Scientific Paper

# Effect of the application of mycorrhizal strains on the morphoagronomic development of *Glycine max* L.

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## Abstract

**Objective**: To evaluate the effectiveness of the application of three strains of arbuscular mycorrhizal fungi on the yield of *Glycine max* L., cultivar Incasoy 27, on a reddish brown fersialitic soil, typical of Las Tunas province.

**Materials and Methods**: The research was conducted under field conditions, from September to December, 2018. For setting up the experiment a randomized block design was used, with five treatments and four replicas. The distance between replicas was 1 m. The seed used was from INCA, with 98 % germination. Five treatments were established: an absolute control, fertilization with NPK and inoculation with mycorrhizal strains INCAM4, INCAM11 and INCAM2. During the crop cycle the variables number of pods per plant, number of grains per plant, weight of 100 grains, yield in t $ha^{-1}$, were evaluated.

**Results**: It was observed that the lower number of pods per plant corresponded to the control treatment, which differed from the others ($p < 0,05$). The highest values were recorded in the inoculated treatments, which did not differ among them (51,4; 60,0 and 62,2 for INCAM 4, INCAM 11 and INCAM 2, respectively). Regarding yield, the best results were obtained in the treatment fertilized with NPK and the inoculation with mycorrhizae, with values of 2,0; 2,5; 2,7 and 2,6 for NPK, INCAM 4, INCAM 11 and INCAM 2, respectively. All the treatments were economically profitable. The inoculated ones stood out.

**Conclusions**: The application of arbuscular mycorrhizal fungi strains influenced positively the morphoagronomic indicators evaluated in *G. max*, which allows to decrease the use of mineral fertilizers in this crop.

**Keywords**: *G. max*, inoculation, yield

## Introduction

Biofertilizers constitute a sustainable economic and ecological alternative in the integrated crop management. They allow to reduce external inputs, improve the quantity and quality of internal resources, as well as guarantee higher efficiency of mineral fertilizers (García-León *et al*., 2016). The latter have become, along with biopesticides, key agricultural inputs in sustainable agricultural production (González-Ramírez and Pupo-Feria, 2017). Among them, mycorrhizal arbuscular fungi (AMF) are widely utilized worldwide. In Cuba, they are applied in the fertilization of economically important crops (Mujica-Pérez and Molina-Delgado, 2017).

The world production of *Glycine max* L. has remarkably increased, due to the demand of the food industry. According to the data of the United States Department of Agriculture (Karr-Lilienthal *et al*., 2004), in the productive cycle 2017-2018, this country was leader of the global production, with more than 123 million tons, followed by Brazil, with 118. The American continent stands out in this ranking, with five countries among the ten main producers.

The balanced use of organic manures and inorganic fertilizers is a relevant factor in agricultural economy and in the quality of farm production. The different regimes of fertilizers influence biomass, and the mycorrhiza phytotypes in agriculturally important soils (Qin *et al*., 2015).

In the Amancio municipality, the agricultural yields of *G. max* are low and vary, approximately, from 0,9 to 1,5 t $ha^{-1}$. In recent times, to counteract the negative effect of chemical fertilization the use of biofertilizers is increased, which allow plants to overcome stress situations against the adverse environmental conditions. It favors their growth, development and yield, and thus contribution is made to the decrease of the use of chemical substances.

This research will provide results that confirm the benefits of the use of organic resources, as strategy to contribute nutrients, maintain or improve the

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organic matter of the soil and favor the crop economy, by allowing the expression of microbiological resources that co-evolved with the plants.

The objective of this work was to evaluate the effectiveness of three AMF strains on the morphoagronomic development of *G. max*, cultivar Incasoy 27, in the Amacio Rodríguez municipality, Las Tunas province.

## Materials and Methods

*Location*. The research was conducted under field conditions, on a typical reddish Brown Fersialitic soil (Hernández-Jiménez *et al*., 2015), in the Cooperative of Credit and Services Mártires de Pino III, of the Amancio Rodríguez municipality, in Las Tunas province, between September and December, 2018. This cooperative is located in the geographic coordinates 24°47'55,1" North latitude and 77°35'23,5" West longitude.

*Characteristics of the soil in the experimental area*. Samples were taken at 20 cm of depth through the experimental sampling technique in grid form (Almendros-Martín *et al.,* 2010) and they were dried and sieved with a 2-mm mesh. The pH ($H_2O$) was determined through the potentiometric method, organic matter by Walkley and Black (1934) and available phosphorus according to Olsen *et al*. (1954) with molecular spectrophotometry (EDULST01-13). The cation exchange capacity (CEC), change cations ($Ca^{2+}$, $Mg^{2+}$, $Na^{+}$, $K^{+}$) and base change capacity (BCC) were found according to Mehlich (1984), modified by NC-65:2000 (ONN, 2000) (table 1).

*Experimental design and treatments*. For setting up the experiment a randomized design was applied, with five treatments and four replicas. Plots with a surface of 11,2 $m^2$ (2,8 x 4, m), with four rows, were used. From them, the two central ones (5,6 $m^2$) were taken as sampling area. The distance between replicas was 1 m and the cultivar INCAsoy-27 was used, from INCA, with germination of 98 %. The treatments consisted in an absolute control, NPK and three mycorrhiza strains: INCAM 4, INCAM 11 and INCAM 2.

*Experimental procedure*. The applied plant management was carried out according to the rules established by the technical instruction manual of the crop (Hernández and Bello, 2010).

Sowing was carried out on September 6, 2018, manually, at 4 cm of depth. Two seeds were placed per nest, with distance between rows of 0,70 m and 0,10 m between plants.

During the crop cycle seven irrigations were applied through the spraying technology in the critical periods of water demand, framed in the pre-flowering, flowering-pod formation and grain filling stages, with irrigation interval of 7 to 8 days, depending on the rainy season.

NPK was applied, at a rate of 10- 8- 8, on the row bottom before sowing. The strains of arbuscular mycorrhizal fungi (*Glomus manioti* sp) were applied as mixture. The seed was covered two hours before sowing, with proportion 2:1 of inoculant/water. It was subject to natural drying, under shade.

*Measurements*. After harvest, the number of pods per plant, number of grains per pod and weight of 100 grains (g), were evaluated. An analytical scale (SARTORIUS, model BS 2202S) was used and the yield was expressed in t $ha^{-1}$. The yield obtained in each plot was considered, and later the yields were calculated in tons per hectare.

*Statistical analysis*. The data of the different measurements were processed through a double-classification variance analysis and mean comparison for Tukey's test, for 5 % of error probability. The information was processed with the statistical program InfoStat, version 2017 (Di Rienzo *et al.,* 2017). To determine the parametric statistical analysis the variance homogeneity test was carried out by Barttle's Test. It was tested whether the data were adjusted to a normal distribution by Shapiro-Wilks Test.

*Economic analysis*. For the economic analysis the values obtained in the yield in t $ha^{-1}$ of the dry seed (14 % moisture) were taken into account. As price of an inoculant bag, $ 25,00 was considered. Meanwhile, the cost of the ton of urea in the market is $ 300,00[1]. The value of the soybean ton in the national market, corresponding to $ 10 600 (Tamayo-León, 2020), was taken as basis. The production cost (PC), production value (PV), profit (P), economic benefit (EB) and relative treatment cost

Table 1. Soil fertility components (0-20 cm).

| Depth | OM | pH | EC | Cmol(+)$kg^{-1}$ | | | | ppm |
|---|---|---|---|---|---|---|---|---|
| cm | % | KCL | $dSm^{-1}$ | $Ca^{2+}$ | $Mg^{2+}$ | $K^{+}$ | $Na^{+}$ | $P_2O_5$ |
| 0-20 | 2,25 | 6,43 | 0,40 | 26,77 | 3,50 | 3,50 | 0,39 | 6,26 |

[1]https://www.indexmundi.com

(RTC), were determined, according to the indications by Recompenza and Angarica (2010).

1. PV ($ ha$^{-1}$): crop yield multiplied by the sale price of one ton of dry weight, at 14 % moisture
2. PC ($ ha$^{-1}$): sum of the expenses incurred due to the application of fertilizer or microbial inoculants as corresponded, plus the cost of the other activities
3. P ($ ha$^{-1}$): difference between the production value and production costs
4. C/P ($): quotient obtained from dividing the production cost and production value

## Results and Discussion

Regarding the performance of the yield components (table 2), the lowest number of pods per plant corresponded to the control treatment, which differed from the others for p > 0,05. The best values were for the inoculated treatments, which did not show differences among them (51,4; 60,0 and 62,2 for INCAM 4, INCAM 11 and INCAM 2, respectively). The treatment with NPK presented moderate values.

Vega (2013) obtained similar results to these ones, regarding the number of pods per plant in this cultivar (INCAsoy-27) on similar soils of the Granma province. However, they are lower than the ones obtained by Zamora and Abdou (2007), who in a study conducted in the cold season in the Granma province, reported values between 21,4 and 64 pods per plant.

AMF colonization promotes the increase of the concentration of soluble carbohydrates and chlorophyll in the leaves and, consequently, the increase of the photosynthetic capacity (Keshavarz *et al.,* 2020), which could have favored the increase of the quantity of pods per plant.

The positive effect of AMF on the agricultural productions is widely acknowledged. Medina-García (2016) state that mycorrhizae improve the water and nutrient absorption capacity from the soil, because their hyphae, by exploring the soil, reach the places that can hardly be reached by plant roots on their own. In addition, AMF increase the hydraulic conductivity of the roots and favor the adaptation of the osmotic balance (Ley-Rivas *et al*., 2015).

Regarding the number of grains per pod, the control showed the lowest results and differed from the other treatments. The highest values were found in the treatments inoculated with the strains INCAM 2 and INCAM 1. The rest showed moderate values.

The results of this research were higher than those reached by Lemes *et al.* (2017). Meanwhile, they differ from the ones obtained by Linares-Ramos (2006) en Guatemala, who did not find statistical differences regarding the number of grains per pod.

It could be observed that the treatments that showed higher grain weight were the inoculated ones, without differing from the fertilized treatment. These results can be corroborated with the reports by Romero (2012), who refers a high weight of 100 grains (between 11,50 and 18,20 g) when evaluating seven soybean cultivars in the Majibacoa municipality. This indicates that there was correspondence among the inoculated treatments, regarding the grain weight and yields.

The literature highlights that the high variability of the yields is related, in recent years, to the role played by the climate and soil conditions in the definition of these indicators for a certain cultivar. This aspect allows to explain how some cultivars respond better than others to the edaphoclimatic conditions of certain locality (Villamar-Burgos, 2017). The results from other studies also indicate the influence of high temperatures (higher than 30 ºC) on the decrease of the yields of some soybean cultivars (Zonetti *et al.,* 2012).

Table 2. Components of the variables related to yield.

| Treatment | Number of pods per plant | Grains per pod | Weight of 100 grains, g | Yield, t ha$^{-1}$ |
|---|---|---|---|---|
| Control | 38,2d | 2,0d | 12,1b | 1,7b |
| NPK | 45,3bc | 2,3c | 14,8ab | 2,0ab |
| INCAM 4 | 51,4 ab | 2,7b | 15,2a | 2,5a |
| INCAM 11 | 60,0a | 3,0a | 15,0a | 2,7a |
| INCAM 2 | 62,2ab | 3, 0a | 15,6a | 2,6a |
| VC% | 3,1 | 7,4 | 3,4 | 3,3 |
| SE ± | 0,02 | 0,02 | 0,07 | 0,02 |
| P - values | 0,3406 | 0,3384 | 0,3525 | 0,2854 |

Means with a different letter in the same column significantly differ for p < 0,05

Lower results than those from this research were reported by Molinet *et al*. (2015). These authors obtained yields below 1 t $ha^{-1}$ with the combined use of EcoMic® and Azotofos® and of EcoMic® independently, on a little differentiated fluvisol soil, in the Granma province, using the same cultivar.

Regarding the yield components, it was proven that for the conditions under which the experiment was developed, the best results were obtained in the fertilized treatment with NPK and the inoculation with mycorrhizae. This is closely related to the other analyzed variables. The contents of organic matter and phosphorus in the soil were low. Hence there is higher response to mineral fertilization.

García *et al*. (2017) coincide on the fact that the use of mycorrhizae optimizes the absorption process of nutritional elements. Thus, when they are applied plant development is stimulated and the productive potential of the plants increases.

The effectiveness of the symbiosis among different microorganisms is known and has been proven. Authors like Sotelo *et al.* (2016), when inoculating soybean seeds, variety G7R-315, with the mixture of the *Rhizobium japonicum* strain ICA 8001 and the AMF strain *Glomus clarum*, without applying fertilizer, observed a positive effect on the vegetative development, with considerable increase of the crop yield.

The tripartite symbioses among legumes, rhizobia and mycos are very common in natural ecosystems. The nitrogen fixation symbiosis is highly demanding and, thus, requires high phosphorus quantity. Often mycorrhizae supply the latter (Romagnoli *et al.,* 2017).

Mujica-Pérez and Molina-Delgado (2017) reported higher results than the ones that were achieved in this research, when studying the application of the AMF strain *G. cubense* on a lixiviated ferralitic red soil, in the Mayabeque province. These authors obtained the highest yields with the use of the mycorrhizal strain combined with Azofert®, liquid based on bacteria of the *Rhizobium* genus.

The economic calculation of the crop (table 3) allows its utilization as an alternative to substitute imports, because the results show profits that vary between 17 380,00 and 27 966,00 pesos per hectare.

All the treatments reached profits in their production. The one that provided the highest profit was that inoculated with the strain INCAM 11, with higher results than $ 27 000,00 pesos per hectare, with a cost of $ 0,02 per produced peso. These results are higher than the ones found by Velásquez (2009), Turruelles (2012) and Romero (2012) for the cultivar INCAsoy-27, in other soil types for Las Tunas province. The sowing of the soybean crop with biofertilizers constitutes a viable alternative for the crop production, in order to increase the agricultural yield and sustainability of agroecosystems.

Reyes-Tana *et al.* (2016) state that the inoculation of biofertilizers that contain rhizospheric bacteria, causes significant increases in the productivity of agricultural crops.

Regarding this research, Turruelles (2012) and Romero (2012) achieved lower profits ($ 6 166,97 and $ 5 571,78 CUP; respectively) for the cultivar INCAsoy-27 on other soil types of Las Tunas province.

The new technologies should be focused on maintaining and preserving the sustainability of the production system through the rational exploitation of natural resources and the application of pertinent measures for preserving the environment (Gragenda-Cabrera *et al.,* 2012). The inoculation and agronomic management of the microorganisms with biofertilizer properties constitute rational technologies, which emerge as innovative and promising practices for the agricultural activity (Moreno-Reséndez *et al.,* 2018).

## Conclusions

The application of arbuscular mycorrhizal fungi strains influenced positively the morphoagronomic indicators evaluated in *G. max*, which allows to decrease the use of mineral fertilizers in this crop.

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Table 3. Economic appraisal of the treatment application.

| Indicator | Control | NPK | INCAM 4 | INCAM 11 | INCAM 2 |
|---|---|---|---|---|---|
| Yield, t $ha^{-1}$ | 1,7 | 2,0 | 2,5 | 2,7 | 2,6 |
| Production value, $ $ha^{-1}$ | 18020,0 | 21200,0 | 26500,0 | 28620,0 | 27560,0 |
| Production cost, $ $ha^{-1}$ | 640,0 | 760,0 | 654,0 | 654,0 | 654,0 |
| Profit, $ | 17380,0 | 20440,0 | 25846,0 | 27966,0 | 26906,0 |
| Cost per peso, $ $ha^{-1}$ | <0,03 | 0,04 | 0,02 | 0,02 | 0,02 |

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**Authors' contribution**

- Aracelis Romero-Arias. Research conception and design, data acquisition and interpretation, manuscript writing and revision.
- Raquel María Ruz-Reyes. Research conception and design, data acquisition and interpretation, manuscript writing and revision.
- María Caridad Nápoles-García. Research conception and design, data acquisition and interpretation, manuscript writing and revision.
- Santa Laura Leyva-Rodríguez. Research conception and design, data acquisition and interpretation, manuscript writing and revision.
- Jorge Ernesto Baez-González. Research conception and design, data acquisition and interpretation, manuscript writing and revision.

**Conflicts of interests**

The authors declare that there are no conflicts of interests among them.

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