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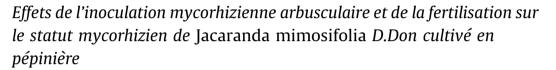
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Effects of arbuscular mycorrhizal inoculation and fertilization on mycorrhizal Statute of *Jacaranda mimosifolia* D.Don cultivated in nurseries



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#### ABSTRACT

The effects of fertilization and the nature of the *inoculum* as well as the variation of the dose intake of the latter on the level of Jacaranda mimosifolia D.Don mycorhization were tested. Young plants were treated with two inoculums presenting different origins, compositions and modes of application: one is a commercial product containing *Glomus* irregulare, and the other is a composite indigenous inoculum resulting from trapping five species of genus Glomus and also from multiplication on mycotrophic plants: leek (Allium porrum L.) and vetch (Vicia sativa L.). For each inoculum, two doses were tested and for each dose of inoculum, four levels of fertilization based on a complete commercial fertilizer (Osmocote) were tested: 0 g/plant, 2 g/plant, 4 g/plant, and 6 g/plant. Three repetitions were performed for each combination treatment of inoculum/fertilizer. One-year-old young Jacaranda plants, being about 40 cm high, were cultured under greenhouse in 10/ 12 cm caliber pots. After six months, all the inoculated plants were mycorrhized. According to endomycorrhizal structures found on their roots, plants receiving doses of composite indigenous inoculum reached a more advanced stage of mycorrhization than those treated with the commercial inoculum. The existence of an interaction effect between the *inoculum* dose and the level of fertilization on *Jacaranda* mycorhization rate was excluded. These two parameters of variation were studied as simple effects. The increase in commercial inoculum dose had a significant positive influence on the level of *Jacaranda* plants mycorrhization (P=0.05). The rate of mycorrhization jumped from 12.69% to 21.92%. Nonetheless, for plants receiving increasing doses of composite indigenous inoculum, the level of mycorrhization has varied randomly. In both instances of inoculum treatments, increasing the dose of fertilizer significantly inhibited endomycorrhizal colonization of Jacaranda roots (P=0.01). Thus, the rate of root colonization decreased from 47.43% to 2.41% for plants receiving the composite indigenous inoculums. It decreased from 32.35% to 3.95% for those treated with the commercial inoculum. Mycorrhization had a positive effect on root dry biomass of Jacaranda, as in the case of unfertilize ave the highest rates of colonization.

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#### RÉSUMÉ

Les effets de la fertilisation et de la nature de l'inoculum ainsi que de la variation de la dose d'apport de ce dernier sur le niveau de mycorhization de Jacaranda mimosofolia D.Don ont été testés. De jeunes plants ont été traités par deux *inocula* d'origines, de compositions diverses et selon des modes d'application différents: l'un est un produit commercial à base de Glomus irregulare et l'autres est un inoculum indigène composite issu d'un piégeage de cinq espèces du genre Glomus et d'une multiplication sur plantes mycotrophes : poireau (Allium porrum L.) et vesce (Vicia sativa L.). Pour chaque inoculum, deux doses ont été testées, et pour chaque dose d'inoculum, quatre niveaux de fertilisation à base d'un engrais complet commercial (Osmocote) ont été testés : 0 g/ plant, 2 g/plant, 4 g/plant et 6 g/plant. Trois répétitions sont réalisées pour chaque traitement de combinaison inoculum/fertilisant. De jeunes plants de Jacaranda âgés d'un an et présentant environ 40 cm de hauteur ont été cultivés sous serre dans des pots de calibre 10/12 cm. Après six mois, tous les plants inoculés étaient mycorhizés. D'après les structures endomycorhiziennes présentes sur leurs racines, les plants ayant reçu des doses d'inoculum indigène composite ont atteint un stade de mycorhization plus avancé que ceux traités avec l'inoculum commercial. L'existence d'un effet d'interaction entre la dose d'inoculum et le niveau de fertilisation sur le taux de mycorhization de Jacaranda a été exclue. Ces deux paramètres de variation ont été étudiés comme effets simples. L'augmentation de la dose d'inoculum commercial a eu une influence significativement positive sur le niveau de mycorhization des plants de Jacaranda (p = 0,05). Leur taux de mycorhization est alors passé de 12,69% à 21,92%. En revanche, chez les plants ayant reçu des doses croissantes en inoculum indigène composite, le niveau de mycorhization a varié aléatoirement. Dans les deux cas de traitements d'inoculum, l'augmentation de la dose de fertilisant a significativement inhibé la colonisation endomycorhizienne des racines de Jacaranda (p = 0.01). Ainsi, le taux de colonisation racinaire a diminué de 47,43 % à 2,41 % chez les plants ayant recu l'inoculum indigène composite, et de 32,35 % à 3,95 % chez ceux traités avec l'inoculum commercial. La mycorhization a eu un effet positif sur la biomasse sèche racinaire de Jacaranda, comme dans le cas des plants non fertilisés et présentant les taux de colonisation les plus élevés.

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#### 1. Introduction

Mycorrhizae are symbiotic associations between plant roots with some fungi soil. Mycorrhizae directly influence mineral nutrition of the plant, water absorption, and resistance to biotic and abiotic stresses [1–4]. The plant generally provides carbohydrates to the fungus. Exchanges between the two hosts may vary according to the type of present mycorrhizal symbiosis [5]. Arbuscular endomycorrhizal fungi belong to the phylum of *Glomeromycota* [6]. They can develop arbuscules and vesicles [7]. These fungi have very little specificity to the host plant. In fact, 130 species of these fungi ensure mycorhization of more than 300,000 plant species [5].

It has been shown that various ligneous tree are able to develop a symbiosis with endomycorrhizal fungi species, such as marula (*Sclerocarya birrea*) and argan (*Argania spinosa*). Studies showed that the inoculation of different varieties of marula by 250 spores *Gigaspora margarita* has allowed one to obtain mycorrhizal young plants at a rate varying by around 40% [8]. Mycorrhization largely improved the resistance of these fruit tree species to water stress, salt well, as well as to flooding conditions. Other research showed that the argan plants colonized with a rate of 70% after their inoculation with different strains of the *Glomus* genus [9]. This mycorhization permitted a gain in dry biomass of aerial and root parts,

respectively equal to 120% and 70%, and improved nutrition elements, such as phosphorus, potassium, calcium, manganese, and copper. Inoculation of the same tree species presented an index of relative mycorrhizal dependency (IRMD) equal to 48% [10]. Jacaranda mimosifolia is also a ligneous tree. It is therefore possible that it withdraws benefits if it is mycorrhized at its nursery production, especially as these mycorrhizal inoculants are commercially available or can be trapped and multiplied under controlled conditions and with mycotrophic plants [11,12]. This requires a better understanding of various characteristics between endomycorrhizal fungi and ornamental ligneous tree, as well as possible interactions between inoculum and fertilizers doses. Indeed, these parameters can affect the level of mycorhization and, consequently, the development of the plant.

The aim of this study is to assess the mycorrhizal status of *Jacaranda mimosifolia* D.Don growing in nurseries, after various arbuscular endomycorrhizal inoculations in the presence of different levels of fertilization.

#### 2. Materials and methods

#### 2.1. Vegetal material and treatments applied

One-year young 40 cm high Jacaranda plants that were obtained using seedlings were inoculated with two

*inoculums* of different origins and compositions, according to different modes of application, and fertilized with increasing doses. These inoculants and fertilizer intakes were made simultaneously when repotting young plants of *Jacaranda* in 10/12 cm size pots, containing a substrate composed of peat and sand, with proportions respectively equal to 2/3 and1/3. The experiment was conducted in a greenhouse at the "Institut national agronomique de Tunisie" under homogeneous conditions; the temperature was maintained constant at 25 °C. After six months of cultivation, mycorhization rates of roots that had undergone different *Jacaranda* treatments were evaluated.

#### 2.2. Fertilizer applied

For each treatment of *inoculum*, four fertilizer levels were tested. Three repetitions were performed for each combination of *inoculum* and fertilizer as a treatment. The fertilizer used is the Osmocote EXACT standard-Scotts, a slow release NPK fertilizer (5-6 months), containing magnesium and trace elements 15+9+12 (+2.5). The detailed composition of the fertilizer is presented in Table 1. The tested doses of fertilizer and their corresponding proportions of nitrogen, phosphorus, and potassium are displayed in Table 1.

#### 2.3. Used inoculums

The *inoculums* used are made of arbuscular endomycorrhizal fungi and differ in their origins, nature and methods of application.

The first tested *inoculum* is in the form of a substrate containing spores and roots colonized by five strains from the genus of *Glomus* : *Glomus constricticum*, *Glomus geosporum*, *Glomus fuegianum*, *Glomus irregulare* and *Glomus* sp. These indigenous arbuscular endomycorrhizal fungi were trapped from soils of three prospected sites of Nahli National Park (Tunisia), on a substrate composed of one volume of perlite, four volumes of clay granule (Terra green) and one volume of vermiculite [13]. Amplification of the *inoculum* was then conducted by greenhouse crops of leek (*Allium porrum* L.) and vetch (*Vicia sativa* L.) during four months on a poor nutrient substrate made with sand

#### Table 1

Composition of fertilizer Osmocote EXACT.

Element	Concentration (%)
Nitrate nitrogen	7
Ammoniacal nitrogen	8
Phosphoric anhydride (P <sub>2</sub> O <sub>5</sub> )	9 soluble in neutral ammonium
	citrate and in water;
	6.8 soluble in water
Potassium oxide (K <sub>2</sub> O)	12 soluble in water
Magnesium oxide (MgO)	2.5 and 1.3 soluble in water
Bore (B)	0.02 soluble in water
Copper (Cu)	0.068 and 0.051 soluble in water
Total iron (Fe)	0.45
Iron soluble in water	0.31
Iron chelated by EDTA	0.22
Manganese (Mn)	0.06 and 0.03% soluble in water
Molybdenum (Mo)	0.025 soluble in water
Zinc (Zn)	0.02 and 0.013 soluble in water

#### Table 2

Proportions in nitrogen (N0<sub>3</sub>), phosphorus ( $P_2O_5$ ) and potassium ( $K_2O$ ) sources corresponding to the applied dose of fertilizer.

Dose of fertilizer Osmocote Exact (g/plant)	0	2	4	6
Quantity brought of nitrogen source (NO3:mg/plant)	0	300	600	900
Quantity brought of phosphorus source (P <sub>2</sub> O <sub>5</sub> :mg/plant)	0	180	360	540
Quantity brought of potassium source (K <sub>2</sub> O:mg/plant)	0	240	480	720

and perlite with variable proportions of 2/3 and 1/3, respectively. Application of this *inoculum* was carried out in terms of percentage of the pot's volume. Two doses have thus been prepared:

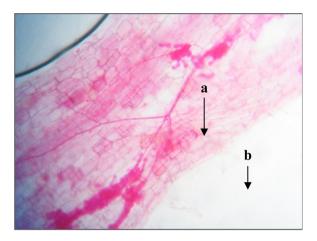
- dose 1: 10% inoculum + 90% substrate (2/3 peat + 1/3 sand);
- dose 2: 20% *inoculum* + 80% substrate (2/3 peat + 1/3 sand).

The second endomycorrhizal *inoculum* tested is a commercial product. It belongs to the brand MYKE PRO, which is produced by Premier Tech Itée, Rivière-du-Loup, QC, Canada. This product contains *G. irregulare* as an active ingredient, with a proportion of 1600 propagules/g, a density of 500 g/L and a granulometry of 0.5 mm. The *inoculum* bag was stored at  $4 \,^{\circ}$ C until use. Doses are expressed in grams, namely 0.06 g and 0.1 g, which were applied for each plant within the pot (Table 2).

#### 2.4. Evaluation of endomycorrhizal colonization

Root fragments are randomly selected at various levels of the underground parts of the different plants for each treatment. Thinning using KOH (10%) and oxidation of the organic matter, which is present on roots, was carried out firstly to eliminate intracellular components and leave only the root and fungal structures. These roots are colored by fushine acid (0.05% in lactoglycerol). Colored roots are preserved in lactoglycerol (25% lactic acid, 25% glycerol, 50% water) to dilute the unfixed colorant and prevent drying. The fushine acid is generally used to color selectively dead tissues or cells. It is, indeed, a colorant capable to be fixed on the chitin of all the fungal structures. A root is considered as endomycorrhized when it presents an endomycorrhizal structure (mycelium, vesicle, and arbuscule) [14]. The evaluation of the rate of roots mycorrhization by endomycorrhizal fungi is achieved through the method of McGonigle and Fitter [15]. For the quantification of endomycorrhizae, microscopic examination (40 to  $200 \times$ ) of 80 1-cm long root fragments was performed for each plant. The root fragments were randomly selected and mounted in parallel by groups of 10 between the slide and cover. Three readings per fragment were conducted to determine the type of the fungus structure.

Each experimental unit included three pots in which one plant was assessed. Since all the assessments were carried out within a green house, it was assumed that all



**Fig. 1.** Arbuscular endomycorrhizal structures observed on the roots of young plants of *Jacaranda mimosifolia* D.Don, after six months of composite indigenous *inoculum* intake ( $400 \times$  magnified images); a: hyphae; b: coils fungal filaments within cells of root fragment. Color online.

experimental units were homogeneous, and a completely randomized experimental design was used according to the following model:

$$y_{ij(k)} = \mu + I_i + F_j + (I \times F)_{ij} + \varepsilon_{ij(k)}$$

where  $y_{ij(k)}$  is the value of the response measured for the *i*th *inoculum* (*I*), the *j*th fertilizer (*F*), in the *k*th pot;  $\mu$  is the overall mean response,  $I_i$  is the effect of the *i*th *inoculum* (*I*),  $F_j$  is the effect of the *j*th fertilizer (*F*),  $(I \times F)_{ij}$  is the interaction effect of the *i*th *inoculum* (*I*) and the *j*th fertilizer (*F*),  $\varepsilon_{ij(k)}$  is the experimental error.

Experiments with both indigenous composite *inoculum* and commercial *inoculum G. irregulare* were performed simultaneously and under the same environmental conditions.

#### 3. Results

# 3.1. Mycorrhizal colonization of Jacaranda roots in the presence of five indigenous strains of Glomus genus present within the substrate

Microscopic observations of the colored roots of *Jacaranda* indicate that all plants having received a proportion of indigenous composite *inoculum* were mycorrhized, for all tested fertilization levels. Fungal structures found in the root tissues are essentially hyphae with fungal filaments bobbins inside certain cells (Fig. 1).

The study of the effect of the interaction between the fertilizer dose and the *inoculum* proportion contained in the pot on the *Jacaranda* root colonization rate (Table 3) shows that it is highly significant (P < 0.01). However, according to Fig. 2, which shows the root colonization rate of *Jacaranda* plants according to the brought doses of fertilizer and the proportions of composite indigenous *inoculums*, some heterogeneity is noted concerning the root colonization rate compared to the proportion of *inoculum* contained in pots. Thus, the recorded rates of colonization evolve independently of the brought

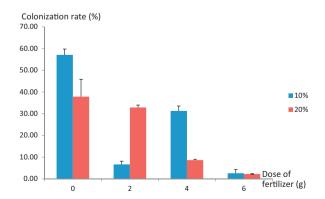
#### Table 3

Effect of variation in the brought proportion of composite indigenous *inoculum* and fertilizer dose on root colonization rates of *Jacaranda mimosifolia* D.Don.

Inoculum proportion contained in the total volume of the pot (percentage of the total volume of the pot)	Fertilizer dose (g)	Colonization rate (%)
10	0	57.10 <sup>a</sup>
20	0	37.85 <sup>b</sup>
20	2	32.88 <sup>b,c</sup>
10	4	31.3 <sup>c</sup>
20	4	$8.64^{d}$
10	2	6.66 <sup>d,e</sup>
10	6	2.59 <sup>e</sup>
20	6	2.22 <sup>e</sup>
F trait	**	
Variation coefficient	14.32	
R <sup>2</sup>	0.99	

Values followed by the same letter are not significantly different. <sup>\*\*</sup> Highly significant difference between treatments (P = 0.01).

proportions of this inoculum. In fact, regarding plants receiving doses of 0g and 4g of fertilizer, the most important endomycorrhizal colonization was observed on the roots of Jacaranda cultivated on substrates containing only 10% of the indigenous inoculum. On the one hand, the colonization rate of unfertilized plants is 57.1% for plants grown on a substrate containing 10% of the indigenous inoculum, while it did not exceed 37.85% for those grown on a substrate containing 20% of the inoculum. On the other hand, for fertilization with 4 g of Osmocote/plant, the colonization rates are 31.29% and 8.64%, respectively, for plants grown on substrates containing 10% and 20% of the indigenous inoculums. However, in case of a fertilization dose of 2 g/plant, the highest colonization rate was recorded with plants receiving the highest dose of inoculum. This rate was equal to 32.87% for an inoculum of 20%, while it was only 6.66% for a 10% inoculum. Finally, for the highest level of fertilization, namely 6 g of fertilizer/plant, the recorded colonization rates with both tested doses of inoculum are low (less than 3%). Therefore, the possibility of the



**Fig. 2.** Mycorrhizal colonization (%) based on the doses brought into fertilizer and *inoculum* indigenous composite proportions contained in pots; 10%, 20%: *inoculum* proportions expressed in terms of volume percentage of the pot. Color online.

#### Table 4

Effect of the fertilization level on root colonization rates of Jacaranda mimosifolia D.Don inoculated with two inoculums with different origins, natures, and intake modes.

Dose of fertilizer (g)	Composite indigenous inoculum		Commercial inoculum		
	Colonization rate (%)	Root dry mass (g)	Colonization rate (%)	Root dry mass (g)	
No fertilization	47.48 <sup>a</sup>	4.35 <sup>a</sup>	32.35 <sup>a</sup>	4.18 <sup>a</sup>	
2	19.77 <sup>b</sup>	2.72 <sup>c,b</sup>	15.82 <sup>b</sup>	2.14 <sup>b</sup>	
4	19.97 <sup>b</sup>	2.99 <sup>b</sup>	17.12 <sup>b</sup>	1.80 <sup>c</sup>	
6	2.41 <sup>c</sup>	2.66 <sup>c</sup>	3.95 <sup>c</sup>	2.32 <sup>b</sup>	
F trait	**	**	**	**	
Variation coefficient	14.33	8.19	44.73	9.28	
<i>R</i> <sup>2</sup>	0.99	0.96	0.78	0.97	

Values followed by the same letter are not significantly different.

\*\* Highly significant difference between treatments (P=0.01).

existence of an interaction effect between the dose of fertilizer and the proportion of indigenous composite *inoculum* on the level of mycorhization in *Jacaranda* was excluded.

The root colonization rate of *Jacaranda* plants varies significantly (Table 4) according to the level of fertilization (P < 0.01). Root dry biomasses of treatments vary in the same way as the rates of root colonization, depending on the levels of fertilization (P < 0.01).

Indeed, the highest and lowest root colonization rates and root dry biomasses are respectively observed in unfertilized and fertilized 6 g/plant treatments. With 2 g and 4 g of fertilizer per plant, the root colonization rates were not significantly different between treatments and root dry biomasses were very similar (Table 4). Generally, the rate of root colonization by indigenous endomycorrhizal complex decreases with increasing the fertilizer dose. Dry root biomasses of treatments vary depending on their levels of mycorhization.

# 3.2. Mycorrhizal root colonization of Jacaranda in the presence of a commercial inoculum G. irregulare in the substrate

Microscopic observations of *Jacaranda* colored roots treated with commercial *inoculum* show that at all levels of fertilization, all the plants are mycorrhized. Arbuscular endomycorrhizal structures observed on the roots are essentially in the form of hyphae with a limited presence of exchange (arbuscules) and reserve (vesicles) structures. Fig. 3 shows a hypha penetrating into a root cell and forming a ramification that constitutes the arbuscule.

Interaction effects between the doses of fertilizer and those of commercial *inoculum* on *Jacaranda* root colonization rate have proved insignificant in this study. Parameters "*inoculum* dose" and "level of fertilization" were then studied as having simple effects on the rate of mycorhization.

The rates of root colonization are evolving positively based on increasing brought doses of *inoculum* (Fig. 4). For all levels of fertilization, the most mycorrhized *Jacaranda* plants are those that received the highest dose of *inoculum*, i.e. 0.1 g/plant. The effect of the *inoculum* dose on the rate of root colonization of young *Jacaranda* plants (Table 5) was significant (P < 0.05). Providing a larger dose in commercial *inoculum*, namely 0.1 g/plant, an increase of 42.1% in

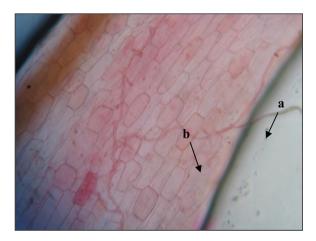
the root colonization rate of *Jacaranda* was observed compared to the situation with a 0.06 g/plant dose.

The rate of root colonization varies significantly based on the level of fertilization (P < 0.01). Thus, it appears that increasing the brought fertilizer dose decreases the level of mycorrhizal *Jacaranda* plants treated with commercial *inoculum* (Table 4). Indeed, the highest and lowest rates of root colonization are respectively observed in unfertilized samples and in those that received 6 g of fertilizer/plant. The rates root colonization treatments with doses of 2 g and 4 g/plant of fertilizer are not significantly different. Root dry biomasses do not evolve exactly as the level of mycorhization. However, non-fertilized but better mycorrhized plants have significantly higher root dry biomasses (P < 0.01) than the rest of the sample (Table 4).

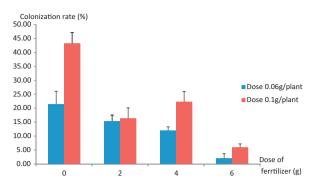
#### 4. Discussion

## 4.1. Quality of root colonization of young Jacaranda plants treated by different inoculums

According to the obtained results, the plants treated with composite indigenous *inoculum* reached a more advanced stage of mycorrhization than those treated with



**Fig. 3.** Observed structures on a root fragment of *Jacaranda mimosifolia* D.Don, six months after inoculation with a commercial *inoculum* containing *Glomus irregulare* ( $400 \times$  magnified image) structures; a: hypha entering a root cell; b: arbuscule. Color online.



**Fig. 4.** Colonization rates (%) according to the brought fertilizer and commercial *inoculum* doses. 0.06 g/plant, 0.1 g/plant: industrial *inoculum* doses. Color online.

the commercial inoculum. Indeed, microscopic observations of the latter showed only small colony dispersed on the mycorrhized roots instead of fungal structures extending over the entire lengths. These roots display rare arbuscules. In addition, the vesicles are almost nonexisting, indicating a recent colonization. On the contrary, the presence of fungal filaments coils on the roots of Jacaranda plants treated with composite indigenous inoculum reflects a more advanced stage of mycorrhization. In fact, these coils represent structures that appear in Paris-type colonization. The colonization of the root by hyphae of arbuscular mycorrhizal (AM) fungi can adopt two morphological types: the *Paris* and the *Arum* ones [16]. Colonization of Arum type has an extensive intercellular evolution of fungal hyphae in the root cortex, with the development of intracellular arbuscules, while colonization of Paris-type is defined by the absence of intercellular phase and the presence of numerous bobbins of intracellular hyphae. Arbuscules are structures that intercalate between the coils [16]. In case of such colonization, the arbuscules are relatively few, small, or even non-existing [16]. Nevertheless, this shows that colonization of Jacaranda plants treated with composite indigenous inoculum was well established.

## 4.2. Effect of the variation of the inoculum brought dose on the mycorrhizal level of young Jacaranda plants

The existence of an interaction effect between *inoculum* dose and the level of fertilization on the rate of

 Table 5

 Effect of the brought dose of commercial *inoculum* on root colonization rates of *Jacaranda mimosifolia* D.Don.

Inoculum dose (g)	Colonization rate (%)
0.06	12.69 <sup>b</sup>
0.1	21.92 <sup>a</sup>
F trait	•
Variation coefficient	44.73
$R^2$	0.78

Values followed by the same letter are not significantly different.  $^{*}$  Significant difference between treatments (*P* = 0.05).

mycorhization in *Jacaranda* was excluded. These two variation parameters were studied as simple effects.

The obtained results showed that the level of lacaranda's mycorhization is closely related to the variation of the brought dose of commercial inoculum. The highest rates of colonization were observed in plants treated with 0.1 g. These results corroborate with the other studies of the tested isolates of *Glomus* sp. on pineapple vitroplants. Their results showed that an increase in *inoculum* doses from 1% to 3% was often linked to a rise in the endomycorrhizal infection [11]. Contrary to Jacaranda plants treated with composite indigenous inoculum, the levels of mycorrhization have evolved independently of the variation in the proportion brought. These different results can be explained by the nature of the used inoculum. In fact, commercial inoculum is a homogeneous product, concentrated in propagules and adapted according to the application, while the composite indigenous inoculum is a product of trapping and multiplication with different mycotrophic plants. It is made of propagules of arbuscular endomycorrhizal fungi and mycorrhizal root fragments mixed in an inert substrate. The latter may present some heterogeneity in the concentrations of propagules and mycorrhized root fragments. The proportions applied to Jacaranda plants may not contain the same quantities of inoculants structures, which could explain the random changes in the levels of mycorhization after varying the proportions brought in this inoculum.

## 4.3. Effect of the level of fertilization variation on the mycorhization rate of young Jacaranda plants

The effect of varying the dose of fertilizer on the mycorhization level of young jacaranda plants has proved highly significant (P = 0.01) in both types of inoculum. In fact, increasing the brought dose of fertilizer reduced the infectivity of arbuscular endomycorrhizal fungi contained in the inoculum, thus, inducing a decrease in the root colonization rate of Jacaranda plants. These results corroborate those obtained with the studies on the interactions between extractible phosphorus soil level and colonization by arbuscular mycorrhizal fungi of white clover (Trifolium repens L.) [17]. The level of root colonization was negatively correlated with extractible phosphorus soil concentration. [17]. Studies on the effect of fertilization at different levels of phosphorus and nitrogen on mycorhization of Allium shoenoprasum with G. caledonium showed that the simultaneous application of high doses of phosphorus and nitrogen led to a decrease in the rate of root colonization. [18]. This effect is more apparent with ammonia's nitrogen than with its nitrate form [18].

Therefore, it is noticed that a too high level of mineral elements in soil inhibits the formation of mycorrhizae. In fact, increasing nitrogen and phosphorus concentration in the soil solution promotes the synthesis of proteins and of phosphorylated compounds (nucleic acids, DNA and RNA, inositol, phosphates, etc.), which causes a decrease in the soluble sugar content in the root. Since the soluble sugar content of the root is a key parameter for the nutrition of mycorrhizal fungi associated with the plant, it also determines the rate of formation of mycorrhizae [19]. According to these results, mycorhization has a positive effect on root dry biomass of *Jacaranda*, as in the case of unfertilized plants. Intake of different doses of fertilizers did not allow them to reach such a level of root system development. These results corroborate other studies on the role of arbuscular mycorhization in the improvement of the morphology and the development of host plant root system, e.g., those reporting the inoculation of "Annona cherimola Mill" with propagules of arbuscular mycorrhizae, which allowed a significant increase in the branching intensity of the first-order laterals [20].

#### 5. Conclusion

At all the levels of fertilization, plants of *Jacaranda* having received a dose of *inoculum* were mycorrhized. Six months after inoculation, the plants treated with the composite indigenous *inoculum* reached a more advanced stage of mycorhization than those treated with the commercial *inoculum*.

For *Jacaranda* plants inoculated with the commercial product, the variation in the level of mycorhization has proved to be closely related to the *inoculum* brought dose. Then, an improvement in the rate of root colonization by arbuscular endomycorrhizal fungi with an increase in the *inoculum* dose is observed. For plants treated with different proportions of composite indigenous *inoculum*, the level of mycorhization varies randomly. This can be explained by the existence of some heterogeneity of the concentrations of the propagules contained in the brought *inoculum* proportions.

For the two types of *inoculum*, a too high level of fertilization inhibited the infectivity of arbuscular endomycorrhizal fungi, then, inducing few mycorrhized *Jacaranda* plants. The best root colonization rates were observed in the unfertilized plants, which have the most important root dry biomasses.

#### **Disclosure of interest**

The authors declare that they have no conflicts of interest concerning this article.

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