**ORIGINAL ARTICLE / ORIGINALBEITRAG** 



# Effect of Phosphorus and Arbuscular Mycorrhizal Fungi (AMF) Inoculation on Growth and Productivity of Maize (*Zea mays* L.) in a Tropical Ferralsol

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### Abstract

Soil fertility in the Lubumbashi region often proves to be limiting factor for crop production due to their low nutrient reserves. The objective of this work was to evaluate the impact of arbuscular mycorrhizae on phosphorus uptake by maize on Ferralsol. The trial was conducted in pots with 30 kg or 60 kg of  $P_2O_5$  ha<sup>-1</sup> and a control. These three levels of phosphorus were combined or not with arbuscular mycorrhizae. The combinations of 30 kg or 60 kg of phosphorus ha<sup>-1</sup> formed very few or almost no blisters in the roots. Cob weight, length, diameter, number of rows and kernel weight varied significantly with phosphorus on both inoculated and uninoculated pots. The inoculated plants had high averages for these yield parameters. Due to the lack of phosphate fertilizer, inoculum alone can be an alternative to phosphorus provided that nitrogen and potassium are added, resulting in small but seed-filled ears compared to the phosphorus (0.3 to 6.1 tons ha<sup>-1</sup>) and less significantly with inoculum (3 to 3.7 t ha<sup>-1</sup>). The combination of treatments showed a significant difference in favour of the 60 kg of phosphorus or 60 kg of phosphorus associated with the inoculum. The highest phosphorus content was obtained on the inoculum treatment alone, which provided 1.4 mg phosphorus g<sup>-1</sup> maize compared to other treatments, which provided 0.69 to 0.71 mg phosphorus g<sup>-1</sup> maize.

**Keywords** Ferralsol · Arbuscular mycorrhizae · Phosphorus · Maize

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# Wirkung von Phosphor und arbuskulären Mykorrhizapilzen (AMF) auf Wachstum und Produktivität von Mais (*Zea mays* L.) in einem tropischen Ferralsol

#### Zusammenfassung

Die Bodenfruchtbarkeit in der Region Lubumbashi erweist sich aufgrund der geringen Nährstoffreserven oft als einschränkender Faktor für die Pflanzenproduktion. Ziel dieser Arbeit war es, die Auswirkungen von AMF auf die Phosphoraufnahme von Mais zu bewerten. Der Versuch wurde in Töpfen mit 30 kg oder 60 kg  $P_2O_5$  ha<sup>-1</sup> und einer Kontrolle durchgeführt. Diese drei Phosphormengen wurden mit oder ohne AMF kombiniert. Die Kombinationen von 30 kg oder 60 kg Phosphor mit dem Inokulum führten zu einer männlichen Blüte des Maises 63 Tage nach der Aussaat. Mais, der mit 60 kg Phosphor ha<sup>-1</sup> behandelt wurde, bildete sehr wenige oder fast keine Blasen in den Wurzeln. Das Kolbengewicht, die Länge, der Durchmesser, die Anzahl der Reihen und das Korngewicht variierten signifikant mit dem Phosphorgehalt sowohl in den beimpften als auch in den nicht beimpften Töpfen. Die beimpften Pflanzen wiesen hohe Durchschnittswerte für diese Ertragsparameter auf. Mangels Phosphatdünger kann Inokulum allein eine Alternative zu Phosphor sein, sofern Stickstoff und Kalium zugesetzt werden, was zu kleinen, aber samengefüllten Ähren führte, verglichen mit der phosphorfreien Kontrolle ohne Mykorrhiza, die zu leeren Ähren führte. Der Ertrag variierte signifikant mit der Zugabe von Phosphor (0,3 bis 6,1 t ha<sup>-1</sup>) und weniger signifikant mit dem Inokulum (3 bis 3,7 t ha<sup>-1</sup>). Die Kombination der Behandlungen zeigte einen signifikanten Unterschied zugunsten von 60 kg Phosphor oder 60 kg Phosphor in Verbindung mit dem Inokulum. Der höchste Phosphorgehalt wurde bei der Inokulation allein erzielt, die 1,4 mg Phosphorg<sup>-1</sup> Mais lieferte, verglichen mit anderen Behandlungen, die 0,69 bis 0,71 mg Phosphorg<sup>-1</sup> Mais lieferten.

Schlüsselwörter Ferralsol · Arbuskuläre Mykorrhizapilze · Phosphor · Mais

# Introduction

Phosphorus (P) is an essential nutrient which is important for plant growth and many physiological processes such as metabolic energy transfers and nucleic acid metabolism (Chang et al. 2008; Eppendorfer and Eggum 1994; Grant et al. 2014; Martin-Prével and Montagut 1966). In tropical soils, the P concentration in the soil solution generally ranges between 0.1 and 10µM (0.003 to 0.3 ppm) and is much lower than the adequate concentration for optimal growth of many crops (Liu et al. 2016). The orthophosphates, H<sub>2</sub>PO<sub>4</sub><sup>-</sup> and HPO<sub>4</sub><sup>2-</sup>, are the primary forms of phosphorus taken up by plants. However, these two forms are often in low concentration, mostly in highly weathered tropical soils. In general, the pH range of 6-6.5 is considered optimal for phosphorus uptake by the plant. In acidic soils (pH<5.5), aluminium cations (Al<sup>3+</sup>) are dominant in soil solution and they can immobilize P to form the poorly soluble AlPO<sub>4</sub> which can limit plant root growth. In basic soils, Ca2+ and Mg2+ dominate the cationic exchange capacity and the P immobilization becomes more important (Enita 2000; Oteino et al. 2015). The role of phosphorus (P) is yet crucial in plant production and it can be supplied as chemical fertilizers, but this is not generally possible for smallholder's farmers in tropics because of their low income. It is then useful to improve phosphorus (P) availability and one opportunity is provided by the inoculation of crops with arbuscular mycorrhizal fungi (AMF). The AMF make symbiosis with root plants and are able to secret phosphatase, which catalyze and release P for the benefit of plants (Grant et al. 2004; Tshibangu et al. 2020a). They also confer robustness to plants and they improve crop yields (Beaudin et al. 2008; Grant et al. 2014; Jan et al. 2014; Tshibangu et al. 2020b). AMF thus improves uptake of immobilized plant nutrients, especially P (Erdinc et al. 2017). The aim of this study was to assess the effects of AMF inoculum on P assimilation by maize and its impact on maize growth and productivity on an acidic and deficient in available phosphorus Ferralsols.

# Material and Methods

#### **Experimental Soil and Plant Material**

The experimental soil was collected on an old maize farm located in the Luano area, Lubumbashi, DR Congo  $(-11,60233^{\circ}S; 27,50897^{\circ}E \text{ to } -11,60234^{\circ}S; 27,50908^{\circ}E,$  altitude 1200m asl). It is a red and deep sandy clay loam soil whose available laboratory data are summarized in Table 1.

Spores were extracted from a 100 g sample of soil by using the standard wet sieving and decantation method ( $45 \,\mu m$  and 1 mm mesh) followed by counting using an Olympus SZ61 stereo microscope according to Walker et al. (2006). AMF morpho-species identification was done under microscope of 40 and 100 X on three to five spores of similar morphology, assembled on a strip in the polyvinyl lactoglycerol. Spore shapes, their colors and sizes, occurrence or not of pedicels, number of spore wall layers and wall surface ornamentation were main criteria used for identifi-

pH <sub>(water)</sub>	Total nitrogen (%)	Organic carbon (%)	Available phosphorus (µg/g soil)	ECC (cmol (+) kg <sup>-1</sup> soil)	Exchanges cations contents (cmol (+) kg <sup>-1</sup> soil)		
					Calcium	Potash	Magnesium
6.3	0.09	0.97	10.3	0.43	6.38	0.52	1.9

Table 1 Experime	ntal chemical se	oil properties
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ECC exchange cationic capacity, cmol centimol

cation (Oehl et al. 2011a; Trappe 1992). The mean density was of 150 spores  $100 \text{ g}^{-1}$  soil with predominance of genus belonging to *Gigaspora, Acaulospora* and *Archaeospora*. The maize variety "Katanga" was used as test crop in the experiment. This local variety blooms from 60 to 65 days after sowing and presents a maximal height of 191 cm depending on growing conditions. Grains are white and have a weight nearing  $168 \pm 25 \text{ g cob}^{-1}$ , i.e. 380 g per 1000 grains. Before sowing, the grains were disinfected with sodium hypochlorite (1%) during 5 min, then with the alcohol (75%) and rinsed with tap water afterwards.

#### **AMF Inoculum Production**

The inoculum was obtained from red Sorghum (Sorghum vulgare L.) cultivated in pots, to have high AMF spore density and root colonization; as described by Mukerji et al. (2002), and Selvaraj and Chellappan (2006). The pots had 28 cm diameter (0.4396 m<sup>2</sup>) and depth 28 cm. Sorghum was sown in a litter substrate at 25 to 30 plants per pot. At 3.5 months later, root and AMF spores from Sorghum crop were extracted and used as inoculum. The litter substrate was collected from Miombo woodland dominated by Brachystegia and Julbernardia. The choice of this substrate is justified by its high porosity. This is a reasonable method of AMF inoculum production contrary to the high cost of commercial inoculum and their scarcity in the Lubumbashi region. The inoculum produced had a spore mean density of 250. 100 g<sup>-1</sup> of substrate and 10 g of colonized roots at the frequency of 70 to 80%. Acaulospora and Ambispora were the most dominant genera.

#### **Chemical Fertilizers**

Di-ammonium phosphate (DAP—18-46-0) and potassium chloride (KCl—60% K<sub>2</sub>O) were used as basal fertilizers and urea (46% N) was added as top fertilizer to cover the N crop demand. The K and N were maintained constant while P was added as DAP in 3 doses:  $60 \text{ kg } P_2 O_5 \text{ ha}^{-1}$ , which is the normal dose, recommended for maize in tropical conditions (Raemaekers 2001),  $30 \text{ kg } P_2 O_5 \text{ ha}^{-1}$  as the half of recommended dose and  $0 \text{ kg } P_2 O_5 \text{ ha}^{-1}$  as the control.

# **Experimental Design**

The experiment was conducted in pots of 28 cm diameter and 28 cm depth in a factorial design  $2 \times 3$  with 5 replications. Two levels of AMF inoculation and 3 levels of phosphorus application were used as treatments:

- Control: unfertilized and un-inoculated
- 30 kg of P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup>, without AMF inoculum
- 60 kg of P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup>, without AMF inoculum
- 100 g AMF inoculum
- 30 kg of P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup>, 100 g AMF inoculum
- 60 kg of P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup>, 100 g AMF inoculum.

The maize plants were watered during the 3 first months (September to November 2017), every 2 days with 1 liter of tap water per pot having 1 plant each. The experiment took place in a shade house from September 2017 to January 2018. The last two months (December 2017 and January 2018) of experiment were rainy. During the experimental period, the prevailing temperature varied between 15 to 32 °C and a mean relative humidity of 79% and 12h sunshine.

#### **Measurements and Data Analysis**

The plant height, the height at the insertion of panicle, days to the flowering, the number of leaves, the leaves chlorophyll content, the cob length, the cob diameter, the cob rows number, the seed weight per cob, the yield and the total P assimilated in grains were measured. The data were statistically evaluated by factorial analysis of variance with mycorrhizal inoculation and P application levels as factors. Means and standard errors were calculated for five replicate value. Means were compared by ANOVA and the Tukey-HSD test at significance level P < 0.05. The Minitab 16 software was used for the statistical analysis.

#### Results

# Vegetative Growth Parameters and Root Colonization by AMF

The effect of inoculation and P application on maize was significant for the number of days before flowering. Plants

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Treatments	Days	Final plant	60 days after sowing			
	before flowering	height (cm)	Number of leaves plant <sup>-1</sup>	Leaves chlorophyll content (SPAD units)	Root colonization frequency (%)	
Mycorrhizal inoculum (AMF)						
Without AMF	73±14 a	149±43 a	10±2 a	41 ± 10 a	$40 \pm 20 \text{ b}$	
With AMF	70±13 a	158±44 a	10±2 a	43±8 a	57±25 a	
Phosphorus (kg P <sub>2</sub> O <sub>5</sub> )						
$0 \text{ kg P}_2 \text{O}_5 \text{ ha}^{-1}$	91±3 A	102±15 C	7±1B	31±5 B	$49 \pm 27 \text{ AB}$	
30 kg P <sub>2</sub> O <sub>5</sub> ha <sup>-1</sup>	$63 \pm 1 B$	$169 \pm 22 \text{ B}$	11±1 A	$47 \pm 4$ A	31±18 B	
$60  \text{kg}  \text{P}_2 \text{O}_5  \text{ha}^{-1}$	61 ± 2 B	$191 \pm 21 \text{ A}$	11±1 A	$47 \pm 4$ A	65±11 A	
AMF  imes Phosphorus						
Without AMF 0 kg P <sub>2</sub> O <sub>5</sub> ha <sup>-1</sup>	93±0 a	$105 \pm 3 \text{ b}$	7±0b	$28 \pm 3 b$	25±11 b	
Without AMF30 kg P <sub>2</sub> O <sub>5</sub> ha <sup>-1</sup>	$63 \pm 0$ b	171±27 a	11±1 a	46±3 a	32±13 b	
Without AMF $60 \text{ kg } P_2O_5 \text{ ha}^{-1}$	$63\pm0$ b	197±19 a	12±1 a	46±4 a	63±8 a	
With AMF 0 kg P <sub>2</sub> O <sub>5</sub> ha <sup>-1</sup>	88±0 a	98±21 b	7±1b	34±5 b	73±8 a	
With AMF $30 \text{ kg P}_2\text{O}_5 \text{ ha}^{-1}$	$62\pm0$ b	166±20 a	12±1 a	47±5 a	$30 \pm 23$ b	
With AMF $60 \text{ kg } P_2O_5 \text{ ha}^{-1}$	$59\pm0$ b	184±23 a	11±1 a	47±5 a	68±13 a	

**Table 2** Growth parameters and root colonization frequency (%) by AMF. SPAD units: soil plant analysis development. The means that do notshare any letters in a group in each column are significantly different after ANOVA and the Tukey multiple comparison test (P < 0.05)

**Table 3** AMF structures in root. AMF structures are present on 25% of root samples (+); mycorrhizae structures are present on 50% of root samples (++); mycorrhizae structures are present on 100% of root samples (++++); mycorrhizae structures are not present (–)

Treatments	AMF structures			
	Hyphae	Vesicles	Arbuscles	
Without AMF 0 kg P2O5 ha-1	++	-	_	
Without AMF30kg P2O5 ha-1	++++	-	-	
Without AMF 60 kg P2O5 ha-1	++++	-	-	
With AMF 0 kg P2O5 ha-1	++++	+	+	
With AMF 30 kg P2O5 ha <sup>-1</sup>	++++	+	-	
With AMF $60 \text{ kg } P_2O_5 \text{ ha}^{-1}$	++++	-	-	

that were inoculated with AMF flowered earlier than noninoculated plants (Table 2). The combination of inoculation with P application increased maize flowering precocity while control plants were the latest to flowering. The effect of inoculation was not significant for plant height, number of leaves and chlorophyll content while, P application significantly increased the number of leaves per plant and leaves chlorophyll content. The effect of AMF inoculation and P application was significant on maize root colonization. Root colonization was more important on inoculated plants than on plants that were not inoculated. For non-inoculated plants, increasing P application proportionally increased their ability to be colonized by soil indigenous mycorrhiza. However, for inoculated plants, the maize which did not receive any P showed the highest rate of root colonization, followed by those that received 60 kg P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup> and the one that received  $30 \text{ kg } P_2O_5 \text{ ha}^{-1}$  had the lower rate of colonization.

At 60 days after sowing, the most common mycorrhizae structures on all treatments were intra or extra-radical hy-

phae. Vesicles were found only on inoculated plants on which there was no P application and those that received  $30 \text{ kg } P_2O_5 \text{ ha}^{-1}$ . In the same way, arbuscles were found only on plants that were inoculated and did not receive any P application (Table 3).

# **Yield Parameters**

Yield results (Table 4) shows that the AMF treatments had a significant effect on yield parameters, except cob height insertion and cob length, which varied more with P treatments than with AMF inoculum. The height at the cob insertion, cob weight, cob length, cob diameter and the number of seed rows varied more with P than with AMF inoculum applied. Obtained averages under P influence were double or threefold higher than the control and AMF treatment alone. Cobs were then inserted between 73 and 103 cm height for 30 or 60kg P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup>, whereas without P cob insertion was observed between 30 cm (for the control) and 40 cm (for inocula alone). The highest cob weight was obtained on treatments with 60 kg P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup> with or without inocula, i.e. 145 to 146 g. The length of these cobs measured on average 15 cm. A fertilization with  $30 \text{ kg } P_2 O_5 \text{ ha}^{-1}$  gave cobs weighing 79 to 107 g with a length of 12 to 13 cm. The lowest cob weight was obtained on inocula alone and the control, respectively 4 to 13 g, and a cob length of 6 to 7 cm. The cob diameter was the same for 30 kg and 60 kg P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup>, being 42 to 45 mm. The AMF only treatment had 24 mm of cob diameter, double compared to the control. Cobs from the 30 and 60 kg P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup> treatments without inoculum contained 13 seed rows, whereas they were empty for the control treatment. AMF combined to 60 kg P ha<sup>-1</sup> had 13 seed rows followed by AMF combined with 30 kg P<sub>2</sub>O<sub>5</sub>

ANOVA and the Tukey multiple comparison test ( $P < 0.05$ )								
	Cob inser- tion height (cm)	Cob weight (g)	Cob length (cm)	Cob di- ameter (mm)	Seed raw number	Grain weight (g)	Yield of grains tons/hectare	Grain phosphorus content (mg of phosphorus g <sup>-1</sup> )
	$70 \pm 35a$	$76 \pm 82a$	$11 \pm 4a$	$33 \pm 15a$	$13 \pm 1a$	$86 \pm 33a$	$3.0 \pm 2.6a$	$0.70 \pm 0a$
	$67 \pm 27a$	$89 \pm 60a$	$11 \pm 4a$	36±11a	$10 \pm 3b$	$71 \pm 48a$	$3.7 \pm 2.5a$	$0.97 \pm 0a$
	$35 \pm 13C$	9±6C	6±1C	$18 \pm 9B$	$7\pm0B$	$12 \pm 1C$	$0.3 \pm 0.3$ C	$0.71 \pm 0A$
	76±19B	$93 \pm 23B$	$12 \pm 2B$	$41 \pm 3A$	$12 \pm 3A$	$71 \pm 21B$	$3.7 \pm 1.1B$	$0.72 \pm 0A$

 $13 \pm 1A$ 

 $13 \pm 1a$ 

 $13 \pm 1a$ 

 $7 \pm 0b$ 

 $11 \pm 4a$ 

 $13 \pm 1a$ 

Table 4 Yield parameters and phosphorus content in maize grains. The means that do not share any letters in a group in each column are significantly different after ANOVA and the Tukey multiple comparison test (P < 0.05)

 $15 \pm 3A$ 

 $6 \pm 1b$ 

 $13 \pm 2a$ 

 $15 \pm 2a$ 

 $7 \pm 1b$ 

 $12 \pm 1a$ 

 $15 \pm 3a$ 

 $45 \pm 2A$ 

 $12 \pm 4c$ 

 $42 \pm 2a$ 

 $44 \pm 2a$ 

 $24 \pm 8b$ 

 $40 \pm 4a$ 

 $45 \pm 3a$ 

ha<sup>-1</sup> with 11 seed rows, cobs from the AMF only treatment had 7 rows. The comparison of the treatments 30kg P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup> with AMF and 30 kg P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup> without AMF considering grain weight, grain yield and grain phosphorus content parameters shows better the influence of AMF on P uptake. The addition of 30 kg P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup> produced 59 g of grains per cob, the AMF inoculation offer 12g of grains per cob, whereas the control treatment had cobs without grains. The results found indicate that the AMF treatments had a highly significant effect on P uptake. The P treatments and their interaction with AMF also had a strong effect. Grains from inoculated plants had a double content of phosphorus in comparison with un-inoculated plants. The highest phosphorus content in maize grains was 1.41 mg phosphorus g<sup>-1</sup> obtained on AMF inoculation alone followed by inoculum +60 kg of  $P_2O_5$  ha<sup>-1</sup> and inoculum +30 kg of  $P_2O_5$  ha<sup>-1</sup>.

 $96 \pm 18A$ 

 $30 \pm 3b$ 

 $78 \pm 26a$ 

 $40 \pm 17b$ 

 $73 \pm 12a$ 

 $89 \pm 23a$ 

 $103 \pm 12a$ 

 $146 \pm 22A$ 

 $4 \pm 2c$ 

 $79 \pm 22b$ 

 $145 \pm 19a$ 

 $107 \pm 16b$ 

 $146 \pm 26a$ 

 $13 \pm 6c$ 

# Discussion

Treatments

*Mycorrhizal inoculum* Without AMF With AMF

Phosphorus (kg P<sub>2</sub>O<sub>5</sub>) 0kg P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup> 30kg P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup> 60kg P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup>

AMF × Phosphorus

Without AMF 0kg P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup>

Without AMF30 kg P2O5 ha-1

Without AMF 60 kg P2O5 ha-1

With AMF 0 kg P2O5 ha-1

With AMF 30 kg P2O5 ha-1

With AMF 60 kg P2O5 ha-1

# **Vegetative Growth Parameters**

Plaxton and Tran (2011) and Stigter and Plaxton (2015) have shown the phosphorus implication in plant physiology already which lead to the amine acid and protein constitution; and its deficiency in the soil can reduce severely the grain's production. AMF contribution to maize growth was strongly observed on male precocity on the plants inoculated and fertilized (59 and 62 days after sowing) in comparison to the control (93 days after sowing). Inoculating crops conferred an advance of 30 days duration at flowering, which is explained by P acquiring which is the first benefit of the symbiosis. Oehl et al. (2011b) demon-

strated that for plants inoculated with AMF and fertilized, flowering occurred earlier compared to those that were not inoculated. The same average was obtained for plant height, leaves number and chlorophyll rate in leaves especially on treatments inoculated and fertilized with 30 or 60 kg  $P_2O_5$ ha<sup>-1</sup>. This joins the allegations of Davies and Linderman (1990) and Aguegue et al. (2017) showing that the only micro-doses of chemical fertilizers (half of the crop demand) are sufficient to promote plant growth when AMF have been applied. Munir et al. (2003) also showed that the addition of phosphorus decreases the mycorrhizal symbiosis because the plant's growth depends on the chemical fertilizer.

 $116 \pm 17A \quad 6.1 \pm 0.9A$ 

 $3.1 \pm 1.0b$ 

 $6.0 \pm 0.7a$ 

 $0.6 \pm 0.0c$ 

 $4.4 \pm 0.8b$ 

 $6.2 \pm 1.0a$ 

 $59 \pm 20b$ 

 $12 \pm 1c$ 

 $83 \pm 15b$ 

 $118 \pm 20a$ 

 $114 \pm 15a$ 

#### **Phosphorus Fertilizer Effect on Root Colonization**

Root colonization was very intense (73%) on inoculated plants without P addition and less intense (25%) on control plants, the difference being 48%. The inoculated plants were more colonized (57%) than un-inoculated plants, which had a mean of 40% root colonization rate. This root colonization rate can be explained by AMF previously existing in the experimental soil. As shown in the methodology, the experimental soil harboured 150 mycorrhizae spores per 100g soil, a spore density sufficient to colonize the roots of a given crop. This is sufficiently proving the ubiquist character of AMF in soils as underlined by Liu et al. (2016), Cozzolino et al. (2013) and Ambrosini et al. (2015). Although most soils contain AMF, inoculation is still very important because it provides new AMF propagules capable of colonizing the newly established crop as quickly as possible. While assessing the root colonization, fungal hyphae as primary structure of symbiosis

 $0.73 \pm 0A$ 

 $0.69 \pm 0b$ 

 $0.71 \pm 0b$ 

 $1.42 \pm 0a$ 

 $0.74 \pm 1b$ 

 $0.75 \pm 0b$ 

were present in all treatments. However, vesicles were only found in treatments with inoculum alone or combined to the small dose of  $30 \text{ kg } P_2O_5 \text{ ha}^{-1}$ . The dose of  $60 \text{ kg } P_2O_5 \text{ ha}^{-1}$ seemed to compromise the vesicle formation on roots. The highest dose of  $P_2O_5$  ( $60 \text{ kg } \text{ ha}^{-1}$ ) reduces fungi activity in the root and compromises arbuscles or vesicles formation, as was also observed by Braunberger et al. (1991).

#### **Yield Parameters and Grains Phosphorus Content**

The height at the cob insertion, the weight of cobs, their length and diameter were not directly influenced by AMF inoculum but rather by phosphorus applications. Fertilization with  $30 \text{ kg P}_2 \text{O}_5 \text{ ha}^{-1}$  seemed to be sufficient to improve the nutritional status of the Ferralsol used as substrate for this experiment, compared to the dose of  $60 \text{ kg } P_2 O_5 \text{ ha}^{-1}$ which is low cost effectiveness. These results are nearing those found by Aguegue et al. (2017) in Ferralsol in southern Benin, indicating that Glomus cubens species combined to a half dose of chemical fertilizers of NPK (7.5 P<sub>2</sub>O<sub>5</sub> kg ha<sup>-1</sup>) allow to get an equivalent production of maize to that obtained using the recommended dose of NPK (15 P2O5 kg ha<sup>-1</sup>) alone. The AMF inoculum alone given cobs well filled with grains, thus much better than plants of the control (without phosphorus neither AMF inoculum) given cobs without grains. The AMF showed P efficiency in the experimental soil; this induced the fruition. Several researchers such as Pandey et al. (2005) and Huang et al. (2017) have demonstrated the mycorrhiza efficiency in plant production. Grain P concentration increased with both inoculation and P fertilization. The highest P concentration was obtained with the treatment of inocula alone. This proves that mycorrhizae can be used as biofertilizers as confirmed by Miransari (2011) and Zhang et al. (2017), and that these fungi enhance chemical P uptake through fungal hyphae. Results of this study showed that the grain P concentration was 2 to 3 times lower than the standard P concentration (210 mg 100 g<sup>-1</sup>) contained in maize grains (Messiaen and Fakorede 2006). This gap could be explained by root concentration in the pot experiment culture that uptakes only mineral fertilizers than field experiments which benefit also the soil minerals.

# Conclusions

ing duration short, but also allowing to obtain sturdy plants and a production of cobs with a considerable length and diameter. However, economically, it would be recommendable to use the dose of  $30 \text{ kg } P_2 O_5 \text{ ha}^{-1}$ . AMF used alone appeared to be an alternative to the lack of P in poor soils, provided that N and K were preliminary added to the soil.

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**Conflict of interest** A. Tshibangu Kazadi, J. Lwalaba Wa Lwalaba, B. Kirika Ansey, J. Mavungu Muzulukwau, G. Manda Katabe, M. Iband Karul, G. Baert, G. Haesaert and R.-P. Mukobo Mundende declare that they have no competing interests.

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