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Biocontrol of plant-parasitic nematodes by soil in soybean cultivation

Controle biológico de fitonematoides via solo no cultivo de soja

Control biológico de fitonematodos vía suelo en el cultivo de soja

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Abstract
Soybean is a commodity widely cultivated in Brazil and represents one of the main export products. Several biotic and abiotic factors have limited the productivity growth of this oilseed, one of them are the nematodes associated with the monoculture. Therefore, the objective of this study was to evaluate the effect of different bioagents in control of plant-parasitic nematodes in soybean, via soil. The experiment was carried in Gurupi-TO in a randomized complete block design, with four repetitions and 14 treatments, including the witness. The treatments consisted of biocontrol agents: *Paecilomyces* sp., *Trichoderma asperirellum*, *Bacillus subtilis* and *Pochonia* sp., evaluated singly and in combinations. The treatments with *Paecilomyces* sp. + *Bacillus subtilis* and the combination of all the microorganisms results in higher averages for most traits when compared to the control treatment. The treatments *Paecilomyces* spp. + *Bacillus subtilis* and *Paecilomyces* sp. + *Trichoderma asperellum* + *Bacillus subtilis* + *Pochonia* sp., were also the most efficient in reduction of the population of nematodes evaluated. For this reason, the combination of biological control agents can reduce the population of phytonematodes.
Keywords: Nematophagous fungi; Bioagents; Promoting.

Resumo
A soja é uma commodity bastante cultivada no Brasil e representa um dos principais produtos de exportação. Diversos fatores bióticos e abióticos têm limitado o aumento da produtividade dessa oleaginosa, dentre eles os nematoides associados à monocultura. Diante disso, objetivou-se com o presente trabalho avaliar o efeito de diferentes agentes biológicos no controle de fitonematoides na cultura da soja, via solo. Foi conduzido um experimento em Gurupi-TO em delineamento experimental de blocos ao acaso, com quatro repetições e 14 tratamentos, incluindo a testemunha. Os tratamentos foram compostos por agentes de biocontrole: *Paecilomyces* sp., *Trichoderma asperellum*, *Bacillus subtilis* e *Pochonia* sp., sendo avaliados isoladamente e em combinações. Os tratamentos contendo *Paecilomyces* sp. + *Bacillus subtilis* e a combinação de todos os microrganismos resultaram em maiores médias para a maioria das características quando comparados ao tratamento controle. Os tratamentos *Paecilomyces* sp. + *Bacillus subtilis* e *Paecilomyces* sp. + *Trichoderma asperellum* + *Bacillus subtilis* + *Pochonia* sp. UFT, também foram os mais

eficientes na redução da população de nematoides avaliada. Portanto, a combinação de agentes de controle biológico pode reduzir a população fitonematoides.
Palavras-chave: Fungos nematófagos; Agentes biológicos; Promoção.

Resumen

La soja es un producto ampliamente cultivado en Brasil y representa uno de los principales productos de exportación. Varios factores bióticos y abióticos han limitado el aumento de la productividad de esta oleaginosa, entre ellos los nematodos asociados al monocultivo. Por tanto, el objetivo del presente trabajo fue evaluar el efecto de diferentes agentes biológicos en el control de fitonematodos en soja, vía suelo. Se realizó un experimento en Gurupi-TO en un diseño de bloques completos al azar, con cuatro repeticiones y 14 tratamientos, incluido el testigo. Los tratamientos estuvieron compuestos por agentes de biocontrol: *Paecilomyces* sp., *Trichoderma asperellum*, *Bacillus subtilis* y *Pochonia* sp., siendo evaluados solos y combinados. Los tratamientos que contienen *Paecilomyces* sp. + *Bacillus subtilis* y la combinación de todos los microorganismos dieron como resultado medias más altas para la mayoría de los rasgos en comparación con el tratamiento de control. Los tratamientos *Paecilomyces* sp. + *Bacillus subtilis* y *Paecilomyces* sp. + *Trichoderma asperellum* + *Bacillus subtilis* + *Pochonia* sp. UFT, también fueron los más eficientes en reducción de la población de nematodos evaluados. Por lo tanto, la combinación de agentes de control biológico puede reducir la población de fitonematodos.
Palabras clave: Hongos nematófagos; Agentes biologicos; Promoción.

1. Introduction

Use Soybean (*Glycine max* (L.), species belonging to the Fabaceae family, is a prominent crop in the world grain scenario, widely cultivated in Brazil, representing one of the main export products, generating thousands of direct and indirect jobs, and boosting other sectors of the economy (Rocha et al., 2018). In the 2021/2022 harvest, Brazil, the largest producer, sowed 40.7 million hectares, an increase of 3.8% in area, with an expected production of 122.76 million tons (CONAB, 2022).

However, some biotic and abiotic factors have significant importance in the cycle of this oilseed, among them the polyphagous plant-parasitic nematodes existing in the soil (Bortolini et al., 2013). To the detriment that these can cause loss of production, and, consequently, the final quality of the product. (Costa et al., 2020). Among the species of nematodes relevant to the culture in Brazil, the genus *Pratylenchus*, considered one of the most harmful and common occurrences in the country (Cruz et al., 2020).

Therefor, to prevent or control polyphagous agents that affect soybean, different strategies can be used such as the biological control, which is the use one or more organisms to reduce or control the population of disease-causing agents in a crop, even being an ecologically desirable option (Machado & Costa, 2018).

Biological control is an alternative that allows the interaction of several natural enemies of plant-parasite nematodes, highlighting fungi, such as *Trichoderma* sp., which is an antagonist that can degrade chitin, this is how it controls nematodes acting on eggs and bacteria, such as *Bacillus* sp, acting as an antagonism, and producing lytic enzymes, siderophores, solubilizing phosphorus and fixing nutrients, making them excellent alternatives for the formulation of bioproducts (Santos et al., 2019)

Another prominent fungus, *Pochonia* sp., which is nematophagous, capable of colonize epiphytically and endophytically roots of several species of mono and dicots, promoting a symbiotic association with these plants (Bordallo et al., 2002). The developing of products based on this fungus has been applied in soybean cultivation areas, aiming at the management of nematodes. In addition to *Pochonia* sp., fungi of the genus *Paecilomyces*, have great nematicide potential, due to their saprophytic characteristics, they are easily established in the soil, capable of quickly parasitizing enormous amounts of eggs and female nematodes, such as *Meloidogyne* sp. and *Heterodera* sp. (Costa, 2015)

Therefore, the objective of the present study was to evaluate the effect of biological agents in the control of plant-parasitic in soybean, via soil.

2. Methodology

The study was carried out in the experimental area of University of Tocantins, University Campus of Gurupi, located in the South area of the state of Tocantins (11°43'45'' S, 49°04'07'' W, 280 m). The climate classification according to Köppen (1948), is Aw', characterized as humid tropical, rainy season in summer and dry season in winter. The average temperature throughout the year varies between 22 ºC and 28 ºC and the annual average rainfall between 1500 mm to 1600 mm.

The experience was conduced in polyethylene packages with a volume of 10 L. The soil used was collected in the 0 – 20 cm layer on the Boabá Farm in São Desiderio (S 12°34.555' W 46°08.405'), classified as dystrophic Red-Yellow Latosol, deep and with a clayey texture (EMBRAPA, 2013). For several years of cultivation, soybean plants showed characteristic symptoms of nematode infection in a dead spot, places from which the soil was collected.

For better physical condition of the soil, a mixture of soil + sand was used, in proportion 3:1 and maintenance fertilization, based on soil analysis.

The result of the physio-chemical analysis of the soil was: pH in $CaCl_2$ = 5.9; M.O (%) = 1.5; P (Mel) = 92.1 mg dm^{-3}; K = 90 mg dm^{-3}; Ca+Mg = 3.4 cmolc dm^{-3}; H+Al = 1.6 cmolc dm^{-3}; Al = 0.0 cmolc dm^{-3}; SB= 3.63 cmolc dm^{-3}; V = 69%; 450, 100 and 450 kg ha^{-1} of sand, silt and clay, respectively. 120 kg ha^{-1} of P_2O_5 in the form of simple superphosphate and 90 kg ha^{-1} of K_2O in the form of potassium chloride were applied, mixed in the soil preparation in a homogeneous way through handling with a hoe. The cultural treatments and irrigation were carried out according to the crop.

Prior to sowing, a small portion of soil was removed, and treatments were applied in the opening formed, returning the removed soil to the top after application.

The soybean cultivar used was M8808 IPRO, as it is widely sown in the region. Seed treatment was carried out by the plastic bag method, composed of insecticide (TIAMETOXAM, 300 ml 100 kg^{-1}), fungicide (CARBENDAZIM + TIRAM, 200 ml 100 kg^{-1}), commercial liquid inoculant (Bradyrhizobium japonicum strains SEMIA 5079 and SEMIA 5080; $7.2x10^9$ cells per ml product 100 ml 100 kg^{-1}) and pro-additives. Sowing was carried out on December 22, 2016 (according to the rain forecast in the municipality of Gurupi-TO). Two plants were kept per pot after thinning performed at 14 days after emergence (DAE).

The experimental design used was random blocks, with four repetitions and 14 treatments, including the control, totaling 56 plots, each plot consisting of three bags filled with infested soil, duly homogenized (Matsuo et al., 2012).

The treatments were composed of soil biocontrol agents via soil, described in Table 1 and the control, without application of nematode control. The active ingredients of the products used were: BIO NEM (Pa) = *Paecilomyces* sp.; QUALITY (Tr) = *Trichoderma asperellum*; RIZOS (Bs) = *Bacillus subtilis*; POCHONIA IN RICE (Pc) = *Pocho*nia sp. UFT. Active ingredients doses ranged from 1 = $2x10^9$ UFC ml^{-1}, 2 = $2x10^9$ UFC g^{-1} and 3 = $1x10^{10}$ UFC g^{-1}.

Table 1 - Treatments, doses of active ingredient and product used to control nematodes.

Treatments	Dose of i.a.	Dosage
1. Pa	1	1 L ha^{-1};
2. Tr	3	100 g ha^{-1}
3. Bs	2	200 mL ha^{-1}
4. Pc	2	80 mg ha^{-1}
5. Pa + Tr	1; 3	1 L ha^{-1}; 100 g ha^{-1}
6. Pa + Bs	1; 2	1 L ha^{-1}; 200 mL ha^{-1}
7. Pa + Pc	1; 2	1 L ha^{-1}; 80 mg ha^{-1}
8. Pa + Tr + Bs	1; 3; 2	1 L ha^{-1}; 100 g ha^{-1}; 200 mL ha^{-1}
9. Pa + Tr + Bs + Pc	1; 3; 2; 2	1 L ha^{-1}; 100 g. ha^{-1}; 200 mL ha^{-1}; 80 mg ha^{-1}
10.Tr + Bs	3; 2	100 g ha^{-1}; 200 mL ha^{-1}
11. Tr + Pc	3; 2	100 g ha^{-1}; 80 mg ha^{-1}
12. Tr + Bs + Pc	3; 2; 2	100 g ha^{-1}; 200 mL ha^{-1}; 80 mg ha^{-1}
13. Bs + Pc	2; 2	200 mL ha^{-1}; 80 mg ha^{-1}
14. Treatments Control		

Pa: *Paecilomyces* sp.; Tr: *Trichoderma asperellum*; Bs: *Bacillus subtilis*; Pc: *Pochonia* sp. Source: Authors.

With the exception for the treatment POCHONIA IN RICE (Pc), the others are commercial products and the dosage used was as recommended by the manufacturers. The Pc treatment was produced at the Microbiology Laboratory (MicroBio) of the University of Tocantins – Gurupi Campus and had its dosage established based on previous experiments. To prepare the treatment, the fungus was placed to grow in a petri dish containing PDA (Potato Dextrose Agar) and incubated at 25°C ± 2 °C with a photoperiod of 12 hours for 21 days.

Polypropylene bags containing 300 g of rice and 300 mL of distilled water were used, which were autoclaved at 121 °C for 1 hour. After cooling, the rice was inoculated with six disks of 5 mm in diameter, containing mycelia and spores of the fungus and PDA medium, and incubated in a BOD – type growth chamber with a temperature of 25 °C ± 2°C and a photoperiod of 12 hours, for 21 days. Every two days, the rice substrate was turned. The inoculated rice was added to the soil as it was. The concentration of Pochonia sp. was determined by the serial dilution method through the qualification of Colony Forming Units (CFU).

However, at 90 DAE, at physiological stage R6, the following characteristics were evaluated: plant height, measured from the neck of the plants to the highest end of the leaves using a measuring tape graduated in cm; stem diameter, obtained with the aid of a digital caliper expressed in cm, determined from the stem region; number of stem, obtained by directly counting the number of stems of each plant; fresh mass of the aerial part, obtained by directly weighing the aerial part; shoot dry mass and root dry mass, obtained by drying the plants in an oven at 65 °C ± 2 °C for 48 hours, until reaching constant mass; total dry mass, obtained by the sum of the dry mass of the arial part ant the dry mass of the roots; relative efficiency, comparative plant in treatments that received microorganisms in relation to those that did not.

The relative efficiency result was converted to percentage through the equation: RE(%) = (DMAPIP / DMAPUP) x 100, where: ER(%) = Relative Efficiency in percentage; DMAPIP = Dry mass of the aerial part of the inoculated plants; DMSUP = Dry mass of aerial part of uninoculated plants.

The samples sent for analysis of second stage juveniles and number of cysts were also collected at 90 DAE and composed by mixing the four replications, where each composite sample had 500 g of soil and 100 g of roots.

The method used to extract the second stage juveniles (J2) from the soil was centrifugal flotation in sucrose solution (Jenkins, 1964). For the extraction of J2 from the roots, the blender technique was used (Bonetti & Ferraz, 1981). For the extraction of the cysts, the methodology proposed by Shepherd (1970) was used.

Subsequently, the nematodes population present in the soil was classified and divided into low risk of loss (LRL), medium risk of loss (MRL) and high risk of loss (HRL) according to Silva and Inomoto (2015).

Table 2 - Reference values used as limit for population density of nematode species pathogenic to soybean in each 100 cm^3 of soil.

Nematode species	Low risk of loss	Medium risk of loss	High risk of loss
Pratylenchus brachyurus	< 100 species	100 to 400 species	> 400 species
Heterodera glycines	No viable cyst in sandy soil; or 0 to 2 viable cysts for soil with more than 35% clay	1 viable cyst in sandy soil; or 3 to 4 viable cyst for soil with more than 35% clay	> 2 viable cysts in sandy; or > 5 viable cysts for soil with more than 35% clay
Meloidogyne javanica and *M. incognita*	< 25 species	25 to 100 species	> 100 species

Source: Adapted de Silva and Inomoto (2015).

At 120 DAE, at physiological stage R9, the following characteristics were evaluated: height of insertion of the first pod, obtained by measuring the length between the neck of the plant until the insertion of the first pod, using a measuring tape graduated in cm; number of pods per plant and number of grains per pod, determined through the direct counting of these indicators in the sample plants; mass of 100 g, determined on an analytical scale accurately to three decimal places; and productivity, determined based on the grain yield of the plants harvested in the experiment with humidity correction to 13%.

After, analysis of variance was performed using the F test for all characteristics and the means were compered using the Scott-Knott test at 5% probability, using the computer application SISVAR 5.6 (Ferreira, 2008).

3. Results and Discussion

Analysis of variance showed results for most treats, with the exception of number of grains per height, diameter and grain yield. For plant height, aerial part dry mass, relative efficiency was significance at the 5% probability level ($p<0.05$). Other characteristics showed significance at the level of 1% probability ($p<0.01$).

For plant height characteristic (Figure 1), treats Pa + Bs, Pa + Bs +Tr + Pc and Bs + Pc were the components with the highest average and were classified in the group with the lowest average for the characteristic height of insertion of the first pod, demonstrating the ability of the plant to present a greater number of pods in its total length, a desirable characteristic for flat and well-prepared soils, since more inclined or poorly prepared soils require at least 10 cm in height, in order to reduce losses during harvest.

The treats containing *Bacillus subtilis* were the ones that highlighted the characteristics height of plants, which may be associated with the ability of the microorganism to produce plant hormones, such as auxins, cytokinins and gibberellins, fix nitrogen and prevent against deleterious caused by environmental stresses (Sivasakthi et al., 2014), however, its effect is potentiated when in association with other microorganisms, since when its was evaluated alone, it was not able to obtain a higher average.

Analyzing the number of stems (Figure 1), the fact that the control treat is a component with the highest average can be explained by the non-infection of nematodes present in the soil until the moment of evaluation of the characteristic, not being able to affect the average of the same, since they can attack the plant in different stages of development.

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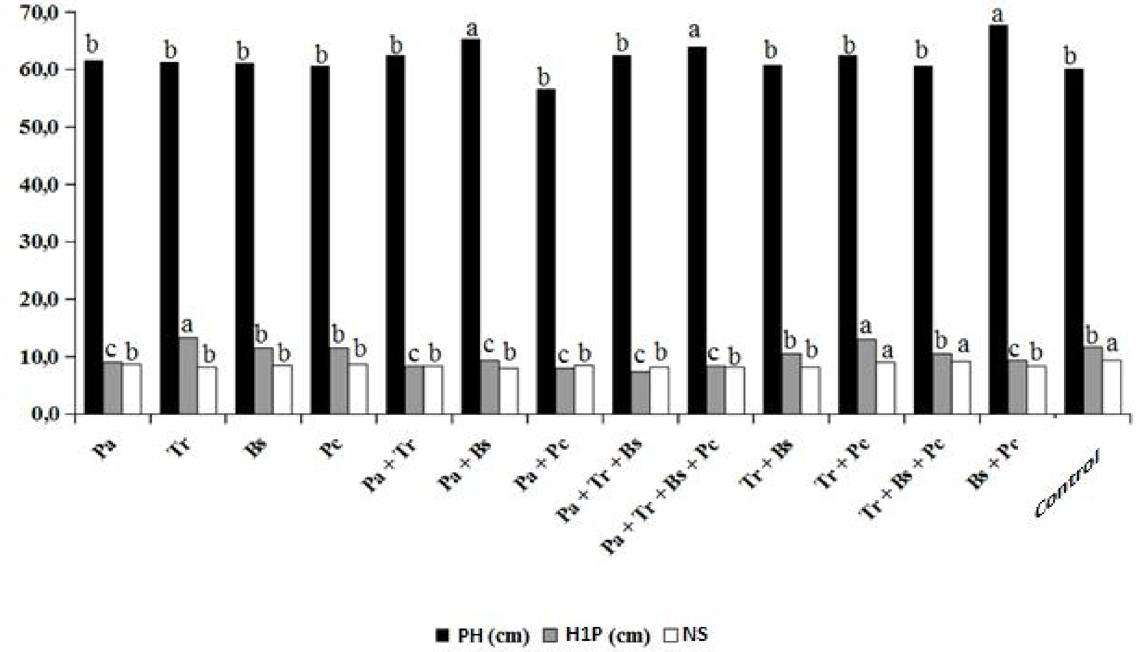
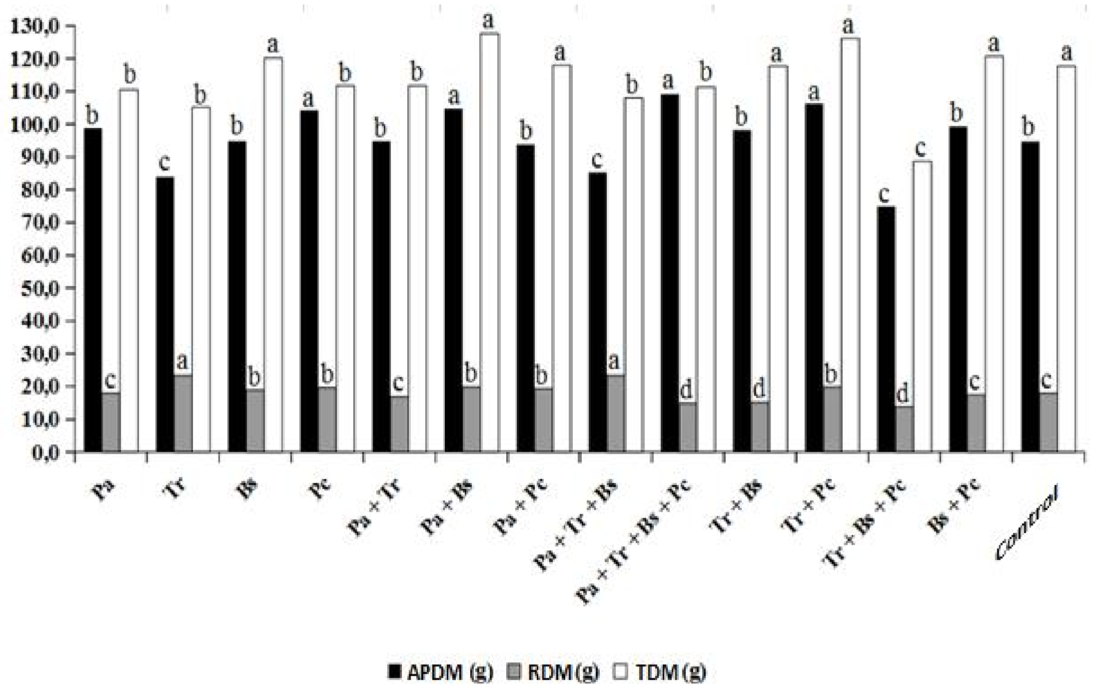
Figure 1 - Average of characteristics plant height (PH), height of first pod (H1P), number of stems (NS) of treats evaluated in soil containing root-knot nematodes, cyst and lesions in soybean. 2016, Gurupi – TO.



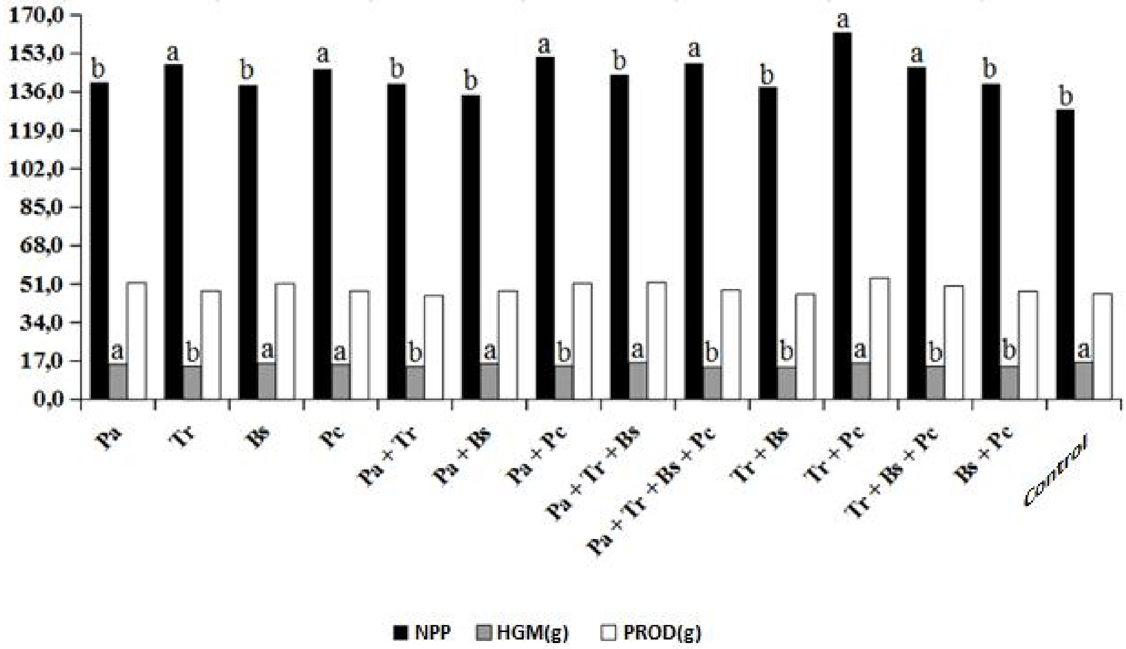
Pa: *Paecilomyces* sp.; Tr: *Trichoderma asperellum*; Bs: *Bacillus subtilis*; Pc: *Pochonia* sp. * Means followed by the same letter within the analyzed variable do not differ statistically from each other, by the Tukey test 5%. Source: Authors.

Figure 2 - Average of the characteristics of aerial part dry mass (APDM), root dry mass (RDM), total dry mass (TDM) of the treats evaluated in soil containing root-knot, cyst and lesions nematodes in soybean. 2016, Gurupi – TO.



Pa: *Paecilomyces* sp.; Tr: *Trichoderma asperellum*; Bs: *Bacillus subtilis*; Pc: *Pochonia* sp. * Means followed by the same letter within the analyzed variable do not differ statistically from each other, by the Tukey test 5%. Source: Authors.

Figure 3 - Averages of the characteristics number of pods per plant (NPP), hundred-grain mass (HGM) and productivity (PROD), of the treats evaluated in soil containing root-knot, cyst and lesions nematodes in soybeans. 2016, Gurupi-TO.



Pa: *Paecilomyces* sp.; Tr: *Trichoderma asperellum*; Bs: *Bacillus subtilis*; Pc: *Pochonia* sp. * Means followed by the same letter within the analyzed variable do not differ statistically from each other, by the Tukey test 5%. Source: Authors.

However, *Trichoderma asperellum* e *Pochonia* sp., treats components with higher averages for number of stems, fungi capable of colonizing the rhizosphere and establish association with the plant, increasing its plant health and improving its tolerance to environmental stresses (Schäfer et al., 2009). Therefore, despite the existence of nematodes in the roots and in the soil, the plants were still able to develop well.

For the characteristic aerial part fresh mass (Figure 2), *Pochonia* sp. stands out, integrating the treats with the highest average. The better development of the aerial part allows a better photosynthetic rate, which may imply in more photo assimilates translocated to the growing or reserve organs in the following stages (Szilagyi-Zecchin et al., 2015), resulting, for example, in heavier grains.

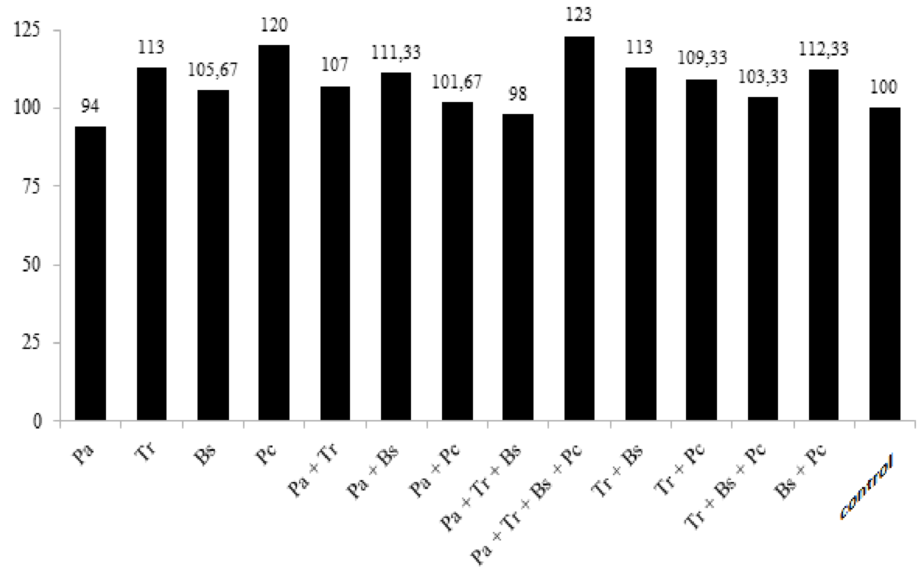
As for root dry mass (Figure 2) it is observed that the treat combining all microorganisms, *Paecilomyces* sp., *Trichoderma asperellum*, *Bacillus subtilis* and *Pochonia* sp., despite having presented a higher average for the dry mass characteristic, did not presented a good development of roots. This fact can be explained by the additional input of indole compounds produced by microorganisms, modifying the endogenous amounts in plants to an optimal level or above the optimum, resulting in the induction or inhibition of root development (Szilagyi-Zecchin et al., 2015).

For the characteristic total dry mass (Figure 2), the treat Tr + Bs + Pc presented the lowest average. This characteristic reflects the other aerial part and root, where the treat was the only member of the group with the lowest average.

The treats made up the group with the highest average for the characteristic number of pods per plant (Figure 3). The pods develop in the axils of each stem of the plant, corroborating the result of the number of stems, since these treats also formed the group with the highest average for this characteristic.

For the relative efficiency characteristic (Figure 4), the group with the highest average was formed by the treats Tr, Pc, Pa + Bs, Pa + Tr + Bs + Pc, Tr + Bs, Tr + Pc e Bs + Pc, with values varying from 123 to 109.33%.

Figure 4 - Relative efficiency (%) of 13 treats containing biocontrol microorganisms, plus the control treat, in soil containing root-knot, cyst and lesion nematodes, in soybean. 2016, Gurupi – TO.



Pa: *Paecilomyces* sp.; Tr: *Trichoderma asperellum*; Bs: *Bacillus subtilis*; Pc: *Pochonia* sp. Source: Authors.

The population values for the three main genera of nematodes present in soil (Table 3) were classified according to Silva and Inomoto (2015) in three levels of risk of loss: low (LRL), medium (MRL) and high (HRL). The number of second instar juveniles (J2) for *Meloidogyne* sp. was HRL for all treats.

Table 3 - Plant-parasitic nematodes species present in roots and soil, and population in second instar juveniles for Meloidogyne sp. (M-J2) and Pratylenchus brachyurus (Pb-J2); and second-stage juvenile population number of viable cysts (VC) and non-viable (NVC) cysts of Heterodera glycines (Hg-J2/VC/NVC). Also, classification as High risk of loss (HRL), Medium risk of loss (MRL) and Low risk of loss (LRL). 2016, Gurupi – TO.

	Roots (10 g^{-1})			Soil (100 cm^{-3})		
Treatments	M-J2	Pb-J2	Hg-J2/VC/NVC	M-J2	Pb- J2	Hg-J2/VC/NVC
Pa	240	504	0	323 HRL	114 MRL	0 LRL
Tr	253	690	0	864 HRL	54 LRL	54; 3 VC and 1 NVC HRL
Bs	253	276	0	627 HRL	114 MRL	19; 2 VC and 0 NVC LRL
Pc	120	520	0	225 HRL	30 LRL	30; 1 VC and 4 NVC LRL
Pa+Tr	147	420	42	646 HRL	51 LRL	34; 0 VC and 1 NVC LRL
Pa+Bs	153	414	0	418 HRL	57 LRL	38 LRL
Pa+Pc	851	575	0	798 HRL	0 LRL	19 LRL
Pa+Tr+Bs	528	432	44	342 HRL	180 MRL	180 MRL
Pa+Tr+Bs+Pc	161	308	0	936 HRL	36 LRL	36; 0 VC E 1 NVC LRL
Tr+Bs	1680	936	0	1071HRL	168 MRL	42 LRL
Tr+Pc	693	462	42	1300 HRL	40 LRL	20; 2 VC and 1 NVC LRL
Tr+Bs+Pc	96	576	0	400 HRL	120 MRL	60; 2 VC and 5 NVC LRL
Bs+Pc	84	315	0	513 HRL	76 LRL	19; 3 VC and 1 NVC HRL
Control treatment	84	252	0	529 HRL	46 LRL	69; 3 VC and 4 NVC HRL

Pa: *Paecilomyces* sp.; Tr: *Trichoderma asperellum*; Bs: *Bacillus subtilis*; Pc: *Pochonia* sp. Source: Authors.

Analyzing the presence of plant-parasitic nematodes in the soil, for *Heterodera glycines*, there was a low risk of loss for most treats, with the exception of the Pa + Tr + Bs treat that presented MRL, and the Tr, Bs + Pc and control treat that presented HRL. Significant reduction of *Heterodera glycines* hatching was observed I vitro after treat with *Bacillus subtilis* (Araújo et al., 2005). The use of different *Bacillus subtilis* isolated inhibited the population of nematodes from 28 to 42% in the bean crop, also increasing root nodulation (Mutua et al., 2011). For *Pratylenchus brachyurus* treat also presented LRL classification.

As for roots, it is observed that the control treat showed low infection compared to other treats. This can be explained by the population variation of nematodes in the collected soil where even with the homogenization performed prior to filling the pots, the same proportion of pathogens was not added to the pots. Even so, it can be said that the treat Pa + Bs e Pa + Tr + Bs + Pc were the most efficient in controlling the nematodes population, both at the root and soil level.

The fungus Pochonia chamydosporia is considered one of the most promising agents for the biological control of nematodes. It is a facultative parasite of eggs and females of cyst nematodes and causes of root lesions (Podestá et al., 2013). Thricoderma species on the other hand, produce enzymes such as polysaccharide-degrading, proteases and lipases. In nature, such enzymes are involved in the degradation of the cell wall of phytopathogens (Hjelford et al., 2001).

In general, endotoxins produced with the reproductive cycle of nematodes, reducing oviposit and hatching of juveniles, in addition to inhibiting the initial penetration of plant parasitic nematodes by altering specific root exudates, such as polysaccharides and amino acids, which modify the behavior of nematodes (Sidhu, 2018).

The combination of biological control agents can increase the efficiency of such agents, especially when they act at different stages of the pathogen's life cycle (Podestá et al., 2013), explaining the control exercised by treats with product combination.

4. Conclusion

The use of treats containing *Paecilomyces* sp. + *Bacillus subtilis* and the combination of all microorganisms resulting in higher averages for the characteristics plant height, aerial part fresh mass, shoot dry mass, total dry mass, number of pods per plant, one hundred grain mass and relative efficiency when compared to the control treat.

The treats *Paecilomyces* sp. + *Bacillus subtilis* and *Paecilomyces* sp. + *Trichoderma asperellum* + B*acillus subtilis* + *Pochonia* sp. UFT, were also the most efficient in promoting lower density of the population of nematodes evaluated.

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