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EVALUATION OF EDAPHICAL FAUNA AND DEVELOPMENT OF COCOA INOCULATED WITH MYCORRHIZAL FUNGI

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ABSTRACT

The soils of the Amazon region are represented mostly by Oxisols and Argisols of high acidity and low fertility. In this sense, efficient management alternatives are sought in this region in the use of natural resources. This work aimed to evaluate the biological quality of the soil and the development of cocoa inoculated with mycorrhizal fungi in the field, as well as parameters of cocoa growth. Samples were collected from each cocoa plantation area to identify soil edaphic organisms. Then, inoculated and non-inoculated cocoa seedlings were planted with Arbuscular Mycorrhizal Fungi (FMAs) of the genus *Glomus*. At 19 months after planting, the seedlings were evaluated for survival rate, stem diameter, plant height and number of leaves. The identified organisms were spiders, nematodes and collemboli in the non-inoculated area and in the inoculated cocoa area, a greater diversity of organisms was found, such as: nematodes, mites, FMAs, aspaspores and colembolos. The inoculated cocoa seedlings perform better in all evaluated parameters. The introduction of biological inputs (FMAs) in the cocoa areas contributed to improving the biological quality of the soil and adapting the seedlings under stress conditions.

KEYWORDS: Cocoa culture; Soil Biology; Production technologies.

AVALIAÇÃO DA FAÚNA EDÁFICA E DESENVOLVIMENTO DO CACAU INOCULADO COM FUNGOS MICORRÍZICOS

RESUMO

Os solos da região Amazônica são representados em sua maioria por Latossolos e Argissolos de alta acidez e baixa fertilidade. Nesse sentido busca-se nesta região alternativas de manejo eficientes na utilização dos recursos naturais. Este trabalho, teve o objetivo de avaliar a qualidade biológica do solo e o desenvolvimento do cacau

inoculado com fungos micorrízicos a campo, bem como parâmetros de crescimento do cacau. Foram coletadas amostras em cada área de plantio do cacau para identificação dos organismos edáficos do solo. Em seguida, foram plantadas as mudas de cacau inoculadas e não inoculadas com Fungos Micorrízicos Arbusculares (FMAs) do gênero *Glomus*. Aos 19 meses após o plantio as mudas foram avaliadas quanto a taxa de sobrevivência, diâmetro do colo, altura da planta e número de folhas. Os organismos identificados foram as aranhas, nematóides e colêmbolos na área não inoculada e na área de cacau inoculado foi encontrada maior diversidade de organismos, como: nematóides, acaros, FMAs, ascósporos e colembolos. As mudas de cacau inoculadas tiveram melhor desempenho em todos os parâmetros avaliados. A introdução de insumos biológicos (FMAs) nas áreas de cacau contribuiu para a melhoria da qualidade biológica do solo e adaptação das mudas em condições de estresse.

PALAVRAS-CHAVE: Cacaucultura; Biologia do Solo; Tecnologias de produção.

INTRODUCTION

The cultivation of cacao has gained more and more prominence in the Amazon region, mainly in the state of Pará, which is currently the largest producer of cocoa in Brazil (IBGE 2017), emerging as a new perspective in the cultivation models in several places in the Amazon, be incorporated as one of the main components of agroforestry systems. Culture has reduced the impacts of migratory agriculture and contributed to the diversification of crops as it incorporates environmental and economic aspects into the production system (SANTOS; SILVA, 2017) and adapts to more diversified systems, composed of different forest species. The expansion of *Theobroma cacao* cultivation in Pará has been encouraged by State government programs with measures to leverage, qualify changes in the technical, economic,

Mantovanelli et al. (2016) state that the Amazon region normally presents poor soils with low nutrient retention due to the low concentration of organic material incorporated in the soil as a result of natural fields that have a high concentration of aluminum, resulting in low values and pH. Knowing that the satisfactory development of cacao requires soils with adequate fertility, deep and well drained, cultivations in these conditions can lead the plants to present nutritional deficiency, resulting in restrictions on the growth of the plants and in the reduction of the sustainability of production (RODRIGUES et al., 2018).

This can be aggravated if producers establish their crops without worrying about factors related to soil conservation, spatial distribution, shading, and seed quality, thus implying problems related to crop instability and cocoa productivity (JESUS et al., 2013).

Therefore, we seek to insert sustainable alternatives in the management of this cultivation, especially those that concern the improvement of soil quality. Carrying out sustainable management of this is extremely important, as the soil is seen as a living and dynamic resource. Based on these conditions, the study of soil quality starts from its capacity to be functional, respecting the limits established by the ecosystem and the forms of land use, in order to preserve and sustain organic production and environmental quality, thus contributing to health of living beings and integrity of ecosystems (BALOTA, 2018; SCHEMBERGUE et al., 2017).

The meso and macrofauna of the soil plays fundamental functions for the maintenance of the ecosystem, being considered a bioindicator of soil quality, since the

anthropic actions, reduce the diversity and abundance of mesofauna organisms. The study of this group is relevant to the knowledge of the state of soil degradation, it is suggested that more studies be done on the diversity of soil mesofauna (BERUDE et al., 2015).

Arbuscular mycorrhizal fungi are a significant part of the soil's microbial biomass and are directly involved in essential processes of the soil-plant interface. The inoculation with fungi provides the highest content of K and S, in addition to the greater growth of the aerial part and roots of the plants, also providing greater efficiency in the use of nutrients, mainly of P, positively influencing their growth and making it more resistant to biotic and abiotic factors (RODRIGUES et al., 2018; NASCIMENTO et al., 2018).

In view of these factors that prioritize the quality of the soil as a condition for the good development of cacao, the FMAs present themselves as a viable alternative for the benefit of this crop, as according to Gomide et al. (2014), these fungi are of paramount importance in the maintenance and dynamics of ecosystems, directly benefiting tropical regions, where the mineral reserve of nutrients in the soil is scarce, resulting in the lack of a functional biological structure in these regions. Being knowledge of these organisms is necessary, as they are fundamental for improving agricultural productivity, since they contribute beneficially to the sustainability of agro-ecosystems (SIQUEIRA et al., 2002).

There is an advance in studies with the inoculation and symbiotic dependence of mycorrhizal fungi in some fruit trees and it is known that the use of AMF species improves the nutritional status in the production of used seedlings and greater vegetative development of the plant (FARIAS et al., 2014; GOMES et al., 2018). However, few studies have evaluated the interaction and mycorrhizal dependence between fungus and cocoa crop. Berude et al., (2015) suggest further studies on mycorrhizal applications and the development of inoculated plants.

Being the cocoa culture of great importance in the Amazon region and the growing search for sustainable alternatives for agricultural production, in this This work aimed to evaluate the soil edaphic fauna and the cocoa growth parameters, after inoculation with mycorrhizal fungi in PA Palmares II in Parauapebas - PA.

MATERIAL AND METHODS

The experimental area corresponded to a 1 ha cocoa monoculture, located in lot 32 of PA Palmares II in Parauapebas - PA. The soils in the lot are characterized as dystrophic Red Yellow Latosols, and the settlement's vegetation cover is composed of forest, pasture, brushwood and fields.

The preparation of the soil for planting cocoa occurred through the cutting and burning system, where the 422 seedlings were planted and inoculated with arbuscular mycorrhizal fungi and 422 seedlings not inoculated with arbuscular mycorrhizal fungi, arranged in 6 lines, with the spacing between rows was 1.5 x 1.5 m and the spacing between plants was 3.0 x 3.0 m and the total area was divided into 0.5 ha inoculated and 0.5 ha uninoculated. The design used was completely randomized, following the recommendations of Pimentel - Gomes (1987).

The plants were inoculated at the time of planting, with a mixture of sand and inoculum of FMAs of the species *Glomus clarum* from the inoculum bank of the Faculty of Agricultural Sciences of Marabá, with approximately 3 g of the inoculum in each

planting hole.

The evaluation of seedlings in the field was carried out nineteen months after planting in the field, and the evaluations took place in the form of an agroecological task force, with the collaboration of undergraduate students from the Agronomy 2011 course, where the parameters of growth and development, such as survival rate, stem diameter, plant height and number of leaves. The data were analyzed with the aid of the SISVAR 5.1 Build 72 software, according to the recommendations of Pimentel - Gomes (1987). Analysis of variance and the Tukey test were used to compare means ($P < 0.05$).

Figure 1 shows the evaluation of the parameters of growth and diameter of the cocoa neck established in the area.

FIGURE 1- Evaluation of growth parameters and diameter of the cocoa neck in PA Palmares, Parauapebas - PA.



Source: MENDES (2013).

10 simple soil samples were collected in the monoculture area of the inoculated cocoa and 10 simple soil samples in the monoculture area of the non-inoculated cocoa, each simple sample corresponding to a volume of soil collected from a random point of the plot, to a depth of 10 cm in an area of 100 m², according to the methodology of Lemos (2000) (Figure 2). The soil samples were homogenized, constituting composed samples, being separated approximately 100 grams of soil from each area in plastic bags that were sent to the Microbiology Laboratory of the Faculty of Agricultural Sciences of Marabá, where they were kept at room temperature for the extraction of the soil organisms and subsequent characterization and identification of mycorrhizal fungi species.

FIGURE 2- Soil sample collection in the area of inoculated and non-inoculated cocoa plantation with mycorrhizal fungi in PA Palmares, Parauapebas - PA.



Source: MENDES (2013)

The technique used to evaluate the organisms present in the soil samples was wet sieving by Gerdeman and Nicolson, (1963) and centrifugation in water and 40% sucrose (Jenkins, 1964). The identification of the genera and species found were made by observing the external morphological characteristics of their formation with the aid of a stereoscopic magnifying glass. Then, microscopic slides were made for further identification and classification.

RESULTS AND DISCUSSION

Biological Quality of Soil

From the biological analysis performed on soil samples from the cocoa monoculture area, the diversity of organisms that indicated soil quality was identified (table 1).

TABLE 1 -Evaluation of the biological quality of the soil after implantation of the inoculated cocoa in PA Palmares I - Parauapebas-PA. Average of 10 repetitions.

| Treatment | Organisms Found |
|------------|---|
| Witnesses | Arachnids Nematodes Collembola |
| Inoculated | Nematodes Acarina Mycorrhizal fungi Ascospores Collembola |

According to Table 1, there is a greater number of species of edaphic fauna, indicators of biological quality, in the area of cocoa inoculated with arbuscular mycorrhizal fungi, when compared to the non-inoculated area. The following organisms are found in the area without inoculation: spiders, nematodes and collemboli and in the inoculated area: nematodes, mites, mycorrhizal fungi, aspaspores and collemboli. Which were not counted, since the objective of this is not to quantify the organisms, but to determine the types of organisms of the macro and meso fauna found in the study area. It is also observed that there was no mycorrhizal contamination in the area that was not inoculated in the analyzed period.

The presence of organisms found in the cocoa monoculture area is due to the fact that this area contains a large amount of organic matter, consequently, a greater presence of these living organisms in the soil is observed, especially the mites and collembolos, as the organic matter helps to compose the bodies of most edaphic organisms, providing high amounts of energy through plant residues and root excretions (BRADY; WEIL, 2013, BERUDE et al., 2015; PRIMAVESI; PRIMAVESI, 2018;).

The data found corroborate with Geremia et al. (2015), Pompeu et al. (2016), Casaril et al. (2019) who observe increases and / or positive responses from the presence of edaphic fauna in areas managed under a greater presence of organic matter. It should be noted that in addition to the fact that organic matter is a source of energy for microorganisms, it serves as a source of nutrients for plants (PRIMAVESI; PRIMAVESI, 2018).

The presence of higher proportions of mites is related to the eating habits of these organisms, which for the most part feed on organic matter or microorganisms that grow on this decomposing material (SILVA; MORAES; KRANTZ, 2004; BEITIA et al. , 2015). Collemboli play a role similar to mites. Berude et al. (2015), Silva et al. (2020) punctuate the detritivorous function of collemboli, contributing to the decomposition of organic matter and control of populations of microorganisms, especially fungi.

It is also noteworthy that the greater occurrence of organisms in the inoculated area, can be attributed to the extension of the external mycelia of mycorrhizal fungi in the rhizosphere of cocoa trees, which can serve as food for organisms such as collems and nematodes (PAULA, (1992); BIANCIOTTO et al., (2012) apud REIS et al., (2010). Silva et al. (2016) observe a greater mycelium length in areas under forest and agricultural vegetation, thus the extension of the mycelia may have increased the availability of food for these bodies.

The area of monoculture of cocoa inoculated with arbuscular mycorrhizal fungi showed a greater variability of organisms present in a single area, thus showing the viability of mycorrhizal fungi in improving soil quality.

Because mycorrhizae are symbiotic and mutualistic associations, they contribute to better phosphorus fixation and consequently decrease in pH, making this soil more conducive to the appearance of other organisms that are indicators of soil quality, such as collemboli that play a fundamental role in incorporating vegetable remains in the soil, thus increasing the availability of organic matter and other nutrients (PRIMAVESI; PRIMAVESI, 2018).

Thus, the identification of these organisms is of fundamental importance to obtain information about the organisms existing in the soil and the interactions they

establish with the environment, in order to intervene in a sustainable way, obeying the limits and potential of each agro-ecosystem.

Cocoa Growth Parameters

At 19 months after planting, cocoa seedlings inoculated with arbuscular mycorrhizal fungi stood out in comparison to uninoculated cocoa seedlings (controls), since in the first case there was an increase in all observed parameters (table 2).

TABLE 2. Evaluation of survival rate and growth parameters of cacao plants in the field 19 months after planting. Average of 422 plants. PA Palmares I. Parauapebas-PA.

| Treatment | Number of live plants | % of live plants | Height of plants (m) | Neck diameter (mm) | Number of sheets |
|--------------------------|-----------------------|------------------|----------------------|--------------------|------------------|
| Witness (not inoculated) | 54 b | 12,79 | 1,06 b | 14,48 b | 10 b |
| Inoculated | 330 a | 78,2 | 1,90 a | 48,48 a | 28 a |
| CV (%) | 32,7 | - | 27,8 | 29,7 | 32,9 |

CV = coefficient of variation.

Averages followed by the same letter do not differ in Turkey test by 5% probability.

Table 2 shows that there was a significant difference in all the variables analyzed, with the beneficial effect of FMA on the development of cocoa plants at 19 months in the field. Since, when establishing a comparison relationship between plants inoculated with AMF and non-inoculated (control), the first presented a superior response in relation to the second: i) 64.41% regarding the percentage of live plants; ii) 84 cm higher in average plant height; iii) increase of 34 mm regarding the average neck diameter; iv) an average of 18 more leaves as the variable number of leaves. Indicating that the addition of *Glomus inocula. clarum* provided better plant development in the field.

Among the groups of plants, fruit trees are known to benefit from the association with arbuscular mycorrhizal, however there are differences in the degree of mycotrophism that is generally reflected in plant growth (SILVA et al., 2009).

The data referring to the height of the plants corroborates those written by Machineski et al., (2009) who found that plants inoculated with the species of FMA (*G. margarita* and *G. clarum*), showed higher growth in height. Soares and Martins (2000) found that different species of AMF inoculants used in yellow passion fruit seedlings provided significant increases in growth and nutrient content in the aerial part of this fruit tree, when compared to treatments without inoculation.

Moreira and Siqueira (2002) state that there is a proportional relationship between the time of exposure of the fungi to the host roots, where the longer the time of exposure, the greater the benefits for the plants. In addition, Berude et al., (2015) when carrying out bibliographic analysis on the characteristics of the mycorrhizal association, observe that the importance of extracellular hyphae, which, when developed in rhizospheres, contribute to the nutritional aspects of plants and, consequently, increase

their initial ability to growth and stabilization

The good performance of this fruit is also related to the species that was used as an inoculant, as Vitorazzi Filho et al., (2017). Gomes et al., (2018) and Pinheiro et al., (2019) observed positive results in the development of fruit species inoculated with the species *Glomus clarum*.

When inoculating citrus rootstock seedlings with fungi *Glomus clarum* and *Glomus etunicatum* Miranda et al., (2018) they observed that the presence of the fungi contributed to the absorption of primary and secondary macronutrients, especially N, P, K and Mg.

Because the area was prepared by cutting and burning, which results in a decrease in soil microorganisms (macro and meso fauna) and consequent loss of microbial diversity, due to the reduction of the food source (REDIN et al., 2011) the higher occurrence of edaphic fauna taxa in the mycorrhizal area may have favored the process of decomposition of organic residues and mineralization of organic matter and formation of biopores, predisposing favorable conditions of vegetative growth to cocoa seedlings.

Lattuda et al. (2019) also found that arbuscular mycorrhizal fungi optimize the adaptation process of native fruit seedlings of the Myrtaceae family to the field, playing an important role of the association in relation to the vegetative development of the species and accelerating the process of adaptation to the field. In addition, the fact that the availability of N and P is the main limiting factor for plant growth and productivity, makes FMAS a biological input with great potential for agriculture (SIQUEIRA et al., 2002), thus proving efficiency of these fungi in the inoculation of fruits in tropical regions, mainly when the plants are in the field in adverse situations of climate, soil fertility and water availability which explains the seedling survival rate of 78.2% seedling survival rate, which in the analyzed period it was favored by the mycorrhizal inoculation with *Glomus clarum*.

CONCLUSION

The introduction of biological inputs (FMAs) in the planting of cocoa contributed to the biological quality of the soil. In addition, the association of arbuscular mycorrhizal fungi- *Glomus clarum* with cocoa seedlings made it possible to adapt them under stress conditions, beneficially influencing the survival rate and all evaluated growth parameters. Thus, it is observed that inoculation with FMAs is a promising technology for adapting fruit species to the field. It is necessary to continue the investigation of the use of biological input in cocoa cultivars, as a way to foster the information gap on the subject in the southeastern region of Pará.

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