



## Effect of an Organic Fertilizer Based on Native *Rhizophagus intraradices* on *Zea mays* L. Yield in Northern Benin

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### Authors' contributions

This work was carried out in collaboration among all authors. Authors ADK, ASY, NAA and LBM designed the study, performed the analysis, wrote the protocol and wrote the first draft of the manuscript. Authors ADK, MA, GD, AA and MRA managed the analyses of the study. Authors ADK, RR, GD, AA and LBM managed the literature searches. All authors read and approved the final manuscript.

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### ABSTRACT

**Aims:** One of the alternatives proposed to improve the sustainability and productivity of agricultural systems is the use of organic fertilizers based on arbuscular mycorrhizal fungi. The objective of the study was to evaluate the effectiveness of the native mycorrhizal fungus *Rhizophagus intraradices* in the field at Ouenou in North Benin for improved maize (*Zea mays* L.) production.

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**Study Design:** The experimental design was a complete random block of three treatments. Mention the design of the study here.  
**Place and Duration of Study:** Endomycorrhizal infection was assessed on roots of corn plants at 78 days post-seed.  
**Methodology:** Corn variety 2000 SYN EE-W was inoculated with the strain in combination with the mineral fertilizer (NPK) half-dose.  
**Results:** Results showed that of the three treatments, the highest heights were observed in plants treated with *Rhizophagus intraradices* +½ dose of NPK (15:15:15) and urea. The same observations were made with respect to leaf number and leaf area of plants and grain yield. Regarding the endomycorrhizal infection of plants inoculated with *Rhizophagus intraradices* +½ dose of NPK (15:15:15) and urea, of the three producers, the observation was that the endomycorrhizal infection was elevated to the level of the second producer (P2) which was of the order of (50%) while the mycorrhizal intensity was lower (24.6%) in the same producer.  
**Conclusion:** The native mycorrhizal fungus *R. intraradices* plus the half-dose NPK plus urea, had a positive impact on most of the various parameters.

**Keywords:** Mycorrhizal fungus; native; *R. intraradices*; *Zea mays* L.; organic fertilizers.

## 1. INTRODUCTION

Cereals have been the staple of many populations for thousands of years [1]. The most important are wheat, millet, sorghum, maize and rice, the production and consumption of which vary from one country to another. In Benin, the agricultural sector contributes about 40% of Gross Domestic Product (GDP) and 80% of export earnings [2]. Maize in Benin in general and in the north of Benin in particular, is no longer just a crop for own consumption, but is also intended for sale [3].

However, for some time now, the yields of maize obtained at the producer level have been well below the potential yields of the popularized varieties [4]. The threat of a food crisis is therefore persistent and the solutions proposed by the leaders tend to promote the intensification of agriculture on the model of the developed countries [5]. One solution to the observed yield decline is to invest in land fertility [6]. For example, the use of mineral fertilizers is one of the solutions proposed to compensate for nutrient losses and nutritional deficiencies observed at the land level [7]. Unfortunately, prolonged intensive use of mineral fertilizers leads to soil degradation [8].

For several years now, many researchers have been looking at this crisis and looking for solutions to address it and support global food security [9].

In response to these, land degradation problems, one of the alternatives proposed to improve the sustainability of agricultural systems is the use of

organic fertilizers based on arbuscular mycorrhizal fungi [10]. Arbuscular mycorrhizal fungi are one of the key groups for ensuring ecosystem sustainability and increasing productivity [11]. They are known to be involved in improving the uptake of mineral elements, particularly phosphorus [12].

The use of biotechnologies and applied scientific and technical innovations is essential for the expansion of culture and the improvement of production methods associated with the concept of sustainable development [13]. Appropriate management of mycorrhizae in agriculture has been shown to substantially reduce mineral fertilizer inputs while maintaining good yields [10]. Studies have shown that plant growth is generally greater in the presence of these fungi arbuscular thanks to their ability to exploit a greater volume of soil than roots of plants [14].

Given the problem of land degradation and the ability of arbuscular mycorrhizal fungi to sustain the ecosystem and increase productivity, the objective of this study is to assess the effectiveness of the mycorrhizal fungus *Rhizophagus intraradices* in a peasant environment in Ouenou, North Benin, for improved maize (*Zea mays* L.) production.

## 2. MATERIALS AND METHODS

### 2.1 Experimental Site

The study was conducted in the district of Ouénou located between 9°47'6" north latitude and 2°38'5" east longitude. It is located in the municipality of N'dali (Fig. 1) and is characterized

by a rainy season between April and October. The average rainfall varies between 900 mm and 1100 mm. The soils encountered in the area are ferruginous tropical [15].

## 2.2 Materials

### 2.2.1 Variety of maize

The corn variety 2000 SYN EE-W was used in the experiment. It is an early 80-day variety developed by the International Institute of Tropical Agriculture (IITA) and the National Institute of Agricultural Research of Benin (INRAB). The yield in a peasant environment is 2.5 t/ha. It is a variety that has good resistance to breakage, streak, rust-American, the spot blotch. It is also tolerant of pests and drought [16].

### 2.2.2 Microorganisms

The microorganism used is *Rhizophagus intraradices*. The inoculum used was a

solid, clay-textured *R. intraradices* strain isolated and characterized from rhizosphere soils of maize in Benin and preserved in the laboratory for Biology and Molecular typing in Microbiology of the Faculty of Science and Technology of the University of Abomey-Calavi.

## 2.3 Experimental Design

The experimental design was a completely randomized random block of three treatments with three replicates. Each treatment covered an elemental plot of 12.8 m<sup>2</sup> made up of 4 lines of 4 m. seeding was done at a spacing of 0.80 m x 0.40 m, giving a density of 31.250 plants/ha. The paths between plots and replicates were 1.8 m and 2 m respectively. Growth and yield data were collected on the useful plot (6.4 m<sup>2</sup>) represented by the two Central lines. The different treatments evaluated are presented in Table 1.

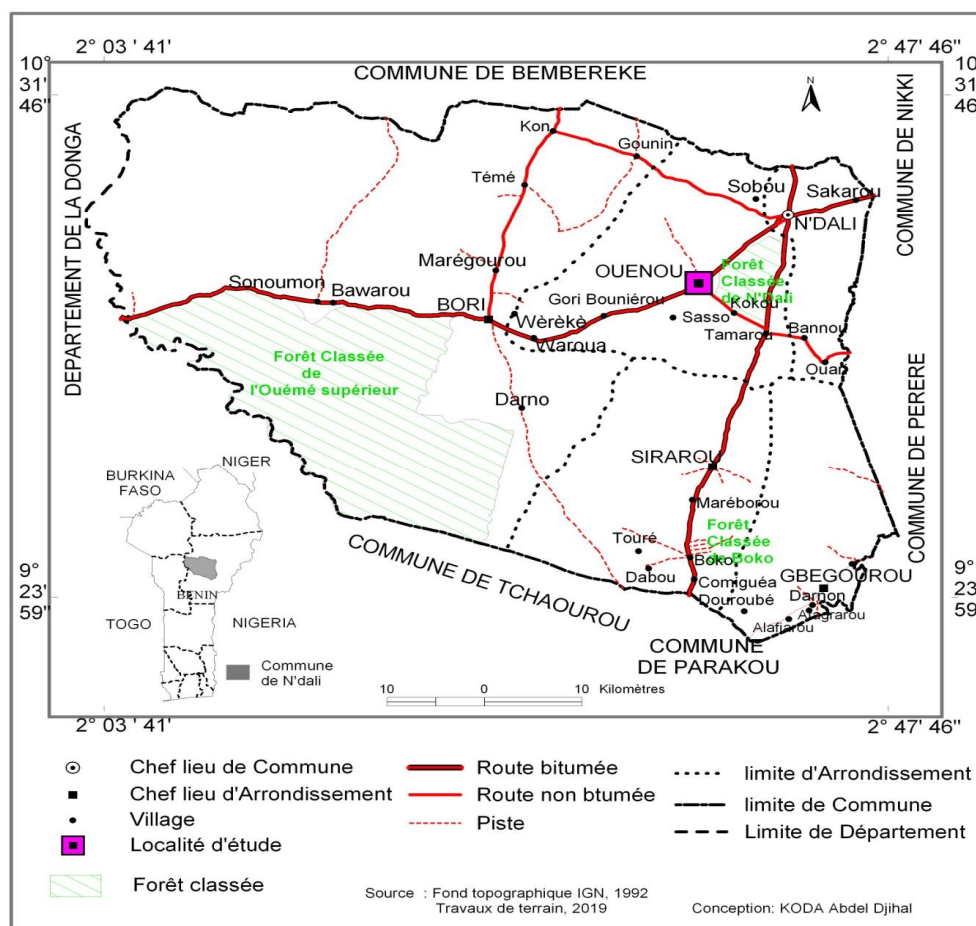


Fig. 1. Geographical location of the experimental station

**Table 1. Used treatments**

N°	Treatments	Code
1	Country Practice	T1
2	<i>Rhizophagus intraradices</i> +½ dose of NPK (15:15:15) and urea	T2
3	100% NPK + urea recommended (200 kg / ha)	T3

(N: 15; P: 15; K: 15) indicates the dose of elements (N, P and K) in 100 kg of NPK fertilizer. The recommended mineral fertilizer dose is 200 kg / ha NPK at sowing and 100 kg / ha urea at flowering [17]

## 2.4 Inoculation of Maize Seeds by the Mycorrhizal Fungus

Inoculation of corn seeds with *R. intraradices* was carried out using the method described [18]. The amount of Mac applied was 10% of the weight of the corn seed with an amount of water equivalent to 600-ml. kg<sup>-1</sup> of Mac. The seeds were coated and then left to air for drying for 12 hours.

## 2.5 Seeding and Maintenance of Plants

Two seeds of corn (inoculated or non-inoculated) were placed in a pot and were immediately closed after making about 5 cm of depth. For the maintenance of the plants, two weeding was carried out, the first weeding was coupled with reducing the plant to one per pot two weeks after sowing, and the least vigorous plant was uprooted. The second weeding was carried out six weeks after planting.

## 2.6 Growth Parameters

Growth parameters such as length, Crown diameter were measured every 15 days during the 80 days, and leaf area was measured on the 60 days. Estimates of arbuscular mycorrhizal infection were made [19] and mycorrhizal rates were estimated [20].

## 2.7 Yield Parameters

In the 90<sup>th</sup> century, the plants on the two central lines of each elementary plot were harvested. Twelve (12) plants were harvested per elementary plot. After spathe remove from corn ears, the total weight of the corn ears on each plot was taken using an electronic scale (Highland HCB 3001. Max 3000g x 0.1 g). After this operation, the ears of each plot were erased. The total weight of the maize kernels and the relative humidity (LDS-1F) of the kernels were taken at the level of each treatment.

The average grain yield of maize plants was determined according to the formula [21]:

$$R = (P \times 10.000) / (S \times 1.000) \times 14 / (\% H),$$

Where:

R is the average maize grain yield expressed as t ha<sup>-1</sup>

P is the weight of maize kernels expressed in KG)

10000 is conversion of ha to m<sup>2</sup>;

S is the harvest area in m<sup>2</sup>;

1000 conversion from ton (t) to kg

% H is the moisture content of the kernels, expressed%

## 2.8 Statistical Analysis

Yield and leaf area data were subjected to a two-factor mixed-effect analysis of variance. Root collar diameter, leaf number, and height were subjected to repeated analysis of variance, and the degree of Root infestation was analyzed through proportion comparison tests. The software R 3.5.2, nlme packages, car, rcompanion and ggplot2 were used to carry out these analyses.

## 3. RESULTS AND DISCUSSION

### 3.1 Results

#### 3.1.1 Height

Repeated analysis of variance revealed similar growth induction performance at all three treatments. Height growth over time has varied increasingly (Fig. 2). Of the three treatments, we note that treatment with 100% NPK+ urea (T3) gave a good result followed by treatment with *R. intraradices* + ½ dose of NPK (15:15:15) and urea (T2) in relation to the witness.

#### 3.1.2 Leaf area

The results of the analysis of variance showed that there was no difference ( $p > 0.05$ ) between the effects of the three treatments on plant leaf area. However, the T3 treatment gave a relatively high leaf area (Fig. 3).

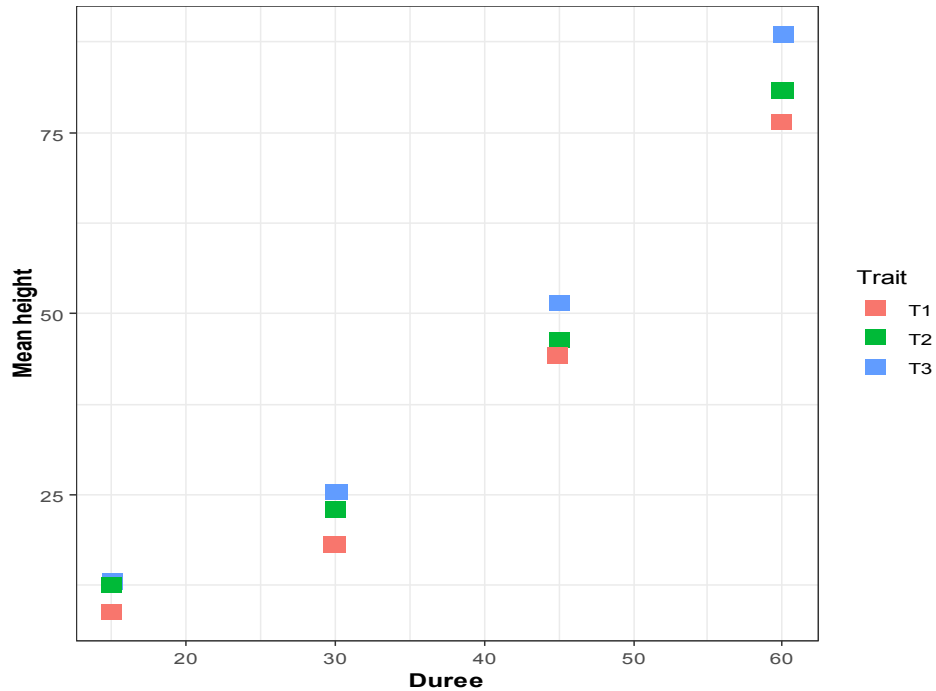


Fig. 2. Interaction plot treatment and duration on the mean height of plants

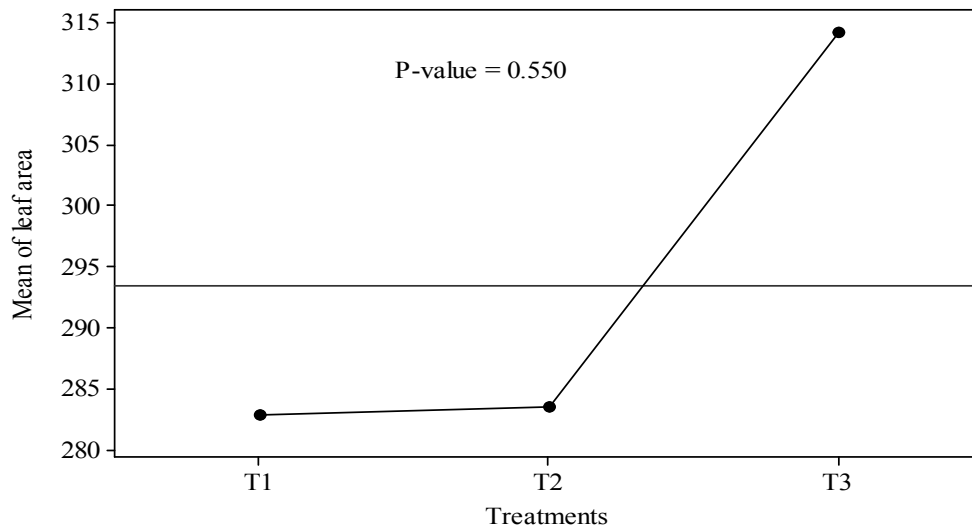


Fig. 3. Average leaf area of production as a function of treatments

### 3.1.3 Diameter

Results from the replicate custom analysis of variance table showed a significant difference ( $p < 0.05$ ) in Mean root collar diameter of plants induced by different treatments. Mean Crown diameter also varied over time ( $p < 0.001$ ). The difference between treatments was very remarkable at 30 and 45 days with T3 treatment

followed by T1 treatment, which induced large-diameter root collar plants (Fig. 4).

### 3.1.4 Number of sheets

The results of the repeated custom analysis of variance showed that the average number of leaves produced by plants under different treatments remained similar between treatments.

In contrast, leaf number varied over time with *Rhizophagus intraradices* +½ dose of NPK (15:15:15) and urea (T3) at the top (Fig. 4).

### 3.1.5 Yield

Analysis of variance results from the mixed-effect model showed that the treatments induced yields similar to the 5% threshold. Nevertheless, the yield induced by farmers' practices (T1) remains low (1.51) compared to *Rhizophagus intraradices*

+½ dose of NPK (15:15:15) and urea (T2) which is 1.74 (Fig. 6).

### 3.1.6 Degree of root infection

Analysis of the infection rate of plant roots among different growers revealed a significant difference in the proportions of infected roots at the producer level with the highest infection rate at the producer level 2 (Table 2). No significant differences were observed in mycorrhizal intensity (F %). visual.

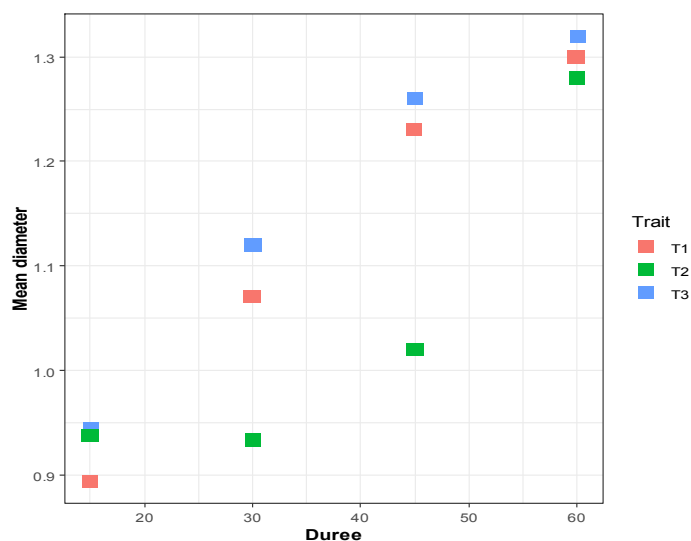


Fig. 4. Interaction plot treatment and duration on the mean diameter of the seedlings

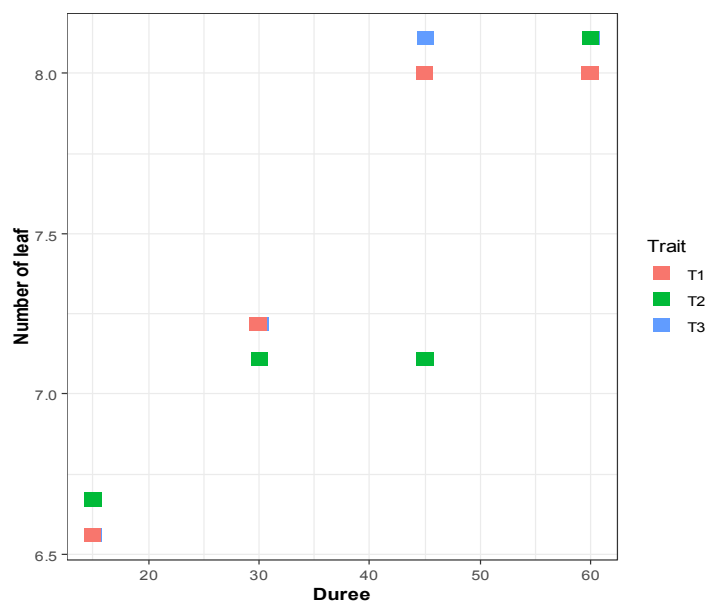


Fig. 5. Interaction plot treatment and duration on the mean leaf number of seedlings

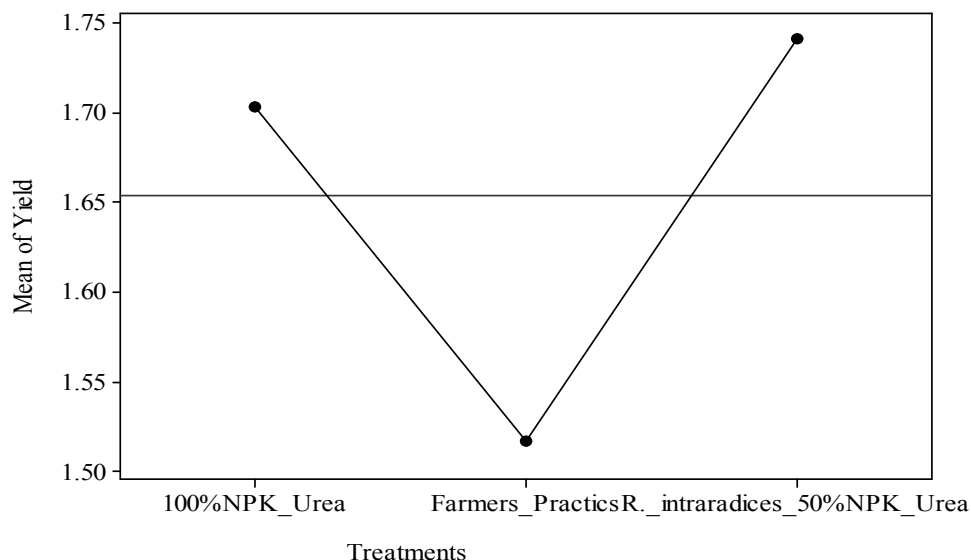


Fig. 6. Average production yield as a function of processing

Table 2. Degree of root infection

Producers	% infection	F % visual
P1	33	18.48
P2	50	27.60
P3	38	23.68
Probability	0.042	0.635

P1: producer 1, P2: producer 2, P3: producer 3, F%: mycorrhizal intensity, Infection %: degree of infection

### 3.2 Discussion

The results of this study showed a temporal height growth performance in the three treatments with T3 (44.5%) followed by T2 (40.6%) compared with the control, an increase of 10.21%. Studies have shown that when phosphorus and trace element availability is limited, mycorrhizal symbiosis enhance the growth of inoculated plants is faster than that of no inoculated plants [22]. Similar results were observed by some authors [23], which showed that corn plants receiving *Rhizophagus intraradice* plus half the dose of NPK induced good results (17.43%). Plant height relative to control plants. Other studies, such as those by Kahiluoto et al. [24] and Valentine et al. [25], also showed a decrease in mycorrhizal plant growth compared with nonmycorrhizal plants. This could be explained by an imbalance in the flow of carbon to the Mac in exchange for mineral elements [26]. Indeed, work conducted by Diagne and Ingleby [27] has shown that high mycorrhizal colonization of roots does not always result in improved plant growth. The beneficial role of Macs on plant growth may be due to

improved uptake, transport, and uptake of mineral elements, primarily phosphorus, by plant tissues [28]. This improvement in the nutritional status of mycorrhizal plants can be due to extra-radical mycelial hyphae that explore a larger volume of soil that is not accessible only to plant roots [29].

Considering the leaf area, *Rhizophagus intraradices* +½ dose of NPK (15:15:15) and urea (T2) gave good results compared with the control (T1). Similar results were observed by previous study [23] showed that *Rhizophagus intraradices* +½ dose of NPK (15:15:15) and urea produced good results of corn plants leaf area compared with the control.

Analysis of variance results for root collar diameter revealed a significant difference ( $p < 0.05$ ) between the mean root collar diameters of plants induced by different treatments. The difference between treatments was very remarkable at 30 and 45 days with T3 treatment followed by T1 treatment, which induced large-diameter root collar plants. This could be explained by an imbalance in the flow of carbon

to the Mac in exchange for mineral elements [26; 36]. Other authors [23; 30] who showed that inoculation carried out on corn by biofertiliser (*Rhizophagus intraradice* + ½ dose of NPK (15:15:15) and urea) have observed contrary results when the observed a good result for root collar diameter.

Previous work on mycorrhizal fungi has shown that they have a positive effect on increasing leaf number [31]. Some studies have suggested that the only explanation is an increase in the rate of photosynthesis of the plant due to Macs [32]. Similar results were observed in our study, among the three treatments used, we noticed that the treatment T2 gave good results with time. Similarly, treatment with *Rhizophagus intraradices* +½ dose of NPK (15:15:15) and urea (T2) induced a good yield compared with the control, an increase of 15.23% followed by the full NPK (15:15:15) and urea (T3). Jangde [33] reported similar results showing a significant increase in the number of grains per panicle in field conditions when Rice was in association with the Macs. Indeed, several studies [34- 36] showed the beneficial effect of symbiosis between AMF and plants in their development, growth and production.

Of the three growers, we noted that only the plants of producer 2 had a good result with respect to root infection, which was 50% and intensity, which was 27.60%. Similar results were observed in the work of Kumar et al. [35] on Poaceae species (*Sporobolus indicus*, *Grevillea pteridifolia*, *Eragrostis tenella*, *Digitaria bicornis*, and *Cynodon dactylon*), which induced mycorrhizal frequencies ranging from 72.8% with *R. irregularis* to 16.17% with *R. irregularis*. This study allowed us to show the beneficial effect of mycorrhizal fungi on the good development and productivity of corn plantings.

#### 4. CONCLUSION

This study allowed us to show that the native mycorrhizal fungus *R. intraradices* plus the half-dose NPK plus urea, had a positive impact on most of the various parameters (height growth, leaf number, and yield). In view of the results obtained, it will be good to popularize this fertilizer at the level of producers.

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#### COMPETING INTERESTS

Authors have declared that no competing interests exist.

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