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Importance and diversity of Mycorrhizae under plantain cultivation in the slash-and-burn and non-burn cropping systems in the forest region of Kisangani, Tshopo Province, D.R. Congo

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Mycorrhization is known to have beneficial effects on growth vigour and protection against certain pathogens in several plant species including plantains, which has so far been little studied in the Kisangani forest region. This study aimed to determine the importance and biodiversity of mycorrhizae under plantains in the slash-and-burn and non-burn cropping systems. This research was carried out in two experimental sites located at Kisangani region in DR Congo, namely: Simi-Simi and Masako villages. Thus, 96 samples each consisting of the plantain roots and the rhizospheric soil, as well as 4 soil samples were analysed. The mycorrhization rate of the roots was calculated and spore description to determine the mycorrhizal taxa was carried out. The overall mycorrhization rate was 40.75%. The mycorrhizal spore number was higher in non-burned fields than burned fields. There was no significant difference between the 2 practices at Masako, but the difference was significant at Simi-Simi. Moreover, vigorous plants revealed higher number of spores than non-vigorous plants. There was a significant difference between the two types of plantains. Genera of mycorrhizal spores identified were *Glomus* (54.96%), *Gigaspora* (27.84%), *Acaulospora* (10.50%) and *Scutellospora* (6.71%). The mycorrhizal spore numbers significantly varied among plantain cultivars. The presence of tree species in the plantain plantations at high density positively influenced mycorrhization to a small extent. The above spore genera showed a positive correlation with pH, nitrogen, clay, sand and phosphorus and negatively with organic carbon. These results showed the possibility of isolating high-performance biofertilizing mycorrhizal strains from vigorous plantain crops.

Key words: Mycorrhizae, plantain, Kisangani, slash field, burned field, non-burned field.

INTRODUCTION

Plantain is among the most used crops in fallow fields, forests and agroforestry in Tshopo Province with several cultivation practices (Yenga, 2014). In the Kisangani region, plantains are often monocropped in hut gardens. However, in other cropping systems (fallow, secondary forest and agroforestry), they are always cultivated in association with other food crops and sometimes a few trees (CHN, 2008). Plantains are the real pillars of agricultural production and food security in Kisangani Region. In addition, plantain is a cash crop which, in most cases is the 3rd source of income for households after cassava, rice or palm oil (Dhed'a et al., 2011). However, like all major crops in the Kisangani region, plantain yields are continuously decreasing due to losses of pest and disease attacks, loss of soil fertility, non-use of agroforestry practices and slash-and-burn agriculture (Mate, 2001). The hope of increasing food production based on plantains in Kisangani Region is related to the expansion of the agricultural area, often at the expense of the forest, where soils are more fertile and crops are less prone to pest attacks (Marien, 2013). Sustainable management of biological resources by fallowing for 5 to 8 years, implementing agroforestry and using mycorrhizal inoculum as biofertilizers could improve soil fertility and, ensure adequate food production. Mycorrhizae are biotrophic organisms which live in symbiosis with the roots of host plants such as plantain banana. They colonize roots and produce mycelium in rhizospheric soil that allow them to take up water and mineral elements from the soil (L'huillier et al., 2010; Egli and Brunner, 2002). This symbiosis facilitates the uptake of phosphorus and other less available nutrients (e.g: iron, zinc) for host plants (Elsen et al., 2003; Kugler, 1986). Mycorrhizae can reduce the use of chemical fertilizers by 15 to 25%. Moreover, mycorrhization protects the crop roots against pathogenic microorganisms resulting in a reduction of pesticide use (Dechamplain and Gosselin, 2002). Also, for banana plants the mycorrhizal symbiosis has big advances by increasing the nutrient and water absorption surface so that nutrients that are not accessible by the roots become available. Fungal species require 100 times less biological material than a plant to cover the same absorption space (Dechamplain and Gosselin, 2002). Many studies show the beneficial effects of mycorrhizal fungi on crops (Jaizme-Vega and Azcón, 1995; Gagné, 2010; D'haene, 2015) and on weaning banana *in vitro* plants (Jefwa et al., 2010; Thienpondt, 2016). There is need to consider the vulnerability of forest region to soil depletion and degradation of sustainable agricultural practices based on e.g: bio-fertilizers. The

general objective of this study is to determine the importance and diversity of mycorrhizae under plantain cultivation in the Kisangani forest region in order to contribute to the search for efficient biofertilizers. More specifically, spore numbers and mycorrhizal colonisation and diversity are used to compare non-burn plantain cropping system to the slash-and-burn system and to study the effect of mycorrhizal colonization on the vigor of plantains. In addition, this study aimed to assess the diversity of mycorrhizal spores in relation to plantain genotypes, plantains are (Musa AAB) the presence and density of tree species in the plantation Edaphic parameters of the test sites are determined and their relation to mycorrhizal colonisation was studied.

MATERIALS AND METHODS

Study environment

This research was carried out in 2 experimental sites, namely: Simi-Simi and Masako. The experimental site of Simi-Simi is located 15 km, from the Western part of the Kisangani City, in the locality of Linoko (00°33'04.6"N, 25°05'15.6"E, and 397 m). The site of Masako is administratively located in the sector of Lubuya-Bera, northeast of the Kisangani town, 14 km on the old Buta road, in the surroundings of the Masako forest reserve (00°36'08.4"N, 25°15'59.9"E, and 429 m). Figure 1 shows the study sites of this research around Kisangani Region.

Plantain roots of plants growing under different management systems were sampled to assess root colonization rates by Arbuscular Mycorrhiza Fungi (AMF), as well as rhizospheric soil to test mycorrhizal spores. Soil samples for determination the physico-chemical parameters of both sites tested are sampled and trees in the fields were also counted and measured to test their influence on mycorrhization.

Experimental set up

In each site, an experimental platform was installed 0.5 ha containing 6 plots of 15 m × 20 m. Four samples of plantain roots and rhizospheric soil per plot were collected; one composite soil sample was also collected in this 0.5ha with diagonal method for physico-chemical analyses. For both sites, an area of 2 hectares, including 2 non-burn and 2 slash -and -burn fields, 14 cultivars of vigorous and non-vigorous plantains were identified and harvested. In this research, both biological and edaphic parameters were assessed. The biological parameters include 96 samples of plantain roots for measuring colonization rate (AMF) and 96 rhizospheric soils for counting mycorrhizal spores. In addition, a quantitative method was adopted to select trees with a diameter at breast height (DBH) ≥ 10 cm (Lejoly et al., 2010; Ndamiyehe et al., 2020) in the experimental fields to assess the influence on mycorrhization. The DBH of the trees measured allowed to calculate the density as well as the basal area. The density was calculated in terms of number of stems per hectare using the formula below:

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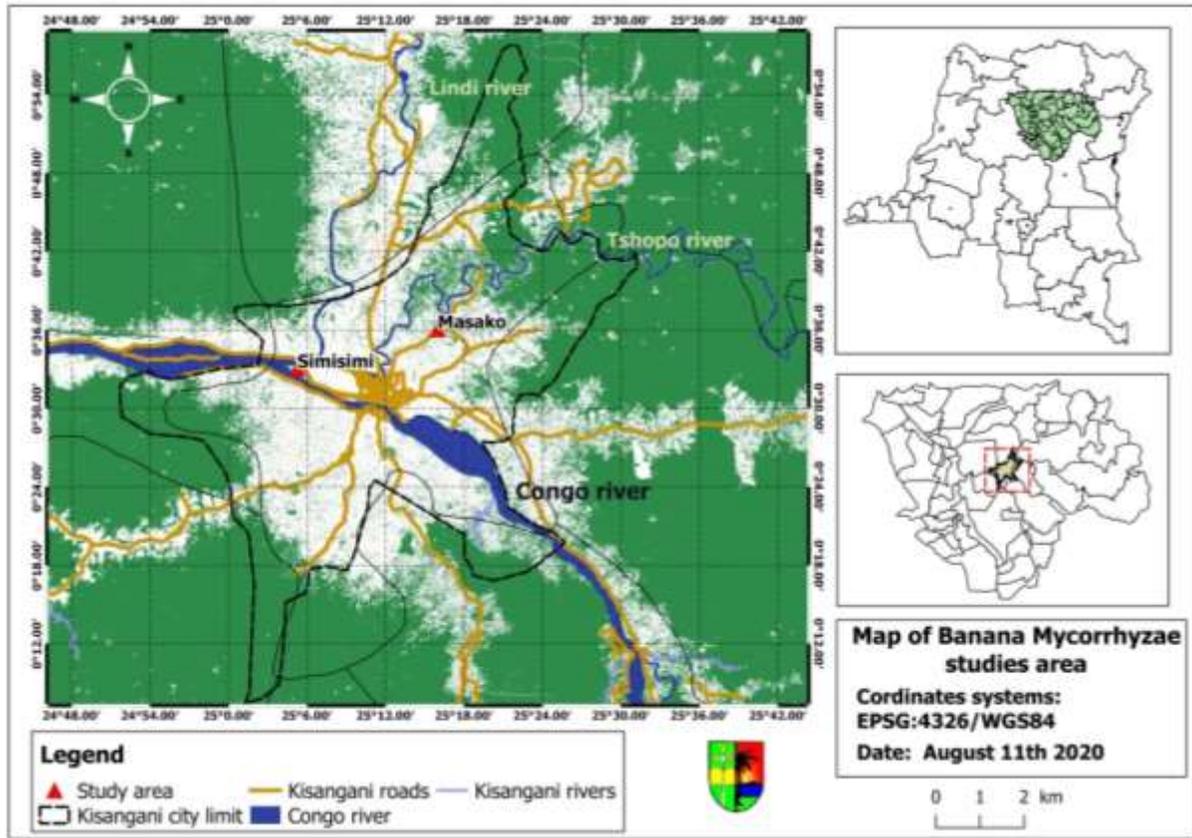


Figure 1. Location of the study sites.

Density (stems/ha) = (number of stems of a given species)/(considered area (ha)), whereas the basal area was calculated from the diameter at breast height : Basal area = $[\pi \cdot (\text{DBH})^2] / 4$.

Soil analyses

The soil parameters studied from four soil samples collected in each experimental field were used to analyze organic carbon (OC), with the Walkley and Black method (Van Ranst et al., 1988; Pauwels et al., 1992), available phosphorus with the Bray 2 Methods (Bray and Kurtz, 1945; Van Ranst et al., 1988), total nitrogen with the Kjeldahl method (Van Ranst et al., 1988; Pauwels et al., 1992), soil pH potentiometry method (Van Ranst et al., 1988) and soil texture with the successive sedimentation method (Van Ranst et al., 1988).

Mycorrhizae scoring systems

Mycorrhiza colonisation was assessed by each level represented as class of mycorrhization. The mycorrhization index was calculated using the formula: $M(\%) = (95n_5 + 70n_4 + 30n_3 + 5n_2 + n_1) / \text{total number of fragments}$. In this formula, n_5 represents the number of mycorrhized fragments scored 5 and n_4 represents the number of fragments scored 4, etc. (Trouvelot et al., 1986; Hamid and Collion, 2004).

Observation of mycorrhizal spores

The spores and hyphae of AMF are structures that allow the infection of new plant roots. These spores are large and can therefore be recovered by sieving and observed with a binocular magnifying glass (Guizon and Selosse, 2010). AMF spores were extracted according to the methods of Walker (2013) and Zézé et al. (2007). Spore families were determined using an identification key according to their shape and colour (Gerdemann and Trappe, 1974). To describe them, the methods of Omar et al. (1979), Schenck and Pérez (1987) and Dodd (2000) were used (Figure 2). Identification of mycorrhizal spores was limited to the genus level.

Data analysis and processing

The data collected were processed in Microsoft Excel 2016; R (v.3.1.3) and Past software helped for the statistical tests used. The environmental and specific parameters were used to perform the Principal Component Analysis (PCA), allowing to see the correlation between the variables studied. The other analyses performed under the R software are Chi-squared to see the dependency between two individuals. Student *t* test was used to compare the means between two variables, ANOVA test after verification after testing normality (Shapiro), to compare the means more than two. The correlation test allows to see the interaction between the variables. The diversity indices (Simpson, Equitability, Shannon and Fisher alpha) were used to have the diversity of genera AMF sporal in two systems of culture (Burn and non-burn). The similarity index was calculated for having the system than was the taxonomic

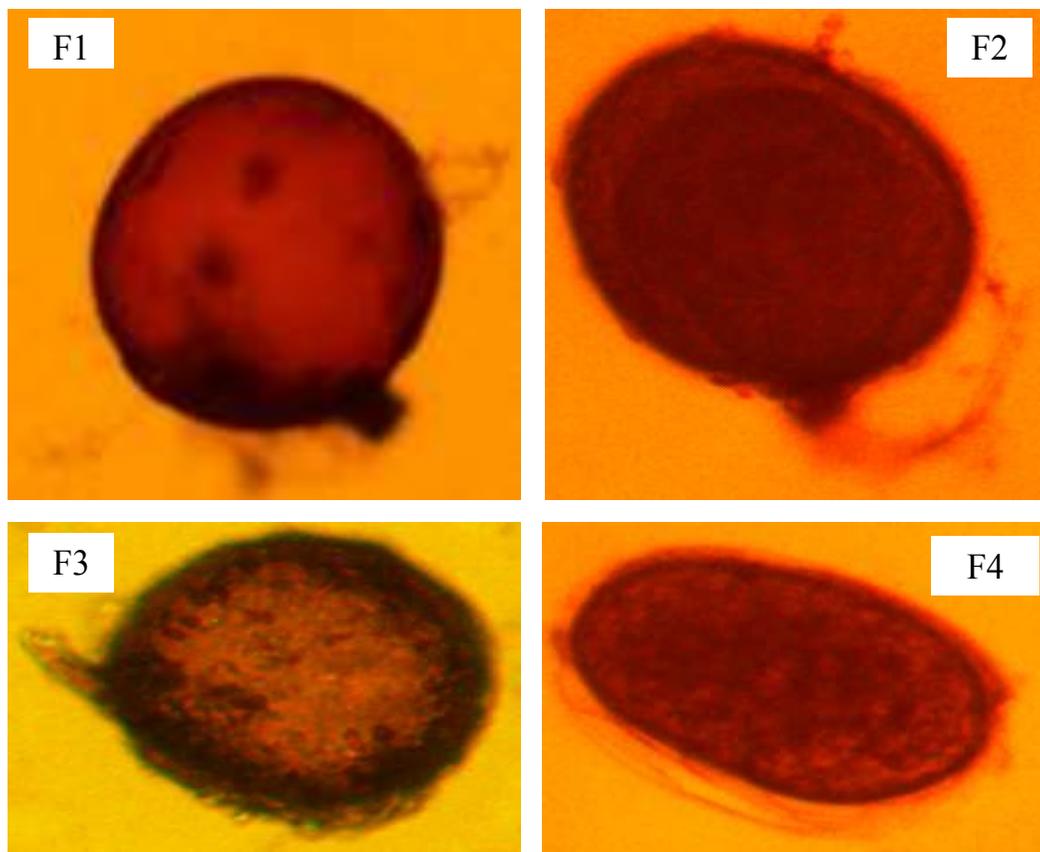


Figure 2. Mycorrhizal spores of the different mycorrhizal taxa observed (F1: *Gigaspora*: size from 200 to 360 μm , shape spherical with hyphae, reddish color); (F2: *Glomus*: size 60 to 180 μm , shape spherical ellipsoidal, yellowish flow); (F3: *Acaulospora*: 100 to 250 μm , shape: subglobular, Yellow-bright color) and (F4: *Scutellospora*: 20 to 100 μm amorphous, reddish form).

abundance of AMF sporal.

RESULTS

Biological parameters on mycorrhizae

Results showed a mean mycorrhization rate of 40.75%. The mycorrhizal spores found in rhizospheric soil of plantain roots belong to different genera (Figure 2). The distribution of *Glomus* (54.96%) was most prevalent followed by *Gigaspora* (27.84%), *Acaulospora* (10.50%) and *Scutellospora* (6.71%).

Abundance and diversity of mycorrhizal spores

Abundance of mycorrhizal spores

Figure 3 illustrates abundance of mycorrhizal spores classified by scale, cultural practice, age and vigour. The number of mycorrhizal spores was higher in non-burned field compared to the burned field on both sites; there

were 944 spores in non-burned field versus 522 spores in the burned fields (Figure 3b). Comparison of the means using a student *t*-test revealed a non-significant difference between the two practices at Masako ($t = -1.4001$, $p\text{-value} = 0.1685 > 0.05$) in contrast to Simi-Simi where there was a significant difference between the two practices ($t = -2.486$, $p\text{-value} = 0.0169 < 0.05$). Vigorous plants (2311 spores) and non-vigorous plants (1002 spores) showed the highest number of spores at both sites which was statistically underpinned by the *t*-test. In relation to the age of the plantains, Figure 3d revealed more spores in the 0.5 year-old fields (1847 spores) than the 2 years old fields (1466 spores). The student *t*-test showed a significant difference ($t = 270.11$, $df = 2$, $p\text{-value} = 1.371e-05 < 0.01$).

Number of mycorrhizal spores according to cultivars

The mycorrhizal spores in relation to plantain cultivars are shown in Table 1. The plantain cultivars mostly represented in the experimental fields were Libanga

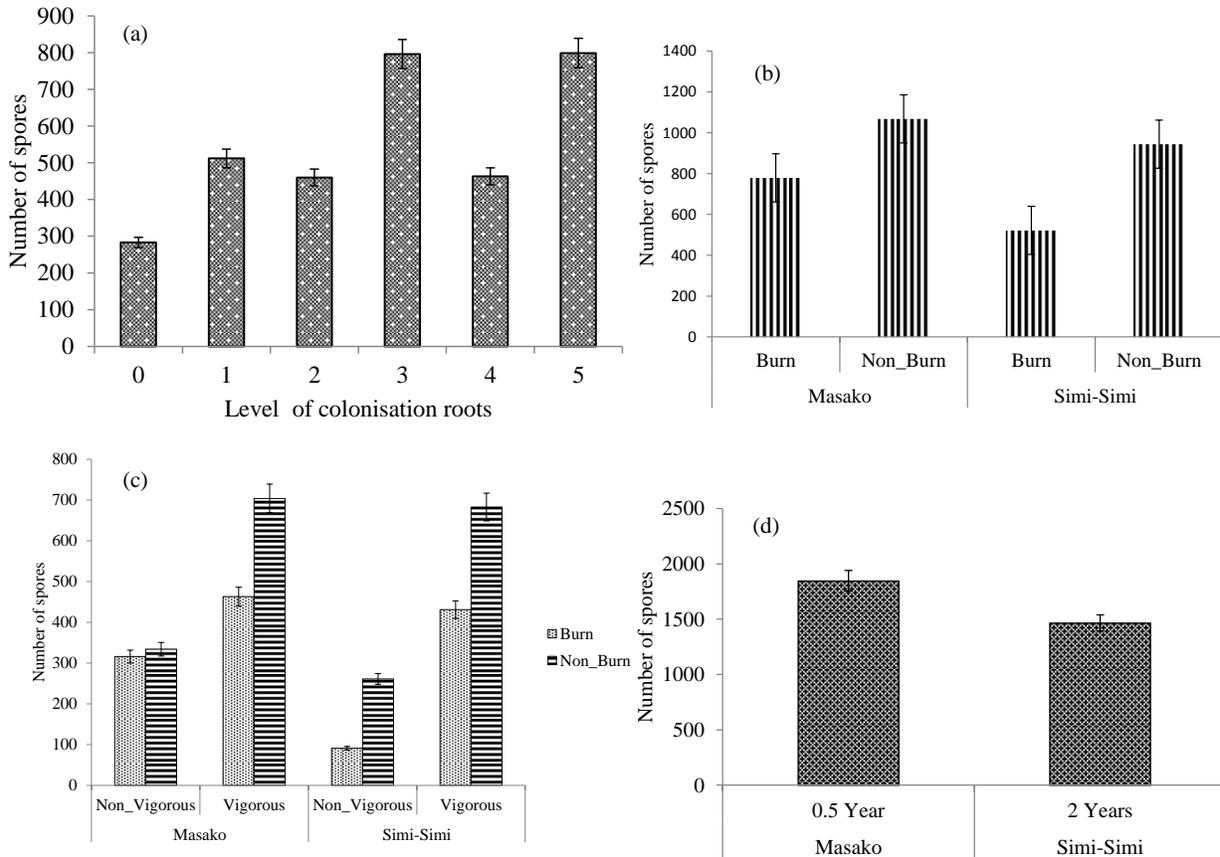


Figure 3. (a) Level (0-5) of plantain root colonization with AMF, (b) number of mycorrhizal spores under burned and unburned fields in Masako and Simi-Simi sites, (c) by vigorous and non-vigorous plants in Masako and Simi-Simi sites. (d) by age of plantain plantation on burned and unburned in Masako and Simi-Simi sites.

Table 1. Mycorrhizal spores of different cultivars of plantains.

Cultivars	Frequency	Mean of sporal mycorrhizae	Range	χ^2	P-value
Libanga lifombo	19	32.05 ± 23.39	7-86	307.21	<0.001
Libanga likale	19	36.79 ± 32.99	1-96	532.68	<0.001
Akoto	16	22.56 ± 21.95	6-84	320.17	<0.001
Tala lola	14	33.07 ± 31.67	2-96	394.39	<0.001
Libanga dark green	6	22.83 ± 16.62	6-47	60.474	<0.001
Litete	6	42.67 ± 11.17	24-53	14.609	<0.05
Lingu	4	33.75 ± 18.15	11-52	29.296	<0.001
Akpasi	3	6.33 ± 3.21	04-10	4.3043	ns
Bosakalaka	3	67.33 ± 28.87	34-84	24.752	<0.001
CRBP	2	82.00 ± 11.31	74-90	1.561	ns
Autres cultivars	4	67.00 ± 30.54	34-99	-	-

lifombo (32.05 ± 23.39 spores), Libanga likale (36.79 ± 32.99 spores), Akoto (22.56 ± 21.95) and Tala lola (33.07 ± 31.56 spores); those less represented were Egbe-omabese, Libanga liabokaykay, Libanga noir and Pita 21. Statistical analyses of Chi-square revealed a significant

difference for spore density in relation to the cultivars (Akoto, Libanga lifombo, Libanga likale and Tala lola (p-value <0.001). It was noted that all cultivars represented by more than 2 plants showed significant differences for AMF density.

Table 2. The test of ANOVA for spore diversity by cultivar.

Cultivars	Frequency	Glomus	Gigaspora	Acaulospora	Scutellospora	F	P-value
Libanga lifombo	19	291	140	33	43	8.928	<0.001
Libanga likale	19	351	182	64	46	12.11	<0.001
Akoto	16	264	106	39	26	9.602	<0.001
Tala lola	14	177	105	38	26	10.17	<0.001
Libanga dark green	6	155	96	35	11	4.105	<0.005
Litete	6	99	41	21	13	1.978	ns
Lingu	4	59	40	29	7	4.11	<0.005
Autres	4	61	32	16	5	-	-
Akpasi	3	11	6	8	3	1.744	ns
Bosakalaka	3	75	32	4	8	1.583	ns
CRBP	2	64	34	20	8	1.659	ns

Table 3. Diversity indices.

Diversity indices	Slash-and-burn	Non-Burn
Taxa_S	4	4
Individuals	1280	1644
Simpson_1-D	0.5711	0.6271
Shannon_H	1.049	1.138
Equitability_J	0.7568	0.8212
Fisher_alpha	0.5111	0.4931

Diversity of mycorrhizal spores according to cultivars

The distribution of mycorrhizal spores in the different genera is shown in Table 2. The mycorrhizal genus *Glomus* was found in the root system of on all cultivars, followed by *Gigaspora*, *Acaulospora* and *Scutellospora*. Statistical analyses revealed a significant difference among the cultivars, Akoto ($p < 0.001$), Libanga dark green ($p < 0.005$), Libanga lifombo ($p < 0.001$), Libanga likale ($p < 0.001$), Lingu ($p < 0.005$) and Tala lola ($p < 0.001$); the cultivars which showed non-significant differences were Akpasi, Bosakalaka, CRBP and Litete.

Diversity indices of AMF in slash -and-burn field and non-burn field

With regard to species richness, Table 3 shows that both cropping systems (slash-and-burn and non-burn) contain the same taxonomic groups (4 genera each). Non-burned fields had more mycorrhizal spores than slash-and-burned fields. Furthermore, Simpson's index revealed that the probability of finding a taxonomic group in both cropping systems is moderately high (tending towards 1). The equitability and Shannon indices revealed a strong structuring of mycorrhizal spore populations ($E = 0.7568 >$

0.5 ; $H = 1.049$ for slash-and-burned fields and $E = 0.8212 > 0.5$; $H = 1.138$ for non-burned fields). Finally, the Fisher-alpha index showed that the slash-and-burned fields (0.5111) are moderately diversified than the non-burned fields (0.4931).

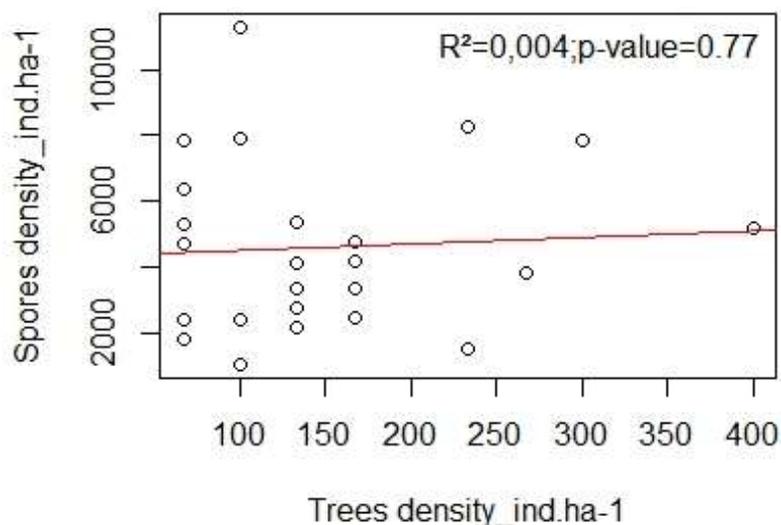
Density of trees and mycorrhizal spores in experiments fields

Table 4 illustrate that tree density was higher in the burned field than in the non-burned field in Masako. In Simi-Simi, the non-burned field showed a higher tree density than the burned field. Furthermore, the basal area was $1.62 \text{ m}^2. \text{ ha}^{-1}$ for the burned field and $3.95 \text{ m}^2. \text{ ha}^{-1}$ for the non-burned field in Masako; while the burned area in Simi-Simi was $1.44 \text{ m}^2. \text{ ha}^{-1}$ and the non-burned area was $2.35 \text{ m}^2. \text{ ha}^{-1}$. Concerning mycorrhizal spores, it was observed that on the Masako site in general 779 spores were presented in the rhizospheric soil in the burned plot versus 1038 spores in for the unburned field; while for the Simi-Simi site 522 and 944 spores were presented for the burned and non-burned plot respectively. The correlation between tree species densities and presence of mycorrhizal spores is shown in Figure 4.

Figure 4 shows there is no correlation between tree and mycorrhizal spore density. The presence of trees in

Table 4. Density of trees, basal area and density of mycorrhizal spores.

Variable	Cultivated on burned field		Cultivated on non-burned Field	
	Masako	Simi-Simi	Masako	Simi-Simi
Density of trees (ind. ha ⁻¹)	42	38	36	90
Basal area (m ² . ha ⁻¹)	1.62	1.44	3.95	2.35
Spores Density (ind. ha ⁻¹)	779	522	1038	944

**Figure 4.** Correlation between tree species density and mycorrhizal spores.

the experimental plots has a positive, but non-statistical significant ($P\text{-value}=0.77 > 0.05$) influence on AMF spores presence.

Physico-chemical soil parameters

The physico-chemical analyses of the soils are presented in Table 5. The table showed that for pH-H₂O and pH-KCl the soils in these 4 fields were less acidic between fields of this research. On the other hand, organic carbon was high in the burned Simi-Simi plot (1.8%) and the lowest in the unburned plots of Masako (1.3%) and Simi-Simi (1.3%). However, available phosphorus was high in the unburned Simi-Simi plot (19.33 ppm), while the lowest value was observed in the unburned Masako plot (13.41ppm). Regarding soil texture, sand presented a higher percentage (64%), in the unburned Simi-Simi plot, while the lowest was observed in the unburned Masako plot (53%) and burned Simi-Simi plot (53%). For silt, 34% was found in burned Masako plot while unburned Simi-Simi plot showed a low percentage (24%), clay showed a high percentage (18%) in the unburned Simi-Simi field, while low percentage (14%) was observed in the 2 Masako plot. Finally, total nitrogen content was high

(0.6%) in the non-burned plot of Simi-Simi, while the lowest nitrogen content (0.4%) was observed in the non-burned Masako plot. Figure 5 presented the correlation between every number representing a plant of banana tree plantain and the physico-chemical elements of soils. This correlation matrix (Figure 5 (a) and (b), with the foot numbers from 1 to 96 that represented the cultivars of the identified plantains) expressed that the first quadrant with the plantain feet (22, 28, 35, 37, 47, 59, 63, 72, 76,77 and 84) correlated positively with organic carbon, while the 2nd quadrant feet (8, 51, 56, 57, 60, 61, 64, 69, 85,86, 87, 89, 90, 94, 95) showed a positive correlation with silt, but were negatively correlated with organic carbon. On the other hand, the 3rd quadrant containing the foot numbers (1, 9, 10, 18, 19, 21, 22, 24, 27, 31, 32, 48, 71, 82) showed a positive correlation with pH_H₂O, pH_KCl, nitrogen (N), clay, sand, phosphorus. On the other hand, the 4th carrying feet (3, 5, 6, 7, 13, 29, 45, 49, 50, 55, 58, 65, 88, 91, 92 and 96) correlated positively with the genera of mycorrhizal spores, namely: *Scutellospora*, *Glomus*, *Acaulospora* and *Gigaspora*, whereas they showed a negative correlation with the 2nd quadrant variables. However, the 2 axes PCA 1 (40.05%) and PCA 2 (20.8%) contributed 60% to AMF colonization. In addition, figures(c) and (d) also expressed the same

Table 5. Physical and chemical analyses of soils.

Analyses	Masako NB	SimNB	Masako B	SimB
pH_H ₂ O	5.52	5.9	6.14	6.35
pH_KCl	4.35	4.76	4.59	5.46
OC (%)	1.3	1.3	1.5	1.8
Available phosphorus (ppm)	13.41	19.33	14.29	13.82
Sand (%)	53	64	54	53
Silt (%)	33	24	34	28
Clay (%)	14	18	14	17
Nitrogen (mg/g)	0.4	0.6	0.5	0.5

SimB and SimNB (Simi-Simi burn and non-burn); Masako B and Masako NB (Masako burn and non-burn).

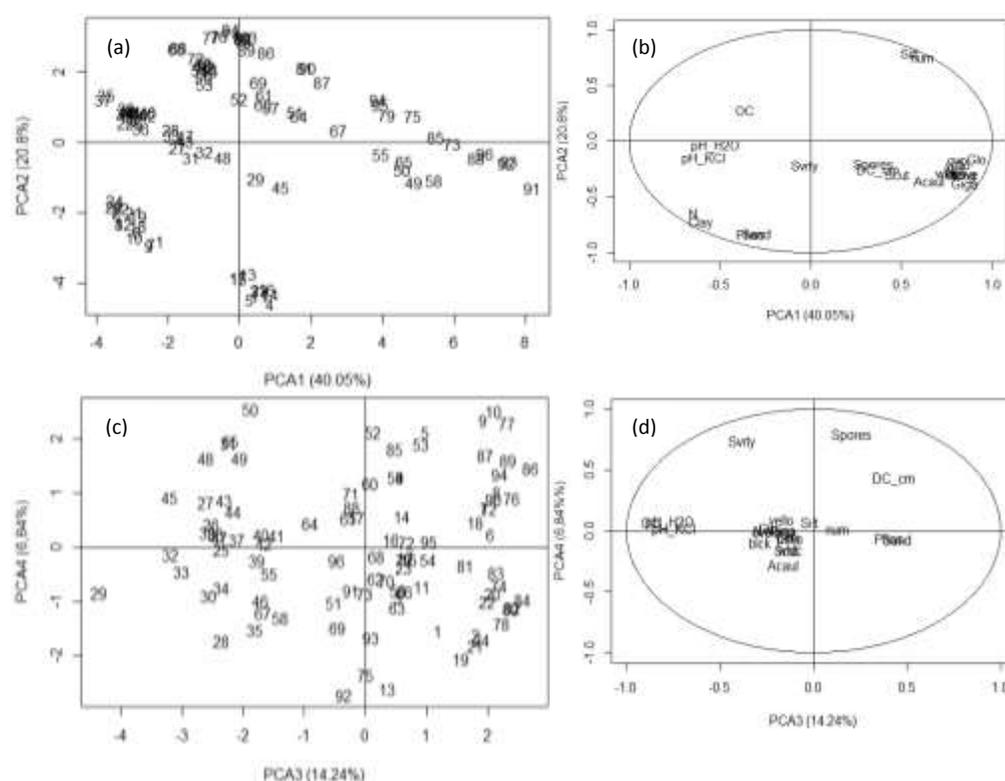


Figure 5. The correlation between every number representing a plant of banana plantain tree and the physico-chemical elements of soils.

effect as above; the feet (26, 27, 37, 38, 40, 41, 42, 43, 44, 45, 48, 49, 50, 64, 65, 71, 77, 88) in the 1st quadrant correlated positively with OC, pH_H₂O and pH_KCl, silt and the yellow coloured of mycorrhizal spores and showed a negative correlation with the 3rd quadrant variables. For the 2nd quadrant, the feet (14, 16, 52 and 53) correlated positively with the spores and the neck diameter and correlated negatively with the 3rd quadrant variables. The 3rd quadrant (25, 30, 33, 34, 35, 39, 46, and 67) correlated positively with passive shape, black colour of mycorrhizal spores and the genera,

Scutellospora, *Acaulospora*, *Glomus* and *Gigaspora* as well as the soil parameters (Silt and Clay) and correlated negatively with 3rd quadrant variables. On the other hand, in the 4th quadrant, the numbered feet (11, 20, 54, 62, 66, 68, 70, 74, 78, 81 and 83) also correlated positively with sand and phosphorus content, whereas these variables correlated negatively with the variables in the 1st quadrant. In addition, the 2 axes PCA 3 (14.24%) and PCA 4 (6.84%) contributed 21.08% to AMF colonization. Overall, the above variables contributed 81.93% representing the 4 components out of the 20 components

contributing to AMF colonization.

DISCUSSION

Biological parameters

Abundance and diversity of mycorrhizal spores

Over the two sites, different management systems and plantain cultivars, the overall mycorrhization rate were 40.75%. Elsen et al. (2008) found on the tissue-cultured plantlets of the banana cultivar Grand Naine (*Musa* sp. AAA, ITC 1256) a frequency of colonization of 100% after artificial inoculation. However, the intensity, which was an indication of the colonization quality, ranged from 13 to 24%. Lebisabo et al. (2019) worked on strains of symbiotic endomycorrhizae of banana and plantain trees (*Musa* sp.) from Kisangani Region (DRC); they found an overall intensity of mycorrhization ranging from 20 to 80%. Sunisa et al. (2020) worked on coffee robusta in Mueang, Chumphon, in Thailand, applying inoculum from *Glomus* sp. and LU3 inoculation of *Glomus intraradices* on clay soil, with a low amount of available phosphorus (7.89 mg kg^{-1}), pH 5.01, spore count (5.24 spores/g), and root colonization (6.83%). The authors used two factors: inoculation with 300 g per plant⁻¹ of AMF on non-colonized plants, and phosphate fertilizers at 0, 100, 200 and 400 g per plant⁻¹. Differences were significant with densities of 64.7 and 64.9%, respectively. However, the 400g plant⁻¹ of rock phosphate fertilizer was revealed in the highest density of root colonization, available phosphorous in the soil and total phosphorous in leaves of 4 and 8 months after inoculation (25.82 and 27.24%). Gamalero et al. (2004) and Das et al. (2007) showed in their research that AMF has a positive influence on rhizobacteria and root colonization of plants. Venneman et al. (2017) worked roots of maize, rice, soybean and Sudangrass in rhizospheric soils at Kisangani Region; the plants host was used to test colonization rate in local endophytic AMF. The results revealed that soybean and rice showed a colonization rate of 17 and 25% respectively, while maize revealed 58% and Sudangrass 83%. Adamou et al. (2013) supported the idea that mycorrhizae are part of symbiotic associations that promote plant growth and development. The present study was based on natural inoculation under plantain field, and other studies were based on the artificial inoculation. So, AMF has the capacity to increase the production of banana tree if inoculation or sources of colonization is used.

Abundance of mycorrhizal spores under burned and unburned conditions

Our results revealed that in the two sites number of mycorrhizal spores was higher in the unburned plot compared to the burned ones. However, the difference

was not significant for the two practices in Masako, but significant in Simi-Simi. This can be explained by the difference in soil texture, the micro-climate of the site and the presence of trees in fields in Masako and Simi-Simi. The duration of burning before the establishment of plantain crops can also explain differences in AMF presence between burned and non-burned. Gnahoua (1993) mentioned that the burning system has a very large negative impact on litter inputs to the soil, as was observed in the Sangoué Forest in Côte d'Ivoire. Thus, there was incineration of slash and litter corresponding to a 60% loss of organic matter and nitrogen inputs. Mate (2001) observed in his study carried out in Masako and Simi-Simi that the reduction of leguminous in old fallow soil by incineration confirmed the harmful effects of burning on the regeneration and dynamics of the Fabaceae family as well as the destruction of soil microorganisms. Abbadie (1984) has shown that slash-and-burn agriculture is one of the main sources of deforestation and soil degradation. In general, slash-and-burn agriculture would always have a negative impact on the forest and the environment because of the successive displacement of farmers to other areas and leaving old areas to fallow. We agreed with other researchers that the practice of slash-and-burn agriculture was not appropriate, as it has much harm on ecosystems and the environment. André-Fortin (2011) supported the idea that agricultural practices have been designed and used without taking into account the existence of mycorrhizae. Yet they are available in all areas and play fundamental roles in all environment site of a plant's growth.

Mycorrhizal spores following vigorous and non-vigorous plantains

The overall calculated mycorrhization rate was 40.75%, but, vigorous plants showed higher number of AMF spores than non-vigorous ones at the 2 sites. It presents a significant difference between the vigorous and non-vigorous plants. Nwaga (2007) found in his research in Cameroon that microbial AMF led to an improvement in crop growth and yields of 50 to more than 200% compared to crops that were not inoculated. However, it was difficult to assess separately the mycorrhizal influence on plantain vigour, especially since Declerck et al. (1995) showed that the prevalent strains that colonise root systems of plantain were not necessarily those that affected yield the most. Sieverding (1990) observed that when fertilization was intense, plants became increasingly independent and that their need for mycorrhization decreased. It was mentioned by many researchers that the application of mineral fertilizers limited the extension of the plant root system. Since this application appeared to be a constraint, the hope was that the evaluation of the separate effects of mineral and organic (compost and manure) fertilization used by local

farmers would be important for the growth of banana culture.

Abundance of mycorrhizal spores in relation to the age of banana plantations

Regarding the age of plantations, Figure (3c) shows more spores at 0.5 years -old in Masako. The finding was the same and 2 years of age in Simi-Simi. Jaizme-Vega and Azcón (1995) explain that the age of banana trees has a positive impact on mycorrhization of this plant. These authors claimed that for the effects of fungi on yield, mycorrhization of banana trees showed under certain conditions positive responses to growth, and a significant nutritional improvement; especially in the early stages of plant development. A multitude of results supporting the same conclusions above have been obtained by Lin and Chang (1987).

Diversity of mycorrhizal spores identified according to cultivars

Regarding mycorrhizal genera, the genus *Glomus* was found on all cultivars, followed by *Gigaspora*, *Acaulospora* and *Scutellospora*. Statistical analyses as a whole revealed a significant difference among the cultivars, Akoto, Libanga dark green, Libanga lifombo, Libanga likale, Lingu and Tala lola; non-significant differences were found between Akpasi, Bosakalaka, CRBP and Litete. On the other hand, the genus *Glomus* from the family Glomeraceae is the most abundant and most frequent AMF known. In research carried out in India, Lakshmiathy et al. (2012) showed a large number of mycorrhizal spore species belonging to the family Glomeraceae. On the 56 and 67 species identified in the pre-monsoon season and post-monsoon season respectively, the same authors mentioned that *G. fasciculatum* was the most abundant, followed by *G. geosporum* and *G. mosseae*. In Cameroon, Mbouapouognini et al. (2007) found a meaningful difference in colonization rate between the Fombot and Bamenda cultivars of *Solanum nigrum* of 45 and 55% for Fombots Bamenda, respectively. The inoculation increased the rate of root colonization significantly in relation to the control. Biomass of Bamenda and Fombo cultivars was increased with 60 and 128% respectively after inoculation. Lebisabo et al. (2019) found the most abundant *Glomus* genus among all identified genera. Many authors on mycorrhizae in the tropic, particularly in Kisangani, have confirmed that the genus *Glomus* is more abundant in the area, corroborating the result found in this research.

Diversity indices

The calculated ecological indices assessed the spore

diversity in the two systems. Indices of ecological diversity showed that there was a low taxonomic diversity of the mycorrhizal groups studied (Table 3). This is explained by the fact that the number of taxa inventoried was low, and the taxonomic groups in the 2 cropping systems were identical. In addition, the identification of mycorrhizal species was difficult due to the lack of molecular markers. Nevertheless, the non-burned fields showed higher diversity index values than the slash-and-burned fields. This can be explained by the fact that a greater number of individuals were inventoried in the non-burned fields. Thiémélé et al. (2017) conducted a survey to collect local varieties in banana production areas in Côte d'Ivoire, and 88 varieties were selected during the agro-morphological characterization at Azaguié. The first 10 varieties revealed the highest technical indices (IT \geq 20). The difference in the results of the present study is that the research was conducted on plantain MFAs, whereas the predecessors were conducted on variety diversity. These results are similar to that of other studies, with the only difference of AMF in the present one.

Density of trees and mycorrhizal spores in experiment fields

The presence of trees in the fields has a positive but weak influence on mycorrhization. The correlation test revealed a non-significant difference ($P > 0.05$), with a very low coefficient of determination ($R^2 = 0.04$). The tree density found by Dupuy (1998) in Central African semi-deciduous moist forests was between 100 and 130 stems/ha of commercial species. Gourlet (2013) found an average of 122.5 stems ha⁻¹ in the Mbaiki Region, in Central African Republic. This difference in tree density results could be explained by the fact that each study was conducted in different sites, with different ecosystems and at different times. The study carried out by Yenga (2014) at the Masako and Simi-Simi sites, covering an area of 9 ha, inventoried 1056 trees of 10 cm DBH trees \geq all species combined. Nshimba (2008) working at Yoko Forest Reserve in Kisangani Region, identified 2534 trees of DHP \geq 10 cm in 1 ha area. The basal area and tree density at the sites corroborated those found by other authors. Moreover, the presence of trees in banana fields would have a huge advantage on growth and sustainable production, as biomass acted in the biogeochemical cycle in favour of crops. Thus, Carbaya (1988) showed that all absorbent short roots of plants were colonized by fungi, particularly mycorrhizae. Their presence, especially ectomycorrhizae on the tree species *Gilbertiodendron dewevrei*, was observed by Onguene and Kuyper (2004) in the tropical rainforest of southern Cameroon. In addition, Kasha et al. (1989) studied the root symbiosis of some important forest species in D.R. Congo and confirmed the presence of

AMF in the short roots of these species. Thoen (1974) followed the same approach as these predecessors drew the same conclusion in the Guinean forest.

Several studies have demonstrated the importance of trees on soil fertility. In fact, trees enrich the soil through the leaves that fall, decompose and are transformed into humus, which constitutes the organic matter useful to the plant. In addition, trees allow the soil to retain its moisture and prevent solar radiation from reaching the soil directly, causing water loss from the soil. On the other hand, in a treeless field, the soil is exposed to the sun's rays, which cause the evaporation of water. The low availability of nutrients forced the banana plants to develop a mycorrhizal symbiosis in order to access the elements useful for their good growth. This would explain the negative correlation between the presence of trees and AMF.

Physico-chemical parameters of soils

The results showed that the soils in these 4 fields of banana are acid. Organic carbon was high in the burned Simi-Simi field and lowest in all the unburned fields. However, available phosphorus was high in the unburned Simi-Simi field, while the lowest was observed in the unburned Masako field. Lakshmiathy et al. (2012) observed under Niligiri conditions in India that high percentages of sand, porosity, total nitrogen, organic carbon and exchangeable bases were positively correlated with the activity of arbuscular mycorrhizal fungi; whereas the percentages of clay, silt, potassium, total phosphorus and exchangeable phosphorus were negatively correlated. Moreover, Nyssens (2012) observed that soil from Martinique with a strong phosphorus fixation was suitable for the development of AMF. The physico-chemical parameters of the soils would greatly influence crop growth and production, as soils are not homogeneous throughout the world. This would allow us to agree with the ideas of previous authors. Karaarslan et al. (2015) confirmed that the presence of phosphorus in the soil affects rapidly the colonization of plant roots. The length of plant roots to bulbs benefits from the infestation of these AMF which has increased soil fertility in Turkey because, before cultivation, the phosphorus content of these soils was low. Kombele (2004) proved that the most important source of nutrients for crops in the humid tropics was organic matter, which, as it decomposed, enriched the soil with various minerals essential for plants. The faster mineralization occurred more, the soil benefited from most nutrients, and the less organic matter was accumulated on the soil surface. The same author added that the marked acidity of forest soils in the Central Congolese Basin favoured the mycological decomposition of soil organic matter and the predominance of unstable organo-mineral complexes. Landon (1991) added that at low pH values, nitrified and nitrogen-fixing bacteria were

destroyed by soil acidity, and nitrification of organic matter was significantly limited, leading to nitrogen deficiency. Segalen (1994) and White (2006) also showed that in many tropical soils with pH below 4.5 such as those in the dense forests studied in the Yangambi forest zone, decomposition of organic matter stopped when aluminium levels were high. Moreover, cropping systems showed that organic carbon contents ranged from 1.25 to 1.75%, while total nitrogen ranged from 4.2 to 5.74%. The pH ranged from 5.52 to 6.35. The relation between physico-chemical parameters and the specific diversity of the mycorrhizae is not yet well known. Otherwise, the mycorrhizal production as the spores can increase with the pH and the organic carbon, and decrease with the increase of the content in phosphorus of soil as mentioned by Menge et al. (1978). In view of the small differences observed, it is therefore important to say that from the point of view of soil chemical characteristics, the differences among cropping systems are not significant.

Conclusion

Sustainable agriculture and preservation of tropical forests are a major priority currently. This study aims to determine the importance and diversity of AMF under slash-and-burn, and non-burn plantain cultivation in order to search for local mycorrhizal strains that can be used as effective biofertilizers in the Kisangani Region. Plantain AMF colonization roots and spore counts were determined. The results showed an overall mycorrhization rate of 40.75%. Moreover, the AMF spore number was higher in non-burned than in slash-and-burned fields. Vigorous plants showed more AMF in both sites than non-vigorous plants. Four mycorrhizal genera (*Glomus*, *Gigaspora*, *Acaulospora* and *Scutellospora*) were identified. The genus *Glomus* was the most abundant. The presence of tree species in a high density under banana plantations positively influenced mycorrhization to a small extent. The soil physico-chemical parameters showed a positive and negative influence for some AMF groups. These results showed the possibility of isolating high performance biofertilizing mycorrhizal strains from vigorous plantain plants for the non-burning cropping system in the Kisangani Region of DR Congo.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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