



## Vermicompost humates as a salinity mitigator in the germination of basil

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**ABSTRACT:** *This study determined the effect of vermicompost humates as salinity mitigator in germination and morphometric characteristics of basil varieties seedlings, tolerant and sensitive to salinity, subjected to vermicompost humates diluted 1/60 (v/v) and 0, 50 and 100 mM NaCl in a completely randomized design with four replications. The rate and percentage of germination, root length, shoot height, root and shoot fresh and dry-weight were measured. The varieties showed differential response, highlighting Napoletano as the most tolerant. Vermicompost humates showed biostimulant effects on variables evaluated, allowing that tolerant variety to improve germination and growth, and increasing the salinity tolerance of the sensitive variety.*

**Key words:** *herbal species, biostimulant, NaCl, morphometric characteristics.*

## Vermicomposto humato como mitigador de salinidade na germinação de basil

**RESUMO:** *O objetivo foi determinar o efeito de humatas de vermicomposto como mitigador de salinidade na germinação e morfometria de mudas de variedades de manjericão, tolerantes e sensíveis à salinidade, submetidas a 0, 50 e 100 mM de NaCl e humatas de vermicomposto diluído 1/60 (v/v) em delineamento inteiramente casualizado com quatro repetições. Foram medidos a taxa e porcentagem de germinação, comprimento da radícula, altura da muda, comprimento da raiz, altura da parte aérea, raiz e biomassa fresca e seca da parte aérea. As variedades apresentaram resposta diferenciada, destacando-se o Napoletano como a mais tolerante. Humatos de vermicomposto mostraram efeitos bioestimulantes nas variáveis avaliadas, permitindo que a variedade tolerante melhorasse a germinação e o crescimento aumentando a tolerância à salinidade da variedade sensível.*

**Palavras-chave:** *bioestimulante de espécies aromáticas, NaCl, características morfométricas.*

## INTRODUCTION

One of the biggest problems that agriculture faces almost all over the world is salinity due to the harm that causes to the functions of plants (CHEN et al., 2008). In arid and semiarid regions, salinity is considered as the main environmental factor to limit the plant productivity (TESTER & DAVENPORT, 2003). Salinity reduces water absorption and restrains plants from growing. The salt concentration in the outer solution of cells results on osmotic drought, toxicity due to excessive absorption of Na and Cl, as well as a nutritional imbalance (KARIMI et al., 2005). Thus, alternative immediate solutions have been studied such as the exogenous. Among them are

humic substances that, according to CALDERÍN et al. (2012) influence the resistance to the salinity of beans. The same authors ascertained that different doses of vermicompost humates had an important effect on plants under conditions of salt stress. In the last two decades many biostimulants have been used in agriculture around the world, which have reduced the use of traditional mineral fertilizers. Overcome the situations of plant stress and environmental adversities, helped the plant growth and increased the agricultural yield (ROUPHAEL & COLLA., 2020). Aqueous extracts of humic substances are one of the alternatives within the group of products used in sustainable agriculture, mainly those that are obtained from recyclable organic sources such as compost and

vermicompost, given their stimulating effects known as “like-auxin” analogous to the phytohormones present in plants (MUSCOLO et al., 2007). The effects of humic substances on plant growth point to the positive influence on ion transport, facilitating the absorption and permeability of membranes; direct action on metabolic processes such as photosynthesis, respiration, and protein synthesis, by increasing or decreasing the activity of various enzymes and hormone-like activity of these substances, which are reflected in biochemical-physiological growth indicators (MUSCOLO et al., 2007). This product like other equivalents, have shown favorable effects in different crops, but the scientific literature does not collect information about its application in herbal plants in abiotic stress. In this context, studies are needed to know the effect of vermicompost humates on plants of economic interest that face saline stress conditions through the analysis of their growth.

The vermicompost is a process by which all types of biodegradable wastes such as farm wastes, kitchen wastes, market wastes, bio-wastes of agro based industries, livestock wastes, and others, are converted while passing through the worm-gut to nutrient rich vermicompost. These vermi-worms act as biological agents to consume those wastes and to deposit excreta in the process called vermicompost, which is capable of improving soil health and nutrient status (ADHIKARY, 2012)

Conversely, I basil (*Ocimum basilicum* L.) belongs to the family Lamiaceae containing aromatic essential oils such as eugenol, methyl eugenol, carvacrol and caryophyllin (HANDA & KAPOOR, 1989) located in the flowers of the plant, has pharmaceutical, aromatic and culinary properties, the aromatic and essential oil compounds present in the plant contain biologically active compounds with insecticidal, fungistatic y antimicrobial properties (OXENHAM et al., 2005) Basil is important economically because the world crop and the production of essential oil has a commercial value as edible of US\$15 million per year (BEGUM et al., 2002). The cultivation of organic basil is economically profitable, and Baja California Sur is the main producer of organic basil in the world; however, it faces the problem of water and saline soils. This study determined the effect of vermicompost humates as attenuating salt stress on germination and seedling morphometry of two varieties of basil, one tolerant and one sensitive to salinity, in order to elucidate the possible difference response of both varieties to the application of humates.

## MATERIALS AND METHODS

### *Site of study*

The experiment was carried out in February 2013 at the plant physiology laboratory in Centro de Investigaciones Biológicas del Noroeste, Mexico, located north of the city of La Paz, Baja California Sur, Mexico at 24° 08' 10" 03 "LN and 110° 25' 3" 31" LO, at 7 m above sea level.

### *Genetic material*

The varieties Napoletano and Sweet Genovese were used. These varieties were selected as tolerant and sensitive to salinity in previous experiments carried out in the germination, emergence, and initial vegetative development stages. In order to evaluate the seed quality of the varieties under study, a germination test was carried out using the methodology proposed by ISTA (2019).

### *Experimental design and treatments*

The experiment was performed in a completely randomized design with a trifactorial arrangement of  $2 \times 2 \times 3$ , where factor A were two varieties, Napoletano and Sweet Genovese, factor B were one concentration of vermicompost humates (1/60 v/v) and sterile distilled water as a control and factor C were three concentrations of NaCl (0, 50 and 100 mM), with four replicates of 30 seeds each. Seeds were previously disinfected by immersion for 5 min in a solution of calcium hypochlorite, containing 5 % active chlorine and subsequently washed with sterile distilled water. The seeds were placed in 150 × 15 mm Petri dishes with filter paper sheet as substrate. The dishes were moistened with 5 mL of the appropriate NaCl solution (0, 50 and 100 mM NaCl) and 3 mL of the appropriate dilution of vermicompost humates were daily added to each dish (1/60 v/v), considered a vegetable biostimulant and / or carrier of nutrients (Ca, Mg, Na, P<sub>2</sub>O<sub>5</sub>, K, N), amino acids, polysaccharides, carbohydrates, inorganic elements, humified substances, beneficial microorganisms, plant hormones and soluble humus. The composition of vermicompost humates by chemical fractions is shown in table 1. The Petri dishes with the seeds inside were incubated in a germination chamber (Lumistell, model IES-OS, series 1408-88-01) at a temperature of  $25 \pm 1$  °C, 80 % humidity and 12 hours continuous light. The germination was recorded daily, and the final percentage was determined at seven days. The germination rate was calculated using the Maguire equation (1962):  $M = n_1 / t_1 + n_2 / t_2 + \dots n_{30} / t_7$ ; where  $n_1, I \dots n_{30}$  are the number of seeds germinated

Table 1 - Effect of the interaction varieties × humates of vermicompost on the rate germination, percentage of germination and morphometric variables of seedlings of two varieties of basil subjected to NaCl-stress. The composition of the vermicompost humates by chemical fractions corresponds is included.

Varieties	Vermicompost	GR	GP (%)	RL (cm)	SH (cm)
Napoletano	0	6.82±0.23c*	67.77±3.44b	1.51±0.23b	1.18±0.16b
Napoletano	1/60	11.16±0.94a	84.16±3.76a	2.11±0.31a	1.65±0.15a
Sweet Genovese	0	9.00±0.59b	47.50±3.60c	0.92±0.16d	0.76±0.11d
Sweet Genovese	1/60	11.68±1.11a	63.61±3.49b	1.26±0.18c	1.04±0.13c
F-value	----	13.42	14.14	17.47	21.60
P-value	----	0.0007	0.0006	0.0001	0.00004
		RFW (mg)	RDW (mg)	SFW (mg)	SDW (mg)
Napoletano	0	15.79±1.62b	1.70±0.11b	172.50±14.57b	9.10±0.55b
Napoletano	1/60	21.08±1.98a	2.45±0.18a	245.83±22.81a	14.35±1.16a
Sweet Genovese	0	10.36±0.71d	0.95±0.05d	112.17±3.63d	6.92±0.43d
Sweet Genovese	1/60	13.08±0.97c	1.21±0.08c	139.17±6.79c	8.03±0.36c
F-value	----	17.91	70.59	61.96	253.97
P-value	----	0.0001	0.0000001	0.0000001	0.0000001
Composition of vermicompost humates by chemical fractions	pH	C (%)	H (%)	O (%)	N (%)
	8.7	53.4	4.85	35.6	3.05
	S (%)	O/C ratio	C/N ratio	Humic acids	Fulvic acids
	0.72	0.62	18.4	4.82	7.17

\*= Averages and standard error values with different letters in the same column does not differ from one another (Tukey HSD,  $P \leq 0.05$ ). GR= germination rate; GP= germination percentage; RL= root length; SH= shoot height; RFW= root fresh-weight; RDW= root dry-weight; SFW= shoot fresh-weight; SDW= shoot dry-weight. Values represent the average of four replicates. The composition of humic acids and fulvic acids in an E4/E6 ratio of its optical coefficient.

at times t1, t2, ... t7 (up to seven days). Seeds were considered germinated when the radicle was about 2 mm in length.

#### Morphometric variables

The germinated seeds were maintained for 14 days and 10 seedlings per replicate were randomly selected, and the root length (cm), shoot height (cm), root and shoot fresh and dry weight (mg) were determined by the destructive method. These variables were determined by dividing each seedling into shoots (stems + leaves) and roots and weighing each separately, using for that purpose an analytical balance. (Mettler Toledo, AG204). Subsequently, the corresponding weights were added, which were expressed in milligrams of fresh matter. The fresh weight of roots and shoots were obtained and then, the tissues were placed in paper bags and disposed in a drying oven (Shel-Lab, FX-5, series-1000203) at a temperature of 80 °C until obtaining constant

weight (approximately 72 hours). After that, they were weighed using an analytical balance (Mettler Toledo, AG204) expressing the weight in milligrams of dry-matter.

#### Statistical analysis

The homogeneity of variance was confirmed for the data set by employing Bartlett's test. Once it was reported that the homogeneity of variance was within acceptable ranges a three-way ANOVA was carried-out with basil varieties as factor A, vermicompost humates as factor B and NaCl concentrations as factor C. The Tukey HSD test where run to test for mean differences at  $P \leq 0.05$ . To comply with the assumptions of normality, the data of the germination expressed as a percentage was transformed by arcsine according to LITTLE & HILLS (1989) and STEEL & TORRIE (1995). The statistical analyzes were performed using the GLM module in Statistica v. 10.0 (STATSOFT, INC., 2011).

## RESULTS AND DISCUSSION

### Rate and percentage of germination

The germination rate (GR), showed significant differences between varieties, vermicompost humates, NaCl, the interactions varieties  $\times$  humates, varieties  $\times$  NaCl and humates  $\times$  NaCl. The interaction varieties  $\times$  humates  $\times$  NaCl, showed no significant differences. The analysis if the interactions showed that in both varieties, the GR increased when the vermicompost humate was applied, being slightly higher in Sweet Genovese in both control and 1/60 dilution (Table 1). Sweet Genovese in two NaCl concentrations revealed the highest GR; however, GR decreased in both varieties as NaCl concentrations increased (Table 2). Although, the interaction varieties  $\times$  humates  $\times$  NaCl showed no significant differences, Napoletano showed the highest GR at 1/60 and 100 mM NaCl and GR increased in both varieties regarding the control at the dilution of 1/60 humates at all NaCl concentrations. Napoletano showed the lowest GR in 0 vermicompost and 100 mM NaCl (Table 3).

The germination percentage (GP) showed significant differences between varieties, vermicompost humates, NaCl, the interactions varieties  $\times$  humates, humates  $\times$  NaCl, varieties  $\times$  NaCl and varieties  $\times$  humates  $\times$  NaCl. The interactions revealed that Napoletano showed greater GP both in control and in the 1/60 dilution of humates and in both varieties GP increased with the application of humates (Table 1). Also, Napoletano showed the highest GP in all NaCl concentrations, but GP decreased in both varieties as NaCl increased (Table 2). The interaction varieties  $\times$  humates  $\times$  NaCl showed that GP was highest in Napoletano in 1/60 humates and 0 mM NaCl; however, in both varieties the GP increased regarding the control at the dilution of 1/60 humates at all NaCl concentrations. Sweet Genovese showed the lowest GP in 0 humates and 100 mM NaCl. Results showed an increase in GP when the vermicompost humates was applied, which coincides with the results reported by CHEN et al. (2004) and those obtained by DAVID et al. (1994) in studies using different doses of vermicompost

Table 2 - Effect of the interaction varieties  $\times$  NaCl on germination rate, germination percentage and morphometric variables of seedlings of two varieties of basil subjected to NaCl-stress.

Varieties	NaCl (mM)	GR	GP (%)	RL (cm)	SH (cm)
Napoletano	0	11.50 $\pm$ 1.48a*	90.83 $\pm$ 3.60a	3.01 $\pm$ 0.20a	2.07 $\pm$ 0.09a
Napoletano	50	8.30 $\pm$ 0.57b	74.16 $\pm$ 3.37b	1.61 $\pm$ 0.07b	1.40 $\pm$ 0.06b
Napoletano	100	7.17 $\pm$ 0.50b	62.91 $\pm$ 3.04c	0.82 $\pm$ 0.07d	0.78 $\pm$ 0.11e
Sweet Genovese	0	13.77 $\pm$ 1.01a	69.16 $\pm$ 3.06b	1.73 $\pm$ 0.08b	1.26 $\pm$ 0.05c
Sweet Genovese	50	10.04 $\pm$ 0.51a	55.83 $\pm$ 3.65d	1.18 $\pm$ 0.06c	1.10 $\pm$ 0.08d
Sweet Genovese	100	7.20 $\pm$ 0.36b	41.66 $\pm$ 3.21e	0.36 $\pm$ 0.05e	0.35 $\pm$ 0.03f
F-value	-----	8.97	18.28	80.52	50.66
P-value	-----	0.0006	0.000003	0.0000001	0.0000001
		RFW (mg)	RDW (mg)	SFW (mg)	SDW (mg)
Napoletano	0	26.37 $\pm$ 1.48a	2.66 $\pm$ 0.18a	290.00 $\pm$ 21.21a	14.62 $\pm$ 1.19a
Napoletano	50	16.62 $\pm$ 0.75b	2.10 $\pm$ 0.17b	195.00 $\pm$ 13.09b	12.46 $\pm$ 1.46a
Napoletano	100	12.31 $\pm$ 0.94c	1.46 $\pm$ 0.08c	142.50 $\pm$ 9.20c	8.10 $\pm$ 0.36a
Sweet Genovese	0	15.12 $\pm$ 0.81b	1.35 $\pm$ 0.08c	138.75 $\pm$ 8.33c	9.18 $\pm$ 0.14b
Sweet Genovese	50	11.51 $\pm$ 0.64c	1.09 $\pm$ 0.05d	135.00 $\pm$ 5.34c	7.22 $\pm$ 0.26b
Sweet Genovese	100	8.53 $\pm$ 0.28d	0.81 $\pm$ 0.02e	103.25 $\pm$ 3.35d	6.02 $\pm$ 0.24b
F-value	-----	57.23	43.30	136.62	70.52
P-value	-----	0.0000001	0.0000001	0.0000001	0.0000001

\*= Averages and standard error values with different letters in the same column does not differ from one another (Tukey HSD,  $P \leq 0.05$ ). GR= germination rate; GP=germination percentage; RL= root length; SH= shoot height; RFW= root fresh-weight; RDB= root dry-weight; SFB= shoot fresh-weight; SDB= shoot dry-weight. Values represent the average of four replicates.

Table 3 - Effect of the interaction varieties × humates of vermicompost × NaCl on the germination rate, germination percentage and morphometric variables of seedlings of two varieties of basil subjected to NaCl-stress.

Varieties	Vermicompost (v/v)	NaCl (mM)	GR	GP (%)	RL (cm)	SH (cm)
Napoletano	0	0	7.58±0.11a*	81.66±2.15b	2.50±0.04b	1.82±0.02b
Napoletano	1/60	0	15.41±0.17a	100.00±0.00a	3.52±0.12a	2.32±0.04a
Napoletano	0	50	6.83±0.15a	65.83±2.09d	1.42±0.02d	1.22±0.02def
Napoletano	1/60	50	9.77±0.32a	82.50±1.59b	1.80±0.04c	1.57±0.02c
Napoletano	0	100	6.06±0.41a	55.83±2.84ef	0.62±0.04f	0.50±0.04g
Napoletano	1/60	100	8.29±0.43a	70.00±1.36cd	1.02±0.02e	1.07±0.04fg
Sweet Genovese	0	0	11.16±0.47a	61.66±1.66de	1.52±0.04d	1.12±0.02ef
Sweet Genovese	1/60	0	16.37±0.23a	76.66±1.92bc	1.95±0.06c	1.40±0.04cd
Sweet Genovese	0	50	8.95±0.48a	46.66±2.35f	1.02±0.02e	0.90±0.04f
Sweet Genovese	1/60	50	11.12±0.47a	65.00±0.96de	1.35±0.05d	1.30±0.04de
Sweet Genovese	0	100	6.87±0.65a	34.16±2.84g	0.22±0.02g	0.27±0.04hi
Sweet Genovese	1/60	100	7.54±0.35a	49.16±1.59f	0.50±0.04f	0.42±0.02gh
F-value	---	---	1.41	14.01	7.76	10.24
P-value	---	---	0.25	0.00003	0.001	0.0003
			RFW (mg)	RDW (mg)	SFW (mg)	SDW (mg)
Napoletano	0	0	22.75±0.85a	2.17±0.07c	235.00±6.45b	11.50±0.28c
Napoletano	1/60	0	30.00±0.91a	3.15±0.06a	345.00±6.45a	17.75±0.25a
Napoletano	0	50	14.75±0.47a	1.65±0.06d	162.50±4.78c	8.67±0.08ef
Napoletano	1/60	50	18.50±0.28a	2.55±0.02b	227.50±8.53b	16.25±0.62b
Napoletano	0	100	9.87±0.12a	1.27±0.04e	120.00±4.08ef	7.15±0.06fg
Napoletano	1/60	100	14.75±0.47a	1.65±0.06d	165.00±6.45c	9.05±0.06d
Sweet Genovese	0	0	13.25±0.75a	1.15±0.06ef	117.50±2.50ef	8.82±0.09ef
Sweet Genovese	1/60	0	17.00±0.40a	1.55±0.02d	160.00±4.08c	9.55±0.06d
Sweet Genovese	0	50	10.02±0.34a	0.95±0.02fg	122.50±2.50de	6.55±0.06g
Sweet Genovese	1/60	50	13.00±0.57a	1.22±0.04e	147.50±4.78cd	7.90±0.14ef
Sweet Genovese	0	100	7.82±0.16a	0.76±0.01g	96.50±2.36f	5.40±0.07h
Sweet Genovese	1/60	100	9.25±0.06a	0.87±0.03g	110.00±4.08ef	6.65±0.06g
F-value	---	---	2.18	3.82	3.41	45.70
P-value	---	---	0.12	0.03	0.04	0.000001

\*= Averages and standard error values with different letters in the same column does not differ from one another (Tukey HSD, P ≤ 0.05). VH= vermicompost humates; GR= germination rate; GP= germination percentage; RL= root length; SH= shoot height; RFW= root fresh-weight; RDW= root dry-weight; SFW= shoot fresh-weight; SDW= shoot dry-weight. Values represent the average of four replicates.

humates. They observed that humates improved the GP in the different NaCl concentrations, inhibiting this abiotic effect. The improvements caused by the vermicompost humates indicated a certain bioprotective effect of these materials on the plant development, when is cultivated under saline stress. The bio stimulating effects of humic substances on germination have been confirmed by LOVLEY et al.

(1996) for the generation of stimuli in the enzymatic activity of seeds, as well as the presence in the humic material of free semiquinone radicals that are able to intervene in the respiratory chains, increasing the energy supply to the cells. According to GOMEZ et al. (1999), REYES-PÉREZ et al. (2014) and PÉREZ et al. (2017) the main mechanism of specific toxicity caused by NaCl is the generation of high

superoxide radicals, which provoke oxidative stress in the mitochondria. The bioprotective effect shown by vermicompost humates on the germination of basil varieties subjected to NaCl concentrations is due to the capture of the free radicals generated by the NaCl toxicity by those present in the humic substances. In this way the oxidative stress of the mitochondria decreases.

#### *Morphometric variables*

The root length (RL) showed significant differences between varieties, vermicompost humates, NaCl, the interactions varieties  $\times$  humates, varieties  $\times$  NaCl, humates  $\times$  NaCl and varieties  $\times$  humates  $\times$  NaCl. The interactions of the factors, revealed that Napoletano showed higher RL in both the control and the 1/60 dilution of humates and both varieties increased RL in the 1/60 dilution of humates (Table 1). Napoletano also showed higher RL, but RL decreased in both varieties as NaCl increased (Table 2). The interaction varieties  $\times$  humates  $\times$  NaCl showed that Napoletano had the highest RL in the dilution of 1/60 humates at 0 mM NaCl; in both varieties RL perfectly increased with respect to the control at the dilution of 1/60 humates at all NaCl concentrations. The lowest RL was showed by Sweet Genovese in 0 of humates and 100 mM NaCl (Table 3). Although, root development is severely affected by saline stress, humic acids from vermicompost humates positively influence growth. This is due to the fact that humic acids promote increases in the permeability of the cell membrane in basil, so that despite the hypertensive conditions of the saline environment, they facilitate the imbibition of the seed, to solubilize the starches and this favor the availability of carbohydrates for root growth. Another effect is due to its function as regulator or growth promoter (NARDI et al., 2000). These results are in agreeing with KULIKOVA et al. (2003) who noted that humic substances facilitate the absorption of nutrients, especially those that are deficient. KHALED & FAWY (2011) reported that the humic acid adsorbed to the plant cells at the surface level, increase its permeability, which favors the absorption of nutrients. The negative initial effect observed in the growth of the seedlings in saline environments was counteracted by the action of the humic substances applied, inducing a sudden subsequent growth. According to the results of this study, it is evident that basil requires a certain number of soluble salts for the growth of the shoots. This requirement, expressed as tolerance, appears to occur once the seedling leaves the autotrophic stage. Although, it shows some sensitivity to salinity at the

interface to the heterotrophic stage; once heterotrophy is reached, tolerance to salt-stress appears.

The root fresh-weight (RFW) showed significant differences between varieties, vermicompost humates, NaCl, the interactions varieties  $\times$  humates, humates  $\times$  NaCl, varieties  $\times$  NaCl. The interaction varieties  $\times$  humates  $\times$  NaCl showed no significant differences. Napoletano showed higher RFW in both the control and the 1/60 dilution of humates, observing that in both varieties, RFW increased when the humate was added (Table 1). The table 2 shows that Napoletano had the highest RFW in the three concentrations of NaCl. Root fresh-weight decreased in both varieties as NaCl increased. Although, the interaction varieties  $\times$  humates  $\times$  NaCl showed no significant differences, Napoletano exposed the highest RFW in the dilution of 1/60 humates and 0 mM NaCl; in both varieties the RFW increased regarding the control in the dilution of 1/60 humates at all NaCl concentrations. The lowest RFW was observed in Sweet Genovese in 0 of humates and 100 mM NaCl and (Table 3).

The root dry-weight (RDW) showed significant differences among varieties, vermicompost humates, NaCl, the interactions varieties  $\times$  humates, varieties  $\times$  NaCl and varieties  $\times$  humates  $\times$  NaCl. Napoletano showed higher RDW in both control and 1/60 dilution of vermicompost humates. It was observed that for both varieties RDW increased when vermicompost humates was applied (Table 1). The table 1 shows that Napoletano exhibited the higher RDW, which decreased in both varieties as NaCl increased. The interaction varieties  $\times$  humates  $\times$  NaCl showed Napoletano with the highest RDW in the dilution of 1/60 humates and 0 mM NaCl; however, in both varieties the RDW increased regarding the control at the dilution of 1/60 humates at all NaCl concentrations. The lowest RDW was for Sweet Genovese in 0 of humates and 100 mM NaCl (Table 3).

Seedling height (SH) showed significant differences between varieties, vermicompost humates, NaCl, the interactions varieties  $\times$  humates, varieties  $\times$  NaCl, varieties  $\times$  humates  $\times$  NaCl. The humates  $\times$  NaCl interaction did not show significant differences. Napoletano showed the highest SH both in control and in the 1/60 dilution of humates and in both varieties the SH increased with the application of humates (Table 1). The table 2 shows that Napoletano exhibited the higher SH, which decreased in both varieties as NaCl increased. In the interaction varieties  $\times$  humates  $\times$  NaCl Napoletano showed the highest SH in the dilution of 1/60 humates and 0 mM NaCl; however, in both varieties SH increased

regarding the control at the dilution of 1/60 humates at all NaCl concentrations. The lowest SH was for Sweet Genovese in 0 of humates and 100 mM NaCl (Table 3). The growth of the shoot was affected as the NaCl concentration was increased; however, the application of vermicompost humates counteracted the negative effect of salinity, which coincide with the results resported by YLDRM et al. (2003).

Shoot fresh-weight (SFW) showed significant differences between varieties, vermicompost humates, NaCl, the interactions varieties  $\times$  humates, varieties  $\times$  NaCl, humates  $\times$  NaCl and varieties  $\times$  humates  $\times$  NaCl. Napoletano showed higher SFW in both the control and the 1/60 dilution of vermicompost humates. In both varieties, SFW increased when the vermicompost humates was applied (Table 1). Napoletano showed the higher SFW, which decreased in both varieties as NaCl concentrations increased (Table 2). The interaction varieties  $\times$  humates  $\times$  NaCl showed that Napoletano had the highest SFW in the dilution of 1/60 humates and 0 mM NaCl; however, in both varieties the SFW increased regarding the control at the dilution of 1/60 humates at all NaCl concentrations. The lower SFW was for Sweet Genovese in 0 of humates and 100 mM NaCl and (Table 3).

The concentration of vermicompost humates promoted the height of the seedling and this dose counteracted the saline stress, since the dilution of 1/60 (v/v) mitigated the negative effect of NaCl-stress. These results are agreeing with those reported by Chen and Aviad (1990) who reported that plants treated with humic acids applied as composites showed higher growth compared to controls. BUNISELLI et al. (1990) in *Zea mays* reported weight gain, plant height, ear length and grain yield when applied humic acids. NARDI et al. (2002) stated that the greatest growth of stem when applying humic substances was due to the activation of cell division in the younger parts, which promotes longer stem lengths. MICELI et al. (2003) reported that those plants grown under salinity conditions showed inhibition in their growth, with reduction of their size. The root dry-weight increased when vermicompost humates was applied; however, decrease as a function of the increase of the NaCl concentrations. In this study, SH decreased as NaCl concentrations increases. These results coincided with those reported by COOPER et al. (1998) who conducted an experiment with *Agrostis stolonifera* L. to determine whether humic substances could increase the biomass, length, and nutrient uptake, the humic acids were sprayed at doses of 0, 100, 200 and 400 mg L<sup>-1</sup>. They reported

that roots dry-matter increased 45 % compared to the control, but the N, Ca Mg and Fe uptake were not significant. Conversely, ROSE et al. (2014) considers that humic substances, regardless of their origin and concentration, improved the formation and number of roots, as well as the dry matter.

Shoot dry-weight (SDW) revealed significant differences between varieties, vermicompost humates, NaCl, the interactions varieties  $\times$  humates, varieties  $\times$  NaCl, humates  $\times$  NaCl and varieties  $\times$  humates of vermicompost  $\times$  NaCl. Napoletano showed higher SDW in both control and 1/60 dilution of vermicompost humates; however, both varieties increased SDW when the humates were applied (Table 1). The table 2 shows that Napoletano had the highest SDW, which decreased in both varieties as NaCl increased. The interaction varieties  $\times$  humates  $\times$  NaCl showed Napoletano with the highest SDW in the dilution of 1/60 humates and 0 mM NaCl; however, in both varieties the SDW increased regarding the control at the dilution of 1/60 humates at all NaCl concentrations. The lowest SDW was for Sweet Genovese in 0 of humates and 100 mM NaCl (Table 3).

In this study the increase of shoot fresh and dry weight when vermicompost humate was applied suggested that humates mitigate the negative effect of the NaCl-stress, since this effect is evidenced in the biomass production. About this, DEGANO (1999) evaluated two sources of salt (NaCl and NaSO<sub>4</sub>) on the growth and fresh-weight of stem of *Tessaria absinthoides* and reported that as salt concentration increased, regardless of source, fresh-weight decreased, attributing this decrease to the distance and length that the internodes had, which gave the plant a stunted appearance. This response is attributed to the osmotic effect caused by the saline solution, which hinders the water regime of the plants and the toxic effect of the ions that interfere in metabolic processes such as the synthesis of carbohydrates and the transport of photosynthetic products, as well as its use in the production of new tissues. Shoot dry biomass indicated the accumulation of nutrients, minerals and metabolites in the foliar structures, whose variation depends on the fertility and mineral condition of the soil and water. MATA-GONZÁLEZ & MELÉNDEZ-GONZÁLEZ (2005) studied the Mexican oregano (*Lippia berlandieri*) under saline stress. They reported that the leaves and stems significantly reduced their dry-weight inversely to the increase of salinity, but not in the case of the root; which although lost weight, this was not significant. This study showed a positive effect of vermicompost humates on the accumulation

of shoot dry-weight. Similar results were reported by DEMIR et al. (1997) who carried out a study using *Cucumis sativus* L. as an indicator plant cultivated under salinity and applying humic acids and reported that stem dry-weight decreased with salinity and that humic acid partially counteracted this effect. Also, they reported a lower content of K, but increases in Ca, Mg and Fe content, while Zn remained constant. Meanwhile, VEOBIDES (2018) argued that the use of humic acids generate a permeability of the cellular membrane at root level that causes an increase in the absorption of nutrients, concentrations that are exhibited in the shoot (leaves + stems) of the plants.

The negative effect of saline stress on plant growth has been discussed. TAYLOR & LOCASSIO (2004) suggested that while saline stress increases, the shoot dry-weight reduce; this phenomenon occurs due to Ca deficiency, which weakens and reduces cell division, as well as others not determined physiological disorders. Conversely, the vermicompost humate mitigates this negative effect by facilitating the absorption of several essential elements, among them Ca, hence the trends observed in the present study.

CHEN & AVIAD (1990) affirmed that the use of humic acid can increase the dry-weight of the leaf regardless of the application form. The results of the present study coincide with those reported by ACEVEDO & PIRE (2004) who carried out studies in *Carica papaya* L., applying vermicompost as a source of humic acid and ammonium sulfate. They evaluated leaf area, plant height, stem diameter and total dry matter, finding increases when the higher dose of vermicompost and the mean dose of vermicompost with ammonium sulfate were applied. The increase in dry-matter is attributed to the humic acid of vermicompost, which contains substances that stimulate growth, especially in young plants. The results of the present study are useful as a starting point for new research, together with the notorious shortage of experiences and previous studies on the growth and development patterns of basil under saline and arid environments. There are several reports of research, several theoretical explanations on the reduction of the size of plants growing in saline environment, as well as various reports of the beneficial action of humic acids on the growth of cultivated plants, but in both cases, theories have not yet been tested.

## CONCLUSIONS

A differential response among varieties for germination rate, germination percentage and

morphometric variables under NaCl-stress and the application of vermicompost humates was observed, emphasizing that Napoletano was the most tolerant, showing the highest values in all variables with the application of the vermicompost humates.

The vermicompost humates have bio stimulating effects on the germination rate, germination percentage and morphometric variables of basil varieties under NaCl-stress, allowing the tolerant variety to improve its germination and growth and the sensitive variety increase its tolerance to salinity.

The use of vermicompost humates can be beneficial for producers who want to increase their production, and it is important for those who produce in more salinized areas.

## ACKNOWLEDGEMENTS

To the Center for Biological Research of the Northwest (CIBNOR) for the support granted through the AGROT1 and 110C (SAGARPA-CONACYT) projects. To Carmen Mercado Guido, Lidia Hiraes Lucero and Laura Elena Manjarrez Esquivel for their technical support. To CONACYT for the grant (245853) granted to the author responsible for the publication through the modality program of national short visits within the framework of the call for national sabbaticals, sabbaticals abroad and short stays for the consolidation of research groups (2014, second stage). To CONACYT for the grant 224216 dedicated for scientific and technological infrastructure 2014.

## DECLARATION OF CONFLICT OF INTEREST

The authors declare no conflict of interest. The founding sponsors had no role in the design of the study; in the collection, analyses, or interpretation of data; in the writing of the manuscript, and in the decision to publish the results.

## AUTHORS' CONTRIBUTIONS

JJRP, FHRE, BMA generated the idea and designed the research. JJRP, FHRE, PPR and EORP developed the research. BMA and LGHM analyzed the data. PPR and LGHM contributed materials, reagents, analysis tools. BMA, FHRE, EORP and LGHM wrote, revised and edited the paper. JJRP, PPR, EORP, LGHM, BMA and FHRE approved the lastet paper version.

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