Article

Germination of *Salicornia bigelovii* (Torr.) under Shrimp Culture Effluents and the Application of Vermicompost Leachate for Mitigating Salt Stress

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Abstract: Attenuators of salt-stress favor the use of effluents, being a low-cost organic product. The present study aimed to evaluate the effect of vermicompost leachate (VL) added to shrimp-culture effluent on seeds and seedlings of *Salicornia bigelovii* (Torr.) under salt-stress, evaluating germination, water relations variables, and biomass. Seeds were irrigated with distilled water (DW) (CE: 0.0027 dS m⁻¹), freshwater (FW) (CE: 1.36 dS m⁻¹), seawater (SW) (CE: 55.83 dS m⁻¹), shrimp residual water (SRW) (CE: 59.85 dS m⁻¹), and with the same water sources but adding VL in 1:20 v/v. The means for the index of germination rate (IGR), germination energy (GE), and germination time (MGT) were higher with DW, DWVL, FW, and FWVL, decreasing with the others ($p \leq 0.05$). In seedlings, the lowest water potential (Ψ_w) and osmotic potential (Ψ_s) were observed with SW, SWVL, SRW, and SRWVL ($p \leq 0.05$), evidencing higher stress but the highest relative water content (RWC). The fresh and dry biomass increased and showed significant differences with SRW, and adding VL (DWVL, FWVL, SWVL, and SRWVL) acted as an effective attenuator of salt-stress. The response of water relations variables suggested an osmotic adjustment for mitigating the salt-stress in seedlings, lowering the Ψ_w and Ψ_s but increasing the RWC.

Keywords: *Salicornia bigelovii*; halophyte; vermicompost; aquaculture effluent; organic crop; salt stress

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1. Introduction

Salinity is a global scale problem affecting around 77 Mha of agricultural soils worldwide and around 20 to 33% of global irrigated land [1]. In Mexico, about 13% of agricultural soils are salt affected, mainly in the northern part of the country [2]. This situation has generated the need to explore crops with commercial agronomic traits that are capable of growing in saline environments, without compromising the use of fresh water [3].

Salicornia bigelovii (Torr.) is a halophyte plant that requires saline water during most of its lifecycle and is reported as a promising crop due to its multiple uses for human food [4], forage [5,6], and biofuel production [7]. *S. bigelovii* is distributed in North America, on the coastal areas of the Atlantic, the Gulf of Mexico, the Caribbean and the coast of South California and adjacent Mexico [8]. In Mexico, it is distributed along the coastal wetlands of Sonora and Baja California Sur, where plants are flooded for intermittent periods due to tidal flows [9]. It is considered as one of the most salt-tolerant halophyte plants due to its adaptation to seawater along its lifecycle [4,10]. This feature allows the use of residual seawater from aquaculture activities as an irrigation source for this novel crop.

Marine and coastal aquaculture effluents contain a high amount of nutrients, organic matter, and salts which cause harmful effects to the environment when they are released untreated into the sea, causing different problems such as eutrophication, contamination, hypoxia, and loss of biodiversity [11,12]. *Salicornia* plant species have been used as a biofilter for aquaculture effluents allowing the use of the nutrients and minerals from these water sources and reducing the risk of contamination of marine ecosystems [13]. It is an obligate halophyte; however, it is prone to develop salinity stress during its early development stages when it is exposed to high-salinity irrigation water such as aquaculture effluents [14].

The effects of saline stress on crop growth and productivity are dependent on the duration, intensity, sensitivity, and the phenological stage of the target crop [15]. Germination and seedling stages are especially vulnerable to saline stress in halophytes [16]. Salt accumulation in the soil solution increases the osmotic pressure, reducing its water potential and radically limiting the absorption of water by seeds and seedlings [17]. As a physiology strategy, these plants develop different mechanisms, mainly an osmotic adjustment reducing the cellular water potential through the synthesis of compatible osmolytes, which favors the absorption of saline water [18]. Former studies about the osmotic adjustment have focused on different species of *Salicornia* [19,20]; however, such information is incipient for *S. bigelovii*, specifically during early life-cycle stages, germination and seedling emergence.

Organic fertilizers such as vermicompost leachate (VL) are used in agriculture because they increase crop yields by improving the availability of nutrients for plants [21]. The presence of antioxidants, phytohormones, and humic acids has been reported in vermicompost leachates [22], contributing to attenuate the harmful effects of saline stress on seedlings [23]. A former study reported that the application of VL increased the growth of tomato seedlings by improving root development, total leaf area, number of leaves and stem thickness when submitted to saline stress by NaCl [24]. In this sense, it has been reported that the foliar application of VL in pomegranate seedlings reduced its Na⁺ concentration when NaCl treatments were applied, and also that chlorophyll degradation and oxidative stress diminished with it, suggesting its effect as an attenuating agent against salt stress [15]. At present, there is a lack of information about the use of VL to improve the germination and survival of *S. bigelovii* seedlings and to mitigate the effect of saline stress caused by marine aquaculture effluents.

The aim of this study was to evaluate the effect of VL on *S. bigelovii* seeds and seedlings under saline stress imposed by seawater aquaculture effluents. In this study, we investigated the use of different qualities of water and doses of VL to improve the germination and seedling stages of *S. bigelovii*.

2. Materials and Methods

2.1. Study Site and Plant Material

The experiment was carried out in the Center for Biological Research of Northwest México S.C. (CIBNOR) located north of the city of La Paz, Baja California Sur, Mexico with coordinates 24°08'10.03" N and 110°25'35.31" W (Figure 1).

S. bigelovii seeds were collected from September to October 2018 from mature plants located on the coast of Comitán, Baja California Sur, with coordinates 24°1' N and 110°2' W, adjacent to CIBNOR. Mature plants were sundried for 72 h (Figure 2a), then grounded to obtain the dry biomass with the seeds. The separation of seeds from dry plant fragments was carried out by pouring ~700 mL of distilled water into a beaker where the seeds that were deposited on the bottom by gravity were collected after 24 h (Figure 2b). Then, the seeds were dried at room temperature and sieved to select the largest seeds with uniform color and without apparent damage. Subsequently, they were sterilized by immersion in sodium hypochlorite (3%) for 30 min and finally rinsed three times with distilled water.

Figure 1. Satellite image from the Center for Biological Research of the Northwest México S.C. (CIBNOR), the red area indicates the seed collection area of *Salicornia bigelovii* (Torr.).

Figure 2. Sundried mature *Salicornia bigelovii* (Torr.) plants: (a) for seed separation, (b) at the CIBNOR Plant Physiology Technical Laboratory.

2.2. Experimental Design

The experiment was established under controlled conditions in a germination chamber (Lumistell, model IES-OS, series 1408-99-01) at 25 ± 1 °C, with a relative humidity of 80%, and a 12/12 h night/day photoperiod.

Eight treatments of different water quality sources were applied: (1) distilled water (DW) (CE: 0.0027 dS m^{-1}), (2) freshwater (FW) (CE: 1.36 dS m^{-1}), (3) seawater (SW) (CE: 55.83 dS m^{-1}), (4) shrimp residual water (SRW) (CE: 59.85 dS m^{-1}), and another group with the same water sources plus the addition of vermicompost leachates (VL) in a dilution 1:20 *v/v* (volume). Watering was provided around every four days and 8 mL of treatment solution was added every time; although this was not always with high precision since evaporation was uneven in the Petri dishes, the added amount maintained a homogeneous volume. A total of 100 seeds were sown over filter paper circles (Whatman no. 4) in sterilized Petri dishes ISTA [25]. Every treatment was established with four replicates forming 32 experimental units, distributed under a completely randomized design.

The chemical composition of treatments was analyzed in the CIBNOR Water Quality Laboratory before the experiments started (Table 1). The electric conductivity (EC), total dissolved solids (TDS), salinity and pH were measured at the Edaphology Laboratory of CIBNOR with a portable conductivity meter Thermo Scientific © model Orion Star A222. Nitrates (NO_3^-) [26] and Nitrites (NO_2^-) [27] were measured with a Lachat ion autoan-

alyzer. Sulfates (SO_4^{2-}) [28] were measured in a portable photometer HANNA model HI 96751 in the Water Quality Laboratory of CIBNOR. Calcium (Ca^{2+}), potassium (K^+), sodium (Na^+) [29] and phosphates (PO_4^{3-}) [30] were measured in a portable photometer HANNA model HI83303 in the Plant Physiology Technical Laboratory of CIBNOR.

Table 1. Physico-chemical analyses of water quality variables of treatments.

Treatments	E.C.	TDS	Salinity	pH	NO_2^-	NO_3^-	PO_4^{3-}	SO_4^{2-}	Ca	K^+	Na^+
	dS/m	ppt	psu		μM			mg/L			
DW	0.0027	0.0018	0.061	5.74	ND	ND	ND	ND	ND	ND	ND
DWVL	0.87	0.348	0.164	6.57	0.952	849.05	12.4	42.75	52	6.2	36
FW	1.36	0.669	0.728	8	<0.1	416	0.481	58.9	114	5.8	240
FWVL	1.84	0.894	0.967	8.33	1.150	1182.6	9.25	99.3	173	20	280
SW	55.83	27.36	36.77	7.39	<0.1	1.32	1.16	2181.8	480	6.2	3348
SWVL	53.92	26.42	35.37	7.7	0.995	809.01	13.15	3250.9	400	20	3218
SRW	59.85	29.33	38.7	6.88	63	1305.75	57.6	3013.6	348	17.1	3894
SRWVL	58.98	28.9	38.06	6.91	98.5	2255.25	66.5	3600	192	20	3801
VL	11.93	5.85	6.65	8.75	*	1500	*	*	110	*	640

DW: Distilled Water. FW: Freshwater. SW: Seawater. SRW: Shrimp Residual Water. VL: Indicates the addition of vermicompost leachates. *: Not quantified. ND: None detected. ($n = 3$).

Vermicompost was produced at CIBNOR experimental field according to the methodology suggested by Acosta-Durán et al. [31]. The vermicomposting process was carried out in 200-L containers cut in half to which 5 holes were made in its base. Subsequently, a 5-cm-thick layer of gravel and an anti-aphid mesh were placed to separate the gravel from the bed where the earthworms developed. Five kilograms of kitchen waste were used as food for the earthworms in the following proportions: banana peels (25%), watermelon peel (15%), papaya peel (15%), coffee grounds (15%), orange peel (10%), cucumber peel (10%), carrot peel (5%), and tomato pieces (5%). The kitchen waste was pre-composted for 21 days before being used as food for the earthworms. The feeding process was carried out using 5-cm-thick layers of pre-composted food every week for 12 weeks. A total of 400 adult *Eisenia foetida* earthworms were placed at the beginning of the feeding process. It was considered that the vermicomposting process ended when a homogeneous material was observed without the presence of remnants of the original material. At this point, the vermicompost was separated to be laid and sheltered in a dry place and away from light for 90 days for its mineralization.

Afterwards, VL were obtained according to the methodology described by Bidabadi et al. [15], where five kilograms of vermicompost were placed in a 10 L container to which five holes were made at the bottom with a 1 mm drill bit. Three liters of distilled water were poured into the bucket and then the leachate was collected in a container.

Physico-chemical analyses were made at the Edaphology Laboratory of CIBNOR (Table 2). EC, TDS, pH, organic matter (O.M.), total nitrogen (TN), NO_2^- , NO_3^- , P, Ca^{2+} and Mg^{2+} were quantified, [32–36].

Table 2. Physico-chemical analyses of vermicompost; CIBNOR, La Paz BCS, México.

Biomaterial	EC	TDS	pH	OM	N_{total}	NO_2^-	NO_3^-	P	Ca^{2+}	Mg^{2+}
	dS m^{-1}	g L^{-1}		%				mg kg^{-1}		
VC	4.83	2.53	8.49	12.5	0.792	12.7	495.7	65.4	130.3	19,370

Note.—VC: vermicompost ($n = 3$).

2.3. Germination Indices

Germination was recorded daily after seeds were sown in Petri dishes. Seeds were considered germinated when hypocotyl reached a length of 2 mm (Figure 3).

$$GP = (\text{No. of germinated seeds} / \text{No. of seeds}) \times 100 \quad (1)$$

$$GRI = (G1/D1) + (G2/D2) + \dots (Gn/Dn) \quad (2)$$

where 'G' is the number of germinated seeds at day 'D'.

Figure 3. Germination images of *Salicornia bigelovii* seeds with FW: (a) 3 DAS, (b) 60 DAS. The germination percentage (GP) and the Germination rate index (GRI) at 20 days after sowing (DAS) were calculated according to Al-Mudaris [37] (Equations (1) and (2)).

The mean germination time was calculated according to Orchard [38] (Equation (3)).

$$MGT = \sum(N \times D) / \sum N \quad (3)$$

where 'N' is the number of seeds germinated at day 'D', and 'D' is the number of days.

The germination energy is a mathematical method used as an indicator of the potential that a seed has to germinate [39] and was estimated according to the Maguire's expression [40] (Equation (4)):

$$GE = (N1/D1) + (N2 - N1)/D2 + (Nj - N1)/Dj \quad (4)$$

where 'N' means the number of germinated seeds on the counting date, and 'D' the number of days.

2.4. Seedling Water Status Relations

In order to determine the internal water status, the water potential (Ψ_w) was measured on healthy and uniform seedlings at the 20th day after sowing (DAS) using a Dewpoint Potential Meter model WP4-T (Decagon Devices Inc., Pullman, WA, US). Four readings per treatment were recorded using enough seedlings to complete a sample to cover the bottom of the sample cup/lids. Every reading corresponds to one sample; units are expressed in MPa. For the osmotic potential (Ψ_s), a sample of 0.5 g of healthy *S. bigelovii* seedlings was collected from each treatment 20 DAS and transported to a deep freezer to maintain them at -20°C . Afterwards, the sage was extracted according to Mogahieb et al. [19], where samples were thawed and centrifuged at $1200 \times g$ for 25 min at 4°C . The osmotic potential of this stage was measured with a Wescor (Logan, UT, USA) Model 5500 Vapor Pressure Osmometer (VPO), using standards of known osmotic potentials for calibration. The disk filter papers used in the VPO equipment were saturated with the sage. A total of three

readings per treatment were made. Data were recorded in mmol/kg and transformed into MPa according to the van't Hoff equation [41] (Equation (5)).

$$\Psi_s = -C \times R \times T \quad (5)$$

where “C” is the molarity of the solution (mol of solute kg⁻¹ H₂O), “R” represents the universal gas constant (0.0831 kg MPa mol⁻¹ K⁻¹), and “T” the temperature (K).

The turgor potential was measured by subtracting Ψ_s from Ψ_w , according to Mogahieb et al. [19].

The relative water content (RWC) was calculated according to the equation proposed by Yamasaki and Diellenburg [42] (Equation (6)) from complete and healthy seedlings 60 DAS.

$$\text{RWC} = (\text{Fw} - \text{Dw}) / (\text{Tw} - \text{Dw}) \times 100 \quad (6)$$

where, Fw is the fresh weight, Dw is the dry weight, and Tw is the turgent weight.

Fresh and dry weights were recorded using an analytical balance (Mettler Toledo, model AG204). Turgent weight was obtained from complete plants which were imbibed in distilled water until they reached a constant weight (24 h). Dry weights were registered after samples were dried in a pre-heated oven (Shel-Lab, model Fx-5, serie-1000203, USA) at 55 °C until a constant weight was reached (48 h); four repetitions per treatment were measured.

2.5. Seedling Biomass

Because the analytical balance used in the present study was not able to record the dry biomass weight of seedlings at the 20th DAS, it was necessary to let the seedlings grow until the 60th DAS. Four seedlings per treatment were transported in individual paper bags to the laboratory for fresh (FB) and dry biomass (DB) recording using an analytical balance (Mettler Toledo, model AG204, USA). After fresh biomass recordings, seedlings were placed in a pre-heated oven (Shel-Lab, model Fx-5, serie-1000203, USA) at 55 °C in each paper bag until they reached constant weight (48 h); four seedlings were used per treatment. Results were expressed in mg.

2.6. Statistical Analyses

Data were transformed to arcsine to meet the normality and homoscedasticity tests. A one-way ANOVA was performed to compare treatment means; Tukey HSD test $p \leq 0.05$ was used for mean significance separation. The statistical analyses were carried out with the software Statistica v. 10.

3. Results

3.1. Effect of Vermicompost Leachate on the Physico-Chemical Properties of Treatments

The addition of vermicompost leachates increased the electric conductivity (E.C.), total dissolved solids (TDS), and salinity for distilled water (DWVL), and freshwater (FWVL) treatments and decreased for treatments with shrimp residual water (SRWVL) and seawater (SWVL). The vermicompost leachate addition increased all treatments' pH, except for SRWVL (Table 1). An increase in nitrates (NO₃⁻), phosphates (PO₄³⁻), sulfates (SO₄²⁻), and potassium (K⁺) was observed in treatments with the addition of VL, where the highest value was recorded in SRWVL. Meanwhile, Ca⁺ and Na⁺ increased only for FWVL and DWVL and decreased for SWVL and SRWVL.

3.2. Germination Percentage, Germination Rate, Mean Germination Time, and Germination Energy

The germination percentage was affected by salt concentration from the water sources with high salinity treatments, with SRW, SRWVL, SW and SWVL registering less than the 50% of germination. Meanwhile, for distillate and freshwater treatments (DW, DWVL, FW and FWVL), the germination percentage was above 70% (Figure 4A). A 12% germination

percentage increase was registered when VL was added in DW treatments, as shown by Tukey HSD analysis. A similar effect was observed for SRWVL, reflecting a germination percentage 13.3% higher than SRW. However, the germination percentage was not increased on FW and SW where VL was added (FWVL and SWVL). The highest germination percentage was registered for DWVL (70%) and lowest on SRW (33%). For the germination rate index, the maximum germination rate for DW, DWVL, FW and FWVL was registered in the first five DAS in comparison to the SW, SWVL, SRW and SRWVL, where it was reached on the seventh day (Figure 4B). The effect of the addition of VL was observed at the maximum germinated values reached along the days. For FW and FWVL, the maximum number of germinated seeds was registered on the third day (12 and 19, respectively). However, the addition of VL increased the number of germinated seeds by 37% compared to the treatment without it. DWVL registered the highest number of germinated seeds on the fifth day (14), compared to the DW whose maximum number of germinated seeds was 11 and was recorded on the sixth day. For SWVL, 10 fully germinated seeds were recorded on the seventh day; meanwhile, with SW, there were eight germinated seeds on the same day.

Figure 4. (A) Germination percentage of *Salicornia bigelovii* (GP), same letters above bars denote no-significant differences, and (B) germination rate (GR) of seeds under different water treatments; vertical lines at the top of bars (A) and on the dots (B) are the standard deviation ($n = 4$).

The germination energy (GE) showed the highest values for DWVL, FW and FWVL, being lower for DW, SWVL and SRWVL; the lowest values were recorded in SW and SRW. GE showed higher values when VL was added to the water sources (Figure 5A). The mean germination time (MGT) decreased in all treatments with VL. This index decreased in 2.7 days with DWVL, compared to DW and SRWVL (Figure 5B).

Figure 5. (A) Germination energy (GE), and (B) mean germination time (MGT) of *Salicornia bigelovii* germinated seeds under different water treatments. Same letters above bars denote no-significant differences; vertical lines at the top of bars are the standard deviation ($n = 4$).

The water potential (Ψ_w) of seedlings was higher with treatments of low salinity (DW and FW) even when VL was added (DWVL and FWVL). Treatments under high salinity showed the lowest values of Ψ_w (approximately -2 MPa), in comparison to the low salinity treatments (DW, DWVL, FW and FWVL); at the same time, SRW reflected the lowest Ψ_w . The addition of VL to SW and SRW improved the values of Ψ_w only in the second one (SRWVL) (Figure 6A). The osmotic potential (Ψ_s) showed similar results as Ψ_w (Figure 6A) for treatments with low salinity, even when VL was added (DW, FW, DWVL and FWVL) showing higher values than those with high salinity content (SW, SRW, SWVL and SRWVL). Ψ_s decreased for SWVL and increased for SRWVL (Figure 6B). For the turgor potential (Ψ_t), the effect of the addition of VL was observed in DWVL and SWVL treatments. For DW, the addition of VL (DWVL) decreased the Ψ_t in 0.63 units. On the contrary, the addition of VL to SW (SWVL) increased the Ψ_t values in one unit.

The application of VL generated a positive effect in the SWVL and SRWVL on the relative water content (RWC) according to the Tukey HSD test. The highest increase was registered in SWVL compared to SW with an increase of 20% (Figure 6C). Data showed that seedlings under low salinity (DW, DWVL, FW and FWVL) registered less than 60% of their RWC compared to the high salinity treatments (SW, SWVL, SRW and SRWVL) whose values of RWC were over 60%.

Figure 6. Water potential (Ψ_w) (A), osmotic potential (Ψ_s) (B) and relative water content (RWC) (C) of *Salicornia bigelovii* seedlings under different water treatments. Same letters above or below bars denote no-significant differences; vertical lines are the standard deviation ($n = 4$).

3.3. Seedling Biomass

Fresh and dry seedling biomass showed significant differences. Lower fresh biomass values were observed in all treatments under high-saline conditions (SW, SWVL, SRW, SRWVL) compared to the low-saline ones (DW, DWVL, FW, FWVL). The fresh biomass production notoriously increased adding VL to FW (FWVL) compared to the same treatment without it (FW) (Figure 7A). For this variable, the best source of water was FWVL, registering the highest values, followed by DWVL > SRWVL > SWVL.

Figure 7. (A) Fresh biomass (FB), and (B) dry biomass (DB) of *Salicornia bigelovii* seedlings under different water treatments. Same letters above bars denote no-significant differences, vertical lines bars at the top of bars are the standard deviation ($n = 4$).

The data showed that treatments with VL promoted increases in dry biomass production, compared to the same treatments without its addition (Figure 7B).

4. Discussion

4.1. Germination Indices

Salicornia bigelovii germination was found to be a process favored by solutions with low salinity concentration, according to the results of the germination percentage in the DW and FW treatments compared to the SW and SRW which showed lower values.

Similar results were reported for *S. bigelovii* [9], *S. europaea* [43] and *S. rubra* [16]. The low germination percentages registered in the SW and SRW treatments could occur due to the high concentration of solutes in both solutions, which increased the osmotic pressure in the medium, causing the development of water stress and inhibiting germination in both treatments, as mentioned by Guillén et al. [44]. This study quantifies the application of VL as an attenuating agent of salt stress on *S. bigelovii* seed germination.

The addition of VL increased the total germination of tomato seeds by 26.67% in response to NaCl treatments compared to the control group, but there was no meaningful measurement of natural saline conditions of shrimp culture effluents which include other salts, besides NaCl [23]. Results of the germination percentage for *S. bigelovii* seedlings using distilled water plus VL (DWVL), and shrimp residual water plus VL (SRWVL) improved in 12 and 13.3% of the total seed germination, respectively.

According to previous reports, VL contains auxins, cytokinins, gibberellins and brassinosteroids [22,45] which are phytohormones that play an important role in plant development and contribute to improving seed germination and emergence [46,47]. A stress-attenuating effect on the germination rate was detected when VL was added to the treatments and fewer days were needed to obtain a bigger number of germinated seeds, compared to treatments without VL. In this sense, an increase in germination rate of tomato and lettuce seeds under vermicompost tea treatments was reported, attributing its effects to the presence of hormones such as gibberellins, which induce the expression of the α -amylase enzyme, responsible for the hydrolysis of starch reserves that are stored in the endosperm [48]. The transformation of starch into soluble sugars gives the embryo the necessary energy and vigor for the germination process [49,50]. Besides, improvement of the addition of leachate was more evident on germination energy where the number of fast-germinating seeds was considerably higher for treatments with VL.

4.2. Seedling Water Status

The obtained results showed that *S. bigelovii* seedlings with high salinity treatments (SW, SWVL, SRW, and SRWVL) developed less water and osmotic potential than seedlings under low salinity (DW, DWVL, FW, and FWVL). Similar results were reported for *S. europaea* [19] and *S. persica* [20] seedlings that were treated with a NaCl-salinity stress gradient, suggesting an increase in proline and glycine betaine synthesis and a greater accumulation of Na^+ within the cellular vacuole as well as a decrease in K^+ and P as salinity levels increased. They also reported that the development of an osmotic adjustment mechanism where proline and glycine betaine act as compatible osmolytes could protect different sensitive enzymes against these ions in the cytoplasm, in addition to using them as compatible osmolytes to reduce their total water potential and maintain their RWC. Under this context, the obtained results about the water status, osmotic potential and RWC variables evidence the development of an osmotic adjustment mechanism by *S. bigelovii* seedlings under high salinity levels. The synthesis of compatible osmolytes or the accumulation of inorganic ions within the cell vacuole deserve further investigations.

4.3. Plant Biomass

Results evidenced the decrease in the fresh biomass of *S. bigelovii* seedlings in the higher salinity treatments (SW, SWVL, SRW and SRWVL), compared to the low salinity treatments (DW, DWVL, FW and FWVL). Similar results were reported for *S. bigelovii* [51], *S. europaea* [19], and *S. persica* [20]. Results in the present study showed that the SRWVL treatment, despite being one of the highest salinity treatments, showed similar fresh weight biomass production values compared to low salinity treatments such as DW, DWVL and FW. Regarding the results in dry biomass production, the SRWVL treatment showed the highest values in this variable, followed by FWVL. It is noticeable that the high salinity treatments SRW and SWVL registered similar dry biomass production values in relation to low salinity treatments such as DWVL and FW, and even higher values compared to the lowest salinity treatment (DW).

The application of N sources is necessary to improve the development of *S. bigelovii* [52], and in some cases, inorganic solutes contribute more than organic solutes to the osmotic adjustment, such as in *S. brachiata* (Roxb.) under natural saline conditions [53]. Such responses could explain the highest values registered in the fresh and dry weight biomass production of seedlings with the application of leachates, which contributed a greater amount of N as evidenced by the treatment analyzes (Table 1). Furthermore, VL holds a large amount of humic acids, which can promote plant growth [21], enhancing the absorption of nutrients [54]. Also, Canellas et al. [46] reported that humic acids isolated from earthworm compost improved maize plant development by favoring the ion transport across the root cell membranes, enhancing the H⁺-ATPase activity. This led to a higher electrochemical proton gradient in the soil solution favoring the plant nutrition. As previously discussed, the presence of different types of phytohormones in VL have been reported [22,23,55], which could come from the degradation of organic matter during the vermicomposting process due to the joint action between microorganisms and earthworms [45]. Among those, cytokinins and auxins are known to play important roles in several developmental processes such as chlorophyll production, shoot apical dominance and root development [56]. The points mentioned above could explain the highest fresh and dry biomass values registered when VL is used; however, further investigation is needed to elucidate these arguments.

It is clear that one of the most critical stages in the life cycle of halophytes is the period of germination and establishment [57], but in this sense, it is worth mentioning that the need to increase the productivity of halophytes may imply the indiscriminate use of fertilizers that can increase salinity. The results motivate detailed studies where composts or their leachates are combined with beneficial microorganisms or other biomaterials [58].

5. Conclusions

The obtained results contribute to reinforce the information on the susceptibility presented by *S. bigelovii* at high salinity levels during its germination and seedling development. However, salinity did not completely inhibit their germination. Water relations variables suggest the development of an osmotic adjustment strategy as a defense mechanism against saline stress by *S. bigelovii* seedlings.

The addition of VL showed an attenuation effect on the salt-stress, since the data analysis showed increases in fresh and dry biomass, as well as higher values in different germination indices such as the germination rate index, germination energy and mean germination time. However, its effect on increasing the germination percentage was clear with low salinity water and was not homogeneous with high salinity water sources.

The evidence suggests that adding VL to a water source improves germination and promotes increases in dry biomass production, but it is worth taking into account the saline quality of the water source to avoid depletion of the osmotic pressure in the soil solution and consequently in the plant tissues.

Finally, as vermicompost is a naturally processed manure which is becoming popular day by day, it is promising to consider in future studies that the combined use of vermicompost or vermicompost leachates with biochar may favor synergies that help minimize the impact of soil salinity on a range of crops.

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