

Use of halophyte plants on saline soils and evaluation of salt removal efficiency

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Abstract. The main purpose of the study was to evaluate of effectiveness of the halophyte plants on saline soils and use as a salt-tolerant plants. The experimental studies carried out on the halophyte plants (*Tetragonia tetragonioides* and *Portulaca oleracea*) and the salt-sensitive crop (*Lactuca sativa*, L) in the greenhouse. Two experimental studies were conducted on the effects of salt on germination, total yield and mineral composition of the halophyte plants and salt-sensitive crop. Three salinity treatments (saline water solution with NaCl: T1 - 5 dS m⁻¹; T2 - 9.8 dS m⁻¹; and T3 - 20 dS m⁻¹) and a control treatment (T0 - 0.6 dS m⁻¹) were used for the experimental study. The both halophyte plants showed the potential salt (ions) removing species with high yields when grown on saline soils. The results showed that the above halophytes varieties contributed to improved quality and yield of the salt-sensitive crop. Thus, this method has proven to be one of the important solutions in preventing and managing salinity and ensuring the sustainability of agricultural systems. Analysis of the obtained results showed that halophyte plants could be used to improve the management and stability of saline soils and grow them as food crops.

1 Introduction

Salinity seriously threatens the provision of high-quality agricultural products [1]. Salinity stress is a key factor that limits crop production world-wide and is a constraint to economic development and to the environment; the economic impacts resulting from salinity are mainly associated with a decrease in the production capacity of land [2]. Soil salinization is one of the major soil degradation threats occurring in large areas of the world. It can be observed in numerous vital ecological and non-ecological soil functions [3]. To improve the salinity tolerance of crops, various traits can be incorporated, including ion exclusion, osmotic tolerance and tissue tolerance [4]. Saline tolerance in plants may be explained by the involvement of functional and structural adaptations, such as growth regulation, osmotic adjustment, and changes in water potential [5]. *T. tetragonioides* exhibited good potential for

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use as a species to remove salt [6]. Growing salt-tolerant crops can give good results on saline soils [7-8].

Greenhouse cultivation is noted for high uptake of minerals, consistent climatic conditions, exclusion of natural precipitation and control of salt accumulation [9]. In the greenhouse industry, methods have been developed for the determination of the nutrient availability and salinity status of the soil and substrate [10]. Purslane (*Portulaca oleracea* L.) is a salt-tolerant annual plant that contains high amounts of beneficial antioxidant vitamins and minerals. The effect of salt stress on the growth and mineral composition of purslane (*P. oleracea* L.) was studied [11]. *P. oleracea* is resistant to salinity, is able to remove ions (400–500 kg ha⁻¹ NaCl) and can be grown in high-salinity soil [12].

The salinity condition in the root zone hinders moisture extraction from soil by plants, because of osmotic potential development in soil water, due to the presence of salts, which ultimately decreases transpiration of plants, and thereby affects crop yield [13–15].

Soil salinity reduces the productivity of many agricultural crops, including most vegetable crops, which present low tolerance to soil salinity. However, a substantial increase in production and consumption of vegetable crops that include edible portions of herbaceous species (roots, tubers, shoots, stems, leaves, fruits, and flowers) is a global priority. In fact, vegetables play an important role in human nutrition and health, particularly as sources of vitamin C, thiamine, niacin, pyridoxine, folic acid, minerals, and dietary fiber. Some of the world's most widespread and debilitating nutritional disorders, such as micronutrient deficiencies are related to low vegetable intake [16]. Generally, vegetables are crops with high productivity per unit of water applied and economic value compared with field crops. This may be a very important advantage for small farmers, because vegetables can grow in small areas, under intensive procedures [17]

Therefore, the main goal of this study was to evaluate the effectiveness of the halophyte species in saline soils - analyzing the effects of salinity on plant growth and biomass, as well as salt removing potential.

2 Materials and methods

2.1 Experimental procedures

The experimental studies carried out in an investigation greenhouse at the University of Algarve, Faro, Portugal.

The first experimental study conducted in May and June, with randomized potted plants. Irrigation water amounts were minimal, but still enough for the plant survival (0.2 L d⁻¹ pot⁻¹). The species of the studied plants were *Tetragonia tetragonioides* and *Portulaca oleracea*, submitted to 3 salinity treatments (T) salinity levels of irrigation water 1 dS m⁻¹ (T0), 10 dS m⁻¹ (T1) and 20 dS m⁻¹ (T2).

The second experimental study carried out from October and November. For the study in this experiment used moderately sensitivity crop (*Lactuca sativa*, L). Lettuce plants sowed and grown, just after *T. tetragonioides* and *P. oleracea* species. The experiment accomplished in randomized pots, distributed randomly in four replications, according to the following six treatments: T0 (T) – previous species *T. tetragonioides* - lower salinity regime T1 (T) – previous species *T. tetragonioides* - middle salinity regime T2 (T) – previous species *T. tetragonioides* – higher salinity regime T0 (P) – previous species *P. oleracea* – lower salinity regime T1 (P) - previous species *P. oleracea* – middle salinity regime T2 (P) – previous species *P. oleracea* – higher salinity regime plants irrigated (0.25 L d⁻¹ pot⁻¹) each three days with tap water until October 20. Nitrogen fertigation was done during the last twenty days of the experiment with NO₃⁻ concentration of 2 mM NO₃⁻ and NH₄⁺ concentration of 2 mM.

2.2 Soil analyses

Table 1 shows physical and chemical parameters of the soil was the data which taken before the experiment. The soil characteristics in Algarve (Portugal) were similar to some observed saline areas in Mirzachuli steppe Uzbekistan. The chemical reaction is slightly alkaline – pH 8.5 and the soil salinity, at the saturation point is moderate – 0.3 dSm⁻¹. The soil texture is heavy – silt loam and silt clay loam. The soil structure is moderate.

Table 1. Physical and chemical parameters of the soil.

Soil parameters				
Physical parameters	Chemical parameters	Macro- and micro elements		
Field capacity (fc) (%) – 23.8	pH (H ₂ O) – 8.5	% g sample ⁻¹		
Wilting point (wp) (%) – 11.9	ECs (dS m ⁻¹) – 0.3	Ca – 27.1	Mg – 6.56	Na – 1.15
Bulk density (g cm ⁻³) – 1.40	Total calcareous (%) – 41	K – 1.17	N – 0.11	–
		ppm g sample ⁻¹		
		Fe – 118	Mn – 40	Zn – 4.5
		P ₂ O ₅ – 31	K ₂ O – 189	–

2.3 Chemical analyses

Dried leaves and stems finally grounded and analyzed by using the dry-ash method. The levels of Na and K determined by flame photometer and the remaining cations (Na, K, Ca, Mg and Fe) assessed by atomic absorption spectrometry. Chloride ions were determined in the aqueous extract by titration with silver nitrate according to [18]. Plant nitrogen determined by the Kjeldhal method. Phosphorus determined by colorimetric method according to the vanadate – molybdate method. All mineral analyses performed in the leaves.

2.4 Statistical analyses

Statistical analysis including Analysis of variance (ANOVA), Duncan’s multiple range test was performed to study the significance of different salinity gradient on different parameters studied. Values were calculated at the $p \leq 0.05$ probability level (all tests performed with SPSS Version for Windows program). Results and discussions

3 Results and discussion

3.1 Effect of salts (the first experiment)

The treatments started with nitrogen concentration ten days after the transplantation of the plants to randomised pots. The analysis of germination plant begun two weeks after the transplantation of the plants to randomised pots. Stem length, number of nodes and number of big leaves were analysed each seven days of vegetation period of species and at the same duration, the electrical conductivity (EC_w) and pH of drainage water obtained during the period of the experiment. Soil electrical conductivity (EC_s) and pH were analysed after the experiment. Each plant on the treatments were harvested and washed gently with tap water for a few minutes, wiped with paper and the Fresh Weight (FW) was measured. The fresh samples were dried in forced drought oven at 70° C for 48 hours and the Dry Weight (DW) was measured. Plants were analysed relatively to total growth and mineral compositions (Na, Cl, N, K, P, Ca, Mg and Fe) of the leaves.

3.1.1 Effect of salt treatments to germination

The salinity treatments had a significant effect on the stem length (Fig. 1) of *Tetragonia tetragonioides* and *Portulaca oleracea*. The stem length of both species showed low variations between T1 and T2 treatments. This is confirmed by the results obtained by [6, 12, 19, 20]. On the other hand, there was a great increase of the stem length on the T0 treatment. The stem length of the all treatments where it was grown *P. oleracea*, were different among them ($p < 0.004$) from the stem length of *T. Tetragonioides*.

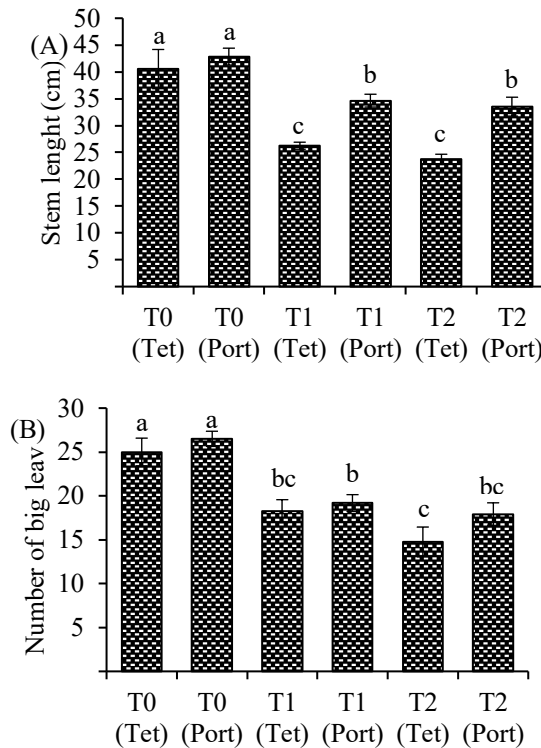


Fig. 1. Effect of salt treatments (NaCl) on the each salt concentration of stem length (A) and number of big leaves (B) of *Tetragonia tetragonioides* and *Portulaca oleracea*. Means \pm S.E., $n = 4$. Bars with different letters are significantly different at $P < 0.05$.

3.1.2 Electrical conductivity of soil (EC_s)

The electrical conductivity of soil (EC_s), saturated with distilled water ($dS\ m^{-1}$) for the different species and treatments (T1, T2 and T3) and species shown in the Table 2. For each treatment, it may be seen that the soil EC_s increased with increasing the salinity of the irrigation water. It may be also seen that the soil electrical conductivity was larger on the case of *T. tetragonioides*, which shows that these species have higher potential capacity to remove salts from the soil than *P. oleracea* species.

Table 2. The electrical conductivity (EC_s) of soil was obtained at the end of the experiment.

Treatments		Electrical conductivity (EC _s , dS m ⁻¹)	
		<i>Tetragonia tetragonioides</i>	<i>Portulaca oleracea</i>
	T0	0.130	1.939
	T1	0.612	2.204
	T2	1.098	2.632

Source: the electrical conductivity (EC_s) of soil was 0.3 dS m⁻¹ before the beginning of experiment.

3.1.3 Fresh and dry weight of plants

The results related to *Tetragonia tetragonioides* and *Portulaca oleracea* have revealed that the fresh weights consistently decline from 526 g plant⁻¹ to 226 g plant⁻¹ (*T. tetragonioides*) and 76.9 g plant⁻¹ to 31.5 g plant⁻¹ (*P. oleracea*) as they were affected by enhanced salinity. Fresh weight of *T. tetragonioides* of all treatments were different ($p < 0.005$) than the fresh weight of *P. oleracea* (Fig. 2A). The biomass production of stem (g DW plant⁻¹) where it was grown *T. tetragonioides*, were significant different between treatments ($p < 0.334$) than the biomass production of stem (g DW plant⁻¹) *P. oleracea* (Fig. 2B). Dry weight of leaves (g DW plant⁻¹) where it was grown *T. tetragonioides*, were significant different between treatments ($p < 0.196$) than dry weight of leaf (g DW plant⁻¹) *P. oleracea* (Fig. 2C). Dry weight of seed (g DW plant⁻¹) where it was grown *T. tetragonioides*, were significant different between treatments ($p < 0.208$) than dry weight of seeds (g DW plant⁻¹) *P. oleracea* (Fig. 2D).

In the present study two agronomic species, *T. tetragonioides* and *P. oleracea* were exposed to salt stress by increasing the NaCl concentration (0, 100 & 200 mM NaCl) of irrigation water. When the salinity increased, the plants dry weight of both species markedly decreased; however, the dry weight of *T. tetragonioides* decreased under salt conditions. The dry weight of *T. tetragonioides* decreased 1.2 and 1.4 and dry weight of *P. oleracea* decreased 1.2 times on 100 and 200 mM NaCl treatment, respectively compared with the control. Generally, the growth of species decreases with increasing salinity, while that of halophytes improves. In the study, the growth of *T. tetragonioides* increased under salt stress, according to the previous data reported on the halophytes *Salicornia europaea* and *Suaeda maritima* [21] and *Alhagi pseudoalhagi* [22], in which salt treatment at low levels improved plant growth. These results indicated that *T. tetragonioides* is a halophyte and so the salt tolerance of this plant was higher than that of *P. oleracea*.

Despite the low dry matter production at the end of the experiment, *P. oleracea* grew without drought, flooding or salts injury symptoms and the plant yield was not greatly affected by salt conditions; moreover, plant tissues accumulated large amounts of sodium and chloride in salt conditions. On the other hand, *T. tetragonioides* produced higher dry matter than *P. oleracea* and showed, therefore, great capacity for ions accumulation.

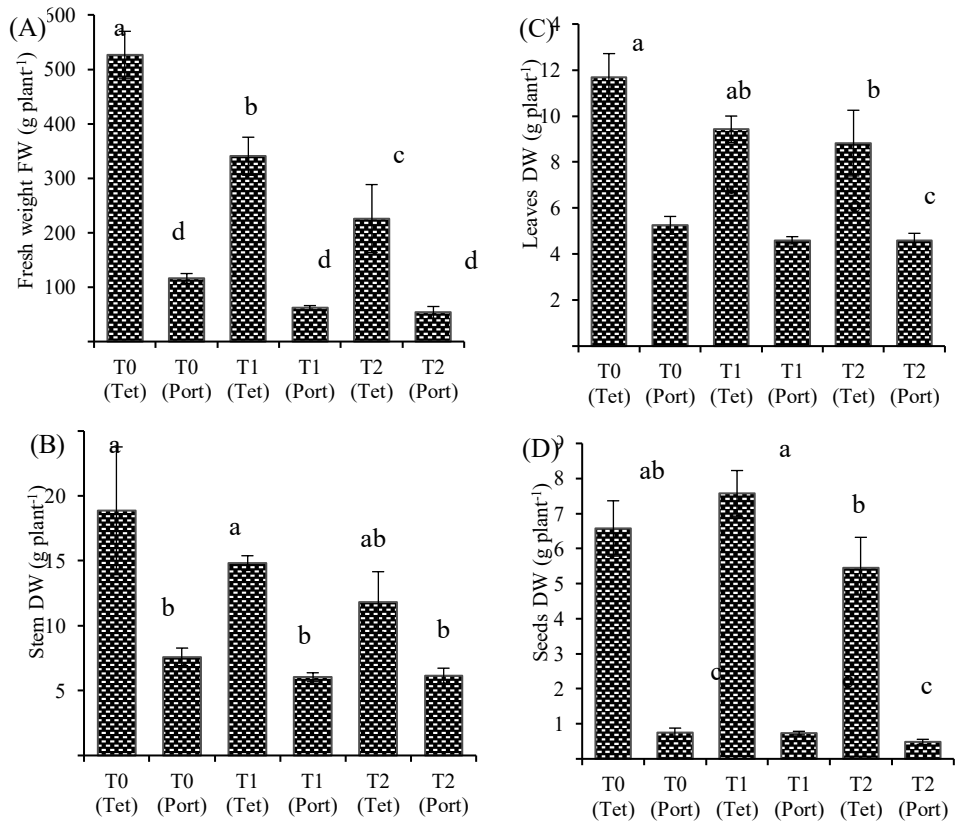


Fig. 2. Effect of salt treatments (NaCl) on the each salt concentration of biomass production of fresh weight (g FW plant⁻¹, A) and dry weight of stem, leaves and seeds (g DW plant⁻¹ B, C, D) of *Tetragonia tetragonioides* and *Portulaca oleracea* (whole plants).

It was shown for both species that the partition among plant organs was affected by the medium salt concentration for both species, as follows: there was an increase of the percentage of dry matter of the leaves in saline conditions and a decrease in seeds; the percentage of dry matter of stems was constant in all treatments. In the other similar present study of many researchers [19, 23], the fresh and dry weight of *T. tetragonioides* was significant increased on 50 mM NaCl treatment end was un-changed on 100 and 200 mM NaCl treatment relative to the control. This is confirmed by the results obtained by [24] who found out an increased yield of salt absorbing species (*T. tetragonioides*). The effect of different concentrations of NaCl on growth, increased salt stress and decreased dry matter biomass length of shoot. This is confirmed by the results obtained by [25].

3.1.4 Mineral compositions of leaves

The salinity had a significant effect on the leaf mineral composition of *Tetragonia tetragonioides* for the majority of analyse mineral elements, as follows (Figure 3): 1) there was a great increase of sodium concentrations ($p < 0.0001$) when water salinity increased (Fig. 3A). 2) Chloride concentrations of the leaves were different between treatments ($p < 0.0001$) Fig. 3B); 3) the total nitrogen leaf content of species showed low variations among

treatments ($p < 0.197$), apparently not related with the salinity.; 4) there was a general decrease of potassium, calcium, and magnesium leaf content of crop; 4) the salinity of irrigation water affected phosphorus and iron leaf content. In the similar previous studies [23, 24], the mineral contents of *T. tetragonioides* which are Mg, P, N decreased, and Na, K, Ca [25], Fe and Cl increased when salts treatments were increased.

The salinity of irrigation water had a significant effect on the mineral composition of *Portulaca oleracea* leaf content for the majority of the analysed mineral elements, as follows: 1) the total nitrogen leaf content showed low variations among treatments; 2) there was a great increase of sodium and chloride concentrations, with the increase of salinity concentration; 3) a low increase of potassium leaf content was shown, with the increase of the saline water; 4) a very low decrease of calcium and magnesium leaf content was verified when the salt concentration of irrigation water was greater; 5) the salinity did not affect iron content of leaf. Similarly in the previously study [23], the results mineral content of *P. oleracea* which are Mg, P, N decreased, and Na, K, Ca, Fe and Cl increased when salts treatments were increased.

On the other hand, the Na concentration of plants (*T. tetragonioides*) increased from 3.5% to 6.3% (% g^{-1} sample of leaf) and (*P. oleracea*) increased from 1.41% to 2.87% (% g^{-1} sample of leaf) in accordance with increased salinity levels. This is confirmed by the results obtained by [23].

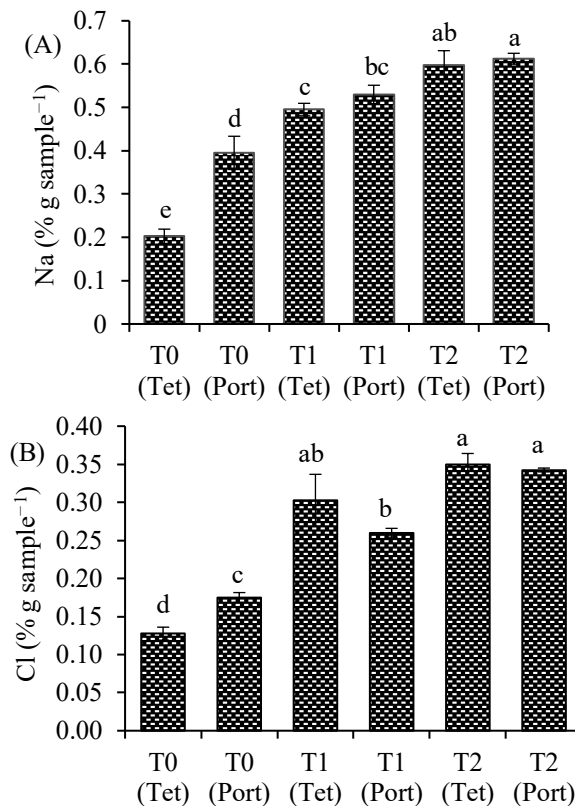


Fig. 3. Effect of salt treatments to the mineral compositions of leaves on the each salt concentration (A, B) of *Tetragonia tetragonioides* and *Portulaca oleracea*.

3.2 Effect of saline soil (the second experiment study)

To conduct of the experiment study was selected leaves vegetable crop which is lettuce (*Lactuca sativa*, L). The experiment study was conducted at the University of Algarve, Campus de Gambelas in the greenhouse during the period of September 25 and November 10 with randomly potted plants. Plants were sowed in the soil pots in which were grown just before *Tetragonia tetragonioides* and *Portulaca oleracea* species. Plants were irrigated each three days with tap water. The total numbers of plants were 24, distributed with accordance of above mentioned salt treatments (3), each treatment was done to four pots with plant sowed in them. Lettuce was irrigated with N nitrogen treatments last twenty days (October 20 and November 10) with concentrations of 2 mM NO₃⁻ and 2 mM NH₄. Electrical conductivity (EC), pH and height leaf of the crop were measured periodically and the mineral composition was determined at the end of the experiment. Plants were harvested in twenty days (November 10) after treatments.

3.2.1 pH of the drainage water

pH of the drainage water of the lettuce's pots of lettuce (grown in the pots of *T. tetragonioides* and *P. oleracea*) is shown in (Fig. 4 A). pH of water showed great increase in treatment T2 (T) and decrease in treatments T0 (T) and T1 (T) of the experimental period. Drainage water of the lettuce's pots shows that the total pH of water has a great increase in the treatment T2 (P) and decrease in the treatment T0 (P) and a great decrease in the treatment T1 (P).

The electrical conductivity (EC_w) of the drainage water (from the pots of *T. tetragonioides* and *P. oleracea*) is shown in (Fig. 4 B). EC_w of the drainage water increased in the treatment T2 (T), a decrease in the treatment T1 (T) and low variations in the treatment T0 (T). EC_w of the drainage water increased in the treatment T2 (P), decreased in the treatments T1 (P) and T0 (P).

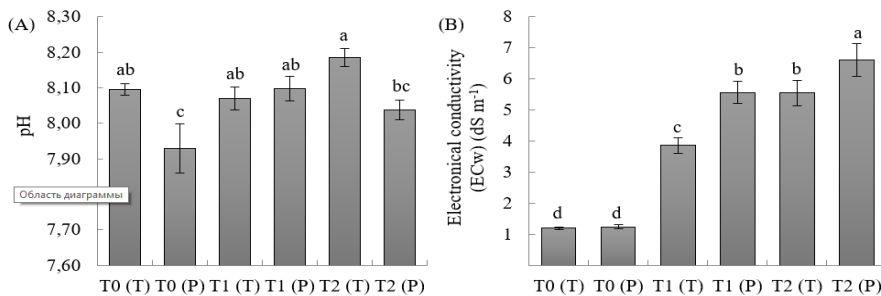


Fig. 4. pH (Figure 4A) and electrical conductivity (EC_w) (Figure 4B) of the drainage water - lettuce (were grown in the pots of *Tetragonia tetragonioides* and *Portulaca oleracea*) to compare of Duncan test. Means ± S.E., n = 4. Bars with different letters are significantly different at P < 0.05.

3.2.2 Fresh and dry weight of plant

Fresh weight of lettuce (were grown in the pots of *T. tetragonioides* and *P. oleracea*) showed no significant differences in each treatment. Fresh weight of lettuce (grown in the pots of *T. tetragonioides*) was lower than the fresh weight of lettuce (grown in the pots of *P. oleracea*) in the treatment T0 (control), but there was shown that the fresh weight is higher on the saline soil in the treatments T1 and T2. Indicated that the soil in which *T. tetragonioides* was grown had less salinity than the soil in which *P. oleracea* was grown (Fig. 5A). Dry weight of lettuce (grown in the pots of *T. tetragonioides*) showed no variations

among treatments, (Fig. 5B). There was a great increase dry matter in treatment T0. Dry weight of lettuce (grown in the pots of *P. oleracea*) showed low variations among treatments. There was a great increase of dry matter in the treatment T0.

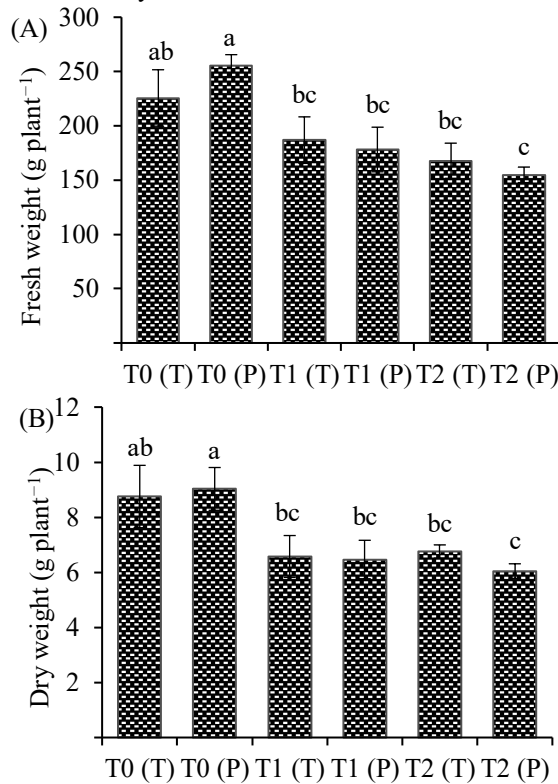


Fig. 5. Fresh (A) and dry (B) weight of lettuce (grown in the pots of *Tetragonia tetragonioides* and *Portulaca oleracea*) on the different treatments of nitrogen concentrations.

In support of the previous observations, [27] demonstrated that foliar application of fertilizer significantly enhanced the growth and yield of citronella plants. High rates of fertilizer can lead high salinity that is high enough to damage plants and reduce growth and yield [28] showed that an increase in salinity of nutrient solution of lettuce plants was associated with a reduction of marketable growth and yield, average plant fresh weight and leaf number per plant. Recently, studying the effects of urea fertilization on cluster bean plants subjected to water stress, found that water stress significantly decreased shoot water potential, fresh and dry mass and maintained a reduction of water content according to the similar previous studies [29-33]. Growth retardation and the loss of fresh and dry weights from stems and leaves of plants under salinity stress have been observed in previous studies [34-35]. In addition, based on fresh and dry weights, it has been demonstrated in several studies that the shoot ratios of many plants increase under salinity stress [6, 36-37].

3.2.3 Mineral compositions of leaves

The significant effect was mineral composition of lettuce (grown in the pots of *T. tetragonioides* and *P. oleracea*) leaf content for the majority of mineral elements analysed (Figs. 6 A and B). The content of lettuce leaf (grown in the pots of *T. tetragonioides* and *P.*

oleracea) showed low variations among treatments. Sodium (Na) in the content of the lettuce leaves (grown in the pots of *P. oleracea*) was different comparing to treatments ($p = 0.0001$) (Fig. 6A). Chloride (Cl) of the lettuce leaves (grown in the pots of *T. tetragonioides*) was different comparing to treatments ($p = 0.0001$) (Fig. 6B).

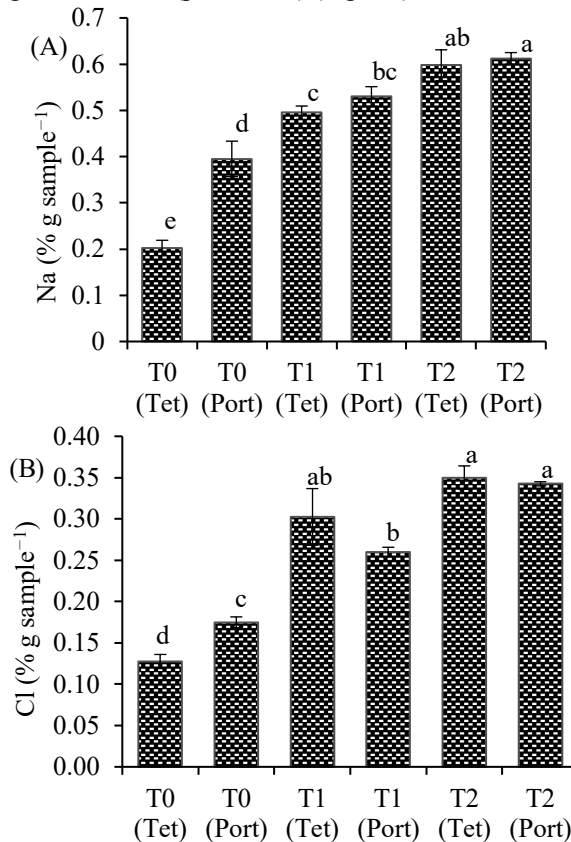


Fig. 6. Mineral compositions of lettuce leaves (T (Tet) that grown in the pots of *Tetragonia tetragonioides* and T (Port) *Portulaca oleracea*) on the each salt concentration (A, B).

4 Conclusion

Tetragonia tetragonioides showed itself as the most potential salt (ions) removing species. Moreover, at the end of the experiment, *T. tetragonioides* was the sole species as a halophytic that had produced significant amounts of dry matter. The causes were: fast rate growth, higher biomass production potential, this species can produce several yields during the year (summer and winter), easy multiplication (seed propagation) and easy crop management, species is tolerant to drought and hot conditions, protection from soil erosion due to excellent soil covering.

The potential of *Portulaca oleracea* as salt removal plant also high, but lower than *T. tetragonioides*. This explained by the larger biomass production of *T. tetragonioides*. On the other hand, for very arid climates, *P. oleracea* may substitute successfully *T. tetragonioides*, once that *P. oleracea* is much more tolerant to drought and salt conditions.

Moreover, these both species can be planted, as ornamentals, in saline soils. These techniques to control salinity showed that agricultural production could be maintained through the reduction of salts application due to the decrease of irrigation amounts, reducing the leaching. In addition, the applied salts through the irrigation can be eliminated by using the salt (ions) removing species. In addition, for very arid climates, *P. oleracea* may substitute successfully *T. tetragonioides*, once that *P. oleracea* is much more tolerant to drought and salt conditions.

The obtained results shows that lettuce (*Lactuca sativa*, L) is not tolerant to drought and hot conditions, it needs large irrigation water amounts. This irrigation water contains a certain quantity of soluble salts, which will be accumulated in the soil and cause reduction of crop yield and its quality. It was shown a good contribution of the above salt removal crops, mainly *T. tetragonioides* halophytic species to the quality and yield of the lettuce, a moderate salt - sensitive crop. Therefore, it has demonstrated that this technique is a clean and environmentally safe tool to avoid salinisation and maintain the sustainability of agricultural systems.

As final remarks, it is concluded that cultivation of these plants on saline soil and in arid climates can be considered as clean and environmentally safe techniques, which combines environmental, economical, and social aspects of problem solving. Hence, these two salt removing species may contribute to increase of the soil sustainability of irrigated areas under climatic changes, and may be used as food crops.

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