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Biomass and minerals in *Lippia graveolens* under three production environments in General Cepeda

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Abstract

Oregano (*Lippia graveolens*) of the family Verbenaceae is an aromatic plant that is distributed in arid and semi-arid regions of Mexico; it is used as a condiment for food, production of cosmetics, drugs and liquors, reasons that make it an export product. In order to determine the best environment and type of fertilization that promotes agronomic variables, we set ourselves the objective of evaluating the production of biomass and concentration of minerals in three growing environments: greenhouse, shade mesh, and open field, and treatments with chemical fertilization with Steiner solution (SS) (0%, 50%, and 100%) and biological fertilization (four strains of *Azospirillum* spp.) in a 3 X 5 factorial arrangement, with a total of 15 treatments in each environment. The work began in July 2019 in General Cepeda, Coahuila. The environment with the highest production values was GRE with: PD (57.56 cm), FW (240.94 g), DW (79.86 g), DSW (34.35 g), and DLW (41.26 g). T3 (100% SS, without *Azospirillum* sp.) resulted in better values of PH (80.78 cm), PD (56.76 cm), FW (260.45 g), DW (103.36 g), DSW (43.36 g), and DLW (53.45 g). For minerals, the SM environment was better in N and K with 14 451.1 and 9 108.1 mg kg^{-1}, respectively, and the GRE for: P (4 206.95), Ca (17 486.5), Mg (1 741.11), Cu (8.24), and Zn (31.49) mg kg^{-1}.

Keywords:

oregano, organic nutrition, protected agriculture, rhizobacteria.

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Introduction

Lippia graveolens, known as Mexican oregano, is an herbaceous plant belonging to the family Verbenaceae. Oregano leaves are used as a condiment for food, making cosmetics, drugs, and liquors; reasons that make it an export product (García-Pérez *et al*., 2012). The collection of wild plants is one of the main economic activities of the inhabitants in marginalized regions of the Mexican Semi-Desert and in particular of Coahuila, where native species are used, such as lechuguilla (*Agave lechuguilla*), piquín chili (*Capsicum annuum*), candelilla, (*Euphorbia antisyphilitica*), oregano (*Lippia* spp.) and others, which are subjected to a semi-process and subsequent commercialization.

In this state, the use of oregano is carried out mainly in natural populations during the rainy season; therefore, its use is limited to a few months and being a resource with a lot of demand, it is essential to promote forms of cultivation that extend the harvest months and its yields without putting the native populations at risk.

On the other hand, its exploitation coincides with the flowering of the plant, altering fruit formation and seed production, so that these populations have decreased in area and density (Orona-Castillo *et al*., 2017; Villavicencio-Gutiérrez *et al*., 2018), putting the sustainability of the resource at risk; therefore, the generation of adequate management techniques will not only contribute to the conservation of the species but also to a better economic return for the inhabitants of these regions.

Mexico is the second largest producer of oregano, after Turkey, and exports about 85% of the national production to the United States of America, 10% goes to the domestic market, and 5% to European and Asian countries (Flores *et al*., 2011). Production is concentrated in the states of Chihuahua, Durango, Coahuila, Guanajuato, Jalisco, Querétaro, San Luis Potosí, and Zacatecas (Rocha, 2014).

Despite the fact that Mexico is the world's second largest producer of oregano, the production comes from wild species, without agronomic management. Therefore, it is urgent to develop research that allows efficient use of inputs for the production of oregano. Protected agriculture can be an alternative to achieve high yield and quality of the harvest to meet the needs of the national and international markets.

The objective was to evaluate the production of fresh and dry biomass of oregano in three growing environments (greenhouse, shade mesh, and open field) and the effect of the application of fertilizers and rhizobacteria on agronomic variables.

Materials and methods

Location of the experimental area

The study was conducted from July to September 2019 in the municipality of General Cepeda, Coahuila, with coordinates: 25° 22' 53.30" north latitude, 101° 26' 55.52" west longitude, at 1 471 masl. According to the Modified Köppen Classification for Mexico, the climate is of the BS0h'(h)x'(w)(e') type, which is arid, semi-warm, with very extreme annual temperature with averages ranging between 19.1 and 27 °C, 385.6 mm of annual precipitation and loamy soil (García, 2004).

Biological material

The plants of *L. graveolens* H.B.K. were obtained from a commercial plantation in the ejido El Amparo, Parras, Coahuila. Naturally germinated plants were selected within the same plantation, they were one-year-old and between 20 and 25 cm tall.

The strains used were three strains of *Azospirillum* spp., isolated in the Department of Horticulture of the Antonio Narro Autonomous Agrarian University (UAAAN, for its acronym in Spanish) and a commercial strain of A*zospirillum brasilense* T. K. D.

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Soil and water analysis

The soil fertility in the experimental plot was diagnosed; it had a loamy texture, density of 1.04 g cm^{-3}, field capacity of 15.3%, permanent wilting point of 9.1%, pH of 8.64, electrical conductivity (EC) of 0.65 dS m^{-1}, total carbonates of 13%, and organic matter of 0.47%; the irrigation water analysis resulted in the following: EC of 1.3 dS m^{-1}, pH of 7.46, and hardness of 43.81; these analyses were carried out at Fertilab Laboratorio Agrícola in Celaya, Guanajuato.

Soil and water inputs were discounted to the concentration of the adjusted Steiner solution (SS). 100% SS= Mg $(NO_3)_2$ (232 mg L^{-1}), KNO_3 (616 mg L^{-1}), KH_2PO_4 (68 mg L^{-1}), HNO_3 (0.084 ml L^{-1}) and H_2SO_4 (0.231 ml L^{-1}); 50% SS= $Mg(NO_3)_2$ (116 mg L^{-1}), KNO_3 (308 mg L^{-1}), KH_2PO_4 (34 mg L^{-1}), HNO_3 (0.042 ml L^{-1}) and H_2SO_4 (0.115 ml L^{-1}).

Setting up the experiment

Three environments were tested: greenhouse (GRE), shade mesh (SM) and open field (OF). Three adjusted SS concentrations (0, 50 and 100%) were tested in each environment. In each concentration of SS, the following were tested: a control without the addition of *Azospirillum* and four strains of *Azospirillum* spp. The 15 resulting treatments were established under a randomized block design with three replications; in the treatments with Azospirillum, 20 ml of a solution with 10^{-6} colony-forming units was added to the base of the stem every 30 days. One hundred thirty-five plants per environment were studied (Table 1).

Table 1. Treatments applied to oregano crops, resulting from the application of chemical nutrition and application of four strains of *Azospirillum* spp., under three production environments.

Steiner solution (%)	**Bacterial strains assessed**				
	Control (C) without *Azospirillum*	**A1**	**A2**	**A3**	**CS**
0 (S0)	T1= S0C	T4= S0A1	T7= S0A2	T10= S0A3	T13= S0CS
50 (S50)	T2= S50C	T5= S50A1	T8= S50A2	T11= S50A3	T14= S50CS
100 (S100)	T3= S100C	T6= S100A1	T9= S100A2	T12= S100A3	T15= S100CS

Steiner solution; A= strains of *Azospirillum*, CS= commercial strain of *Azospirillum*.

Variables evaluated and their measurement

At 60 days after transplantation (dat), plant height (PH) and plant crown diameter (PD) were measured in cm. The aerial part of the plant was harvested at 25 cm above ground level and weighed at the time of cutting to obtain the fresh weight in g (FW); the dry weight of the aerial part (DW) was obtained by placing the plant material in brown paper bags and drying them at 40 °C to constant weight; the dry stem weight (DSW) and the dry leaf weight (DLW) were obtained by manually separating the stems from the leaves. A SIGMA scale with an accuracy of 0.01 g was used.

Concentration of minerals in dry leaves

This analysis was carried out in the Tissue Culture and Mineral Analysis Laboratory of the Department of Horticulture of the UAAAN. The concentration of nitrogen (N) was taken considering 0.05 g of sample and was performed with the Kjeldahl method (Kjeldahl, Novatech, Avante Tecnología, Jal, MX) (Bremmer and Mulvaney, 1982). Phosphorus (P), potassium (K), calcium (Ca), magnesium (Mg), copper (Cu) and zinc (Zn) were determined by the calcination method, taking 0.5 g of sample (Alcántar and Sandoval, 1999).

A GBC Scientific Equipment XplorAA Dual atomic absorption spectrophotometer was used to determine minerals. Dry biomass, acid digestion in $HClO_4$, and a spectrometer (Biomate 5, Thermo Electron Scientific Madison, WI, USA) were used for phosphorus determination (Hanway and Heidel, 1952).

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Statistical analysis

The information was analyzed with the statistical program of SAS® V.9.0 (SAS Institute Inc., 2002) to obtain the analysis of variance (Anova) in each variable and a comparison of means with Tukey's test ($p \leq 0.05$).

Results and discussion

Effect of the production environment

The Anova showed highly significant differences ($p \leq 0.01$) between environments, showing that this factor affected all the agronomic variables evaluated. In the same agronomic parameters, the same significance was present in the treatments evaluated, except for FW, where only significant differences were observed ($p \leq 0.05$); results that indicate that at least one treatment affected the behavior of the agronomic variables differently (Table 2).

Table 2. Analysis of variance performed on agronomic variables of *L. graveolens* under three production environments.

Source of variation	Degrees of freedom	Means squares					
		PH	PD	FW	DW	DSW	DLW
Env	2	5 427.5**	2 532.9**	190 768.2**	6 122.3**	2 002.6**	1 147.6**
Rep (Env)	6	127.3**	80.1**	1 756.1ns	367.7ns	88.1ns	66.1ns
Treat	14	158.6**	97.1**	13 393.2*	1 901.9**	375.8**	500.7**
Env x Treat	28	200.9**	84.1**	16 951.1**	1 984.7**	51 297 482*	492.1
Error	56	79.3	42.4	5 681	721.9	160.6	177.9
CV		12.161	13.014	44.632	38.784	44.318	36.852

Env= environments; Rep= replication; Treat= treatments; ns, *, **= not significant, significant with $p \leq 0.05$ and significant with $p \leq 0.01$; R^2= coefficient of determination, CV= coefficient of variation.

The oregano plants of the SM environment were significantly ($p \leq 0.05$) taller (84.53 cm), exceeding those obtained in OF by 25.95% and those obtained in GRE by 14.1%. The plants in GRE presented higher PD (57.56 cm), significantly exceeding (p# 0.05) those of OF by 26% and those of SM by 13.29%. In GRE, during the day, there was a higher temperature than that recorded in OF or SM, from which it is inferred that the higher temperature favored the vegetative development of the plant (Table 3).

Table 3. Mean values of agronomic variables studied in three production environments.

Environments	Agronomic variables					
	PH (cm)	PD (cm)	FW (g)	DW (g)	DSW (g)	DLW (g)
OF	62.59 c	42.56 c	114.29 b	56.77 b	21.28 b	31.16 b
SM	84.53 a	49.91 b	151.39 b	71.14 a	30.14 a	36.13 ab
GRE	72.59 b	57.56 a	240.94 a	79.87 a	34.36 a	41.26 a

Means with equal letters indicate that there are no significant differences (Tukey $p < 0.05$).

The FW obtained in GRE was 110.8% higher than the value obtained in the OF plants, and 59.1%higher than those developed in SM. Oregano plants from the GRE and SM environment were statistically equal in DW (Table 3); a similar behavior was observed in DSW from GRE and SM plants, although GRE plants outperformed OF plants by 61.5%.

The DLW is a useful and marketable parameter of oregano; in this regard, there were significant differences between environments, the DLW obtained in GRE was 32.41% higher than that recorded in OF (Table 3).

The values obtained in these growing environments are lower than those obtained in *L. palmeri* in OF (Corella-Bernal and Ortega-Nieblas, 2013), where they obtained 132 g of leaves with 35% moisture. In *Lippia graveolens*, Villa-Castorena *et al*. (2011) reported that, in OF, they obtained plants with a height of 90 cm and DLW of 50 g per plant at 140 dat, which represents an average growth of 0.64 cm per day and 0.36 g of average increase in weight per day; in contrast, in the present work, the average growth per day was 1.04 cm and the average increase in weight per day was 0.52 g. Although growth is not linear as a function of time, it can be indicated that, in GRE, the PH and DLW increased more efficiently.

In this regard, in *L. graveolens*, Martínez-Hernández *et al*. (2017) report a DLW of 1.42 g per plant, produced in greenhouses in different substrates, a factor to be considered in a GRE production scheme. These data indicate that the DLW depends on the species, the age of the plant, the type and size of the plant's leaf, as well as the production environment. Dunfort and Silva-Vázquez (2005) reported a FW of 56.6 g, DW of 28.1 g, DSW of 8 g, and DLW of 13.8 g per plant in greenhouses, averages that are lower than those obtained in this research.

If it is considered that the production of DLW is the most important thing, it will be necessary to select the environment and the planting density. In OF, 5 000 plants ha^{-1} have been established (Corella-Bernal and Ortega-Nieblas, 2013), obtaining up to three cuts per year, reporting a yield of 2 240 kg of dry leaf.

On the other hand, in this work, the estimated production of DLW (31.16 g) is 1 869.6 kg ha^{-1} in OF, and in GRE, it is 41.16 g per plant with three cuts per year, estimating a production of 2 475.6 kg ha^{-1} with a density of 20 000 plants ha^{-1}, exceeding the yields estimated by other authors.

Flores Hernández *et al*. (2011) agree with these results and report that, in *L. graveolens* grown in OF in La Comarca Lagunera, a FW of 22 t ha^{-1} and a DW of 13 t ha^{-1} were obtained in five-year-old plants, which resulted in an average production of 2 640 kg ha^{-1} of useful matter.

Nonetheless, in wild plants of *L. graveolens*, Osorno-Sánchez *et al*. (2009) reported an average production of 1 t km^{-2} of dry leaf per year in the municipality of Peñamiller, Querétaro, lower than that obtained in this research.

For their part, in a greenhouse and with a commercial nutrient solution (Fertiplus),Valdés Oyervides *et al*. (2012) had 158.12 g plant^{-1} in FW and 74.6 g of DW per plant. Murillo-Amador *et al*.'s (2013) results agree with those obtained in the present research, where SM has higher production than OF.

Response of agronomic variables to chemical nutrition and rhizobacteria

In PH, treatment 3 (100% SS without rhizobacteria) was significantly equal to 13 treatments, registering 80.78 cm and being 27.7% higher than treatment 8 (50% SS, A3 strain), it had the lowest PH. Complete chemical fertilization of T3, as well as rhizobacteria (A3) without chemical nutrition, induced the highest oregano PH (Table 4).

Table 4. Mean values of agronomic variables in response to chemical nutrition and rhizobacteria.

Treatments	Agronomic variables					
	PH (cm)	PD (cm)	FW (g)	DW (g)	DSW (g)	DLW (g)
	70.98 ab	50.32 ab	161.26 ab	70.49 ab	29.41 ab	37.59 ab
2. S50C	70.67 ab	46.4 ab	114.96 b	49.62 b	19.94 b	26.62 b
3. S100C	80.78 a	56.76 a	260.45 a	103.36 a	43.36 a	53.45 a

Treatments	Agronomic variables					
	PH (cm)	PD (cm)	FW (g)	DW (g)	DSW (g)	DLW (g)
4. S0A1	77.51 ab	56.04 a	216.9 ab	89.95 ab	33 ab	45.35 ab
5. S50A1	72.37 ab	50.85 ab	158.57 ab	68.86 ab	28.27 ab	36.83 ab
6. S100A1	73.47 ab	50.34 ab	180.12 ab	66.46 ab	27.61 ab	32.74 ab
7. S0A2	72.7 ab	49.11 ab	142.68 ab	60.98 ab	24.55 ab	31.33 b
8. S50A2	63.25 b	44.51 b	120.62 b	53.71 b	17.8 b	28.72 b
9. S100A2	73.63 ab	51.11 ab	194.41 ab	74.35 ab	31.47 ab	37.32 ab
10. S0A3	78.39 a	47.93 ab	168.9 ab	71.14 ab	31.4 ab	37.27 ab
11. S50A3	74.58 ab	50.69 ab	193.29 ab	80.16 ab	34.87 ab	44.32 ab
12. S100A3	73.33 ab	50.44 ab	143.65 ab	56.06 b	23.67 ab	28.35 b
13. S0CS	75.74 ab	48.49 ab	163,47 ab	63.37 a	28.01 ab	31.88 ab
14. S50CS	72.86 ab	51 ab	186.32 ab	76.87 ab	33.02 ab	41.04 ab
15. S100CS	68.32 ab	46.18 ab	127.51 b	53.53 b	22.45 b	29 89 b

Means with the same letter in the same column are significantly equal (Tukey $p \leq 0.05$).

Treatments 3 and 4 presented the highest PD but were significantly equal to 13 treatments; the first one was 27.5% higher than treatment 8 (50% SS, A3 strain). In the PH and PD variables, they showed a lower average than that obtained by Corella Bernal and Ortega-Nieblas (2013), who, in their second cut, reported a PH of 85 cm and 78 cm, respectively, applying only supplemental irrigation.

In FW, treatment 3 was the one that exhibited the highest value with 260.45 g and was significantly higher than treatments 2, 8, and 15, which it exceeded by 126.5%, 115.9%, and 104.2%, respectively; it was also the one that presented the highest DW with 103.36 g, being significantly higher than treatments 2, 8, 12, and 15, which it exceeded by 108.36%, 92.4%, 84.4%, and 93.1%, respectively (Table 4).

In DSW, treatment 3 had a value of 43.36 g and was significantly higher than treatments 2, 8, and 15, which it exceeded by 117.4%, 143.6%, and 93.1%, respectively. In DLW, treatment 3 registered the highest average (53.45 g), a value significantly higher than treatments 2, 7, 8, 12, and 15, which surpassed by 100.8%, 70.6%, 86.1%, 88.5%, and 78.8%, respectively, but significantly equal to 9 treatments. In the aforementioned variables, treatment 3 exhibited the best behavior; however, treatment without SS and rhizobacterium A1 was significantly the same as treatment 3, representing an alternative for oregano production (Table 4).

If a density of 20 plants ha^{-1} is established, an estimated production of DLW of 1 069 kg ha^{-1} would be reached with T3; on the other hand, by using T4, a DLW of 907 kg could be obtained with a single cut, showing a difference in DLW of 162 kg; nevertheless, there is the advantage that using rhizobacteria results in a production that is cheaper and less harmful to the environment.

In this sense, Juárez-Rosete *et al*. (2019) obtained a FW of 52.23 g and a DW of 7.99 g plant^{-1} by applying 75% Steiner solution; values that are low compared to those recorded in treatment 3 of this work, where the FW was 4.5 times higher and the dry weight was 13 times higher. This difference may be due to the age of the plant as these researchers used plants that were about three months old.

Leaf mineral content in response to the production environment

The foliar content of minerals showed significant differences (*p*# 0.01) between environments, which indicates that this variable is affected by the production environment. In GRE, the concentration of P, Ca, Mg, Cu, and Zn exceeded the value observed in SM and OF, but the concentrations of N and K were higher in SM, while OF showed the lowest values of minerals (Table 5).

Table 5. Comparison of means in production environments for the concentration of minerals in dry leaves (Tukey *p*≤ 0.05).

Production environment	Contents of mineral elements in oregano leaves (mg kg^{-1})						
	N	P	K	Ca	Mg	Cu	Zn
OF	12 522.2 b	3 293 b	8 283.6 b	14 007.1 b	1 546.2 b	5.36 b	25.24 b
SM	14 451.1 a	2 538.7 c	9 108.1 a	11 545.3 c	1 438.2 b	5.79 b	22.14 c
GRE	10 624.4 c	4 206.9 a	8 113.3 b	17 486.5 a	1 741.1 a	8.24 a	31.49 a

Macronutrients: N= nitrogen; P= phosphorus; K= potassium; Ca= calcium. Micronutrients: Mg= Magnesium; Cu= copper; Zn= zinc. Means with the same letter in the same column are significantly equal (Tukey *p*≤ 0.05).

The N content in leaves of plants developed in SM was significantly higher than the values recorded in the plants in the GRE condition (Table 5); the temperature inside the greenhouse could be a factor that influenced the lower amount of N in the plants, contrary to what was reported by Nkansah and Ito. (1995), who mention that, as the air temperature increases, nitrogen absorption increases.

The concentration of P in dry leaves in GRE plants was 4 206.96 mg kg^{-1}, a concentration that significantly exceeded the concentration observed in SM and OF by 65.7% and 27.7%, respectively, coinciding with Nkansah and Ito (1995), who mention that phosphorus is absorbed in greater quantities at higher temperatures.

The concentration of K in dry leaves in SM condition was significantly higher than that recorded in GRE and OF (Table 5). Nkansah and Ito (1995) point out that, in tomatoes, higher temperatures favor calcium absorption; in this work, the highest temperature and the highest concentration of calcium were recorded in GRE (Table 5).

The concentration of microelements, such as Mg, Cu, and Zinc, was significantly higher in GRE plants (Table 5). Antal *et al*. (2015) report that in field plants of *Origanum vulgare*, they showed a higher content of potassium and magnesium, whereas the concentration of calcium was lower than those reported in this study. These differences could be due to the genotype, as well as the soil type of the region of origin and growing conditions.

In this study, the Cu content in leaves registered a concentration similar to that reported in *O. vulgare* by Antal *et al*. (2015). In contrast, the concentration of Zn was higher. The content of Cu in oregano of GRE was significantly higher than that of SM or OF, significantly surpassing the latter by 53.7%. A higher concentration of Zn was recorded in GRE, significantly exceeding the concentration observed in SM and OF by 42.2 and 24.76%, respectively (Table 5).

Leaf mineral content in response to treatments

There were significant differences (*p*≤ 0.05) between treatments for six of the minerals studied, indicating that at least one treatment significantly affected the mineral content in oregano plants (Table 6).

Table 6. Concentration of mineral elements in oregano leaves, response to the application of chemical and organic nutrition treatments (Tukey *p*≤ 0.05).

Treatments	Mineral elements (mg kg^{-1})						
	N	P	K	Ca	Mg	Cu	Zn
	11 433 ab	2 636 g	8 393 abcd	13 847 b	1762 a	6.3 ab	24.1 b
2. S50C	13 922 ab	2 914 efg	9 507 a	14 128 b	1529 a	6.9 ab	26.2 ab
3. S100C	15 011 a	3 132 def	7 097 e	21 263 a	1778 a	5.9 ab	25.5 ab
4. S0A1	14 078 ab	2 838 fg	7 580 de	15 249 b	1557 a	5.9 ab	23.2 b
5. S50A1	13 300 ab	3 254 cde	8 879 abc	11 946 b	1507 a	5.6 b	25.8 ab

Treatments	Mineral elements (mg kg^{-1})						
	N	P	K	Ca	Mg	Cu	Zn
6. S100A1	11 278 ab	3 405 bcd	8 521 abcd	13 001 b	1389 a	6.7ab	28.4 ab
7. S0A2	11 433 ab	3 230 cde	8 260 cd	11 677 b	1411 a	5.8 b	21.6 b
8. S50A2	11 278 ab	3 669 ab	8 336 bcd	12 176 b	1636 a	7.1 ab	26.0 ab
9. S100A2	10 033 b	3 478 abcd	9 139 abc	12 288 b	1497 a	7.6 a	31.7 a
10. S0A3	13 611 ab	3 513 abc	7 593 de	15 820 b	1437 a	5.8 b	24.9 ab
11. S50A3	12 989 ab	3 564 abc	8 580 abcd	14 870 b	1626 a	6.7 ab	28.3 ab
12. S100A3	11 589 ab	3 794 a	9 022 abc	13 936 b	1526 a	6.4 ab	25.4 ab
13. S0CS	13 300 ab	3 408 bcd	8 491 abcd	15 577 b	1678 a	5.9 ab	23.2 b
14. S50CS	13 144 ab	3 635 ab	9 450 ab	13 250 b	1472 a	7.1 ab	28.4 ab
15. S100CS	11 589 ab	3 723 ab	8 676 abcd	16 169 b	1826 a	7.1 ab	31.5 a

Means with the same letter in the same column are significantly equal (Tukey $p \leq 0.05$).

Treatment 3 significantly outperformed treatment 9 by 49.6% in the concentration of N in dry leaves (Table 6). In these treatments, the difference between both treatments was the addition of the A2 strain; these results show that the rhizobacteria induced a reduction of N in leaves, and chemical fertilization had a positive effect on the N of leaves, similar to that reported by Juárez-Rosete *et al*. (2019) in *Origanum vulgare* L. sp. Hirtum, where a higher concentration of N was obtained with the 125% Steiner solution.

Treatment 12 showed the highest concentration of P; however, it was significantly equal to treatments 8, 9, 10, 11, 14, and 15 (Table 6). This shows that the interaction of SS and the type of strain used influences the amount of P in leaves. The results obtained are similar to those reported by Fukalova *et al*. (2021) in leaves of *O. vulgare*, whereas Juárez-Rosete *et al*. (2019) reported concentrations of 5 100 mg kg^{-1} in the same oregano species.

The authors Raffi and Charyulu (2021) mention that the application of *Azospirillum* contributes to the solubilization of phosphorus and facilitates the absorption of nutrients by the plant, coinciding with the results of this work, where treatments without bacteria and with the lowest chemical nutrition exhibited lower concentration of P. The highest concentration of K in oregano leaves was found with treatment 2, a value significantly higher than those obtained in treatments 3, 4, 7, 8, and 10, which it surpassed by 33.9%, 25.4%, 15.1%, 14%, and 20.1%, respectively (Table 6). Fukalova *et al*. (2021) obtained three times the concentration of K in *O. vulgare* with 2.8% of the weight of its tissues, whereas Juárez-Rosete *et al*. (2019) recorded a K amount of 55 700 mg kg^{-1}.

The availability of K is affected by the amount applied of this macroelement, the nature of the soil clays, and potassium saturation, which cause a redistribution of K in interchangeable and fixed forms (Conti, 2004). The highest amount of Ca in leaves was found with treatment 3, significantly exceeding 7 by 82.1%. In *O. vulgare*, Fukalova (2021) registered a lower concentration of Ca in its tissues, whereas in Mg, no significant differences were observed between treatments.

Treatment 9 presented the highest concentration of Cu in dry leaves, significantly surpassing treatments 5, 7, and 10 by 35.7%, 31%, and 31%, respectively. The results observed in this study differ from those reported by Fukalova (2021), who obtained a concentration of 4 000 mg kg^{-1} in *O. vulgare* (Table 6). The same treatment 9 also exhibited the highest Zn content, significantly outperforming treatments 1, 4, 7 and 13 by 31.53, 36.5, 46.7, and 36.6%, respectively. This indicates that chemical nutrition favored the higher concentrations of Zn in the leaf. The results indicate that none of the rhizobacteria evaluated had an effect on Zn content in oregano leaves; nevertheless, Arzanch *et al*. (2012) report that the application of *Azospirillum* increases the concentration of Zn in the plant.

Conclusions

In greenhouses, the largest crown diameter, total fresh weight, total dry weight, dry stem weight, and dry leaf weight were induced, resulting in greater production of biomass, which is what gives value to the oregano harvest, recommending its production in greenhouses.

The 100% Steiner solution registered the highest values in the agronomic variables studied, but all treatments with rhizobacteria and without Steiner solution were significantly equal to the application of 100% Steiner solution; therefore, the application of rhizobacteria is recommended for oregano production.

The highest content of N and K was induced in shade mesh, and the highest content of P, Ca, Mg, Cu, and Zn in the greenhouse, indicating that the production environment can favorably modify the mineral composition of the edible part of oregano.

The *Azospirillum* sp. strains, A1 and the commercial one, studied through environments and doses of chemical nutrition, registered the highest values in mineral elements in leaves.

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