### Organic amendments on soil characteristics and melon yield

Jonás Alan Luna-Fletes<sup>1,§</sup>
Oscar Raúl Mancilla-Villa<sup>1</sup>
Elia Cruz-Crespo<sup>2</sup>
Álvaro Can-Chulím<sup>2</sup>
Oscar Germán Martínez-Rodríguez<sup>3</sup>
Rubén Darío Guevara-Gutiérrez<sup>4</sup>

- 1 Departamento de Producción Agrícola-Centro Universitario de la Costa Sur-Universidad de Guadalajara. Av. Independencia 151, Autlán de Navarro, Jalisco, México. CP. 48900. (oscar.mancilla@academicos.udg.mx).
- 2 Unidad Académica de Agricultura-Universidad Autónoma de Nayarit. Carretera Tepic-Compostela km 9, Xalisco, Nayarit, México. CP. 63780. (ccruzc2006@yahoo.com.mx; canchulim@yahoo.com.mx).
- 3 Campo Experimental Santiago Ixcuintla-INIFAP. Carretera Internacional México-Nogales km 6, Santiago Ixcuintla, Nayarit, México. CP. 63300. (ogmartinez.r@gmail.com).
- 4 Departamento de Ecología y Recursos Naturales-Centro Universitario de la Costa Sur-Universidad de Guadalajara. Av. Independencia 151, Autlán de Navarro, Jalisco, México. CP. 48900. (ruben.guevara@academicos.udg.mx).

Autor para correspondencia: jonas.luna@academicos.udg.mx

#### Abstract

Agricultural soils with inefficient agronomic management lose their ability to support plants; it is considered that one strategy is the use of organic amendments to improve soil characteristics and the productivity of crops, such as melon. The objective was to evaluate compost (CMT) and goat manure (GM) mixed with *Trichoderma* sp. (Tsp) and *Bacillus subtilis* (Bs) in the physicochemical properties and macronutrient contents of the soil and the quality and yield of melon. The experiment was conducted from December 2023 to April 2024 in Tonaya, Jalisco, Mexico. The experimental design used was a completely randomized design with six treatments: CMT + Tsp, CMT + Bs, GM+ Tsp, GM + Bs, CMT, and GM, and one control (no amendment). It was observed that, compared to the control, all treatments increased the levels of organic matter, field capacity, porosity, cation exchange capacity, N, P, K, S, Ca, and Mg of the soil. The CMT, GM + Tsp and CMT + Tsp treatments increased the mass and size of fruits compared to the control, whereas for °Brix, only the CMT + Tsp treatment outperformed the control. The estimated yield decreased by up to 21% with the GM treatment and by 30% with the control compared to the rest of the treatments. It is feasible to use these amendments to improve the quality of agricultural soils and increase the quality of fruits and melon yields.

### **Keywords:**

Bacillus subtilis, Cucumis melo L., Trichoderma sp., physicochemical properties of the soil.

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## Introduction

Organic waste is used to produce fertilizers with high amounts of nutrients and organic matter that are used as amendments to improve agricultural soils (Murillo-Montoya *et al.*, 2020). The organic amendments that have been most studied in the last two decades are composts due to the benefits they provide to the soil, such as improving nutritional content, reducing erosion, increasing the microbial population, and improving water retention, which favors crop production (Trinidad and Velazco, 2016; Cervantes-Vázquez *et al.*, 2022).

In this regard, Macías-Duarte *et al.* (2020) reported an increase in organic matter of 68%, nitrogen ( $NO_3$ ) of 42%, and phosphorus of 26% when applying compost plus wheat straw to the soil with *Olea europaea* crops compared to the control (unamended). Another material that can be an alternative is goat manure due to its affordable cost, regional availability, and the fact that it implies less process for its application.

Nonetheless, the information is limited regarding the effect that this type of manure causes on the soil and on the production and quality of the crops; as a reference we have the studies of Cervantes-Vázquez *et al.* (2022), who showed that the application of bovine manure improved the properties and nutritional contents of the soil and increased the production of *Citrullus lanatus*. In a soil cultivated with *Aniba rosaeodora* and amended with biochar plus poultry manure, Cavalcante-Ferreira *et al.* (2024) found higher levels of phosphorus, calcium, magnesium, zinc and manganese compared to the soil without amendment.

In addition to the above, *Trichoderma* sp. not only has the ability to control phytopathogenic fungi, but also to promote plant growth and increase soil quality (Candelero *et al.*, 2015) since it improves the assimilation of nutrients by crops, increases the supply of nutrients in the soil, and has the ability to release hormones; these characteristics make this microorganism an excellent biological amendment (González-León *et al.*, 2023).

In *Musa paradisiaca* crops, Romero-Cún and Loayza-Agurto (2023) found that when *Trichoderma spirale* plus chopped rachis from the same plant were applied to the soil, the height of the plant and leaf area increased compared to the control (without application), and the same happened for the content of organic matter in the soil. Likewise, *Bacillus subtilis* is a bacterium that biocontrols plant pathogenic fungi, which endophytically associates with roots and has the ability to fix atmospheric nitrogen, solubilize phosphates and produce siderophores, which increases crop production and improves soil characteristics (González-León *et al.*, 2023).

Melon (*Cucumis melo* L.) is considered a crop of economic importance in Mexico due to its high production, with Michoacán being the largest producing state in 2023 with 144 824 t and Jalisco has an annual production increase of 10% due to the business opportunity that this crop represents in the state (SIAP, 2023). Based on the above, the study aimed to evaluate the organic amendments of compost and goat manure mixed with the microorganisms of *Trichoderma* sp. and *Bacillus subtilis* in the physicochemical characteristics, macronutrient concentrations of the soil, and in the quality of fruits and melon yield.

### Materials and methods

## Characteristics of the experimental site

The experiment was conducted in a plot located in the Municipality of Tonaya, Jalisco, Mexico (geographical coordinates 19° 52′ 0″ north latitude and 103° 55′ 5″ west longitude and altitude of 778 m). The municipality has a semi-dry warm climate, with an average annual temperature and precipitation of 16.6 °C and 617.7 mm, respectively (IIEG, 2022). The soil had a clayey texture, 1.26 g cm<sup>-3</sup> of bulk density, 2.65% of organic matter, 7.1 of pH, 0.96 dS m<sup>-1</sup> of electrical conductivity, 26 mg kg<sup>-1</sup> of N, 22.25 mg kg<sup>-1</sup> of P, 193.63 mg kg<sup>-1</sup> of K, 6 582.35 mg kg<sup>-1</sup> of Ca and 759.65 mg

kg<sup>-1</sup> of Mg; these determinations were made in the Soil, Water and Plant Analysis Laboratory of the University Center of the South Coast of the University of Guadalajara.

## **Experimental design and treatments**

The experimental design used was a completely randomized design with six treatments [compost (CMT) + *Trichoderma* sp. (Tsp), CMT + *Bacillus subtilis* (Bs), goat manure (GM) + Tsp, GM + Bs, CMT and GM] and a control (no amendment). Four replications were used for each treatment, and the experimental unit consisted of three 20 linear m rows with 50 plants each row.

# Characteristics and supply of organic amendments and plant growthpromoting microorganisms

The compost was formed from vegetable residues and cattle and goat manure in a 2:1 v/v ratio and the dried goat manure was collected directly from a goat farm in Autlán de Navarro, Jalisco, Mexico. *Trichoderma* sp. (*harzianum*, *viride*, and *asperellum*) was supplied with the commercial product Trichodef<sup>®</sup>, which presented a concentration of 1 x 10<sup>12</sup> colony-forming units per g and for *Bacillus subtilis*, the product Baci-Soil<sup>®</sup> was used with a concentration of active ingredient of 10%, both products in solid form.

On the same day of the application of the amendments, the compost or goat manure was mixed with *Trichoderma* sp. or *Bacillus subtilis* depending on the treatment at a ratio of 1000:1 v/v. Then, the soil received a dose of 10 t ha<sup>-1</sup> of each treatment, which was incorporated directly into each furrow (effective area) with an inverted clod-breaking harrow (Swissmex<sup>®</sup>, Rotostone RSH-M105, Mexico). Some chemical properties and macronutrient contents of the amendments are shown in Table 1.

				production.				
ORA	рН	EC (dS m <sup>-1</sup> )	N	P	K	Ca	Mg	Na
		_						
CMT	7.89	2.64	980.14	169.13	3 961.9	5 826.42	1 667.9	276.2
GM	8.84	5.24	52.48	150.61	6 750.79	5 872.44	1 698.34	2 852.05

# Genetic material and crop management

The seed used corresponded to Super Vida hybrid melon of Sakata Seed® (cantaloupe type) (with 90% germination), which was sown in 200-cavity expanded polystyrene trays (one seed per cavity) in a greenhouse, where the average minimum and maximum temperature was 20 and 30 °C, respectively, with average relative humidity of 77%. The trays were filled with peat moss substrate at 40% moisture, this moisture content was maintained until the germination of the seeds; subsequently, each tray was irrigated daily with 1.5 L of water combined with 1 g L<sup>-1</sup> of triple 17 NPK until transplantation to the field (20 days after sowing).

A seedling was transplanted into a row per hole of the white plastic mulch (90 µm caliber) with a distance between plants of 0.4 m and between rows of 1.6 m, for a planting density of 15 625 plants ha<sup>-1</sup>. After transplanting, irrigation and fertilization began; to do this, a drip fertigation system was used, the maximum irrigation volume was 40 m<sup>3</sup> ha<sup>-1</sup> per day (in the production stage), and the fertilization dose used was 180N, 100P, 200K, 60Ca, and 20Mg, which was supplied with fertilizers of calcium nitrate, monopotassium phosphate, potassium nitrate, and magnesium sulfate; the plants were fertigated every two days throughout the crop cycle.



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### Variables evaluated

### Physicochemical properties and macronutrient concentrations of soil

At 100 days after transplantation (dat), a composite sample of soil was collected per replication and treatment, and these were analyzed in triplicate for physicochemical properties and macronutrient concentrations in the Soil, Water and Plant Analysis Laboratory of the University Center of the South Coast of the University of Guadalajara using the methodologies of the Official Mexican Standard NOM-021-SEMARNAT-2000 (SEMARNAT, 2002); the parameters evaluated were field capacity (FC) (method AS-06), bulk density (BD) (method AS-03), pH (method AS-02); electrical conductivity (EC) (method AS-18); cation exchange capacity (CEC) (method AS-12); organic matter (OM) (method AS-07).

Exchangeable sodium percentage (ESP) (method AS-21); N-inorganic (N) (method AS-08); Olsen phosphorus (P) (method AS-10); potassium (K), calcium (Ca), magnesium (Mg), and sodium (Na) (method AS-19); sulfur (S) (method AS-20). Likewise, the total porosity (TP) was determined by the equation TP= (RED-BD/RED)\*100. Where: RED is the actual reference density (2.65 g cm<sup>-3</sup>), according to McPhee *et al.* (2015).

### Fruit quality and melon yield

At 90 dat, a random sample of 40 ripe fruits was collected per treatment, their fruit mass (FM) was evaluated with a digital scale (Tanita<sup>®</sup>, KW-002, Japan), °Brix with a refractometer (Hanna<sup>®</sup>, HI96801, USA), pulp firmness (FMS) with an analog penetrometer (Wagner<sup>®</sup>, FT30, USA), and equatorial (ED) and polar diameter (PD) with a tape measure (Truper, Mexico). To obtain the estimated yield, the number of commercial fruits produced per plant was counted, this data was multiplied by the fruit mass and the number of plants ha<sup>-1</sup> and the result was extrapolated to t ha<sup>-1</sup>.

## Statistical analysis

All the data were analyzed with the statistical software of SAS  $^{\circ}$  (Version 9.0 for Windows) using an Anova (analysis of variance) and comparison of means by Tukey's test ( $p \le 0.05$ ).

### Results and discussion

# Physicochemical properties of soil

The control obtained the lowest value of FC (57.06%) and OM (2.76%) compared to all treatments. TP increased to a greater extent with the CMT + Bs (57.36%), GM + Tsp (58.49%), CMT (58.87%) and GM (57.74%) treatments compared to the control (53.21%). Soil salinity expressed in EC and ESP increased with the application of the GM + Tsp, GM + Bs and GM treatments, with values of 3.07 dS m<sup>-1</sup> and 2.16%, 3.3 dS m<sup>-1</sup> and 1.98%, 3.13 dS m<sup>-1</sup> and 2.31%, respectively, compared to the control (1.95 dS m<sup>-1</sup> and 2.31%) (Table 2).

Table 2. Effect of organic amendments alone and combined with plant growth-promoting microorganisms on the physicochemical properties of the soil cultivated with melon.

Treatment	FC (%)	BD (g cm <sup>-3</sup> )	TP (%)	рН	EC (dS m <sup>-1)</sup>	CEC (Meq 100 g <sup>-1</sup> )	OM (%)	ESP
CMT + Tsp	61.7a	1.17	55.85c	7.55	2.53ab	54.95 b	7.6a	1.39c
CMT + Bs	61.83a	1.13	57.36abc	7.43	2.55ab	54.49 b	7.26a	1.38c
GM + Tsp	60.6a	1.1	58.49ab	7.69	3.07a	55.55b	6.45a	2.16ab
GM + Bs	62.4a	1.16	56.23bc	7.4	3.3a	54.81b	5.98a	1.98ab
CMT	61.6a	1.09	58.87a	7.65	2.82ab	53.24b	7.93a	1.49bc



Treatment	FC (%)	BD (g cm <sup>-3</sup> )	TP (%)	рН	EC (dS m <sup>-1)</sup>	CEC (Meq 100 g <sup>-1</sup> )	OM (%)	ESP
GM	62.86a	1.12	57.74abc	7.67	3.13a	59.58a	8.14a	2.31a
Control	57.06b	1.24	53.21d	7.26	1.95b	49.99c	2.76b	1.2c
MSD	3.29	0.3	2.59	2.61	1.05	2.59	2.65	0.72
Significance	**	ns	**	ns		**	**	**

Means with different letters within each column are significantly different (Tukey,  $p \le 0.05$ ). \*\* = statistical significance at  $p \le 0.01$ ; ns= not significant; MSD= minimum significant difference; FC= field capacity; BD= bulk density; TP= total porosity; EC= electrical conductivity; CEC= cation exchange capacity; OM= organic matter; ESP= percentage of exchangeable sodium.

The control obtained the lowest value of CEC (49.99 Meq 100 g<sup>-1</sup>) compared to all the treatments evaluated, with the GM treatment obtaining the highest value, with 59.58 Meq 100 g<sup>-1</sup> (Table 2). These results demonstrate the modification of most of the physicochemical properties of the soil mainly due to the effect of organic amendments, where the increase in water retention capacity (FC), porosity, cation exchange capacity, and organic matter is highlighted; this favors the productive quality of agricultural soils.

The increase in these characteristics in soil is due to the fact that goat manure and compost by origin provided organic matter and cations (Ca, Mg, K, and Na) in high quantities. In this regard, Trinidad and Velazco (2016); Macías-Duarte *et al.* (2020) indicate that with the application of compost or manure, the percentages of organic matter increase, thereby increasing the stability of aggregates and the amount of macro and micropores in the soil, which favors water retention and filtration.

On the other hand, Guerrero-García (1996) mentions that the increase in CEC is partly influenced by the increase in cations from organic amendments that can be exchanged between the solution and soil colloids, but also by the activity of humic substances from organic matter that forms large clay-humic complexes.

Similar responses were reported by Macías-Duarte *et al.* (2020), who demonstrated that the application of compost of vegetable waste and bovine manure to the soil increased OM by 70% and CEC by 27% compared to the unamended control; for their part, Lagos and Huertas (2019) reported higher total porosity (70.41%) in soil with the supply of guinea pig (*Cavia porcellus*) manure compost compared to the control without application (67.17%).

In the case of the increase in EC and ESP in the soil treated with goat manure alone or in mixture with *Trichoderma* sp. or *Bacillus subtilis*, this can be attributed to the higher EC (5.24 dS m<sup>-1</sup>) and concentration of Na (3 117.34 mg kg<sup>-1</sup>) of goat manure compared to compost (5.24 dS m<sup>-1</sup> and 542.15 mg kg<sup>-1</sup>, respectively) (Table 1), which influenced the increase in these chemical characteristics of the soil.

One of the main disadvantages of using manure (from cattle and goats) as organic amendments is the high concentration of soluble salts that they contain and contribute; therefore, the constant use of manure and high doses of it applied to the soil can cause problems of salinization of the soil, which can affect the productivity of crops (Trinidad and Velazco, 2016).

### Soil macronutrient concentrations

Table 3 shows that the CMT treatment increased the concentration of N (187.59 mg kg<sup>-1</sup>) and P (185.51 mg kg<sup>-1</sup>) compared to the rest of the treatments, and even to the control, which had values of 50 and 32.46 mg kg<sup>-1</sup>, respectively. The concentration of K, S and Na was higher with all treatments compared to the control, where the highest concentrations of these elements were obtained by the GM treatment, with 167.04, 133.16 and 321.53 mg kg<sup>-1</sup>, respectively.



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Table 3. Effect of organic amendments alone and combined with plant growth-promoting microorganisms on soil macronutrient concentrations with melon cultivation.

Treatment	N	Р	K	Ca	Mg	S	Na			
_	(mg kg <sup>-1</sup> )									
CMT + Tsp	137.59b	126.96e	897.47b	8 023g	846.88g	86.15c	159.22e			
CMT + Bs	133.33b	161.02b	630.8e	8 795.4d	1000.66d	74.47d	173.06d			
GM + Tsp	102.84d	128.55de	493.99f	8 929.92c	1036.07c	114.92b	276.14b			
GM + Bs	123.76c	147.1c	640.53d	8 656.1e	1099.58b	88.53c	212.06c			
CMT	187.59a	185.51a	727.54c	8 119.82f	863.73f	70.77d	171.81d			
GM	95.74e	131.45d	1167.04a	9 437.78a	1105.61a	133.16a	321.53a			
Control	50f	32.46f	418.38g	9 231.98b	869.29e	37.48 e	151.12f			
MSD	5.18	4.43	4.44	4.92	4.4	4.32	4.71			
Significance		**	••	**	••	••	••			

Means with different letters within each column are significantly different (Tukey  $p \le 0.05$ ). \*\*= statistical significance at  $p \le 0.01$ ; MSD= minimum significant difference.

In the case of Ca and Mg, the highest levels of both elements compared to the control (9 231.98 and 869.29 mg kg<sup>-1</sup>, respectively) were shown by the GM treatment, with values of 9 437.78 mg kg<sup>-1</sup> Ca and 1 105.61 mg kg<sup>-1</sup> Mg; however, the control exceeded the treatments of CMT + Tsp (8 023 mg kg<sup>-1</sup>). CMT + Bs (8 795.4 mg kg<sup>-1</sup>), GM + Tsp (8 929.92 mg kg<sup>-1</sup>), GM + Bs (8 656.1 mg kg<sup>-1</sup>) and CMT (8 119.82 mg kg<sup>-1</sup>) in the concentration of Ca and the treatments of CMT + Tsp (846.88 mg kg<sup>-1</sup>) and CMT (863.73 mg kg<sup>-1</sup>) in the concentration of Mg.

The increase in soil macronutrient concentrations can be explained by the macronutrient contribution of organic amendments. A relationship and trend of increase were observed between the macronutrient concentrations of soils treated with organic amendments and their nutrient contents since, according to (Table 1), CMT supplied the soil with a greater amount of N (980.14 mg kg<sup>-1</sup>) and P (52.48 mg kg<sup>-1</sup>) compared to GM (52.48 and 150.61 mg kg<sup>-1</sup>, respectively), whereas GM contributed more K (3 961.9 mg kg<sup>-1</sup>), Ca (5 826.42 mg kg<sup>-1</sup>), Mg (1 667.9 mg kg<sup>-1</sup>), and Na (276.2 mg kg<sup>-1</sup>).

This caused CMT to obtain higher levels of N and P, and GM to obtain higher levels of K, Ca, Mg, and Na in soil. This coincides with the study by Jiménez-Ortiz *et al.* (2019) because they reported that the application of bovine manure compost in *Zea mays* increased the levels of N, P, and K in soil by up to 36% compared to the unamended control, which they attributed to the contribution of macronutrients by the compost.

The microorganisms *Trichoderma* sp. and *Bacillus subtilis* decreased the supply of macronutrients available in the soil, which was possibly due to nutrient immobilization by them since, according to Rincón-Castillo *et al.* (2012), as microorganisms decompose the organic matter contained in the amendments or in the soil, they require nutrients, such as N, P, K, Ca, Mg and S, in soluble inorganic forms for their metabolic reactions and the synthesis of nucleic acids, enzymes, amino acids, and proteins for biomass formation, so nutrients are converted to non-soluble organic forms.

## Fruit quality and melon yield

Fruit mass increased by up to 24% with all treatments compared to the control, where the plants treated with CTM (1 452 g), GM+ Tsp (1 328.3 g) and CMT + Tsp (1 425.9 g) obtained fruits with greater mass; in the case of °Brix, CMT + Tsp, with a value of 9.44 °Brix, was the only treatment that statistically surpassed the control (7.86 °Brix) (Table 4).



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Table 4 Effect of organic amendments alone and combined with plant growth-promoting microorganisms o fruit quality and melon yield.								
Treatment <sup>*</sup>	FM (g)	°Brix	FMS (kg cm <sup>-2</sup> )	ED (cm)	PD (cm)	EY (t ha <sup>-1</sup> )		
CMT + Tsp	1 425.9ab	9.44a	6.33	21.58a	22.89a	69.73a		
CMT + Bs	1 282.2bc	8.6ab	5.61	20.87ab	22.33ab	60.1b		
GM + Tsp	1 328.3abc	8.29ab	5.92	21.32a	23.04a	62.02ab		
GM + Bs	1 265.9c	8.93ab	5.48	20.87ab	22.27ab	59.33b		
CMT	1 452a	8.53ab	5.69	21.58a	23.15a	70.27a		
GM	1 265.1c	8.78ab	6.22	21.1ab	22.5ab	55.17bc		
Control	1 101.3d	7.86b	6.4	20.19b	21.4b	49.53c		
MSD	145.2	1.17	0.96	1.1	1.33	8.87		
Significance	**	••	ns	**		**		

Means with different letters within each column are significantly different (Tukey,  $p \le 0.05$ ). \*\*= statistical significance at  $p \le 0.01$ ; ns= not significant; MSD= minimum significant difference; FM= fruit mass; FMS= pulp firmness; ED= equatorial diameter of fruit; PD= polar diameter of the fruit; EY= estimated yield.

Compared to the control, plants treated with CMT, GM + Tsp, and CMT + Tsp produced fruits with higher equatorial diameter and polar diameter, with values ranging from 21.32 to 23.15 cm. Regarding the estimated yield, this decreased by up to 21% when GM was supplied and by 30% with the control compared to the rest of the treatments; the highest estimated yields were obtained with the treatments of CMT (70.27 t ha<sup>-1</sup>), GM + Tsp (62.02 t ha<sup>-1</sup>), and CMT + Tsp (69.73 t ha<sup>-1</sup>) (Table 4).

The increase in yield was directly related in most cases to the increase in fruit mass and the obtaining of fruits with greater mass in general is due to the improvement of the physicochemical and macronutrient characteristics of the soil caused by organic amendments (CMT or GM) alone or in mixture with *Trichoderma* sp. or *Bacillus subtilis*, as explained and reported by González-Salas *et al.* (2021) in melon grown in soil with the combined application of bovine manure and growth-promoting bacteria.

In addition, these results corroborate the beneficial effect of *Trichoderma* sp. and *Bacillus subtilis* on yield, which can be attributed to the fact that these microorganisms promote the growth of plants and fruits by the synthesis of phytohormones, solubilize nutrients and organic compounds, produce secondary metabolites and control phytopathogenic fungi, which increases the production and quality of crops, as noted by Barbosa-Santos *et al.* (2020).

Results that agree with those obtained by Cantú-Nava *et al.* (2021) because they found an increase in the yield of *Carya illinoinensis* of 27.7% with the application of *Bacillus subtilis*, *Bacillus cereus*, *Pseudomonas fluorescens*, and *Trichoderma harzianum* compared to the control without inocula.

It should also be noted that the estimated yield obtained with the experimental treatments exceeded up to 2.14 times the average yield recorded in Mexico in 2023, which was 32.82 t ha<sup>-1</sup> (SIAP, 2023); this was possibly due to the high planting density (15 625 plants ha<sup>-1</sup>) analyzed in this research.

According to Monge-Pérez and Loría-Coto (2017), °Brix are closely related to the flavor and sweetness of melons; therefore, for melon fruits to be considered of commercial quality, they must contain 9 to 12 °Brix. This shows that CMT + Tsp was the only treatment that produced fruits with acceptable commercial quality by obtaining an average value within the optimal °Brix range.

The non-optimal results of °Brix obtained with the other treatments can be explained by a possible delay in fruit ripening since, according to González-Loaiza *et al.* (2014), °Brix increase when fruits reach a higher degree of maturity due to the increase in the concentration of soluble solids due to the hydrolyzation of soluble starches and pectins during the ripening process.

In addition, the positive effect of organic amendments and *Trichoderma* sp. on the equatorial and polar diameter of fruits was observed, as reported by Adame-García et al. (2023) in fruits of

Capsicum annuum and Solanum lycopersicum with the inoculation of Trichoderma sp. This result could be partly attributed to the contribution of macronutrients and humic acids by compost and goat manure (Cruz-Crespo et al., 2015), but also to the fact that Trichoderma sp. promotes plant growth by the production of phytohormones (Candelero et al., 2015), which contributed to the increase in fruit size.

## **Conclusions**

Using compost or goat manure without mixing with *Trichoderma* sp. or *Bacillus subtilis* greatly favored the increase of the physicochemical properties and macronutrient concentrations of the soil; therefore, these amendments may be feasible for use in agricultural soils, considering the high contribution of salts from manure. The application of compost in combination with *Trichoderma* sp. was the most viable treatment for melon crops since it increased not only the mass, equatorial diameter, polar diameter, and estimated yield of fruits compared to the control, but also the °Brix, an important variable that indicates higher fruit quality.

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