

Study of spore dynamics in the rhizosphere of millet over time

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Abstract

A study examining the influence of farming practices on soil fertility was conducted between 2015 and 2017 in Lélédjé, Moli Haoussa and Alambaré, three villages in the peripheral zone. The aim of this study was to compare variations in CMA biodiversity over time according to soil fertility management practices. Experiments were carried out in farming environments from 2015 to 2017 using randomised block designs in nine fields. Soil and root samples were taken during the tillering, stem elongation and grain filling stages. A morphological characterisation of arbuscular mycorrhizal fungal spores was carried out. The results revealed that *Glomus* and *Gigaspora* are the two genera of spores produced by these cultivation practices. The density of *Glomus* and *Gigaspora* is statistically higher in the mulching and control treatments (*Glomus*: 30.25; *Gigaspora*: 15.88). The analysis of variance showed no significant difference between years for the *Glomus* genus. However, unlike *Glomus*, *Gigaspora* was abundant in 2017 (17.25) compared to 2015 and 2016 (11.61 and 12.25, respectively). In view of the results obtained, *Glomus* appears to perform better than *Gigaspora*, as its abundance in mulching treatments and its resistance to environmental fluctuations make it preferable to *Gigaspora*. Mulching is therefore a farming practice for managing soil fertility that allows for good maintenance of CMA propagules. This could be an alternative to the use of mineral fertilisers, which are too expensive, unsuited to our environmental conditions and beyond the means of Nigerien farmers.

Keywords: soil fertility; mycorrhiza; *glomus*; *gigaspora*; Niger; farmer

1. Introduction

Like other West African countries, Niger's natural resources, which are its main source of production, are constantly affected by the cumulative effects of recurrent droughts (Harouna et al., 2005), resulting in chronic food insecurity for the population (Abdoul Habou et al., 2016). Many experiments and approaches have been tried, including water and soil conservation techniques, agroforestry, integrated soil fertility management, etc., most of which require large initial investments and often drastic changes to the agricultural landscape and practices (SET, 2011). The solutions usually recommended, such as the application of chemical fertilisers, the use of new seed varieties or water management, are not suitable for this vulnerable region of the world, as they are not accessible to all and can lead to environmental degradation (Becerra, 2012). It is therefore necessary to seek out effective technologies that can be applied by producers to compensate for the lack of fertilisers. Arbuscular mycorrhizal fungi (AMF), which live in symbiosis with plant roots, also offer the same benefits. Several authors have demonstrated the importance of these symbiotic fungi in improving the mineral nutrition of plants in poor soils (Bolan et al., 2011; Adjanohoun, 2012; Pérez, 2014). AM formation modifies plant/soil relationships. It is known to play a role (at the plant level) as a biofertiliser, bioprotector and bioregulator (Gianinazzi and Wipf, 2010). Despite the importance of AMF in soil fertility, very few studies in Niger have examined the influence that the technologies implemented by producers may have on the development of soil fungal biodiversity. The aim of this study is to characterise the AMF naturally present in the rhizosphere of millet according to different soil fertility management practices, in particular grazing, mulching and the addition of organic manure, with a view to

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proposing to farmers the best farming practices that can promote the presence of symbiotic microorganisms. The overall objective of this study is to compare the effect of farming practices on the dynamics of AMF populations over time.

2. Materials and methods

2.1. Study area

The study was conducted in the peripheral area of Niger's W Park in the rural commune of Tamou. This commune is located in the extreme south of the Say department, 75 km from Niamey (Map 1). It has a tropical climate strongly influenced by the Sahel, with recurrent climatic instability (Ambouta, 2002). In this study, three villages in the Tamou area (Moli Haoussa, Alambaré, and Lélédjé) were chosen as experimental sites. They were selected on the basis of their proximity to W National Park.

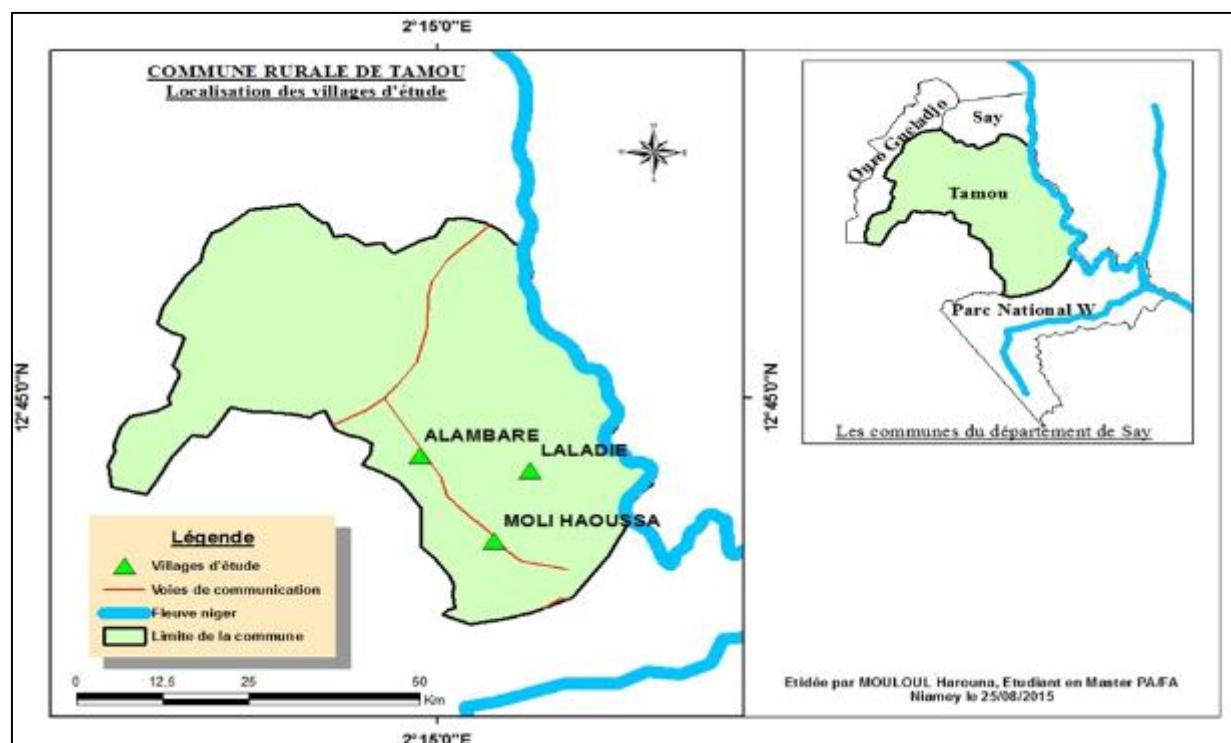


Figure 1 Map of villages in the study area (Source: Maidoukia, 2015)

2.2. Data collection

In each village, an experimental system consisting of 16 m^2 plots arranged in scattered blocks, with one block per field, was set up. The plots were arranged according to the amendments (grazing, mulching, organic manure application and control) applied by the farmers. The application only concerned the first year of the study (2015), and the plots were monitored over three years (from 2015 to 2017). The plant material used was millet (*Pennisetum glaucum* (L.) R. Br), a local variety (somno) and the main crop grown by farmers in the W park area. To carry out the various observations and measurements during the trials, three (3) plots were randomly selected from each field. The measurements and samples taken from these plots concerned the number of tillers and the number of ears per plot. Each year, soil and root samples were taken from the base of three millet plants in the first 10 centimetres at the tillering, booting and heading stages. The samples were sent to the Ecole Normale Supérieure laboratory to assess the physicochemical parameters of the soil and the quantities of spores using the Walker method (1982). The genus of the spores was identified by soil fertility management practices based on the criteria proposed by Schenck and Smith (1982).

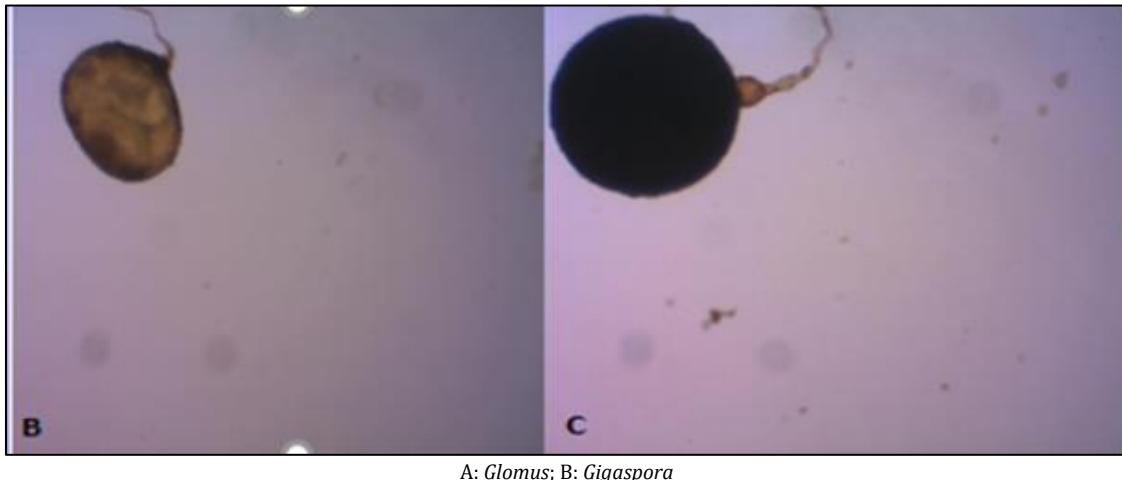
3. Statistical treatments

The data collected for the various parameters were subjected to analysis of variance (ANOVA) using Minitab software, version 16, and the means of the variables were compared using Tukey's test at a probability threshold of $p = 5\%$.

4. Results and discussion

4.1. Biodiversity of arbuscular mycorrhizal fungi

Observation under a binocular microscope revealed the presence of two genera of spores: *Glomus* spp. (Figure 2: A) and *Gigaspora* spp (Figure 2: C). These two genera have already been found in studies conducted by Ibrahim et al. (1995); Laminou (2010) and Sangaré (2017) on the mycorrhization of plants in Niger.



A: *Glomus*; B: *Gigaspora*

Figure 2 Diversity of arbuscular mycorrhizal fungus spores

These spores differ in colour (dark orange, brown, ochre, and black), shape (spherical oval), size (most spores are between 125 and 315 μm in size) and attachment hyphae. Spores of the genus *Glomus* are distinguished from those of the genus *Gigaspora* by the presence of an attachment bulb, which is characteristic of the genus *Gigaspora* (Figure 2). Characteristic structures such as colour, shape, the presence of a spore sac, germination shield and suspension bulb enable spores to be identified (Bâ et al., 1996). Size is an indicator for classifying spores. Large spores are generally *Gigaspora* sp., while small spores are from *Glomus* sp. (A.zézé et al., 2007).

4.2. Variation in spore density over time

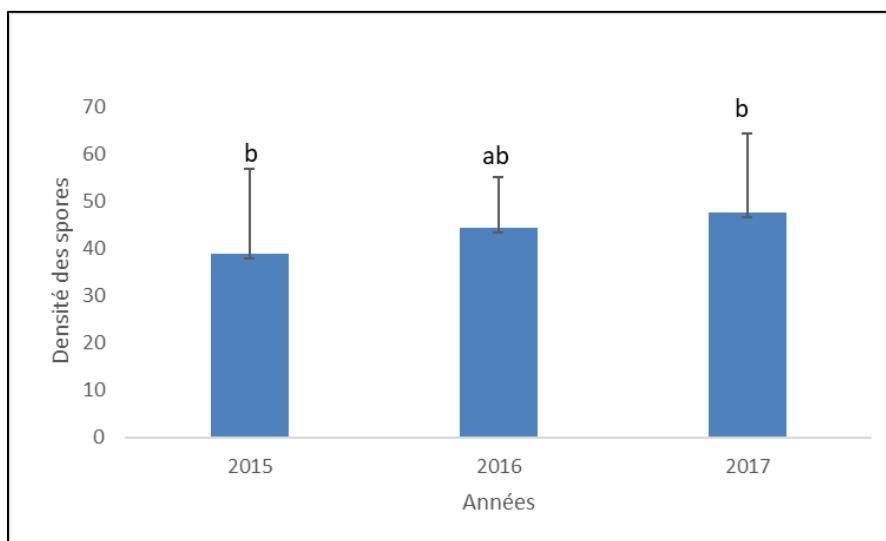


Figure 3 Spore density by year

Analysis of variance at $p = 5\%$ (Figure 3) showed no significant difference between the CMA density of the three years of the study (2015, 2016 and 2017). These results contradict those obtained by Sangaré et al. (2017), who obtained low spore densities in the last year of their study. In this study, the amendments applied in 2015 could be the reason for the

low density of spores collected. Indeed, according to Meddich et al. (2017), CMA abundance is higher in poor soils. When fertilisation is intense, plants' dependence on mycorrhization tends to decrease dramatically, and changes in soil inherent to cultivation practices have a decisive influence on mycorrhizae (Nyssens, 2012).

4.3. Averages that do not share any letters are significantly different.

The establishment of symbiosis is largely controlled by the fertilisation regime in the field. Indeed, over-fertilisation with P leads to a decrease in root colonisation by AMF (Garbaye, 2013). Furthermore, it has also been shown that by varying N and P fertilisation together, root colonisation and the number of CMA spores also vary (Jensen and Jabobsen, 1980).

4.4. Variation in spore genera over time

Figure 4 shows the density of spore genera over the years. Analysis of variance ($p = 5\%$) shows no significant difference between years for the genus *Glomus*. However, for the genus *Gigaspora*, there is a significant difference between years.

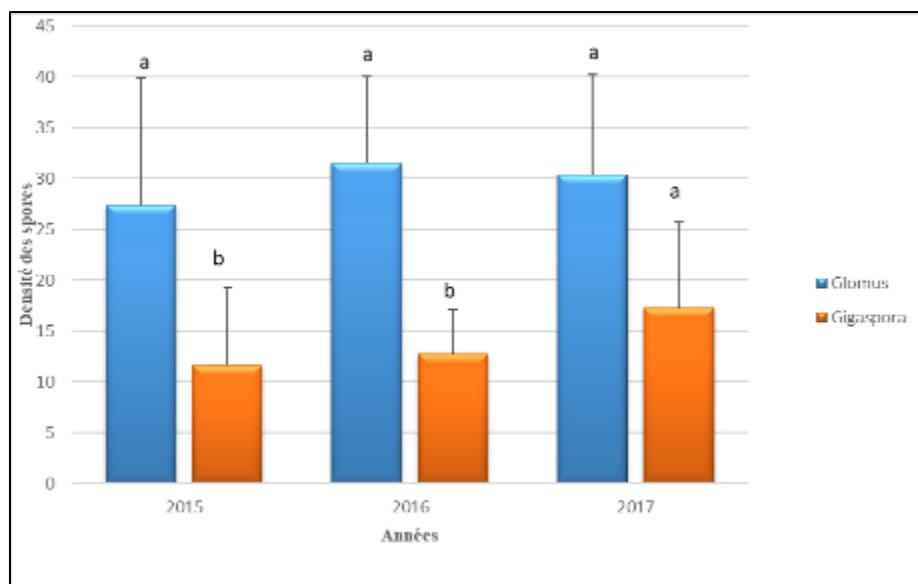


Figure 4 Variation in spore genus density over time

4.5. The averages that do not share any letters are significantly different

A l'inverse des *Glomus*, les *Gigasporas* ont été abondants en 2017 avec (17,25) par rapport aux années 2015 et 2016 qui sont respectivement de (11,61) et (12,25). Ces résultats sont en accord avec ceux obtenus par Sangaré et al. (2016) qui ont trouvé des densités faibles de *Gigasporas* par rapport à *Glomus* dans les sols de Sadoré en 3 ans (de 2013 à 2015) de monoculture de mil. La prédominance du genre *Glomus* sur *Gigaspora* peut être due à sa capacité de germination rapide et à faire face aux contraintes liées à son milieu. Ce qui rejoint la suggestion faite par Stutz et al. (2000) que *glomus* contient des isolats adaptés aux fluctuations des conditions environnementales. D'autres facteurs pourraient aussi être à l'origine de l'abondance des *Glomus* tels que la nature du couvert végétal ou encore du cycle de vie des *Glomales*. Les systèmes agricoles conventionnels, principalement basés sur la monoculture répétant année après année la culture d'une même plante, pourraient conduire à la sélection d'un nombre réduit d'espèces de CMA (Burrows and Pfleger, 2002). Johson (1992) avance que la monoculture ne sélectionne que des espèces peu ou pas avantageuses pour la culture. Selon lui, cela expliquerait en partie la chute des niveaux de production agricole dans ce type de culture. Il est aussi reconnu que les rotations de culture permettent de maintenir une certaine biodiversité dans la communauté de CMA. Ces résultats statiques montrent une faible performance du genre *Gigaspora* par rapport à *Glomus* en ce qui concerne la variation de la biodiversité des genres de CMA en fonction du temps

4.6. Changes in mycorrhization parameters over time

Figure 5 shows the changes in mycorrhization parameters over the three years of the study. This figure shows that there is no significant difference between the years of the study in terms of mycorrhization frequency.

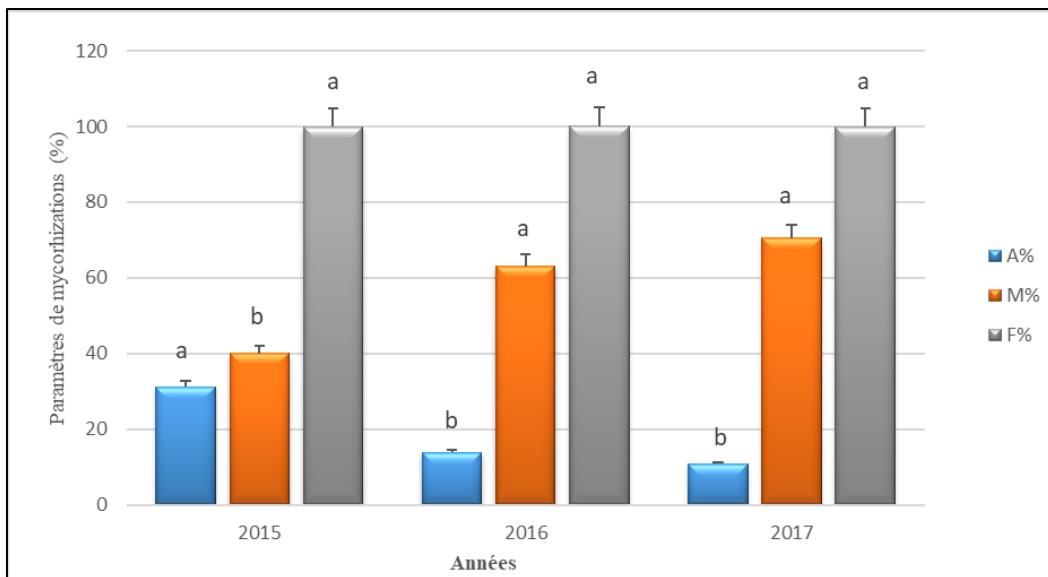


Figure 5 Mycorrhization parameters over time

4.7. Averages that do not share any letters are statistically different

However, analysis of variance showed significant differences between years for intensity and arbuscular rate. For mycorrhization intensity (M %), high mycorrhization rates were recorded in 2016 and 2017 with 63.01% and 70.53% respectively; the lowest mycorrhization rate was obtained in 2015 with 40.05%. The frequency of mycorrhization was close to 100% for all years of the study. Analysis of variance for the arbuscular rate also showed significant differences between the years of the study. Despite low percentages of mycorrhization intensity in 2015, the arbuscular rate (31.19%) was better than those of 2016 and 2017, which were 13.09% and 10.81% respectively. The study showed that high mycorrhization intensity and frequency do not translate into high arbuscular rates in the roots. These results are comparable to those of Knopf et al. (2016), who showed that changes in mycorrhization intensity and arbuscular rates are not related.

5. Conclusion

The results of the study of microbial biodiversity variation over time did not show a significant change in MFA density over the three years of the study (2015, 2016 and 2017). However, it was observed that, unlike Glomus, the density of the genus Gigaspora was significantly higher in 2017 compared to 2015 and 2016. With regard to mycorrhization parameters, only mycorrhization intensity (M %) and arbuscular rate (A %) showed significant differences between years. High mycorrhization rates were recorded in 2016 and 2017. Despite low percentages of mycorrhization intensity in the first year of the study, it was observed that the arbuscular rate was better than in the last two years. The results of this study showed that the Gigasporas genus performed poorly compared to Glomus in terms of variation in CMA genus biodiversity over time, and that high mycorrhization intensity and frequency did not translate into a high arbuscular rate. The study also showed that mulching has a positive effect on spore rates and abundance and that, like grazing and manure, mulching also had a positive impact on grain and straw yields.

Compliance with ethical standards

Disclosure of conflict of interest

No conflict of interest to be disclosed.

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