

Field Evaluation of Growth and Yield of Two Local Rice Varieties (Tox-728-1 and Madjitolngar) in Response to Indigenous Mycorrhizal Inoculation in South-Chad

Yoradi Nadjilom¹, Steve Takoukam Toukam¹, Minista Issa², Albert Ngakou^{1*}

¹Department of Biological Sciences, Faculty of Science, University of Ngaoundere, Ngaoundere, Cameroon

²Cereal Office, Garoua, Cameroon

Email: *alngakou@yahoo.fr

How to cite this paper: Nadjilom, Y., Toukam, S.T., Issa, M. and Ngakou, A. (2020) Field Evaluation of Growth and Yield of Two Local Rice Varieties (Tox-728-1 and Madjitolngar) in Response to Indigenous Mycorrhizal Inoculation in South-Chad. *American Journal of Plant Sciences*, 11, 1175-1192.

<https://doi.org/10.4236/ajps.2020.118083>

Received: June 24, 2020

Accepted: August 3, 2020

Published: August 6, 2020

Copyright © 2020 by author(s) and Scientific Research Publishing Inc. This work is licensed under the Creative Commons Attribution International License (CC BY 4.0).

<http://creativecommons.org/licenses/by/4.0/>



Open Access

Abstract

This study was carried out to find out how dependent are two local rice varieties (Madjitolngar and Tox-728-1) to inoculation with selected endogenous arbuscular mycorrhizal fungi (AMF) in a field where they were isolated. The multi-indigenous endomycorrhiza spores previously isolated and identified were the active ingredient in the production of bioinoculants used for this purpose. Spores massively multiplied from the rhizosphere of each rice variety in each of the four locally collected soils substrates were harvested to constitute 08 AMF inoculants (Kema = T1; Lama = T2; Latox = T3; Ndjatox = T4; Koloma = T5; Kolotox = T6; Ndjama = T7; Ketox = T8). These inoculants were field tested on the two rice varieties at Kelo, under a complete randomized block design, comprising 10 treatments (8 inoculants, 01 positive control = T9, 01 negative control = T10), each of which was repeated thrice. The analysis of data indicates that AMF-inoculated plants were taller, developed more tillers/plant, and produced more rice grains/panicle than non-AMF-inoculated plants for both studied rice varieties. The rice variety Madjitolngar yielded more grains (7.5 t/ha) than the Tox-728-1 variety (5.8 t/ha). Moreover, inoculants Koloma (T1), Latox (T3) and Kolotox (T6) on the one hand, Koloma (T1) and Ketox (T8) on the other hand, were best suited for the improvement of growth and yield of the rice varieties Madjitolngar and Tox-728-1 respectively, tested under field conditions at Kelo. In this study, the two rice varieties have shown a dependency to endomycorrhizal symbiosis at Kelo, and therefore, an industrial-scale production of efficient endomycorrhizal inoculants is necessary to sustainably boost the productivity of this important crop in Chad.

Keywords

Endomycorrhiza Inoculants, Grain Growth and Yield, Maditolngar and Tox-728-1 Rice Variety, Chad

1. Introduction

Arbuscular mycorrhizal fungi (AMF) and plant roots-symbiosis can modify plant-soil interactions and improve plant growth under soil stress conditions [1]. Among the changes likely to occur, an alteration in carbon production and in the mineral composition of the host plants has been highlighted [2] [3]. Increasing the concentrations of available minerals and reducing mineral toxicity in plants under acidic environments could be achieved by the use of arbuscular mycorrhizal fungi (AMF) [4] [5] [6] [7]. Such beneficial effects of AMF have been particularly attributed to the increased surface of root absorption beyond the normal zone of root absorption [8]. According to this mechanism, plants absorb mineral elements that would not have been available to be uptaken by plant roots [6]. Karagiannidis and Hadjisavva-Zinoviadi [9] have shown that certain species of mycorrhiza can increase up to 11.6 folds the biomass of cultivated plants and 5.4 folds their yield in comparison to non-inoculated plants. In addition, they positively affect environmental contaminants through their ability to capture nutrients from the soil, causing a reduction in the use of chemical fertilizers, while ensuring good yields [10] [11]. It would therefore be very beneficial to promote this symbiosis in cereal crops, especially rice, which is one of the most important crops in the world.

Rice is a cereal belonging to the poaceae or grass family, cultivated in tropical, subtropical and warm temperate regions. Rice has been cultivated almost 10,000 years since the Neolithic Revolution [12], and is a fundamental element in the diet of many African population and livestock. It is cultivated in various cropping regimes: lowland rice, cultivated without flooding, which is distinguished from flooded rice where water level is not controlled, and irrigated rice cultivation where water is controlled. According to Agrimonde [13], food needs for population in Sub-Saharan Africa, and rice in particular, will be multiplied 4 folds by the year 2050. Ranked as second cereal after wheat based on the cultivated areas, rice is the most consumed cereal worldwide [14]. In Chad for instance, the cultivated rice is white in color, and is consumed in several forms including grains, or pasta with various soups alone and/or supplemented with tubers or banana. Unfortunately, the local production of this crop remains weak and does not cover the population demands in the country. In this context, the Chadian government has undertaken several projects to develop and improve the cultivation of this cereal in several regions, including Tandjile, Mayo Kebbi and the two Logones. Among the measures taken into consideration, the provision of chemical fertilizers to rice growers at low prices occupies a place of

choice, although they are still more expensive to the common farmer. Furthermore, the inappropriate use of chemical fertilizers is likely to cause serious food and environmental pollution [15]. Considering these observations, an intensification of agriculture based on agro-ecological principles that respect the environment is increasingly recommended [16]. In this context, a strategy based on the valorisation of arbuscular mycorrhizal fungi may be one of the appropriate solutions. Several studies have demonstrated that the use of arbuscular mycorrhizal fungi as biofertilizer or microbiological inoculants in agriculture can give spectacular results in the field, in particular the improvement of biomass production and crop yields [17] [18]. Many studies have highlighted the interest of indigenous strains in that they are adapted to the environment conditions, and therefore, is able to establish adequate and appropriate symbiosis [19] [20]. A previous research conducted in the Tandjile region-Chad has revealed the existence of more efficient indigenous AMF affiliated to local rice varieties than to Nerica rice varieties [21]. Therefore, these indigenous strains associated with rice rhizosphere in growing areas have recently been characterized [22], but their efficacy on the improvement of rice production has not yet been investigated. The aim of this study was to assess the impact of endomycorrhizal inoculants formulated from the above mentioned indigenous AMF on growth and yield attributes of two local rice varieties (Madjitolngar and Tox-728-1) grown South-Chad.

2. Materials and Methods

2.1. Description of the Study Site

The experimental field was located within a plain at Kelo plain in the south of Chad in the Tandjilé region, whose coordinate is N0 9° 18'50.7". The site is characterized by silty clay soils formed on recent alluviums. The climate is of the Sudanese type, characterized by the alternation of a wet season (May-October) and a dry season (November-April). The physico-chemical properties of the experimental soil are shown in **Table 1**.

2.2. Biological Materials

Rice seeds were the local rice of the Madjitolngar and Tox-728-1 varieties appreciated by the local population. These are known as irrigated and lowland rice varieties, with a short cycle extending at 90 days at maturity.

These seeds (**Figure 1**) were graciously provided by the Agricultural research station in the Tandjilé region of Chad.

Table 1. Physico-chemical properties of the experimental soil site (kelo).

Parameters	Carbon (%)	Organic matter (%)	pH	Total nitrogen (%)	Phosphorus (mg/l)	Potassium (mg/l)	C/N ratio
Elemental contents	0.098	0.170	5.03	1.19	0.22	2.8	0.082



Figure 1. Rice varieties used in this study: (a) Tox 728-1; (b) Madjitolngar.

AMF spores were isolated from soils in four regions of Chad by Nadjilom *et al.* [22], using the two above cited rice varieties as trapping plants. Endomycorrhizal inoculants were obtained by massive multiplication of the isolated spores per rice variety and for each of the four soil sampling sites, using the modified method as described by Brundrett [23].

Rice of the Madjitolngar and Tox-728-1 varieties were cultivated on substrates made up of sand and sterile clay sampled at 1 m depth. Substrates were sterilized at 120°C for 1 hour. After cooling, the substrates were distributed in 2 kg nursery plastic pots, in which 150 - 200 spores of AMF were inoculated. Rice seeds were coated with fungal spores of the same origin, and sown separately. For AMF spores associated with each rice variety and for each soil site, seeded pots were established in order to optimize the multiplication of the different spores types. Potted rice plants were watered with tap water up to maturity. The amount of substrate used was relatively little enough to put the plants in conditions of nutritional stress. This increases and accelerates the production of spore in large quantities by the host plants [7]. At maturity, the substrates comprising roots and mycorrhizal propagules were collected and dried in the shade at room temperature for 5 days, then AMF spores of each rice variety per sampled soil multiplied in triplicate test plants were mixed to form a composite inoculum. Inoculants obtained were endomycorrhizal complexes including soil, spores, and fragments of mycelium and roots. Spore density of these inoculants was evaluated after extraction according to the method of Gerdemann and Nicholson [24]. This density was estimated at more than 10 spores/g of soil. In order to optimize the quality of mycorrhizal complex, efficient and ecologically adapted to the sampling site conditions of the study area, 8 different inoculant receipts were formulated (4 soil \times 2 rice varieties as illustrated on **Figure 2**. These include: Kema (spores extracted from Kelo soil with the Madjitolngar variety); Ketox (spores extracted from Kelo soil with the variety Tox-728-1); Ndjatox (spores extracted from the soil of Ndjamena with the variety Tox-728-1); Ndjama (spores extracted from the soil of Ndjamena with the variety Madjitolngar); Koloma (spores extracted from Kolobo soil with the Madjitolngar variety); Kolotox (spores extracted from the soil of Kolobo with the variety Tox-728-1); Lama (spores extracted from Laï soil with the Madjitolngar variety); Latox (spores extracted from Lai soil with the variety Tox-728-1).



Figure 2. Mycorrhizal inoculum specific to rice produced for field assay. (a) For the Maditolngar variety (Ndjama = T7; Kema = T1; Lama = T2; Koloma = T5); (b) for the variety Tox-728-1 (Ndjatox = T4; Ketox = T8; Latox = T3; Kolotox = T6).

2.3. Experimental Design, Treatments and Sowing

Each field trial (Madjitolngar and TOX 728-1) was laid out in a randomised complete bloc design (Figure 3 and Figure 4), comprising 10 treatments made up of the 8 inoculants treatments, a negative control (TN) and a positive control (TP). The experimental unit was a rectangular surface area (2×3) m², separated 1m apart. The experiment was repeated in 3 experimental blocks per rice variety. Each of the experimental field was extended on (41×22) m² = 902 m², the two being separated 5 m apart. Seeds were sown on elementary unit plots on 15 columns and 8 rows with 20 cm between and 25 cm within rows, for a total of 120 plants per unit plot.

At maturity, rice grains were harvested on 20 plants randomly selected per elementary plot, *i.e.* 60 plants per treatment and per variety. On these selected plants, the number of panicles per plant and the number of grains per panicle and per treatment were determined by manual counting. The panicle length expressed in cm on the same 20 plants per plot and per treatment was measured using a double decimeter. An ADAM PGW 153i electric scale (Max 150 g, sensibility = 0.001 g) was used to weigh the mass of 1000 grains per plot and per treatment, as well as for the total mass of grains per treatment. Knowing the surface area of the experimental unit (902 m²) and the grain mass per treated plot, the yield per hectare (10,000 m²) was estimated by extrapolation for each rice variety and for each treatment [18].



Figure 3. Determination of plant size at 60 days after sowing.

2.4. Statistical Analysis

Collected data were statistically analysed using the “statgraphics” program which performs the Analysis of Variance (ANOVA) to determine if treatments have a significant influence on the evaluated parameters. Duncan’s multiple comparison test was used to judge the difference between treatment at the indicated level of significance. The Microsoft Excel program was used to plot graphs.

3. Results and Discussion

3.1. Variation in Growth Parameters between Treatments at 60 Days after Sowing

Figure 4 illustrates the on plant size responses of rice of the Madjitolngar and Tox-728-1 varieties between treatments evaluated at 60 days after sowing.

For Madjitolngar variety, it appears that plant height differed from one treatment to another, with those from treatment T4 being significantly ($p < 0.0001$) taller (0.94 cm) than all the others. In contrast, plants from treatments T9 and T10 were weaker (0.65 cm) and similar. Meanwhile, as for rice variety Tox-728-1, plant height from treatment T1 (1.73 cm) was 2.65 folds significantly ($p < 0.0001$) greater than those of treatments T2 (0.64 cm), T3 (0.63 cm), T4 (0.64 cm) and T9 (0.65), 2 folds greater than those of treatments T8 (0.86 cm), T5 (0.82 cm), T10 (0.76 cm), and 1.9 folds greater than the height of plants raised from treatments T6 (0.91 cm), T7 (0.91 cm). Concerning the tillering of the rice variety Madjitolngar (**Figure 5**), plant from treatment T2 significantly ($p < 0.0001$) developed more tillers (43), whereas the weakest number of tillers accounted for treatments T10 or negative control (9) and T9 or positive control (14). For the Tox728-1 variety, treatment T4 significantly ($p < 0.0001$) enhanced the number of tillers emerging from a single plant (40), compared to those of treatments T1 (7) and T2 (9), although tillering was surprisingly improved for negative control plants.

3.2. Influence of Treatments on Growth Parameters at 90 Days after Sowing

The variation of the growth parameters of rice plants of the Madjitolngar and Tox-728-1 was effective between the different treatments 90 days after sowing is

displayed on **Table 2**, and that reveals that these parameters vary between treatments. At 90 DAS the greatest plant height (1.27 cm) for the rice variety Madjitolngar accounted for plants inoculated with spores isolated from Lai soil under Madjitolngar rhizosphere (treatment T2), or under Tox-728-1 rhizosphere (Treatment T3) was significantly ($p < 0.0001$) more elevated than that of the negative control plants (0.86 cm), represented by treatment T10.

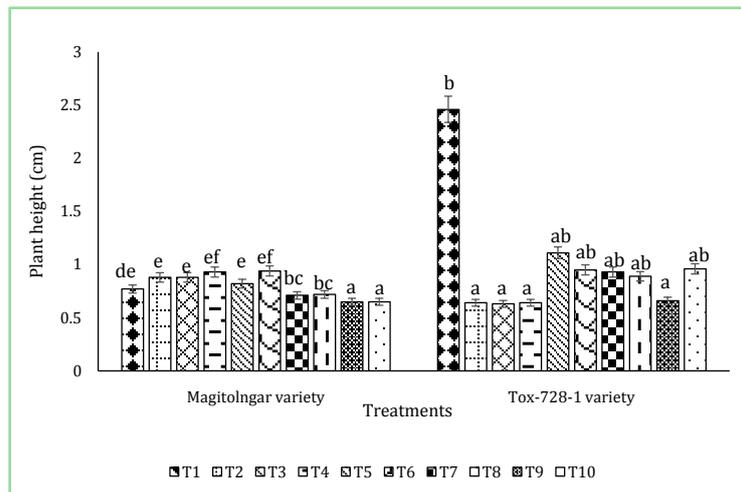


Figure 4. Effects of treatments on plant height at 60 days after sowing on rice varieties Madjitolngar and Tox-728-1.

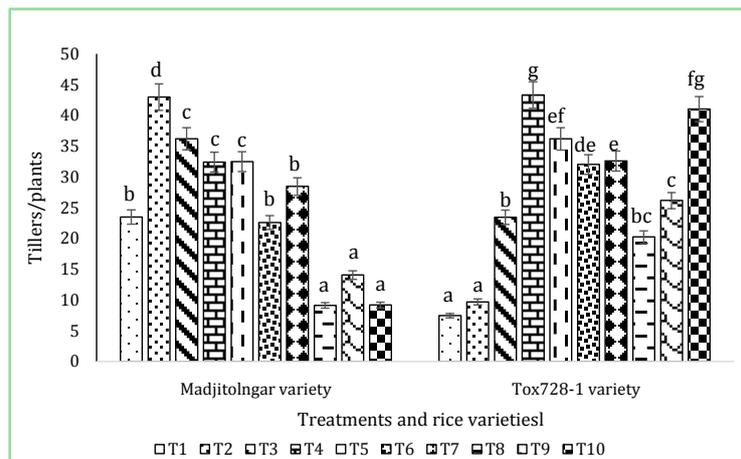


Figure 5. Effects of treatments on tillering at 60 days after sowing on rice varieties Madjitolngar and Tox728-1. T1: Koloma (spores extracted from Kolobo soil with the Madjitolngar variety); T2: Lama (spores extracted from Lai soil with the Madjitolngar variety); T3: Latox (spores extracted from Lai soil with the variety Tox728-1); T4: Ndjatox (spores extracted from the soil of Ndjamena with the variety Tox728-1); T5: Kema (spores extracted from Kelo soil with the Madjitolngar variety); T6: Kolotox (spores extracted from the soil of Kolobo with the variety Tox728-1); T7: Ndjama (spores extracted from the soil of Ndjamena with the variety Madjitolngar); T8: Ketox (spores extracted from Kelo soil with the variety Tox728-1); T9: Positive Witness; T10: Negative control; $P < 0.0001$, $F = 36.10$, $LSD: 0.95$; $ddl 9$; $n: 60$. For each rice variety, bars affected with the same letter are not significantly different between treatments at the indicated level of probability.

The number of tillers emerging from positive (T9) and negative (T10) control plants was the same, and has increased from 9 at 60 DAS to 21 at 90 DAS, but has remained significantly ($p < 0.0001$) weaker compared values from other treatment such as treatments T3 with 41 tillers. Similarly, the highest plant length and width were recorded from treatment T3 (37.66 cm and 1.16 cm) and were significantly ($p < 0.0001$) more elevated than those from treatments T9 (29.09 cm) and T10 (0.83 cm).

For rice of the Tox-738-1 variety, a significant difference ($p = 0.42$) was observed between treatments as far as the plant size is concerned (averagely 1.02 cm) at 90 days after sowing as shown on **Table 2**. In contrast, treatment T1 significantly ($p < 0.0001$) developed more tillers (41) per plant than all the other treatments such as T2 (25), T6 (27), T9 (25) and T10 (25). The lowest leaf length values were registered in plant sampled from treatments T9 (25.48 cm) and T10 (25.28 cm), whereas the highest was measured in plants from treatment T1 (31.77 cm). Moreover, the rice plants leaves width from treatment T1 (1.27 cm) was significantly ($p < 0.0001$) the highest, compared to 0.97 cm obtained from T9 plants. **Figure 6** illustrates a partial view of Madjitolngar and Tox-728-1 rice varieties fields at 90 days after sowing.



(a)



(b)

Figure 6. Rice plants of the Madjitolngar (a) and Tox728-1 (b) varieties inoculated with mycorrhizal spores at 90 days after sowing.

Table 2. Differences in growth parameters of rice varieties Madjitolngar and Tox-728-1 at 90 days after sowing.

Parameters	Madjitolngar Variety			
	Treatments	Plant height	Tillering	Leave lenght
T1	1.02 ± 0.01c	31.11 ± 0.76b	34.87 ± 1.68de	1.01 ± 0.081bc
T2	1.27 ± 0.04e	35.35 ± 0.45c	34.29 ± 1.04cd	1.16 ± 0.06d
T3	1.27 ± 0.08e	41.66 ± 1.86d	37.66 ± 2.2e	1.16 ± 0.013d
T4	1.07 ± 0.01c	32.46 ± 0.25bc	31.25 ± 0.92ab	1.07 ± 0.03cd
T5	1.14 ± 0.01d	34.2 ± 2.01bc	33.16 ± 1.65bcd	1.04 ± 0.08bc
T6	1.05 ± 0.04c	35.88 ± 0.37c	35.52 ± 1.62de	1.04 ± 0.02bc
T7	1.03 ± 0.02c	24.25 ± 1.05ab	31.19 ± 0.10ab	1 ± 0.05bc
T8	1.01 ± 0.02c	31 ± 0.85b	31.55 ± 0.54abc	1 ± 0.10bc
T9	0.94 ± 0.05b	21.05 ± 0.87a	29.09 ± 0.66a	0.97 ± 0.11b
T10	0.86 ± 0.01a	21.06 ± 0.98a	29.15 ± 0.76a	0.83 ± 0.12a
P-value	<0.0001	<0.0001	<0.0001	<0.0001
F-ratio	31.62	14.59	6.69	6.56
	Tox-728-1 Variety			
T1	1.04 ± 0.01b	41.55 ± 0.92c	31.77 ± 0.71d	1.27 ± 0.11d
T2	1.02 ± 0.007ab	24.8 ± 0.92a	29.00 ± 0.77c	1.17 ± 0.13bcd
T3	1.02 ± 0.011ab	31.9 ± 1.27b	28.1 ± 0.21bc	1.09 ± 0.12ab
T4	1.03 ± 0.01ab	40.76 ± 1.17c	27.82 ± 0.50bc	1.22 ± 0.007cd
T5	1.02 ± 0.01ab	34.38 ± 1.78b	27.31 ± 0.17abc	1.15 ± 0.10bc
T6	1.01 ± 0.008ab	27 ± 0.86a	28.18 ± 0.55bc	1.18 ± 0.13bcd
T7	1.04 ± 0.01b	35.33 ± 1.66b	26.67 ± 0.12ab	1.14 ± 0.12bc
T8	1.002 ± 0.01a	31.96 ± 1.5b	28.76 ± 0.57bc	1.13 ± 0.10bc
T9	1.006 ± 0.01ab	25.1 ± 1.07a	25.48 ± 0.14a	0.97 ± 0.07a
T10	1.03 ± 0.01ab	24.96 ± 0.96a	25.28 ± 0.12a	1.17 ± 0.01bcd
P-value	0.42	<0.0001	<0.0001	0.0001
F-ratio	1.02	14.65	5.35	3.72

T1: Koloma (spores extracted from Kolobo soil with the Madjitolngar variety); T2: Lama (spores extracted from Lai soil with the Madjitolngar variety); T3: Latox (spores extracted from Lai soil with the variety Tox728-1); T4: Ndjatox (spores extracted from the soil of Ndjamena with the variety Tox728-1); T5: Kema (spores extracted from Kelo soil with the Madjitolngar variety); T6: Kolotox (spores extracted from the soil of Kolobo with the variety Tox728-1); T7: Ndjama (spores extracted from the soil of Ndjamena with the variety Madjitolngar); T8: Ketox (spores extracted from Kelo soil with the variety Tox728-1); T9: Positive Witness; T10: Negative control. For each rice variety, and for each growth parameter values in a column affected with the same letter are not significantly different between treatments at the indicated level of probability.

In this study, the evaluation of plant size and tillering at 60 and 90 DAS has shown that mycorrhizal treatments generally stimulated better growth than the non-mycorrhized and chemical fertilizer applied treatments. The length and

width of rice leaves were also higher in the mycorrhized than in the non-mycorrhized treatments. These good growth performances of rice plants have been attributed to the beneficial effects of mycorrhiza in increasing the rhizospheric surface explored by plants roots, as well as the optimization of nutrients uptake by plants [1] [25]. According to Hamza [26], the infection of plant roots with mycorrhiza facilitates better root development, thus increasing the plants' vigor. Previous research on mycorrhization of rice has shown beneficial effects at nursery stage on growth and nutrient uptake [27]. Under field conditions, improvement in the growth parameters (height of the plants, the number of tillers per plant) of rice under mycorrhization was revealed by Jangde [28]. In Togo, the assessment of growth and production potentials of rice (*Oryza sativa* L.) variety IR841 inoculated with four AMF strains have indicated an increased AMF-plant size compared to that of control-plants [15]. Several authors have noticed the beneficial effect of the symbiosis between AMF and plant-root, notably their development, growth and production [29] [30]. Mycorrhization was shown to reduce the negative interaction between certain nutrients in the soil, by making them much more available to seedlings, thus promoting their growth [31] [32]. In the nursery experiment, Douira *et al.* [33] reported enhanced size and stem diameter of olive plants respectively 1.3 and 1.7 folds after AMF inoculation compared to the same parameters on non-inoculated plants.

3.3. Impact of Treatments on Yield Parameters at 90 Days after Sowing

There were changes in all yield parameters between rice varieties and between treatments at 90 days after sowing (Table 3).

Considering the rice Madjitolngar variety, the number of panicles was consistently ($p < 0.0001$) lower in T9 plants (24), T7 plants (23) and T10 plants (24) compared to the other treatments, particularly, treatment T3 (41) with the highest value (Table 3). Concerning the panicle length, treatments T2 or T3 (26 cm) had significant ($p < 0.0001$) higher responses, with treatment T7 having the shortest panicle length (22.58 cm). The number of grains/panicle from treatments indicated significantly ($p < 0.0001$) higher values for T3 and T6 (averagly 181), compared to values from other treatments, the lowest value (149) being recorded from treatment T7.

For rice variety of the Tox-728-1, the number of panicles/plant was significantly ($p = 0.004$) differed in treatments T3 and T10 (24), compared to the elevated value (33) evaluated in treatment T1. The length of the panicles/plant was consistently very little ($p = 0.040$) for T7 plants (5 cm) and T9 plants (4 cm), instead of 23 cm for T1 plants. The effect of the treatments on the number of grains/panicle was less pronounced for treatment T7 with 129, as compared to significant ($p < 0.0001$) greater effect observed in treatment T1 (168 grains/panicle). In this work, the weight of 1000 seeds (Table 4) varied between 20.47 - 29.08 g for the local rice variety Maditolngar, against 19.37 - 24.34 g for the local

Tox-728-1 variety, the lowest values being attributed to the control plants, while the highest accounted for AMF-inoculated plants.

Table 3. Variation of yield parameters between treatments on rice of Madjitolngar and Tox-728-1 varieties at 90 days after sowing.

Parameters	Madjitolngar Variety		
	Treatments	Number/panicles	Length/panicles
T1	30.56 ± 0.74b	24.19 ± 0.22bcd	170.11 ± 10.61cd
T2	34.45 ± 0.49c	26.08 ± 0.18e	160.36 ± 5.65abc
T3	40.78 ± 1.83d	25.77 ± 0.22e	180.78 ± 5.94d
T4	31.61 ± 2.48bc	24.68 ± 0.17cde	159.61 ± 12.24abc
T5	33.61 ± 2.03bc	23.35 ± 0.14abc	152.53 ± 20.63ab
T6	35.1 ± 0.38c	25.49 ± 0.24de	180.01 ± 4.85d
T7	23.65 ± 1.02a	22.58 ± 0.10a	148.83 ± 9.61a
T8	30.53 ± 0.84b	23.94 ± 0.17abc	156.53 ± 12.89ab
T9	24.46 ± 0.81a	22.97 ± 0.14ab	159.83 ± 14.80abc
T10	24.39 ± 0.93a	24.04 ± 0.25abcd	163.3 ± 18.95bc
P-value	<0.0001	<0.0001	<0.0001
F-ratio	14.39	4.56	5.93
	Tox-728-1 Variety		
T1	32.83 ± 1.79d	23.50 ± 1.78c	167.71 ± 10.49d
T2	29.43 ± 1.83bcd	7.90 ± 0.79abc	143.96 ± 16.22bc
T3	24.63 ± 1.65a	10.35 ± 1ab	141.6 ± 16.88abc
T4	27.81 ± 1abc	6.79 ± 0.67bc	149.58 ± 18.46bc
T5	25.96 ± 1.49ab	9.59 ± 0.95abc	137.78 ± 19.11ab
T6	31.2 ± 2.25cd	8.78 ± 0.87bc	151.31 ± 14.21bc
T7	27.31 ± 1.61abc	5.62 ± 0.56a	129.5 ± 19.06a
T8	27.25 ± 1.91abc	11.67 ± 1.16c	152.35 ± 12.68c
T9	28.85 ± 1.94abcd	3.81 ± 0.38a	147.85 ± 13.65bc
T10	24.3 ± 9a	23.91 ± 0.4bc	144.1 ± 4.98bc
P-value	0.004	0.047	<0.0001
F-ratio	2.70	1.92	4.21

T1: Koloma (spores extracted from Kolobo soil with the Madjitolngar variety); T2: Lama (spores extracted from Lai soil with the Madjitolngar variety); T3: Latox (spores extracted from Lai soil with the variety Tox-728-1); T4: Ndjatox (spores extracted from the soil of Ndjamena with the variety Tox-728-1); T5: Kema (spores extracted from Kelo soil with the Madjitolngar variety); T6: Kolotox (spores extracted from the soil of Kolobo with the variety Tox-728-1); T7: Ndjama (spores extracted from the soil of Ndjamena with the variety Madjitolngar); T8: Ketox (spores extracted from Kelo soil with the variety Tox-728-1); T9: Positive Witness; T10: Negative control. For each rice variety and for each yield parameter values in a column affected with the same letter are not significantly different between treatments at the indicated level of probability.

Table 4. Variation in yield expressed in weight of 1000 seeds (g) of the two varieties of rice between treatments.

Treatments	Weight of 1000 grains (g)	
	Madjitolngar variety	Tox-728-1 variety
T1	29.08 ± 0.10j	22.11 ± 0.07de
T2	2353 ± 0.11d	20.26 ± 0.07ab
T3	22.51 ± 0.11c	24.34 ± 0.06f
T4	25.54 ± 0.11f	22.05 ± 0.07cd
T5	24.47 ± 0.11e	21.76 ± 0.07bc
T6	27.74 ± 0.16h	23.31 ± 0.07c
T7	21.49 ± 0.11b	19.69 ± 1.21a
T8	28.95 ± 0.20i	21.26 ± 0.05b
T9	26.62 ± 0.15g	22.47 ± 0.02def
T10	20.47 ± 0.11a	19.37 ± 0.09a
P-value (LSD)	<0.0001 (0.84)	<0.0001 (1.21)

T1: Koloma (spores extracted from Kolobo soil with the Madjitolngar variety); T2: Lama (spores extracted from Lai soil with the Madjitolngar variety); T3: Latox (spores extracted from Lai soil with the variety Tox-728-1); T4: Ndjatox (spores extracted from the soil of Ndjamena with the variety Tox-728-1); T5: Kema (spores extracted from Kelo soil with the Madjitolngar variety); T6: Kolotox (spores extracted from the soil of Kolobo with the variety Tox-728-1); T7: Ndjama (spores extracted from the soil of Ndjamena with the variety Madjitolngar); T8: Ketox (spores extracted from Kelo soil with the variety Tox-728-1); T9: Positive Witness; T10: Negative witness. For each rice variety, values affected with the same letter are not significantly different between treatments at the indicated level of probability.

Figure 7 illustrates the yield expressed in t/ha of Tox-728-1 and Maditolngar rice varieties as influenced by treatments. For each of the two varieties, there was a significant difference between treatments. For the two Madjitolngar and Tox-728-1 rice varieties, all the inoculants significantly ($p < 0.0001$) improved the yield compared to the negative control. Rice plants which did not receive an inoculum (those of the negative control) significantly ($p = 0.03$) produced lower grains yield (4.4 t/ha) compared to treatments which were inoculated at sowing with a specific inoculants like T1 (7.15 t/ha), T8 (6.64 t/ha) and T6 (6.53 t/ha). Thus, inoculant Koloma (T1), Latox (T3), were suited to rice plants of the Maditolngar variety, whereas Koloma (T1) and Ketox (T8) were appropriate for cultivation of rice Tox-728-1 variety under field conditions at Kelo.

The number and length panicles in the two rice varieties were significantly higher in the mycorrhized than non-mycorrhized treatments, in agreement with previous results that have reported increased number of panicles in AMF-inoculated rice plants [15]. On the two studied rice varieties, a greater number of grains were harvested from mycorrhizal than non-mycorrhized plants, confirming similar results obtained by Jangde [28].

However, the number of grains obtained in this study was higher than 55 grains from AMF-inoculated rice revealed by Gnamkoulamba *et al.* [15]. Other studies on other crops such as palm have shown enhanced the yield in AMF-inoculated plants [34]. Concerning the 1000 grains/weight, if other

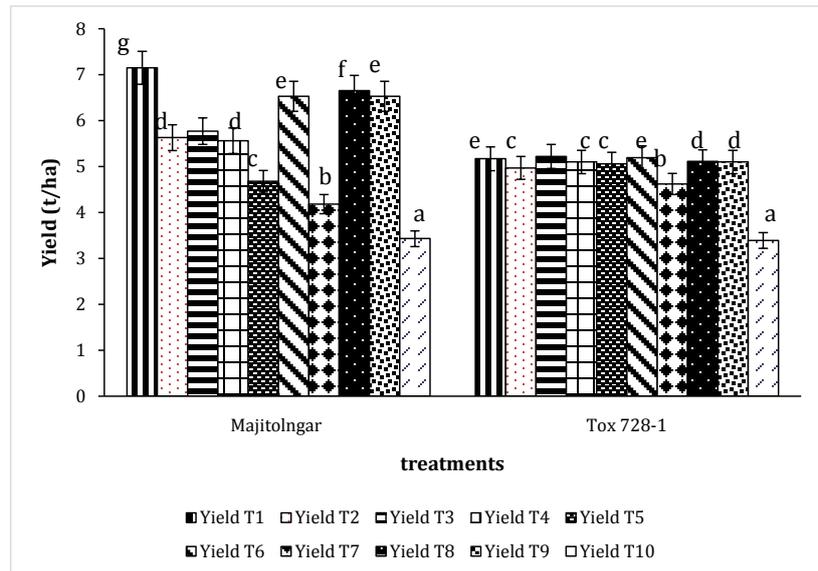


Figure 7. Variation in yield (kg/ha) of the two varieties (Madjitolngar and Tox-728-1) of rice between treatments complétez les lettres qui manquent. T1: Koloma (spores extracted from Kolobo soil with the Madjitolngar variety); T2: Lama (spores extracted from Lai soil with the Madjitolngar variety); T3: Latox (spores extracted from Lai soil with the variety Tox-728-1); T4: Ndjatox (spores extracted from the soil of Ndjamena with the variety Tox-728-1); T5: Kema (spores extracted from Kelo soil with the Madjitolngar variety); T6: Kolotox (spores extracted from the soil of Kolobo with the variety Tox-728-1); T7: Ndjama (spores extracted from the soil of Ndjamena with the variety Madjitolngar); T8: Ketox (spores extracted from Kelo soil with the variety Tox-728-1); T9: Positive Witness; T10: Negative content. For each rice variety, bars affected with the same letter are not significantly different between treatments at the indicated level of probability.

AMF-treatments (T5, T6 for Madjitolngar variety, T4, T6 for Tox728-1 variety) were equal to the chemical fertilizer treatment, other were above enough compared to the negative control treatment (T10), referring to as plots that received neither chemical fertilizers nor mycorrhiza. Enhanced yields have been attributed to high number of panicles developed by AMF-plants as previously claimed by Guei *et al.* [35] (2003). The weight of 1000 grains obtained during this study was closed to between 26 and 33 g obtained by Ondo Ovono *et al.* [36] in some cultivated rice varieties. Related work carried out by Ngakou *et al.* [18], Emadzadeh *et al.* [37], have revealed consistent 1000 grains weight harvested on AMF-inoculated Nerica rice varieties. According to Emadzadeh *et al.* [37], the main factor involved in the grains filling process is sunlight, which acts uniformly and at the same time on all plants growing on an experimental surface area. The weight of 1000 grains of Madjitolngar variety (29.73 g) was closed to that of Nerica N6 variety, while that of Tox-728-1 (23.32 g) was not too far from 24.22 g corresponding to 1000 g grains of Nerica NL28 obtained by Ngakou *et al.* [18]. The 1000 grains weight of Nerica N6 ranging between 31.03 - 33.76 g and 29.48 - 31.89 g was recently reported by Gevrek *et al.* [38]. A positive and significant correlation was observed between the weight of 1000 grains per experimental unit and the grain yield expressed in t/ha. The yields obtained with the

most performant AMF-inoculants in the Madjitolngar variety were higher, while those of the most performant AMF-inoculants in the Tox-728-1 variety were lower and similar to those obtained by Natebaye [39] on Nerica FKR62, which was 5.8 and 6.8 t/ha, respectively for AMF and chemical fertilizers treatments, compared to 4.8 t/ha for the control treatment. These results confirm the compatibility of indigenous AMF-inoculants with the local rice varieties, but at various degrees. For each inoculum used, mycorrhization had a positive impact on the grain yield of the two rice varieties, treatments T1 (for Madjitolngar variety) and T3 (for Tox variety) harboring the highest yield of 7.15 and 5.21 t/ha respectively. Inoculation of rice with suitable mycorrhiza under wetland rice varieties has been reported to be beneficial to plants in terms of yield, corresponding to half the normal dose of phosphate fertilizer used [40], in agreement with the acceleration of N and P transfer from shoots and/or soils to grains in flooded conditions by mycorrhiza [27] [41]. Other results on the positive responses to mycorrhizal inoculation of lowland rice plants [42] [43], and of many cereals including millet [44], corn [45], have been reported. Our results are either higher than 4.2 t/ha obtained for the Nerica 11 variety, or approach 7.15 t/ha for the Nerica 14 variety revealed in Gabon [36].

4. Conclusion

Arbuscular mycorrhizal fungi (AMF) in an agricultural ecosystem are necessary for proper management of beneficial symbiosis. At the end of this study, it is discovered that among the several selected AMF-inoculants produced, inoculants referring to as Koloma (T1), Latox (T3) and Kolotox (T6) on the one hand, Koloma (T1) and Ketox (T8) on the other hand, were respectively best suited for the improvement of growth and yield of the rice varieties Madjitolngar and Tox-728-1 tested under field conditions at Kelo. Although field trials are still scheduled for other sampling sites from which AMF-spores were isolated, the two rice varieties have shown a dependency to endomycorrhizal symbiosis at Kelo. Hence, an industrial-scale production of the efficient endomycorrhizal inoculants is necessary to sustainably boost the productivity of this important crop in South-Chad.

Acknowledgements

The authors are thankful to the Malla Farm company of Tandjile-Chad from where the two rice varieties (Madjitolngar and Tox-728-1) were gratuitously provided.

Conflicts of Interest

The authors declare no conflicts of interest regarding the publication of this paper.

References

- [1] Smith, R.F. and Read, D.J. (1997) Vesicular Arbuscular Mycorrhiza in Agriculture

- and Horticulture. In: Smith, S. and Read, D., Eds., *Mycorrhizal Symbiosis*, 3rd Edition, Academic Press, Cambridge, 454-459.
- [2] Berdeni, D., Cotton, T.E.A., Daniell, T.J., Bidartondo, M.I., Cameron, D.D. and Evans, K.L. (2018) The Effect of Arbuscular Mycorrhizal Fungal Colonisation on Nutrient Status, Growth, Production and Canker Resistance of Apple (*Malus punila*). *Frontiers in Microbiology*, **9**, 1461. <https://doi.org/10.3389/fmicb.2018.01461>
 - [3] Thirkell, J.T., Pastok, D., and Field, J.K. (2019) Carbon for Nutrient Exchange between Arbuscular Mycorrhizal Fungi and Wheat Varies According to Cultivar and Changes in Atmospheric Carbon Dioxide Concentration. *Global Change Biology*, **26**, 1725-1738. <https://doi.org/10.1111/gcb.14851>
 - [4] Linderman, R.G. (1988) Mycorrhizal Interactions with the *Rhizosphere microflora*: The Mycorrhizosphere Effect. *Phytopathology*, **78**, 366-371.
 - [5] Linderman, R.G. (1992) Vesicular-Arbuscular Mycorrhizae and Soil Microbial Interactions. In: Bethlenfalvay, G.J. and Linderman, R.G., Eds., *Mycorrhizae in Sustainable Agriculture*, ASA Inc., Madison, Wisconsin, 45-70. <https://doi.org/10.2134/asaspecpub54.c3>
 - [6] Marschner, H. (1991) Mechanisms of Adaptation of Plants to Acid Soils. In: Wright, R.J., Baligar, V.C. and Murrmann, R.P., Eds., *Plant-Soil Interactions at Low pH. Developments in Plant and Soil Sciences*, Vol. 45, Springer, Dordrecht, 683-702. https://doi.org/10.1007/978-94-011-3438-5_78
 - [7] Sylvia, D.M. and Williams, S.E. (1992) Vesicular-Arbuscular Mycorrhizae and Environmental Stress. In: Linderman, R.G. and Bethlenfalvay, G., Eds., *Mycorrhizae in Sustainable Agriculture*, American Society of Agronomy, Madison, Wisconsin, 101-124. <https://doi.org/10.2134/asaspecpub54.c5>
 - [8] Strullu, D.G. (1991) Mycorrhiza of Trees and Cultivated Plants. 3rd Edition, Lavoisier, Paris, France, 249 p.
 - [9] Karagiannidis, N. and Hadjisavva-Zinoviadi, S. (1998) The Mycorrhizal Fungus *Glomus mosseae* Enhances Growth, Yield and Chemical Composition of a Durum Wheat Variety in 10 Different Soils. *Nutrient Cycling in Agroecosystems*, **52**, 1-7. <https://doi.org/10.1023/A:1016311118034>
 - [10] Dodd, J.C., Burton, C.C., Burns, R.G., and Jeffries, P. (1987) Phosphatase Activity Associated with the Roots and the Rhizosphere of Plants Infected with Vesicular-Arbuscular Mycorrhizal Fungi. *New Phytologist*, **107**, 163-172. <https://doi.org/10.1111/j.1469-8137.1987.tb04890.x>
 - [11] Dodd, J.C., Boddington, C.I., Rodriguez, A., Gonzalez-Chavez, C. and Mansur, I. (2000) Mycelium of Arbuscular Mycorrhizal Fungi (AMF) from Different Genera: Form, Function, and Detection. *Plant and Soil*, **226**, 131-135. <https://doi.org/10.1023/A:1026574828169>
 - [12] Lizhi, G., McCarthy, E.M., Ganko E.W. and McDonald, J.F. (2004) Evolutionary History of *Oryza sativa* LTR Retrotransposons: A Preliminary Survey of the Rice Genome Sequences. *BMC Genomics*, **5**, 1-18.
 - [13] Dorin, B., Treyer, S. and Paillard, S. (2009) Agrimonde—Scenarios and Challenges to Nourish the World in 2050. Springer, Netherlands.
 - [14] Siddiqui, S.U., Kumamaru, T. and Satoh, H. (2007) Pakistan Rice Genetic Resources-I: Grain Morphological Diversity and Its Distribution. *Pakistan Journal of Botany*, **39**, 841-848.
 - [15] Gnamkoulamba, A., Tounou, A.K., Tchao, M., Tchabi, A., Adjevi, A.K.M. and Batawila, K. (2018) Field Evaluation of the Growth and Production Potentials of Rice (*Oryza sativa* L.) Varieties IR841 inoculated in the Nursery with Four Strains of

Arbuscular Mycorrhizal Fungi. *European Science Journal*, **14**, 1857-7881.

- [16] Griffon, M. (2010) For Ecological Intensive Agriculture. The Towel of Aigues, 112 p.
- [17] Ngakou, A., Nwanga, D., Nebane, C.L.N., Ntonifor, N.N., Tamò, M. and Parh, I.A. (2007) Arbuscular-Mycorrhizal Fungi, Rhizobial and *Metarhizium anisopliae* Enhance P, N, Mg, K and Ca Accumulation in Fields Grown Cowpea. *Journal of Plant Science*, **2**, 518-529. <https://doi.org/10.3923/jps.2007.518.529>
- [18] Ngakou, A., Mbaiguinam, M., Nadjilom Y. and Tokam, N.M. (2013) Agro-Morphological and Physical Paddy Seed Attributes of Nerica and Local Rice Varieties as Affected by Mycorrhizal Inoculation and Compost Application under Upland Conditions. *International Journal of Agricultural Science and Research*, **3**, 43-62.
- [19] Abbas, Y., Arahou, M., Duponnois, R. and Abourou, M. (2013) Effect of Arbuscular Mycorrhiza on Growth and Nutrition of *Tetraclinis articulata* (Vahl) from Three Origins. *Annal of Research in Forestry in Maroc*, **42**, 7-16.
- [20] Enkhtuya, B., Rydlova, J. and Vosathka, M. (2000) Effectiveness of Indigenous and Non-Indigenous Isolates of AM Fungi in Soils from Degraded Ecosystems and Man-Made Habitat. *Applied Soil Ecology*, **14**, 201-211. [https://doi.org/10.1016/S0929-1393\(00\)00057-3](https://doi.org/10.1016/S0929-1393(00)00057-3)
- [21] Nadjilom, Y. (2012) Agro-Morphological Characterization of Four Rice (*Oryza* spp.) Varieties Grown in Tandjile Region in Response to Organic and Mycorrhizal Fertilization. Master Thesis, Department of Biological Sciences, Faculty of Science, University of Ngaoundere, Ngaoundere, 57 p.
- [22] Nadjilom, Y., Toukam, T.S., Tobolbaï, R. and Ngakou, A. (2019) Morphological and Structural Characterization of Rhizospheric Endomycorrhiza Communities Associated with Rice Grown in the Sahelian Zone (Chad). *International Journal Plant Soil Science*, **31**, 1-14. <https://doi.org/10.9734/ijps/2019/v31i530222>
- [23] Brundrett, M., Bougher, N., Dell, B. and Malajczuk, N. (1996) Working with Mycorrhizas in Agriculture and Forestry. Canberra, Australia, 37 p.
- [24] Gerdeman, J.W. and Nicolson, T.H. (1963) Spore of Mycorrhizal Endogone Species Extracted from Soil by Wet Sieving and Decanting. *Transaction of the British Mycological Society*, **46**, 235-244. [https://doi.org/10.1016/S0007-1536\(63\)80079-0](https://doi.org/10.1016/S0007-1536(63)80079-0)
- [25] Cardenas, R.E. (2010) Does Rycorrhization Favor the Access to Nitrogen Forms Complexes? Study on the Nutrition of *Pinus pinea*. National Centre for Scientific Recherche, University François Rabelais de Tours, Tours.
- [26] Hamza, N. (2014) Application of Arbuscular Mycorrhiza in Gardening Crops: Case Study of Water Melon (*Citrullus lanatus*). Master Thesis, University of Ferhat Abbas Setif, Setif.
- [27] Solaiman, M.Z. and Hirata, H. (1996) Effectiveness of Arbuscular Mycorrhizal Colonization at Nursery-Stage on Growth and Nutrition in Wetland Rice (*Oryza sativa* L.) after Transplanting under Different Soil Fertility and Water Regimes. *Soil Science and Plant Nutrition*, **42**, 561-571. <https://doi.org/10.1080/00380768.1996.10416325>
- [28] Jangde, N. (2013) Mycorrhizal Study in Selected Cultivars of Rice. Ph.D. Thesis, The Rajendra Agricultural University, Bihar, Pusa.
- [29] Elyazeid, A.A., Abou-Aly, H., Mady, M. and Moussa, S.A.M. (2007) Enhancing Growth, Productivity and Quality of Squash Plants Using Phosphate Dissolving Microorganisms (Bio Phos-Phor) Combined with Boron Foliar Spray. *Research Journal of Agriculture and Biological Science*, **3**, 274-286.

- [30] Manzo, O.L., Ibrahim, D., Campanella, B. and Pau, R. (2009) Effects of Mycorrhizal Inoculation of the Substrate on Growth and Water Stress Tolerance of Five Sand Dune Fixing Species: *Acacia senegal* (L.) Willd; *Prosopis chilensis* stunz and *Bauhinia rufescens* Lam. *Tropical Geology and Ecology*, **33**, 115-124.
- [31] Zougari-Elwedi, B., Sanaa, M., Labidi, S. and Lounès-Haj, S.A. (2012) Evaluation of the Impact of Arbuscular Mycorrhization on the Mineral Nutrition of Plantlets of *Phoenix dactylifera* L. var. Deglet Nour. *Study and Management of Soils*, **19**, 3-4.
- [32] Abbas, Y. (1998) Arbuscular Mycorrhiza in Arid Zones: Biodiversity and Role in the Tolerance of *Trifolium alexandrium* to Saline Stress. Doctorate Thesis, University of Cadi Ayyad, Morocco.
- [33] Douira, A., Rachid, B., Ahmed, O., Abdelmajid, M., Cherkaoui, E.M., Abdelkarim, F.M., Amina, O.T. and Mohamed, C. (2014) Effect of a Composite Endomycorrhizal Inoculum on the Growth of Olive Trees under Nurseries Conditions in Morocco. *International Journal Pure and Applied Bioscience*, **2**, 1-14.
- [34] Ngonkeu, M.E.L. (2009) Soil Tolerance of Certain Maize Varieties to L'aluminium and Manganese and Study of the Fonctional Diversity des Arbuscular Mycorrhizal Fungi in Cameroon. Department of Microbiology and Biochemistry, Faculty of Science, University of Yaounde, Yaounde.
- [35] Guei, G.G., Abamu, F., Traoré, K. and Naman, S. (2003) Genetic Variability in Morphological and Physiological Traits within and among Rice Species and Their Interspecific Progenies. *African Agronomy*, **16**, 15-32.
<https://doi.org/10.4314/aga.v16i1.1636>
- [36] Ondo Ovono, P., Loumbe, M.M., Kevers, C. and Dommes, J. (2013) Field Evaluation of Agro-Morphologic Characteristics of Certain Varieties Nerica Rice Tested in the South-East in Gabon. *African Agronomy*, **25**, 13-23.
- [37] Emadzadeh, B., Razavil, S.M.A. and Farahmandfar, R. (2010) Monitoring Geometric Characteristics of Rice during Processing by Image Analysis System and Micrometer Measurement. *International Agrophysics*, **24**, 21-27.
- [38] Gevrek, M.N., Atasoy, G.D. and Yigi, F.A. (2012) Growth and Yield Response of Rice (*Oryza sativa*) to Different Seed Coating Agents. *International Journal of Agricultural Biology*, **14**, 826-830.
- [39] Natebaye, D. (2010) Contribution of Mycorrhizae to Production of Two Nerica (*Oryza sativa* × *Oryza glaberrima*) Rice Varieties Cultivated in the Field at Dang Ngaoundere, Cameroon. Master Thesis, Department of Biological Sciences, Faculty of Science, University of Ngaoundere, Ngaoundere.
- [40] Gupta, N. and Ali, S.S. (1993) VAM Inoculation for Wetland Rice. *Mycorrhiza News*, **5**, 5-6.
- [41] Solaiman, M.Z. and Hiratam, H. (1995) Effects of Indigenous Arbuscular Mycorrhizal Fungi in Paddy Fields on Rice Growth and N, P, K Nutrition under Different Water Regimes. *Soil Science and Plant Nutrition*, **41**, 505-514.
<https://doi.org/10.1080/00380768.1995.10419612>
- [42] Gangopashyay, S. and Das, K.M. (1984) Interaction between Vesicular Arbuscular Mycorrhiza and Rice Roots. *Indian Phytopathology*, **35**, 34-38.
- [43] Iqbal, S.H., Tanquir, S. and Ahmad, J.S. (1978) The Effect of Vesicular Arbuscular Mycorrhizal Associations on Growth of Rice (*Oryza sativa*) under Field Conditions. *Pakistan Journal of Biological Science*, **24**, 357-365.
- [44] Rao, Y.S.G., Bagyaraj, D. and Rai, P.V. (1983) Selection of Efficient VA Mycorrhizal Fungus for Finger Millet: Glass House Screening. *Zentralblatt für Mikrobiologie*, **138**, 400-413. [https://doi.org/10.1016/S0232-4393\(83\)80038-9](https://doi.org/10.1016/S0232-4393(83)80038-9)

- [45] Lu, S. and Miller, R.H. (1989) The Role of VA Mycorrhizae in the Absorption of P and Zn by Maize in Field and Growth Chamber Experiments. *Canadian Journal of Biological Science*, **69**, 97-109. <https://doi.org/10.4141/cjss89-009>