

Agronomic efficiency of the inoculant FT10 (*Trichoderma asperelloides*) on four lettuce varieties

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Abstract

The present study aimed to evaluate the agronomic efficiency of FT10 (*Trichoderma asperelloides*) on four lettuce varieties development and productivity throughout the year. Four greenhouse experiments were carried out to verify the effect of three doses of the bio-product on plant growth and yield on loose-leaf, butterhead, iceberg and salad bowl cultivars. A randomized block design was used, with six replications. The treatments were: 1) control, 2) 300 mL ha⁻¹ of the commercial inoculant ICB Nutrisolo® *Trichoderma*, 3) 0.05 kg ha⁻¹ of the inoculant FT10, 4) 0.10 kg ha⁻¹ of FT10, and 5) 0.15 kg ha⁻¹ of FT10. The plant height and number of leaves were assessed 35 days after transplanting (DAT) of lettuce seedlings and the plant dry weight and yield were evaluated 42 DAT. The data obtained were compared with Duncan's test. The majority of the evaluated parameters on lettuce varieties were found to be affected by the higher doses of FT10. Significant increases in yield in up to 21% for loose-leaf and 16% for butterhead were observed with the dose of 0.05 kg ha⁻¹ and 22% for iceberg and salad bowl with the dose of 0.10 kg ha⁻¹ of the bioproduct. It can be concluded that FT10 has the potential to promote plant growth and increase the productivity of lettuce cultivars if applied an appropriate dose of the product.

Keywords: inoculant, biostimulant, beneficial microorganism, *Trichoderma*

Introduction

The need for higher agricultural yields has led to the excessive use of inorganic fertilizers, creating environmental problems and compromising food security and quality. The use of beneficial microorganisms and biostimulants to maintain production levels and reduce dependency on fertilizers is a highly desirable strategy for ensuring sustainable agriculture (Azarmi et al., 2011; Ye et al., 2020).

Beneficial microorganisms are capable of colonizing plant roots and improving their growth and productivity. Among plant growth-promoting fungi, *Trichoderma* species are globally recognized and used in a wide variety of agricultural crops (Bettiol et al., 2009). These free-living symbionts influence plant health, growth and production. Their effect on plant development is attributed to different mechanisms of action, such as increasing minerals solubilization and uptake of nutrients,

plant hormone synthesis, volatile organic compound production, improved abiotic stress tolerance and the suppression of harmful microorganisms (Machado, 2012; Fiorentino et al., 2018).

According to Lelio et al. (2021), *Trichoderma* species or strains can have a diverse impact on different crop species, or even genotypes of the same crop. Furthermore, the crop genotype influences significantly the rhizosphere colonization by *Trichoderma* strains. Therefore, the selection of a *Trichoderma* strain suitable to be widely used in agriculture depends not only on targeted use, but also on the ecological adaptability of the strain to the various environments, availability of water and nutrients, climatic conditions and crop genotypes that influence its effectiveness in the field.

Leafy vegetables represent 35% of the area of vegetable species produced in Brazil, and lettuce stands out for being the most consumed and produced due to

their culinary characteristics and cultural acceptance, besides it can be produced all year long. The production volume is over than 1.5 million tons a year and the estimated retail market reaches 8 billion of BRL (R\$). Besides their economic importance, this crop presents an important social role since it is cultivated mainly by micro and small farmers (Souza et al., 2019; Dalastra et al., 2020).

In this context, the use of *Trichoderma* species as inoculants could contribute to maintain or improve the lettuce productivity and decrease the need for synthetic fertilizers.

This study aimed to evaluate the agronomic efficiency of three doses of FT10 (*Trichoderma asperelloides*) in the development and production of lettuce's cultivars throughout the seasons.

Material and Methods

The four greenhouse experiments were conducted in the 2019/2020 growing season, in the city of Ponta Grossa, Paraná state (PR), Brazil (25°09'45.95"S, 50°11'23.63"W, altitude of 807 m). The climate in this area is classified as humid subtropical (Cfb, according to Köppen's classification), with an average annual temperature of 17.5°C and rainfall of 1495 mm. The climatological data presented in the table 5 were collected during the execution of the experiments, from July of 2019 to May of 2020, at the meteorological station located in the experimental farm area of 3M.

The plant growth commercial substrate used in the experiments was from the Carolina Soil Company, with the following composition: sphagnum moss, expanded vermiculite, lime, gypsum and NPK fertilizer (traces), the potential of hydrogen (pH): 5.5 +/- 0.5, electrical conductivity (EC): 0.7 +/- 0.3, density: 145 kg/m³, water holding capacity (WHC): 55%, maximum moisture content: 50%, reactivity – stable inert product, nontoxic, physical nature: solid.

Four studies were carried out with loose-leaf lettuce (cv. 'Amanda'), in July, September, November, and April of the 2019/2020 growing season.

Parallely, in September was also included in the experiment the butterhead (cv. 'Regiane') variety, in November the iceberg (cv. 'Angelina') variety and in April the salad bowl (cv. 'mimosa') variety, to compare the efficiency of the FT10 inoculant in other lettuces types.

Botanical varieties of lettuce used in the experiments: 1. Amanda Loose-leaf cultivar is large, crisp and vigorous, with excellent leaf health, vivid light green coloring, high yields, low wastage and excellent postharvest consistency; 2. Regiane butterhead cultivar has large, bright green cone-shaped plants, with a

vigorous root system and an average cycle of 56 days (early); 3. iceberg cultivar 'Angelina' has bright green leaves, vigorous, uniform plants with a well-formed base and dense head, and an average cycle length of 70 days and 4. Salad bowl (cv. 'Mimosa') are large, light green and easily marketable, with ruffled leaves and an average growth cycle of 56 days.

Five treatments were used: 1) control absolute, 2) 300 mL ha⁻¹ of the agricultural inoculant ICB Nutrisolo® (BioAgritec Ltda, based on *T. harzianum*, *T. asperellum* and *T. koningiopsis* at the concentration of 1 x 10¹¹-CFU mL⁻¹, liquid formulation) as a standard commercial product, 3) 0.05 kg ha⁻¹ of the inoculant FT10 (water soluble powder formulation based on *Trichoderma asperelloides* at the concentration 1 x 10¹⁰ CFU.g⁻¹), 4) 0.10 kg ha⁻¹ of FT10, and 5) 0.15 kg ha⁻¹ of FT10. The treatments were applied to the planting furrow at a spray volume of 150 L ha⁻¹, before the one-month-old seedlings of the lettuce cultivars were transplanted, using a CO₂ pressurized backpack sprayer, equipped with a bar and spray nozzle (JLP 110.02 ADI), at a working pressure of 35 kgf cm².

A completely randomized design was used, with six replications, each plot consisting of a 5-liter pot with five lettuce plants.

Plant height was determined with a tape measure at 7, 14, 21, 28 and 35 days after transplanting (DAT) in two plants per plot, and the number of leaves on the same days, in three plants per plot. At 42 DAT, one plant was randomly selected to assess shoot (SDW) and root dry weight (RDW), after being dried in an oven (Solab SL-100/1080) at 65°C, until a constant weight. The yield was calculated at 42 DAT, by collecting three plants from each experimental plot and measuring fresh weight in grams, converted into kg ha⁻¹.

The survival of *Trichoderma* spp. isolates was assessed at the end of each experiment by randomly collecting samples of the substrate from each plot of the treatments. The substrate was analyzed by serial dilution, followed by plating of aliquots of the dilutions in commercial Potato Dextrose Agar (PDA) medium containing antibiotics (0.17 g of ampicillin and 0.05 of pentabiotic per 1 L of culture medium), and Triton X-100 as a reducing agent. Three repetitions were performed for each dilution. The fungal cultures were placed in a BOD incubator for three days at 25 ± 2°C, under a 12-hour light-dark photoperiod. The number of colony-forming units (CFUs) per gram of substrate was counted.

The data of all experiments were submitted to analysis of variance and, when significant, means were compared by Duncan's test at 5% significance. When

the F-test was not significant at 5% but significant at 10%, the treatment means were compared by Duncan's test at 10% significance, using SASM-Agri® software [System for analysis and mean separation in agricultural assays - Canteri et al. (2001)].

Results and Discussion

A significant increase in the number of leaves was observed at an FT10 dose of 0.05 kg ha⁻¹ for cv. 'Amanda' in 1st (08/21/2019), 3rd (12/10/2019) and 4th (05/12/2020) experiments when compared to the control treatment (Table 1). At doses of 10.0 and 0.15 kg ha⁻¹, the number of leaves rose significantly in all four experiments. For the standard commercial product (ICB Nutrisolo *Trichoderma*) and control, no significant increases were observed in the 2nd (10/11/2019) and 3rd experiments.

There were no significant differences in loose-leaf lettuce height in the 2nd and 4th experiments; however, significant variations were observed for the 1st and 3rd

experiments. The largest increases in plant height (4.2 to 5.5%) were obtained for the doses of 10.0 and 0.15 kg ha⁻¹ of FT10 and with ICB Nutrisolo *Trichoderma* (Table 1). In the 3rd experiment the greatest increase (14.5%) was observed for 0.15 kg ha⁻¹ of FT10 assessed on 12/10/2019, with no statistical differences with the other treatments.

For iceberg lettuce (cv. 'Angelina'), evaluated on 10/11/2019, the number of leaves rose significantly (13.3%) at FT10 dose of 0.10 kg ha⁻¹. A significant increase of 8.3% also was observed for salad bowl lettuce (cv. 'Mimosa') at 0.5 kg ha⁻¹ on 10/12/2019 and the highest value obtained for butterhead lettuce (cv. 'Regiane') on 05/12/2020 was 8.9% at FT10 dose of 0.15 kg ha⁻¹ always comparing with the control treatment.

With respect to plant height, no significant difference among the treatments and control were observed for the iceberg, salad bowl and butterhead varieties (Table 2).

Table 1. Effect of *Trichoderma* spp. on the average number of leaves and plant height of looseleaf lettuce, cv. 'Amanda', 35 days after transplanting (DAT), in four experiments at different times of year. Ponta Grossa- PR. 2019/2020 growing season.

Treatments	Doses***	08/21/2019		10/11/2019		12/10/2019		05/12/2020	
		Height (cm)**	No. of leaves*	Height (cm)**	No. of leaves*	Height (cm)*	No. of leaves*	Height (cm)*	No. of leaves*
Absolute Control	-	20,35c	12,97d	22,35a	10,12b	20,57b	10,45b	27,55a	12,60b
ICB Nutrisolo <i>Trichoderma</i>	300	21,20ab	14,48c	21,37a	10,00b	22,45ab	11,50a	28,48a	13,30ab
FT10	0.05	20,47bc	15,55b	24,47a	11,28ab	22,05ab	11,63a	28,38a	13,67a
	0.10	21,47a	16,45a	23,81a	11,50a	22,23ab	11,38a	29,35a	13,63a
	0.15	21,32a	14,33c	23,96a	11,67a	23,55a	11,98a	28,55a	13,95a
Mean	-	20,96	14,76	23,19	10,91	22,17	11,39	28,46	13,43
C.V.	-	3,84	4,68	9,61	9,67	6,63	6,62	6,71	5,21

*Significant according to the F-test at 5% and **10% probability level

Means followed by the same letter in the column do not differ according to Duncan's test;

C.V.(%): Coefficient of Variation

*** Dosage unit: ICB Nutrisolo in mL.ha⁻¹ and FT10 in kg.ha⁻¹

Table 2. Effect of different doses of two *Trichoderma* spp.-based products on the number of leaves and plant height of iceberg cv. 'Angelina', curly endive cv. 'Mimosa' and butterhead cv. 'Regiane' lettuce, 35 days after transplanting (DAT). Ponta Grossa - PR. 2019/2020 growing season.

Treatment	Doses***	Iceberg		Curly endive		Butterhead	
		11/10/2019	10/12/2019	11/10/2019	10/12/2019	12/05/2020	12/05/2020
		Height (cm)**	No. of leaves*	Height (cm)	No. of leaves*	Height (cm)	No. of leaves*
Absolute Control	-	23,77a	11,62b	23,95a	15,90b	26,15a	17,65b
ICB Nutrisolo <i>Trichoderma</i>	300	24,65a	11,57b	24,77a	16,43ab	27,03a	18,50ab
FT10	0.05	23,42a	12,70ab	24,38a	15,95b	26,08a	17,57b
	0.10	24,72a	13,17a	24,65a	16,83ab	27,33a	18,53ab
	0.25	25,37a	12,98a	25,30a	17,22a	27,10a	19,22a
Mean	-	24,38	12,41	24,61	16,47	26,74	18,29
C.V.	-	6,76	7,72	7,13	4,76	4,71	4,1

*Significant according to the F-test at 5% and **10% probability level

Means followed by the same letter in the column do not differ according to Duncan's test;

C.V.(%): Coefficient of Variation

*** Dosage unit: ICB Nutrisolo in mL.ha⁻¹ and FT10 in kg.ha⁻¹

The *Trichoderma* spp. isolates had a greater impact on the number of leaves than plant height in all

four lettuce varieties studied (Table 1 and Table 2). The number of leaves rose up to 26.8, 15.3, 14.6 and 10.7%

in loose-leaf lettuce for FT10 dose of 0.15 kg ha⁻¹ in July, September, November, and April, respectively, and in 13.3, 8.3 and 8.9% for iceberg, salad bowl and butterhead lettuce at 10.0 or 0.15 kg ha⁻¹ FT10 doses.

According to Oliveira et al. (2004), the number of leaves and fresh weight of lettuce plants may be influenced by factors such as the cultivar, photoperiod and temperature in the growing environment, as observed here in experiments with different lettuce cultivars and varieties in four different seasons. The largest number of leaves obtained at 35 DAT resulted in a higher yield at the end of the experiment, as reported by Araújo et al. (2011).

Partially corroborating the findings obtained here, previous studies report the positive effect of *Trichoderma* on plant height and number of leaves. Martins Filho et al. (2019) found that *Trichoderma aureoviride* inoculation associated with organic fertilizer (cattle manure) significantly improved lettuce height and number of leaves in relation to the control, resulting in increases of more than 131% at the end of the crop cycle. Chaves (2015) reported a significant rise in the height and number of leaves of 'Elba' and 'Solaris' loose-leaf lettuce seedlings treated with three *Trichoderma* spp. isolates 30 days after sowing. Silva et al. (2015) studied 12 *Trichoderma* spp. isolates and observed a 34% increase in the height of butterhead lettuce (cv. 'Regina') at 21 days DAT, while Pereira et al. (2019) found that treatment with *Trichoderma* spp. resulted in respective gains of 45 and 14% in the height and the number of leaves of iceberg lettuce (cv. 'Maureen') at 40 DAT.

The effect of *Trichoderma* spp. in promoting plant growth and yield in lettuce and other agricultural crops has been reported by several researchers (Chagas et al., 2017; Fiorentino et al., 2018; Pereira et al. 2019).

The shoot and root dry weight and yield results of four experiments with the Amanda loose-leaf lettuce cultivar are shown in Table 3.

For the experiment in which transplanting occurred in July and assessment on 08/29/2019, there were significant differences in SDW in relation to the control, with increases of 48.2 and 21.3% for FT10 doses of 10.0 and 0.5 kg ha⁻¹, respectively. It is important to note that the increases obtained resulted in higher yields, with the greatest increase (11.5%) recorded for 0.10 kg ha⁻¹ of FT10, followed by ICB Nutrisolo *Trichoderma* (10.2%). There was no significant difference between treatments for root dry weight in this experiment (Table 3).

In the second experiment, with transplantation in September and assessment on 10/18/2019, SDW rose

by 69.5 and 61.7% with 10.0 and 0.15 kg ha⁻¹ of FT10, respectively, representing a significant difference in relation to the control, but not to the other treatments. These gains led to higher yields, albeit only statistically significant at a dose of 0.05 kg ha⁻¹. Once again, root dry weight remained unchanged, regardless of the dose applied (Table 3).

In the third experiment was observed a variation in shoot dry weight between treatments and a significant difference at FT10 dose of 0.15 kg ha⁻¹, and a 24.7% increase in SDW was observed in relation to the control (Table 3).

The highest yields were obtained for 10.0 and 0.15 kg ha⁻¹ of FT10 and ICB Nutrisolo *Trichoderma* application, with respective increases of 15, 21 and 17% in these treatments when compared to the control. Unlike the previous experiments, RDW rose significantly (27.8%) at 0.10 kg ha⁻¹ of FT10.

In the fourth experiment FT10 application caused changes in the RDW and SDW of looseleaf lettuce, with non-significant increases of 14 and 19% in the latter at doses of 10.0 and 0.15 kg ha⁻¹, respectively, in relation to the control treatment. The highest significant yield (14%) was obtained for 0.15 kg ha⁻¹ of FT10 (Table 3).

Similar SDW percentages were recorded for the iceberg, salad bowl and butterhead varieties (20.2, 26.7 and 17.2%), with the largest increases observed at 0.15 kg ha⁻¹ of FT10 (Table 4).

The average yield of 9.5 metric tons obtained in the four experiments with greenhouse-grown loose-leaf lettuce in different seasons of the year partially agrees with results found in the literature. Torales et al. (2014) reported yields between 8.56 and 11.28 t ha⁻¹ for loose-leaf lettuce grown in the field, depending on the cultivar and plant spacing used, while Gonçalves et al. (2017) obtained higher values of 16.38, 4.37, 16.69 and 17.72 tons ha⁻¹ for the Itapuã, Paola, Vera and Verônica cultivars, respectively, under greenhouse conditions.

As previously mentioned, climate conditions affect the growth and performance of lettuce, with the best yields occurring at cooler times of the year, at ideal temperature and relative humidity ranges of 18 to 23°C and 60 to 75%.

The Ponta Grossa region (PR), where this study was conducted, exhibits similar climate conditions, as shown in Table 5. According to the climate data obtained, the temperature was only below the optimal range for lettuce in the experiment carried out in July, with an average of 14°C, which explains the lower yield recorded during this period. The average temperatures

in the remaining experiments were within the ideal range for lettuce production. Additionally, yield is highly dependent on the movement of nutrients within plants,

with rapid crop development generally accompanied by increased nutrient absorption (Cavalheiro et al., 2015; Silveira, 2016).

Table 3. Root and shoot dry weight (in g) and yield (kg ha⁻¹) of looseleaf lettuce, cv. Amanda, treated with different doses of *Trichoderma* spp.-based products at different times of year, under greenhouse conditions. Ponta Grossa - PR. 2019/2020 growing season.

Treatments	Doses	08/28/2019			10/18/2019			12/17/2019			05/19/2020						
		Dry weight		Yield	V	Dry weight		Yield	V	Dry weight		Yield	V				
		(g)	(g)	(kg.ha ⁻¹)* (%)	(g)	(g)	(kg.ha ⁻¹)* (%)	(g)	(g)	(kg.ha ⁻¹)* (%)	(g)	(g)	(kg.ha ⁻¹)* (%)				
	Root	Shoot*	Root*	Shoot*	Root*	Shoot*	Root*	Shoot*	Root**	Shoot*	Root**	Shoot*					
Absolute Control	-	0,67a	24,62d	7630c	-	3,37a	22,47b	9110b	-	1,94b	25,52b	9480b	-	1,25b	22,68a	8984b	-
ICB Nutrisolo	300	0,72a	32,07b	8410ab	10	1,98a	30,29ab	9160b	1	2,25ab	29,28ab	11065a	17	1,41b	26,96a	9555ab	6
Trichoderma																	
	0.05	0,70a	26,92cd	7895bc	3	3,00a	30,23ab	10875a	19	1,97b	28,50ab	9820b	4	1,28b	23,29a	9002b	0
	0.10	0,72a	36,48a	8510a	12	3,93a	38,08a	9365b	3	2,48a	29,85ab	10895a	15	1,35b	27,01a	9483ab	6
	0.15	0,74a	29,87bc	8005abc	5	3,78a	36,33a	10445ab	15	2,35ab	31,83a	11490a	21	1,75a	26,14a	10261a	14
Mean	-	0,71	29,99	8090,00	3,21	31,48	9791	2,20	28,99	10550	1,41	25,21	9457				
C.V.	-	12,43	12,09	5,46	44,50	28,09	10,63	15,12	13,99	7,45	22,48	17,13	6,56				

*Significant according to the F-test at 5% and **10% probability level

Means followed by the same letter in the column do not differ according to Duncan's test;

C.V.(%): Coefficient of Variation

*** Dosage unit: ICB Nutrisolo in mL.ha⁻¹ and FT10 in kg.ha⁻¹

Table 4. Shoot and root dry weight (in g) and yield (kg ha⁻¹) of iceberg, cv. Angelina, curly endive, cv. Mimosa and butterhead lettuce, cv. Regiane, treated with different doses of *Trichoderma* spp.-based products at different times of year, under greenhouse conditions. Ponta Grossa - PR. 2019/2020 growing season.

Treatments	Doses	Iceberg 10/18/2019			Curly endive 12/17/2019			Butterhead 05/19/2020					
		Dry weight (g)		Yield (kg.ha ⁻¹)*	V(%)	Dry weight (g)		Yield (kg.ha ⁻¹)*	V(%)	Dry weight (g)		Yield (kg.ha ⁻¹)*	V(%)
Absolute Control	-	3,48bc	32,49a	11800b	-	5,00a	26,31c	9935b	-	1,05a	23,94a	8878b	-
ICB Nutrisolo	300	4,30a	34,21a	13360ab	13	4,90a	32,33ab	11345ab	14	1,14a	26,22a	9377b	6
Trichoderma													
	0.05	3,45c	31,30a	11885b	1	4,58a	28,06bc	10095b	2	1,05a	22,83a	8687b	-2
	0.10	3,91abc	37,60a	14435a	22	4,07a	33,17a	12095a	22	1,17a	26,47a	9187b	3
	0.15	4,17ab	39,08a	12870ab	9	3,96a	33,35a	11875a	20	1,32a	28,05a	10273a	16
Mean	-	3,86	34,94	12870	4,50	30,64	11069	1,14	25,50	9281			
C.V.	-	14,09	25,83	9,88	21,68	11,62	10,37	23,12	20,10	6,80			

*Significant according to the F-test at 5% and **10% probability level

Means followed by the same letter in the column do not differ according to Duncan's test;

C.V.(%): Coefficient of Variation

Table 5. Monthly {(average (Av.), maximum (max.) and minimum (min.)} temperature and monthly average relative humidity (RH) in the four seasons studied.

Month/year	T. min.	average (°C)	T. max. average (°C)	T. average (°C)	RH% average	Precipitation average
July/19		7,5	22,4	14,95	82,1	0,9
August/19		8,7	22,8	15,75	81,5	0,6
September/19		13,2	24,8	19	76,9	1,7
October/19		15,4	28,6	22	77,2	1,9
November/19		16	28,2	22,1	79,9	26,4
December/19		16,6	27,7	22,15	82,7	3,9
January/20		18,5	27,8	23,15	85,2	2,5
April/20		14,5	25,7	20,1	76,3	1,8
May/20		8,9	21,5	15,2	80,9	2,2

For iceberg, butterhead and salad bowl lettuce evaluated on 10/18/2019, 12/17/2019 and 05/19/2020, the yield was lower than that reported in the literature,

probably because the plants were harvested at 42 DAT, long before the 70, 56 and 56 days recommended for these cultivars, respectively.

All four lettuce varieties treated with *Trichoderma asperelloides* showed a significant increase in growth and yield in the four seasons assessed, demonstrating the potential of FT10 doses of 10.0 and 0.15 kg ha⁻¹ as a biostimulant/inoculant.

The need for a sustainable increase in crop yields has prompted the growing application of biostimulants in global agriculture, making this strategy a vital technological innovation (Yakhin et al., 2017). The use of certain *Trichoderma* species with predominantly growth-promoting action as inoculants in agriculture could have significant economic, social and environmental impacts on Brazil, as observed in developing countries that have invested considerable financial resources in limiting the use of highly soluble and polluting inorganic fertilizers.

In lettuce crops, Lima (2019) found that *Trichoderma* spp. isolates promoted significant growth in different loose-leaf cultivars, as observed here. For the BRS Leila cultivar, RDW and SDW rose by 44 and 45%, respectively, when *T. virens*, *T. koningiopsis* and *T. harzianum* were applied, with values of 30 and 36.71 for the *T. asperellum* and *T. asperelloides* isolates in the BRS Lélia cultivar and 75 and 78.45 for *T. virens*, *T. asperellum* and *T. asperelloides* application in cv. 'BRS Mediterrânea'. Pereira et al. (2019) studied the effect of three *Trichoderma* spp. strains on the growth and yield of iceberg lettuce (cv. 'Mauren') and recorded yields 37 and 66% higher for the *T. harzianum* strains IBLF006 and ESALQ 1306, respectively. Fiorentino et al. (2018) analyzed the impact on lettuce of biostimulants based on *Trichoderma virens* (GV41) and *T. harzianum* (T22) strains associated with different doses of nitrogen fertilizer and found that inoculation with *Trichoderma* spp. significantly improved yield at all the nitrogen concentrations tested. In the absence of nitrogen fertilization, increases of 34 and 24% were obtained for total and marketable fresh weight, respectively, with GV41 application and moderate improvements of 16 and 17% for isolate T22. Colla et al. (2015) studied the combined application of *T. atroviride* and the mycorrhizal fungus *Glomus intraradices* to lettuce grown in the field and observed a 61 and 57% rise in SDW and RDW, respectively. When *Trichoderma* was applied in conjunction with *Glomus intraradices* under greenhouse conditions, dry weight rose by 167 in relation to the control, which was attributed to the increase in P, Mg, Fe, Zn and B uptake. In field conditions, SDW and RDW increased by 61 and 57%, respectively, with application the microorganisms.

López-Bucio et al. (2015) attributed the phytostimulation mechanism of *Trichoderma* to multilevel

communication with root and shoot systems as it releases auxins, small peptides, volatile compounds and other active metabolites into the rhizosphere, promoting root branching and the absorption of nutrients such as phosphorus, iron, manganese and zinc.

The survival capacity of *T. asperelloide* throughout the experiment was confirmed by plating diluted suspensions of the substrate at the end of the experiments. Populations of up to 4x10⁴ CFU.g⁻¹ were found in the substrate at 42 DAT. The beneficial effects of these microorganisms depend on their establishment in the growing environment and adaptability, among other factors. According to Pereira et al. (2019), increased lettuce growth and yield are heavily dependent on the ability of the *Trichoderma* isolate to establish a relationship with the plant species tested after colonizing the rhizosphere, benefitting the plants through a variety of mechanisms of action. The results obtained here were similar to those reported by

Oskiera et al. (2017), who observed a persistent *T. atroviride* and *T. harzianum* populations of 10³ CFU.g⁻¹ to 10⁵ CFU.g⁻¹ in soil cultivated with lettuce, using conventional sample and molecular plating tests (multiplex-PCR), for up to two years. Harman (2000) reported that the most effective growth-promoting strains can establish lasting interactions with plants, since their effects persist through all or most of the growth cycle. This root-colonizing ability is an important characteristic that allows the beneficial agent to compete with the microbiota for space and/or nutrients and benefit plants (Harman, 2000; Samuels, 2006).

Conclusions

Based on the results of the greenhouse experiments, it can be concluded that the doses 0.10 and 0.15 kg ha⁻¹ of the bioproduct FT10 (*Trichoderma asperelloides*) applied to the furrow before the lettuce seedlings transplant has the potential to improve the development and yield of lettuce cultivars studied.

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