



# ARBUSCULAR MYCORRHIZAL FUNGI INFLUENCE ON GROWTH OF CREOLE MAIZE AND LARVAL DEVELOPMENT OF *Spodoptera frugiperda* (Lepidoptera:Noctuidae) †

## [LOS HONGOS MICORRIZICOS ARBUSCULARES INFLUYEN EN EL CRECIMIENTO DE MAIZ CRIOLLO Y EN EL DESARROLLO DE LAS LARVAS DE *Spodoptera frugiperda* (Lepidoptera:Noctuidae)]

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### SUMMARY

**Background:** The fall armyworm *Spodoptera frugiperda* is one of the main pests of maize in Mexico. Its control and management have been performed mainly with pesticides. One of the alternatives is to incorporate arbuscular mycorrhizal fungi (AMF) because of the benefits they provide to plants improving their growth and promoting their defense system against this pest. **Objective:** Evaluate the effect of AMF in creole maize growth in reducing leaf damage and development of *Spodoptera frugiperda* larvae. **Methodology:** In greenhouse conditions, creole maize plants were inoculated with 80 spores of *Funneliformis mosseae*, *Rhizophagus intraradices* and without AMF. Height and stem diameter were determined in mycorrhized plants at 15, 30, and 45 days after plant inoculation. After 45 days, plants were infested with three third-instar larvae of *S. frugiperda* and left to feed for 12 days; fresh weight and cephalic capsule width and size were measured. Once larvae were removed, leaf damage was determined in plants by means of a visual scale, leaf area and mycorrhizal colonization. **Results:** Root colonization of maize plants by AMF had a significant effect (Tukey's  $p \leq 0.05$ ) in creole maize growth expressed in plant height at 30 days and at 45 days in stem diameter only in plants inoculated with *F. mosseae* (Tukey's  $p \leq 0.05$ ). Leaf damage by the fall armyworm was similar between inoculated and uninoculated with AMF. Larvae that consumed plant leaves inoculated with *R. intraradices* showed greater fresh weight compared to those inoculated with *F. mosseae*. Moreover, width of the cephalic capsule and size were similar between larvae fed with plants inoculated with and without AMF. **Implications:** The results provide new perspectives and considerations to incorporate AMF in management of fall armyworm in creole maize. **Conclusion:** These results show that AMF partially promote plant growth of creole maize; leaf damage is similar between plants with and without AMF. Insect weight increased depending on AMF species, which influenced their development.

**Keywords:** maize; insect development; arbuscular mycorrhizal fungi; plant growth; herbivory.

### RESUMEN

**Antecedentes:** El gusano cogollero *Spodoptera frugiperda* es una de las principales plagas del maíz en México. Su control y manejo se realiza principalmente con insecticidas y una de las alternativas es incorporar a los hongos micorrízicos arbusculares (HMA) en el manejo del gusano cogollero por los beneficios que proporcionan a las plantas mejorando su crecimiento y promoción del sistema de defensa contra insectos plaga. **Objetivo:** Evaluar el efecto de los hongos micorrízicos arbusculares en el crecimiento de plantas de maíz criollo, en la reducción del daño foliar y el

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desarrollo de larvas de *Spodoptera frugiperda*. **Metodología:** En invernadero, plantas de maíz criollo fueron inoculadas con 80 esporas de *Funneliformis mosseae*, *Rhizophagus intraradices* y sin HMA. A las plantas micorrizadas se les determinó la altura y diámetro del tallo a los 15, 30 y 45 días después de inoculadas las plantas. Después de 45 días, las plantas se infestaron con tres larvas del tercer instar de *S. frugiperda* y se dejaron alimentar por 12 días, a las larvas se les determinó el peso fresco, el ancho de la cápsula cefálica y el tamaño. Una vez retiradas las larvas, a las plantas se les determinó el daño foliar mediante una escala visual de daño, el área foliar y el porcentaje de colonización micorrízica. **Resultados:** La colonización de las raíces de las plantas de maíz por los HMA tuvo un efecto significativo (Tukey,  $p \leq 0.05$ ) en el crecimiento de maíz criollo expresado en altura de planta a los 30 días y a los 45 días en el diámetro del tallo únicamente en plantas inoculadas con *F. mosseae*. El área foliar fue significativamente mayor en plantas inoculadas con *R. intraradices* que con *F. mosseae* y sin HMA (Tukey,  $p \leq 0.05$ ). El daño foliar por el gusano cogollero fue similar entre plantas inoculadas y no inoculadas con HMA. Las larvas que consumieron follaje de plantas inoculadas con *R. intraradices* presentaron el mayor peso fresco con respecto a las inoculadas con *F. mosseae*. Además, el ancho de la cápsula cefálica y el tamaño fue similar entre las larvas alimentadas con plantas inoculadas con HMA y sin HMA. **Implicaciones:** Los resultados proveen nuevas perspectivas y consideraciones para incorporar a los HMA en el manejo del gusano cogollero en maíces criollos. **Conclusiones:** Estos resultados muestran que los HMA promueven parcialmente el crecimiento vegetal del maíz criollo, el daño foliar es similar entre plantas con y sin HMA. El peso de los insectos se incrementa dependiendo de la especie de HMA, lo que influye en su desarrollo. **Palabras clave:** maíz; desarrollo de los insectos; hongo micorrízico arbuscular; crecimiento de las plantas; herbivoría.

## INTRODUCTION

Maize is one of the most important crops in Mexico with a sown surface of approximately 7.4 million ha (SIAP, 2019). The greatest part of sown surface is temporal in small production units where creole and native maize and few agricultural materials are used (Turren *et al.*, 2012). In the main production zones, pests are within the factors that limit maize production, such as the fall armyworm (FAW) *Spodoptera frugiperda*, which without any control measure may cause from 45% up to 100% in maize crop yield reduction (Gutiérrez-Moreno *et al.*, 2019). The main control tool of this insect has been the use of pesticides, which according to Mota-Sánchez and Wise (2020), FAW has developed resistance to the majority of the pesticides used in Latin America.

Integrated Pest Management (IPM) is a strategy that includes several control methods to reduce pest damage and decrease the number of pesticides. In the case of maize, natural enemies, resistant plants, biorational pesticides and those with low impact on natural enemies have been used to reduce FAW populations (Blanco *et al.*, 2014). Additionally, beneficial microorganisms have been proposed, such as arbuscular mycorrhizal fungi (AMF) in IPM because of the benefits provided to plants by improving growth and defense against diverse pests, and reducing -as far as possible- ingredients, such as chemical fertilizers and pesticides (Vannette and Hunter, 2009; Gianinazii *et al.*, 2010; Borowicz, 2013). For example, Yan *et al.* (2021) found that AMF increase pesticide accumulation in maize plants and improve control efficacy against *S. frugiperda*. Moreover, AMF may not only positively influence plant growth traits but also provide defenses against FAW by selectively attracting beneficial insects and modulate interaction between FAW and its pathogens

(Kaur *et al.*, 2020; García-Gomez *et al.*, 2021). Thus, AMF could be applied as a component in integrated crop management systems.

Arbuscular mycorrhizal fungi colonize the great majority of plants that exist in natural and agricultural ecosystems (Wang and Qiu, 2006). Plant symbiosis with AMF increases their yield and improves mainly phosphorus acquisition and other nutrients as nitrogen, which translates into a positive effect on plant growth and reproduction (Smith and Read, 2008). Furthermore, plant tolerance to abiotic stress is improved (Aroca *et al.*, 2007), which may modify plant defense against herbivores (Gange and West, 1994; Bennett *et al.*, 2006; Jung *et al.*, 2012). Several studies have reported AMF influence on plant defense expression and herbivory, suggesting some tendencies. For example, plants inoculated with AMF frequently benefit aphids and specialist defoliators and generalist insects, such as caterpillars and beetles are usually adversely affected by the presence of mycorrhizae (Hartley and Gange, 2009). Insect survival, weight, amount of food they consume, and development time are some of the physiology and life history features of generalist insects that plants inoculated with AMF may affect negatively (Roger *et al.*, 2013; Shrivastava *et al.*, 2015; Rivero *et al.*, 2021).

The fall armyworm is a generalist herbivore that may be potentially affected by symbiosis between AMF and agricultural crops as maize (Rabin and Pacovsky, 1985; Kaur *et al.*, 2020). According to Koricheva *et al.* (2009), the effect on generalist herbivores depends on the AMF species and type of plant, which may jointly empower defense against insects. Thus, the hypothesis is that when AMF is inoculated in creole maize plants, generalist insects, as *S. frugiperda* have lower development. Moreover, Rosenthal and Dirzo (1997) found that creole maize used in the majority of the

production zones shows natural resistance against herbivores. This characteristic allows empowering defense against herbivores when they are inoculated with AMF, which also contributes to FAW integrated pest management by reducing damage to maize plants. Additionally, native and creole maize have also been observed to respond positively to AMF inoculation under conditions of low or zero chemical inputs (Gavito and Varela, 1995; Sangabriel-Conde *et al.*, 2014). Studying the interaction between mycorrhizae, plants, and insects in the field is extremely challenging because of the difficulty in manipulating AMF populations in large-scale. Greenhouse experiments allow us to control most of the factors involved in the interactions and to observe the maximum effect of the mycorrhizae on herbivores. Therefore, the objective of this study was to assess mycorrhization effect with two AMF species in creole maize growth, leaf damage and larval development of *Spodoptera frugiperda* fed mycorrhized creole maize in greenhouse conditions.

## MATERIALS AND METHODS

### Plant material

Seeds from creole white grain maize -from the Municipality of Epitacio Huerta, Michoacán, México- were used. These seeds were sanitized with sodium hypochlorite at 5% for five minutes and washed thrice with sterile distilled water. Subsequently, seeds germinated in interfolded white paper towels and then incubated in darkness at 25°C for six days until the coleoptile had been formed.

### Mycorrhizal inoculant

The AMF species used were *Funneliformis mosseae* strain QR-F1 and *Rhizophagus intraradices* strain QR-R1 obtained from the AMF collection of Centro de Investigación y Asistencia en Tecnología y Diseño del Estado de Jalisco (CIATEJ), Unidad Zapopan. These AMF species were propagated previously in pots using sorghum as trap plant in greenhouse conditions for six months.

### Arbuscular mycorrhizal fungus inoculation and experimental design

After the coleoptile was formed, three-day old maize plants were transplanted into black polyethylene pots (diameter 12 cm, height 24 cm) filled with 2.5 kg of autoclaved substrate (121 °C, 1.0546 kg cm<sup>-2</sup>, 6 h). The substrate was made up of a mixture of soil, sand, and agrolite (ratio 6:3:1 v/v). At the same time as transplanting, 80 spores of *F. mosseae* and *R. intraradices* were directly added to the roots, and 10 g of sterile sand was added to plant roots without AMF. Three treatments (*F. mosseae*, *R. intraradices* and one without AMF) were generated. These treatments were

established in complete randomized design in greenhouse conditions. The experimental unit consisted of one maize plant placed in a pot. Ten replicates were performed per treatment. The inoculated plants remained in the greenhouse for 57 days at an average temperature of 22 °C and were irrigated every other day with tap water. Starting from the third week after inoculation, 0.065 g of nitrogen were applied weekly in the form of urea per plant.

### Insects

Larvae of *S. frugiperda* was established in the insectarium located in CIATEJ Unidad Zapopan, Jalisco, México, starting from a stock provided by the Entomology Laboratory of Instituto de Investigaciones Agropecuarias y Forestales (IIAF) from Universidad Michoacana de San Nicolás de Hidalgo, Michoacán, México (19°46'06.89" N, 101°09'01.37" W). To obtain sufficient larvae, 15 females and 15 males were placed in paper bags and fed with honey at 15%. Each 24 h, the bags with eggs -deposited by the females- were replaced. Insects were maintained in a growth chamber at 25 °C, 65% of relative humidity and photoperiod of 16:8 h (light:darkness). The eggs were placed in plastic 0.5 L containers until larvae emerged. Larvae were fed with the artificial diet based on wheat germ (3.21 g), yeast extract (3.43 g), cornflour (12.84 g), ascorbic acid (0.45 g), Methyl P-hydroxybenzoate (0.11 g), Benzoic acid (0.13 g), bacteriological agar (1.83 g), Formalin 40 % (0.05 mL) and Distilled water (78 mL) proposed by Poitout and Bues (1974) until the third instar stage. Third-instar larvae were used for the feeding assays on maize plants with and without (w/o) AMF.

### Insect feeding assay

At 45 days after inoculating plants with AMF, all three treatments were infested with three third-instar *S. frugiperda* larvae per plant. Larvae were placed on plant leaves. Previously, the insects were starved for five hours to promote immediate herbivory. After placing larvae on the plants, they were covered with a white transparent cloth to avoid their movement to other plants. For each one of the treatments with and w/o AMF, ten plants were infested. Plants with insects were maintained in greenhouse conditions with average temperature of 22 °C for 12 days.

### Plant growth, leaf damage by the fall armyworm and microbiological variables

Starting from 15 days up to 45 days after inoculation with AMF, plant height was measured with a flex-o-meter (Truper®, Mexico) and measurements of stem diameter were made using a digital caliper (Neiko®, USA). The leaf area was recorded after removing insects at 57 days that the end of the experiment.

Briefly, the leaves of each one of the plants were cut off from the stem and the images of leaves were obtained using a digital camera (CANON®, México). The images of leaves were captured with a ruler reference and leaf area was estimated from digital photographs with ImageJ software.

Leaf damage by FWA was visually evaluated in plants with or w/o AMF after removing insects. All plants were rated for leaf feeding injury on a 0-9 scale according with Davis *et al.* (1992), where 0 = no visible injury, 1= only pinholes on whorl leaves, 2= pinholes and small circular lesions on whorl leaves, 3= small circular lesions and a few small elongated lesions of up to 1.3 cm in length present on whorl and furl leaves, 4= several small to mid-size 1.3 to 2.5 cm in length elongated lesions present on a few whorl and furl leaves, 5= several large elongated lesions greater than 2.5 cm in length present on whorl and /or furl leaves, 6= several large elongated lesions present on several whorl and furl leaves and/or several large uniform to irregular shaped holes eaten from furl and whorl leaves, 7= many elongated lesions of all size present on several whorl and furl leaves plus several large uniform to irregular shaped holes eaten from the whorl and furl leaves, 8= many elongated lesions of all sizes present on most whorl and furl leaves plus many mid to large sized uniform to irregular shaped holes eaten from the whorl and furl leaves and 9 = whorl and furl leaves almost totally destroyed.

On the other hand, mycorrhizal colonization percentage of maize plant roots was determined as variable after removing insects at 57 days. For this purpose, root samples were taken from each pot, washed thoroughly to remove debris and placed into plastic histology cassettes. The roots were cleared in 10% KOH, acidified in 0.1 N HCl and stained in 0.03% trypan blue in lactoglycerol (Phillips and Hayman, 1970). Roots from each sample were mounted with lactoglycerol, covered with a cover slide and view with a compound microscope at 400 X. Percent colonization for each sample was estimated by gridline-intersection method (Mc Gonigle *et al.*, 1990).

#### Growth variables of *Spodoptera frugiperda* larvae

Larvae fed with the plant leaves inoculated with and w/o AMF were removed after 12 days. Larvae were collected manually with entomological low-pressure tweezers, placing each one of the insects in a disposable cup of 1 oz (Primo®, Mexico). The cups were covered, labeled and transferred to the laboratory to determine *S. frugiperda* larva weight, cephalic capsule width (CCW), and size. In the laboratory larvae were weighed in an analytical balance (BP121S, Sartorius, DE) and CCW was measured with a digital

caliper (Neiko®, USA). Additionally, size of larvae was determined placing them on a millimetric sheet to measure insect length. Larval fresh weight and CCW have been used as indicators of plant quality effect on herbivores (Roger *et al.*, 2013; Vogelweith *et al.*, 2015). Also, in *S. frugiperda*, CCW has been measured to assess the effect of food quality in larval size (Szczepaniec *et al.*, 2013).

#### Data analysis

The effect of AMF inoculation on creole maize plant and larvae of *S. frugiperda* traits was tested using one-way analysis of variance (ANOVA). Prior to statistical analysis, data were tested for variance homogeneity (Bartlett) and normality (Shapiro–Wilk). Plant leaf area and height data were square root transformed, AMF root colonization data were arcsine square root transformed, and larval cephalic capsule width (CCW) and size data were squared transformed prior to analyses to meet homoscedasticity assumptions. In the case of significant differences among AMF a multiple Tukey's comparison of means test with a level of significance of 5% ( $p \leq 0.05$ ) was performed to identify such differences. The data of leaf damage were analyzed using Kruskal-Wallis test. All data analyses were performed using STATGRAPHICS Centurion XV software.

## RESULTS

#### Effect of AMF inoculation on plant growth

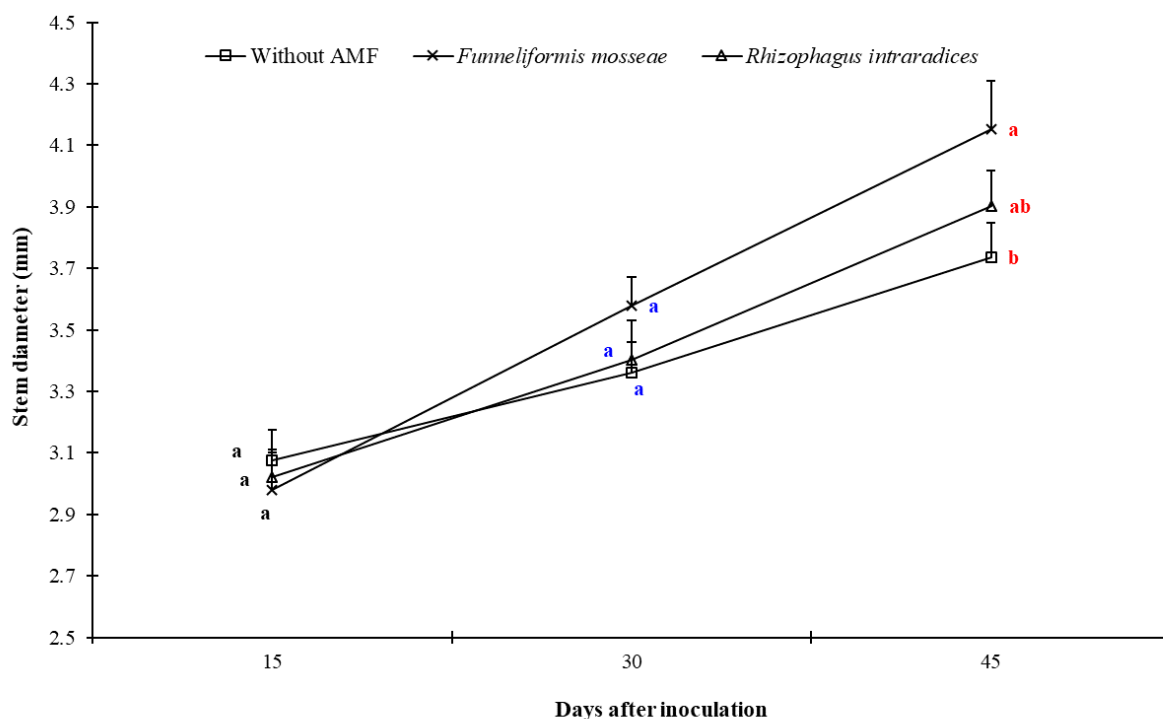
The microscopic analysis of root fragments collected from creole maize plant inoculated with AMF confirmed that *F. mosseae* and *R. intraradices* significantly enhanced the percentage of root fragments colonized by AMF in relation to plants non-inoculated (Table 1). However, root colonization by *F. mosseae* and *R. intraradices* were similar. On the other hand, plant height did not show significant differences between inoculated plants with or w/o AMF at 15 and 45 days (Tukey's test,  $p \geq 0.05$ ). Only significant differences were presented in plants inoculated with *F. mosseae* and *R. intraradices* at 30 days. However, the differences between plant inoculated with AMF and plants non-inoculated were slight (Table 1).

The stem diameter of maize plant did not show significant differences between inoculated plants with or w/o AMF at 15 and 30 days (Figure 1). However, at 45 days, plants inoculated with *F. mosseae* promoted slight stem diameter than *R. intraradices* and w/o AMF (Tukey's test,  $p \leq 0.05$ ). Only plants inoculated with *R. intraradices* promoted greater leaf area than plants inoculated with *F. mosseae* and w/o AMF (Table 1; Tukey's test,  $p \leq 0.05$ ).

**Table 1. Effect of arbuscular mycorrhizal fungi (AMF) on plant growth, leaf area and mycorrhizal colonization level in roots of creole maize plants under greenhouse conditions**

Treatment	Plant height (cm)	57 days after inoculation	
	30 days after inoculation	Leaf area (cm <sup>2</sup> )	AMF root colonization (%)
Without AMF	12.9±2.9 b	169.0±61.1 b	0 b
<i>Funneliformis mosseae</i>	15.5±1.4 ab	151.6±76.2 b	45.0±7.4 a
<i>Rhizophagus intraradices</i>	15.8±2.7 a	290.7±142.5 a	47.1±8.7 a

Values represent median ± standard error (SE). Different letters indicate significant differences according to Tukey's ( $p \leq 0.05$ ).



**Figure 1.** Effect of creole maize inoculation with arbuscular mycorrhizal fungi (AMF) on plant stem diameter through time in greenhouse conditions. Bars represent the median + standard error (SE). Different letters for each measurement point indicate significant differences according to Tukey's test ( $p \leq 0.05$ ).

### Foliar damage by the fall armyworm

The susceptibility of creole maize plants inoculated with AMF to FAW was measured by observed of leaf damage by FAW compared with maize plants non-inoculated. No differences were observed in leaf damage by FAW between plants inoculated with or w/o AMF (Kruskal-Wallis test,  $p=0.1844$ ). Leaf damage by FAW in plants with *F. mosseae*, *R. intraradices* and w/o AMF was on average 7 in Davis scale, where 7 is many small and mid-sized elongated lesions plus several large >2.5 cm elongated lesions on whorl and furl leaves.

### Effects of AMF inoculation on FAW growth

Cephalic capsule size and length of *S. frugiperda* larvae did not show significant differences between larvae that consumed inoculated plants with or w/o AMF (Table 2). In average larvae that fed on creole maize inoculated with and w/o AMF had similar length and CCW (Table 2). According to data of CCW and size, *S. frugiperda* larvae developed up to the fifth instar when feeding on maize plants with and w/o AMF for 12 days. However, larval weight was significantly different between larvae feeding on maize plants inoculated with or w/o AMF (Table 2), whereas those feeding on inoculated maize plants with *R. intraradices* showed greater weight than larvae inoculated with *F. mosseae* (Tukey's test,  $p \leq 0.05$ ).

**Table 2. Size, cephalic capsule width (CCW) and weight of larvae feeding on creole maize plants with or without (w/o) arbuscular mycorrhizal (AMF) fungi for 12 days.**

Treatment	Larva size (mm)	CCW (mm)	Larva fresh weight (mg)
w/o AMF	21.24±0.50 a	2.39±0.06 a	154.48±7.77 ab
<i>Funneliformis mosseae</i>	19.82±0.48 a	2.42±0.06 a	136.29±6.34 b
<i>Rhizophagus intraradices</i>	20.78±0.96 a	2.56±0.05 a	168.07±8.50 a

Values represent median ± standard error (SE). Different letter indicate significant differences according to Tukey's ( $p \leq 0.05$ ).

## DISCUSSION

This study examines the effect of arbuscular mycorrhizal fungi in creole maize growth, leaf damage and development of *S. frugiperda* larvae. AMF was found to partially influence on growth variables, such as height and plant diameter, leaf area and insect weight. With respect to leaf damage caused by the fall army worm, no difference was found between inoculated and uninoculated plants with AMF. With these findings, the main hypothesis that AMF influence on plant growth, damage reduction, and development of *S. frugiperda* larvae is partially supported under the conditions of this study. Seemingly, the insects that fed on colonized plants by *R. intraradices* had greater weight. These findings add up to the knowledge on how AMF may influence positively on the development of generalist herbivores (Goverde *et al.*, 2000; Kempel *et al.*, 2010; Bernaola *et al.*, 2018).

Several studies have demonstrated that AMF have positive effects on maize plant growth, especially creole maize (Gavito and Varela, 1995; Sangrabriel-Conde *et al.*, 2014; Reyes-Tena *et al.*, 2015), as well as reports from an opposite effect up to null differences in plant growth with and without AMF (Faye *et al.*, 2013; Lauriano-Barajas and Vega-Frutis, 2018; López-Carmona *et al.*, 2019), which is consistent with growth data in terms of height and stem diameter in this study. The plants colonized with *R. intraradices* and *F. mosseae* showed a slight increase in height at 30 days when compared with those without AMF. However, at 45 days no significant differences were observed between plants with or without AMF. On the other hand, only slight differences were observed in stem diameter in plants colonized by *F. mosseae* at 45 days then in those with *R. intraradices* and without AMF. Faye *et al.* (2013) mentioned that more than six weeks are need to observe significant growth difference in maize plants inoculated with AMF, and in this experiment, measurements were performed up to six weeks. Other aspects that may explain the results in this study is that plant growth with AMF depends on plant genotype and AMF species (An *et al.*, 2010). For example, creole maize has been documented to have better growth and yield increase when inoculated with native AMF species (Johnson *et al.*, 2010).

The effect of AMF in herbivory depends greatly on AMF species and that of insects (Jung *et al.*, 2012). For example, a meta-analysis revealed that *R. intraradices* is one of the AMF species with a high number of reports on negative effects in the development of generalist insect more than other species (Koricheva *et al.*, 2009). However, the results in this study with the generalist insect *S. frugiperda* showed a greater fresh weight when larvae were fed with plants colonized by *R. intraradices* which contradicts the report of Koricheva *et al.* (2009). Bernaola *et al.* (2018) and Real-Santillán *et al.* (2019) found something similar to what was reported in this study that AMF have a positive effect on the fall armyworm larval weight. On the other hand, when larvae consume creole maize leaves colonized by *F. mosseae* showed lower weight compared to plants with *R. intraradices*. These data agree with Harley and Gange (2009) who set out that AMF promoted a defense against generalist herbivores, as well as reduction in larval weight (Roger *et al.*, 2013). It has been shown that maize defense compounds such as protease inhibitors and benzoxazinoids are altered by AMF colonization of plant roots that affect weight gain of Lepidoptera larvae (Ramírez-Serrano *et al.* 2022; Stratton *et al.* 2022). Low weight in immature insect stages could affect development time, energy availability for the adult stage, survival, and consequently, greater predation probability by natural enemies. A possible explanation is that AMF may not only offer resources to plants to counteract herbivory but also turn it into a greater source of food for larvae and increase their nutritional value (Vannette and Hunter, 2011). According to the results in this study, the plants colonized with *R. intraradices* possibly had a better nutritional quality for the fall armyworm, which should be studied in future research.

Cephalic capsule width (CCW) has been measured in Lepidoptera larvae for differences in instar stages. However, Szczepaniec *et al.* (2013) and Vogelweith *et al.* (2015) measured CCW to assess food quality in growth of the immature Lepidoptera stages. For example, Szczepaniec *et al.* (2013) found a smaller CCW in larvae feeding on related wild maize species, which showed greater resistance to herbivores than with creole maize plants. Additionally, Wiseman *et al.* (1991) found that *Helicoverpa zea* larvae fed on diet



with corn genotype resistance to herbivory had a significant reduction in the CCW. Data in this study showed that CCW and size of larvae feeding on maize plants with or w/o AMF did not show differences. A possible explanation is that *S. frugiperda* was found in its last instar stage when insects do not show a considerable variation in larval size and CCW.

Several studies have reported contrasting effects of symbiosis between AMF and plants against herbivores; the positive effects may be explained by the increase in nutritional value and negative effects by improving plant defense (Pozo *et al.*, 2020). Data of leaf damage suggest that creole maize with AMF did not improve plant defense against the fall armyworm because they showed average damage of 7 in Davis scale and according to Singh *et al.* (2022) creole maize resistance against the fall armyworm should be from 3 to 5 in Davis scale. A possible explanation is that the creole maize used in our experiments has some degree of tolerance to FAW since herbivory did not affect reducing plant growth. According to Lima *et al.* (2022), it is very common that creole maize to show a degree of tolerance against FAW. On the other hand, data of larval development that fed on creole maize colonized with *R. intraradices* agrees with the positive effects of AMF in insect larvae by improving the quality of their food. These results suggest that AMF may influence FAW condition to make them nutritionally valuable to natural enemies. For example, Hoffmann *et al.* (2011) found increased population growth rates of the predatory mite *Phytoseiulus persimilis* when their *Tetranychus urticae* prey was fed on AMF-colonized bean plants. Therefore, AMF may be used to contribute to the management of the FAW by attracting natural enemies such as predators and parasitoids. Nonetheless, more studies in depth with other varieties of maize and other species of AMF should be performed to know if AMF influence on FAW condition and recruitment of natural enemies.

## CONCLUSION

To conclude, the results in this study show that AMF, *R. intraradices*, and *F. mosseae* had a slight effect on creole maize growth. Leaf damage by *S. frugiperda* larvae was similar between plants inoculated with or without AMF, and larvae that consumed leaves of inoculated plants with *R. intraradices* showed greater fresh weight than with *F. mosseae*. Nevertheless, more studies should be performed with other creole maize and native AMF species to confirm positive or negative effects of AMF on the fall armyworm to contribute to its management with the use of beneficial soil microorganisms.

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