0.054 0.893ns	0.726 0.044**	-0.795 0.001**	-0.576 0.109ns	-0.427 0.211

** indicates significant correlations. ns indicates non significant correlations; F (%): endomycorrhizal frequency, I (%): endomycorrhizal intensity, R(%): Specific richness, D (%): Specific density; P (%): Total phosphorus content; sand (%): Sand content

4. CONCLUSION

At the end of this study, rice [*Oryza sativa* (L.)] was found to be dependent on endomycorrhizal symbiosis in the south of Chad. Eleven endomycorrhiza species belonging to six genera were found in the rhizosphere of rice growing in different soils sampled at N'Djamena, Kelo, Kolobo and Laï in Chad. These include: *Septoglomus* (*S. constrictum*, *S. deserticola*); *Rhizophagus* (*R. aggregatus*, *R. fasciculatus*, *R. intraradices*); *Acaulospora* (*A. lacunosa*, *A. rugosa*, *A. trappei*); *Claroideoglossum lamellosum*; *Glomus pansihalos*; *Diversispora epigae*. Although *S. constrictum* was the most dominant and frequent species found in all the localities, *R. aggregatus* was the less frequently encountered specimen. The identification of these multi-native endomycorrhizal spores in the soils is a potential opportunity for production of endomycorrhiza inoculants to boost the production of rice varieties in the south of Chad.

DATA AVAILABILITY STATEMENT

Data used to support the findings of this study are available from the corresponding author upon request (raw data, tables, figures or any other supplementary document). There is no restriction to any reader on data access.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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