Effects of salt and drought stress on seed germination and seedling growth in *Portulaca*

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Abstract

The effects of salt and water deficit on seed germination and early seedling growth were studied in six Portulaca accessions (P. oleracea L. ssp. oleracea, P. grandiflora Hook., P. grandiflora cvs. 'Sundial Mango', 'Double Pink' and 'Double White', and P. halimoides L.). The aims of the study were to evaluate their responses to abiotic stress and to establish the relative degree of tolerance of the selected taxa. Seeds were germinated in the presence of increasing concentrations of NaCl or polyethylene glycol 6000 (to mimic drought conditions), generating the same osmotic potentials: 0, -0.25, -0.5 and -0.1 MPa. Germination percentage, mean germination time, seedling fresh weight and vigour index, and radicle, hypocotyl and cotyledon lengths were determined. Seed germination and seedling development decreased in all taxa in the presence of PEG and NaCl, in a concentration-dependent manner, although the effect of salt was stronger than that of PEG, at the same osmotic potentials. Germination and seedling parameters varied for the different accessions but, in practical terms, considering only the final germination percentages under stress, P. halimoides L. appears to be the taxon most tolerant to water deficit, whereas P. grandiflora Hook. would be the most tolerant to salt stress..

1. Introduction

Drought is probably the greatest environmental constraint affecting agricultural production worldwide (BOYER [1]). Crop yields in arid and semiarid regions are largely dependent on irrigation, which is, in turn, causing the progressive salinisation of arable land by accumulation in the soil of toxic ions dissolved in irrigation water. Salinity is also regarded today as an important problem for agriculture: it has been projected that salinity will seriously affect 30% of the world's cultivated land area within the next 25 years, and more than 50% by the end of this century (WANG & al., [2]). The effects of climate change will no doubt worsen the present situation in the coming decades. Salt stress can affect seed germination, vegetative growth, and reproductive development of plants, and therefore reduce crop productivity (MUNNS & al., [3]). It is now well established that salinity inhibits plant growth by altering different cellular and whole plant processes, at the physiological, biochemical and

molecular levels, and can even affect, in some cases, anatomical and morphological characteristics of the plants (TESTER & DAVENPORT [4]). Crops grown in salt-affected soils may suffer from osmotic imbalance, ion toxicity, and mineral deficiency, leading to reduced growth and productivity (MUNNS, [5]; NETONDO & al., [6]). Soil salinity, apart from ion toxicity, includes an osmotic stress component, causing water deficit ('physiological drought') and cellular dehydration (INCHAROENSAKDI & al., [7]; ROBINSON & al., [8]).

The most promising strategy to increase crop yields and food production in this scenario would be the genetic improvement of crop stress tolerance by both, conventional breeding and genetic engineering techniques (FITA & al., [9]). All our major crops are sensitive to relatively low degrees of salt stress and water deficit, generally much more sensitive than their wild predecessors – domestication and centuries of breeding, by selecting for traits such as rapid growth, biomass accumulation, fruit or grain production caused, as a trade-off, the reduction of resources invested in defence mechanisms (SERRANO & GAXIOLA [10]). Yet some minor crops, which have not been subjected to such intensive breeding, could show higher tolerance. Therefore, they could be grown in marginal or salinised soils, using low-quality, slightly brackish water for irrigation, thus contributing (modestly) to the goal of increasing agricultural production. One example of such crops is purslane (*Portulaca* sp.), a vegetable crop that has aroused great interest in recent years for its nutritional value, since it is an excellent source of omega-3 fatty acids, essential amino acids, vitamins C and E and other antioxidant compounds (FRANCO & al., [11]; UDDIN & al., [12]). Purslane has also been traditionally cultivated as a medicinal plant (RAHDARI & al., [13]) and as an ornamental, appreciated for the beauty of its flowers that come in a variety of bright colours. At least some cultivars of common purslane (*P. oleracea*) have been shown to be resistant to relatively high salinities (ALAM & al., [14]), and to drought (RAHDARI & al., [13]; D'ANDREA & al., [15]); in fact, there are some studies on the physiological mechanisms of response of *P. oleracea* – a C_4 plant – to water stress, which include the transient induction of CAM-like metabolism (D'ANDREA & al., [15]). However, despite this general interest, no systematic study comparing salt and drought tolerance of different purslane species and cultivars has been carried out until now.

The specific aim of this work was to analyse the responses to salt and drought stress of several purslane accessions, to select those with higher tolerance, which could eventually be used for commercial cultivation, as a food, medicinal and ornamental crop, especially for 'saline agriculture' and sustainable development. The analysis has been performed during seed germination and early seedling growth, generally a bottleneck in plant development as the phase of the life cycle most sensitive to stress.

2. Materials and Methods

Seeds of six different purslane taxa (*P. oleracea* L. ssp. *oleracea*, *P. grandiflora* Hook., *P. grandiflora* cv. "Sundial Mango", *P. grandiflora* cv. "Double Pink", *P. grandiflora* cv. "Double White", and *P. halimoides* L.) were used in the experiments. Seeds of all accessions were similar in size, with mean seed weights of 2.3 to 3.0 mg (determined by weighing 25 seeds of each). Three replicates of 25 seeds of each accession were germinated on cotton beds covered with two layers of Whatman No. 1 filter paper, in 9-cm-diameter Petri dishes. To test the effect of osmotic stress, the filter paper was saturated with 30 mL of PEG 6000 solutions of increasing concentrations: 0 (control), 29.25, 48.06, and 75.99 g/L, corresponding to osmotic potentials of 0, -0.25, -0.5 and -0.1 MPa, respectively. Similarly, the effect of salt stress was checked adding to the filters 30 mL of NaCl solutions of the same osmotic potentials (0, 57.2, 114.6, and 227.8 mM NaCl, respectively). The solutions were prepared

according to MICHEL's [16] equations. The dishes were sealed with Parafilm, placed in a germination chamber and incubated for 14 days with alternating day/night temperatures of 30°C/18°C. Germinated seeds were counted every day; a seed was considered as germinated in the moment of radicle protrusion. At the end of the experiments (day 14) final germination percentages were calculated; in addition, all germinated and non-germinated seeds were scanned and the ImageJ Programme for image processing was used to measure radicle, hypocotyl and cotyledon lengths. To assess germination velocity, mean germination time (MGT) was calculated as MGT= Σ D n/Σn, where D is the days from the beginning of the germination test, and n is the number of seeds newly germinated on day D (ELLIS & ROBERTS [17]). Seedling Vigour Index (SVI) was also calculated by the formula: SVI = Germination percentage x [Mean root length (mm) + Mean hypocotyl length (mm)], as described by ABDUL-BAKI & ANDERSON [18].

Statistical analyses were done using IBM SPSS Statistics 19. The significance of the differences between treatments was tested by applying a one-way ANOVA, at a confidence level of 95%. When the ANOVA null hypothesis was rejected, Tuckey's Post hoc test was performed to determine the statistically significant differences at $P<0.05$.

3. Results and discussion
3.1 Effects of osmotic stress on seed germination and seedling development
Seeds of the selected *Portulaca* wild species (*P. oleracea* L. ssp. *oleracea*, *P. grandiflora* Hook. and *P. halimoides* L.) showed very high germination percentages (93-96%) under control conditions (germination in water), which did not change much in the presence of PEG at osmotic potentials of -0.25 MPa or -0.5 MPa; only under the highest level of osmotic stress tested (-1.0 Mpa), slight (but statistically significant) decreases in germination percentages were observed, ranging from less than 3% in *P. halimoides* L., to 10% in *P. grandiflora* Hook and 14% in *P. oleracea* L. ssp. *oleracea* (Table 1). Control values were much lower for the commercial cultivars of *P. grandiflora*, 'Double Pink' and 'Double White' (ca. 43%) or 'Sundial Mango' (71%), but no significant change in germination percentages was observed in the presence of PEG at the highest concentration tested; in fact, a slight stimulation of germination could be measured at low or moderate levels of osmotic stress, -0.25 MPa or -0.5 MPa (Table 1).

Germination velocities in the controls roughly corresponded to final germination percentages, with the highest values (lowest MGT) calculated for the wild species (1.4-1.5 days), and higher MGT, between 2.3 and 5.3 days, for the *P. grandiflora* commercial cultivars. Increasing the osmotic pressure of the medium led, in general, to a significant increase in MGT (that is, reduced germination velocity), except for *P. halimoides* L. and *P. grandiflora* cv. 'Sundial Mango', which showed an MGT reduction (increased germination velocity) of ca. 17% and 18%, respectively, at -1.0 MPa osmotic potential, as compared to the control (Table 1).

In most *Portulaca* accessions, total seedling weight decreased with increasing PEG concentrations, with quantitative differences between taxa: weight reductions in relation to the corresponding controls ranged from 13% in *P. halimoides* L. to about 57% in wild-type *P. grandiflora* in the presence of the highest PEG concentration tested (Table 1). However, under the same conditions two of the *P. grandiflora* commercial cultivars, namely 'Sundial Mango' and 'Double White', did not show any significant weight reduction; in fact, for both cultivars some increase in fresh weight was measured at low or moderate osmotic stress levels (-0.25 or -0.5 MPa) (Table 1). A similar variation pattern was observed regarding seedling vigour index (SVI): a general reduction with increasing osmotic stress, from ca. 13%

in *P. halimoides* L. to 37% in *P. grandiflora* Hook, and no significant change in *P. grandiflora* cvs. 'Double Pink' and 'Double White', always in relation to the seeds germinated in water (Table 1).

Table 1. Effects of increasing osmotic potentials, generated by PEG in the germination medium, on the indicated seed germination parameters of the six selected Portulaca taxa. Values shown are means ± SE (n = 3). For each parameter and accession, different letters in a row indicate significant differences between treatments according to Tuckey's HSD test (P<0.05).

Parameter	Genotype	Osmotic stress (PEG)			
		-0 MPa (Control)	-0.25 MPa	-0.5 MPa	-1 MPa
Germination percentage (%)	*P. oleracea* L. ssp. *oleracea*	94.7 ± 1.3 d	87.0 ± 7.4 b	90.7 ± 4.8 c	81.3 ± 8.1 a
	P. grandiflora Hook.	93.3 ± 1.3 c	91.0 ± 5.0 b	93.3 ± 4.8 c	84.0 ± 4.0 a
	P. grandiflora "Sundial Mango"	71.0 ± 8.1 a	73.3 ± 4.0 b	78.7 ± 7.0 d	74.7 ± 1.3 c
	P. grandiflora "Double Pink"	43.0 ± 12.0 a	48.0 ± 4.0 b	52.0 ± 8.3 c	48.0 ± 2.3 b
	P. grandiflora "Double White"	42.7 ± 3.5 a	50.0 ± 6.0 c	45.3 ± 7.4 b	42.7 ± 8.7 a
	P. halimoides L.	96.0 ± 2.3 b	99.0 ± 1.3 d	97.3 ± 2.7 c	93.3 ± 3.5 a
Mean germination time – MGT (days)	*P. oleracea* L. ssp. *oleracea*	1.5 ± 0.1 a	1.8 ± 0.2 b	1.9 ± 0.2 d	1.8 ± 0.2 c
	P. grandiflora Hook.	1.4 ± 0.1 a	1.4 ± 0.1 a	1.7 ± 0.1 b	2.0 ± 0.1 c
	P. grandiflora "Sundial Mango"	3.0 ± 0.0 b	2.4 ± 0.1 a	3.0 ± 0.3 b	2.4 ± 0.1 a
	P. grandiflora "Double Pink"	2.3 ± 0.1 a	2.9 ± 0.3 c	2.8 ± 0.2 b	2.9 ± 0.1 c
	P. grandiflora "Double White"	5.3 ± 0.4 a	6.3 ± 0.3 d	5.9 ± 0.3 c	5.6 ± 0.2 b
	P. halimoides L.	1.4 ± 0.1 b	1.6 ± 0.1 c	1.6 ± 0.1 c	1.1 ± 0.0 a
Seedling weight (mg)	*P. oleracea* L. ssp. *oleracea*	14.3 ± 0.9 c	9.4 ± 1.1 b	9.3 ± 0.3 b	8.6 ± 0.8 a
	P. grandiflora Hook.	22.2 ± 0.5 d	14.9 ± 3.5 c	10.2 ± 0.4 b	9.6 ± 0.4 a
	P. grandiflora "Sundial Mango"	14.0 ± 0.8 a	14.6 ± 0.7 b	19.3 ± 0.5 c	14.1 ± 0.6 ab
	P. grandiflora "Double Pink"	14.0 ± 0.8 b	14.3 ± 4.7 c	9.9 ± 0.9 a	10.1 ± 0.7 a
	P. grandiflora "Double White"	9.6 ± 4.4 a	14.3 ± 0.9 c	10.6 ± 1.6 b	9.7 ± 0.0 a
	P. halimoides L.	18.0 ± 1.3 d	16.5 ± 0.9 c	14.6 ± 1.4 a	15.3 ± 1.2 b
Seedling vigour index (SVI)	*P. oleracea* L. ssp. *oleracea*	1412.6 ± 60.3 c	1008.9 ± 115.6 a	1091.6 ± 180.5 b	983.3 ± 147.6 a
	P. grandiflora Hook.	1181.5 ± 20.0 c	857.5 ± 33.3 b	741.9 ± 89.7 a	741.3 ± 59.5 a
	P. grandiflora "Sundial Mango"	1238.3 ± 196.6 b	1376.6 ± 79.3 d	1317.5 ± 185.7 c	1030.1 ± 11.7 a
	P. grandiflora "Double Pink"	853.8 ± 204.0 a	917.0 ± 69.7 b	1141.7 ± 169.9 c	894.6 ± 39.0 b
	P. grandiflora "Double White"	451.9 ± 91.5 a	529.1 ± 122.3 b	653.2 ± 166.9 c	442.8 ± 98.4 a
	P. halimoides L.	1390.0 ± 73.1 b	1564.5 ± 221.4 c	1198.0 ± 148.8 a	1212.6 ± 51.4 a

A general inhibition of seedling development with increasing PEG concentrations was confirmed for the wild *Portulaca* species (*P. oleracea* L. ssp. *oleracea*, *P. grandiflora* Hook and *P. halimoides* L.), as well as for *P. grandiflora* cv. 'Sundial Mango', when seedlings dimensions were determined after 14 days of germination: all four showed a reduction of radicle, hypocotyl and cotyledon lengths when comparing the -1 MPa and control treatments (Fig. 1). In most cases the reduction in size, although statistically significant, was relatively small; for example, the largest reduction in radicle length was measured in *P. grandiflora* Hook (31%) followed by *P. oleracea* ssp. *oleracea* (21%), whereas the relative decrease in cotyledon length with respect to the corresponding controls varied between 5% (*P. grandiflora* cv. 'Sundial Mango') and 37% (*P. grandiflora* Hook). The behaviour of the cultivars 'Double Pink' and 'Double White' of *P. grandiflora* was somewhat different, showing no significant change or even a slight increase in radicle and cotyledon lengths with increasing PEG concentrations; in the case of cv. 'Sundial Mango', cotyledon length – but not other parameters – was also significantly higher at -0.25 MPa and -0.5 MPa than in the control germinated in water (Fig. 1).
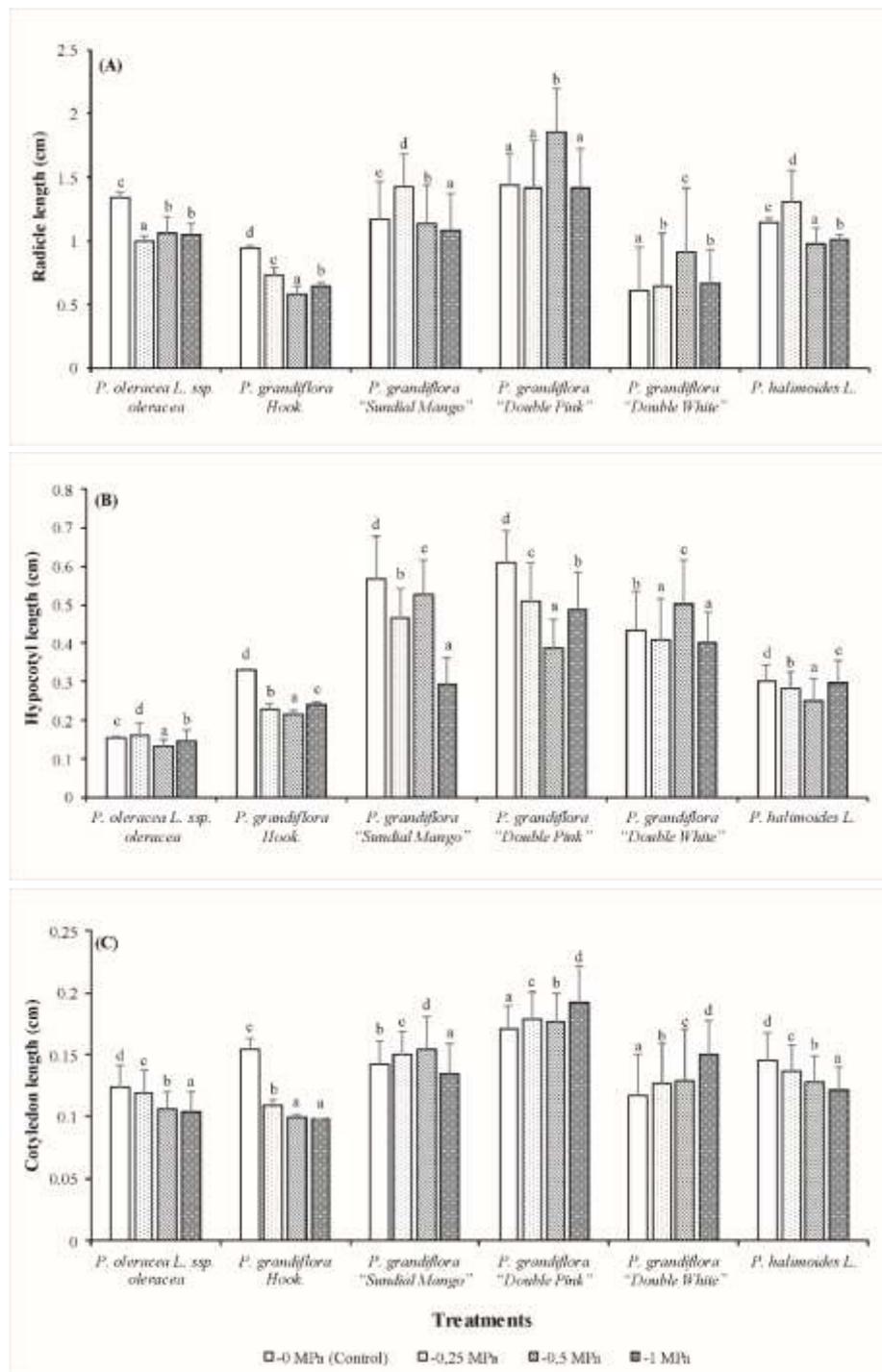
Figure 1. Radicle (A), hypocotyl (B) and cotyledon (C) lengths of seedlings of the six Portulaca taxa, after germination for 14 days at the indicated osmotic potentials (generated by increasing PEG concentrations). Different lowercase letters above the bars indicate significant differences between treatments, for each genotype, according to the Tuckey's HSD test (P<0.05).
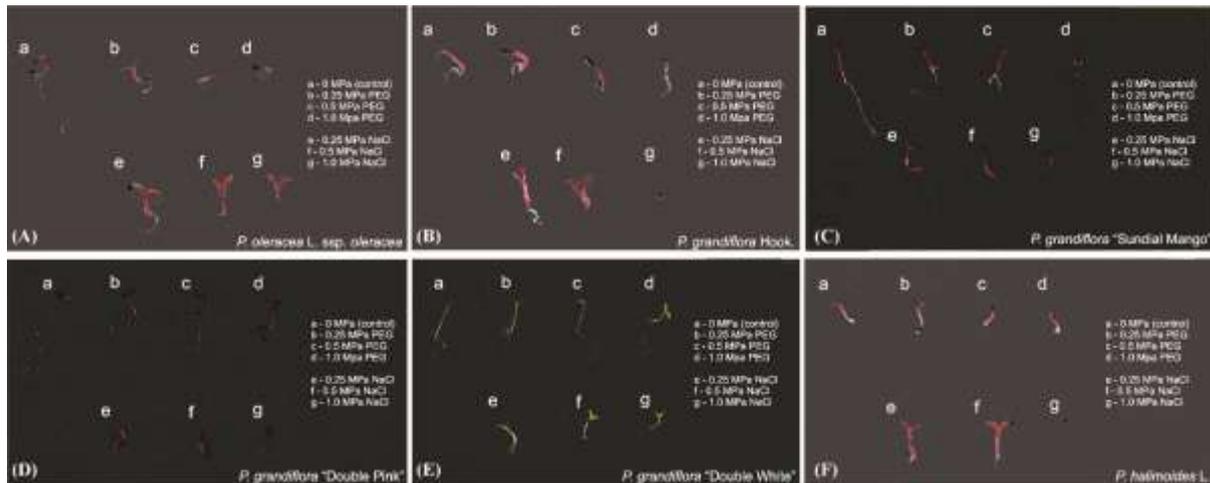
Figure 2. Representative seedlings of the six Portulaca taxa, germinated for 14 days at the indicated osmotic potentials, generated by increasing PEG or NaCl concentration

3.2 Effects of salt stress on seed germination and seedling development

The *in vitro* germination tests described above were repeated in the presence of NaCl, at the concentrations required to provide the same osmotic potentials (-0.25, -0.5, -1.0 MPa) than in the experiments with PEG. Salt stress affected seed germination and early seedling growth in a similar way than osmotic stress, qualitatively, but generally with much stronger quantitative effects. At the highest salt concentration tested, the most resistant accession appeared to be *P. grandiflora* Hook, with ca. 79% final germination percentage – about 16% inhibition with respect to the control – followed by *P. halimoides* L. (63% germination), whereas the most salt-sensitive taxon was *P. grandiflora* cv. 'Double White', with a final germination percentage below 3%, or 94% inhibition as compared with the seeds germinated in water (Table 2). As for PEG, in the presence of the lowest salt concentration (-0.25 MPa) the commercial cultivars of *P. grandiflora* showed a small increase of germination over the controls (Table 2).

Germination velocity was also generally reduced in the presence of increasing NaCl concentrations, as reflected in the increase of the calculated MGTs; here again, *P. halimoides* L. seeds were those germinating most rapidly, followed by *P. oleracea* ssp. *oleracea*, while the largest relative increases of MGT were calculated for *P. grandiflora* Hook and its cvs. 'Sundial Mango' and 'Double Pink' (Table 2).

The strong inhibition of seed germination at high external NaCl concentration led to a significant reduction of seedling weight with respect to the controls in all *Portulaca* accessions, ranging from 20% in *P. grandiflora* cv. 'Sundial Mango' to 80% in the wild *P. grandiflora* Hook species. Calculated values of 'seedling vigour index' decreased by more than 90% of the corresponding controls in all *Portulaca* taxa (Table 2).

As for the PEG experiments, seedlings dimensions were determined after 14 days of germination in the presence of increasing NaCl concentrations (Fig. 3); some examples are shown in Fig. 2. The growth of the seedlings was also inhibited by germination in the presence of salt, with a general, concentration-dependent reduction of the radicle and hypocotyl lengths in all taxa; in fact, at the highest NaCl concentration tested seedlings of most accessions (*P. grandiflora* Hook and its cvs. 'Double Pink' and 'Double White', as well as *P. halimoides* L.) did not develop at all after radicle emergence (Figs. 2 and 3). Under the

same conditions, the other two taxa – *P. oleracea* ssp. *oleracea* and *P. grandiflora* cv. 'Sundial Mango' – showed a strong reduction of radicle length with respect to the control (88% and 94%, respectively), while the latter taxon was more affected by salt in terms of inhibition of hypocotyl length; interestingly, a slight (but significant) salt-dependent increase in cotyledon length was detected in these two accessions (Fig. 3).

Table 2. Effects of increasing osmotic potentials, generated by NaCl in the germination medium, on the indicated seed germination parameters of the six selected Portulaca taxa. Values shown are means ± SE (n = 3). For each parameter and accession, different letters in a row indicate significant differences between treatments according to Tuckey's HSD test (P<0.05).

Parameter	Genotype	Salt stress (NaCl)			
		-0 MPa (Control)	-0.25 MPa	-0.5 MPa	-1 MPa
Germination percentage (%)	*P. oleracea* L. ssp. *oleracea*	94.7 ± 1.3 c	93.3 ± 1.3 c	82.7 ± 15.3 b	44.0 ± 8.3 a
	P. grandiflora Hook.	93.3 ± 1.3 d	90.7 ± 4.0 c	86.7 ± 3.5 b	78.7 ± 10.4 a
	P. grandiflora "Sundial Mango"	70.7 ± 8.1 c	81.3 ± 3.5 d	66.7 ± 3.5 b	34.7 ± 4.8 a
	P. grandiflora "Double Pink"	42.7 ± 11.6 c	50.7 ± 7.0 d	36.0 ± 2.3 b	8.0 ± 2.3 a
	P. grandiflora "Double White"	42.7 ± 3.5 c	45.3 ± 3.5 d	18.7 ± 3.5 b	2.7 ± 1.3 a
	P. halimoides L.	96.0 ± 2.3 b	98.7 ± 1.3 c	100.0 ± 0.0 c	62.7 ± 4.8 a
Mean germination time – MGT (days)	*P. oleracea* L. ssp. *oleracea*	1.5 ± 0.1 a	1.9 ± 0.0 b	2.3 ± 0.7 c	2.5 ± 0.5 d
	P. grandiflora Hook.	1.4 ± 0.1 a	2.0 ± 0.1 b	2.0 ± 0.0 b	3.6 ± 0.3 c
	P. grandiflora "Sundial Mango"	3.0 ± 0.1 ab	2.8 ± 0.2 a	3.2 ± 0.4 b	7.5 ± 0.9 c
	P. grandiflora "Double Pink"	2.3 ± 0.1 a	3.3 ± 0.4 b	3.3 ± 0.2 b	5.7 ± 2.4 c
	P. grandiflora "Double White"	5.3 ± 0.4 d	4.8 ± 0.3 b	5.1 ± 0.3 c	4.7 ± 2.9 a
	P. halimoides L.	1.4 ± 0.1 c	1.3 ± 0.1 b	1.3 ± 0.1 b	1.2 ± 0.2 a
Seedling weight (mg)	*P. oleracea* L. ssp. *oleracea*	14.4 ± 0.9 b	16.6 ± 0.6 c	17.2 ± 3.0 c	5.3 ± 2.8 a
	P. grandiflora Hook.	22.3 ± 0.5 d	20.6 ± 0.5 c	13.9 ± 0.7 b	4.4 ± 0.9 a
	P. grandiflora "Sundial Mango"	13.5 ± 0.8 b	28.8 ± 1.0 d	19.8 ± 0.3 c	10.7 ± 1.0 a
	P. grandiflora "Double Pink"	14.0 ± 0.8 d	9.6 ± 0.3 c	8.7 ± 1.3 b	3.9 ± 0.9 a
	P. grandiflora "Double White"	9.6 ± 4.4 b	19.7 ± 2.2 c	8.9 ± 0.9 b	5.6 ± 1.7 a
	P. halimoides L.	17.6 ± 1.3 b	24.6 ± 2.6 c	25.5 ± 2.1 c	4.2 ± 0.1 a
Seedling vigour index (SVI)	*P. oleracea* L. ssp. *oleracea*	1412.6 ± 60.3 d	744.8 ± 21.9 c	575.9 ± 80.3 b	136.6 ± 25.0 a
	P. grandiflora Hook.	1181.5 ± 19.7 d	606.6 ± 61.4 c	403.3 ± 42.8 b	0.0 ± 0.0 a
	P. grandiflora "Sundial Mango"	1238.3 ± 196.6 c	1371.9 ± 148.8 d	450.7 ± 29.7 b	121.4 ± 19.9 a
	P. grandiflora "Double Pink"	853.8 ± 204.0 c	946.4 ± 102.0 d	362.1 ± 11.0 b	8.2 ± 4.2 a
	P. grandiflora "Double White"	451.9 ± 91.5 c	484.2 ± 128.5 d	108.8 ± 10.4 b	0.0 ± 0.0 a
	P. halimoides L.	1390.0 ± 73.1 c	1254.1 ± 90.7 b	1192.2 ± 128.1 b	0.0 ± 0. a

Despite the interest of purslane species as (minor) vegetable, medicinal and ornamental crops, very little is known about their tolerance to abiotic stress, with the exception of some accessions of common purslane, *Portulaca oleracea*. This species has been shown to be relatively resistant to drought or salinity (ALAM & al., [14]; REN & al. [19]), and it has even been proposed that it can be used as a salt-removing crop (KILIC & al., [20]). There are also some data on seed germination of *P. oleracea* under salt stress, showing that NaCl, at a concentration of ~100 mM, reduces the percentage of germination by 50% (CHAUHAN & JOHNSON [21]); yet, most studies in this species have focused on other factors possibly affecting seed germination, such as temperature, light or pH (SINGH [22]; CHAUHAN & JOHNSON [21]). In any case, there is no previous systematic study comparing the behaviour of different *Portulaca* taxa in response to water deficit and salt stress, such as that presented here.
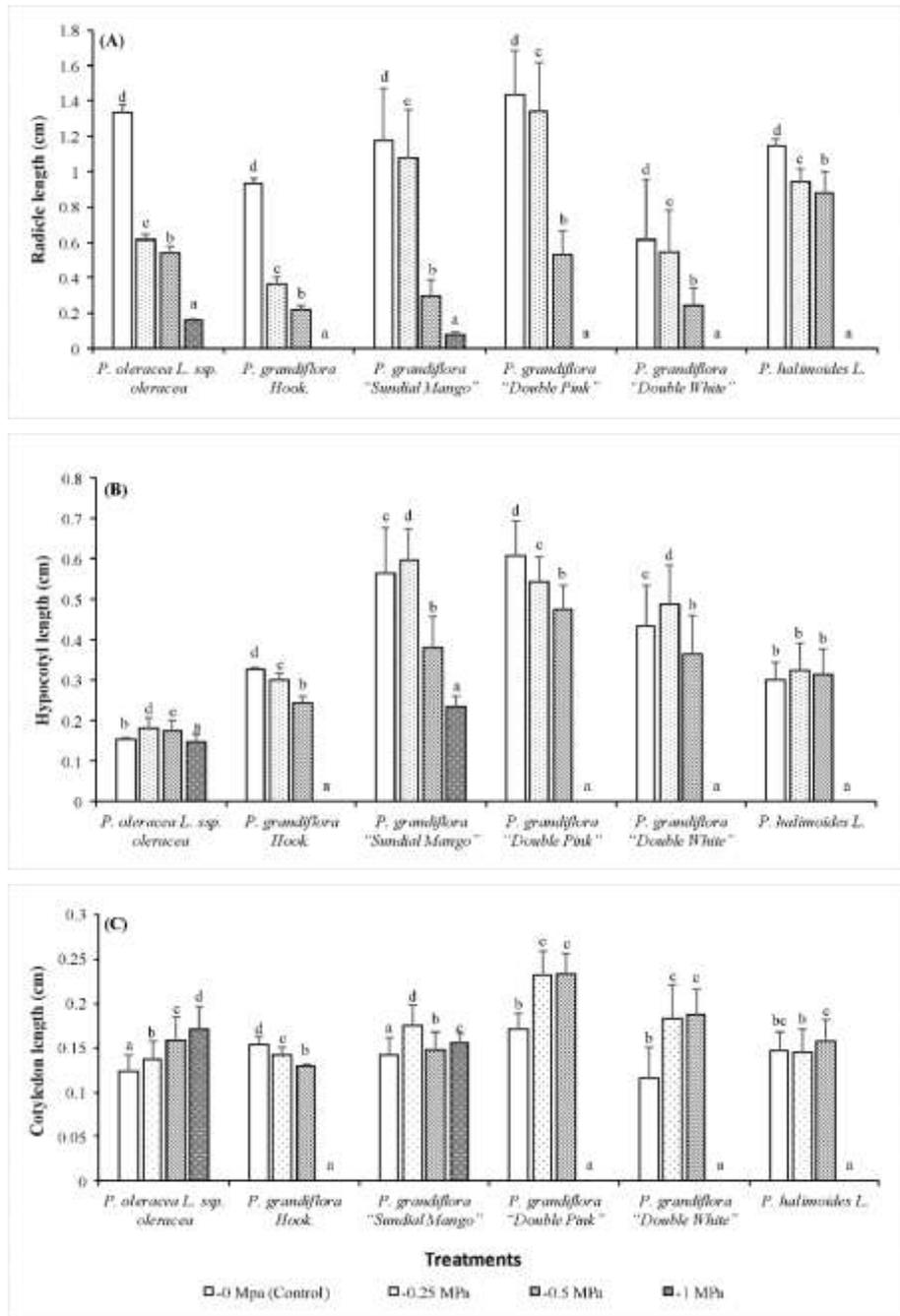
Figure 3. Radicle (A), hypocotyl (B) and cotyledon (C) lengths of seedlings of the six Portulaca taxa, after germination for 14 days at the indicated osmotic potentials (generated by increasing NaCl concentrations). Different lowercase letters above the bars indicate significant differences between treatments, for each genotype, according to the Tuckey's HSD test (P<0.05).

The results of our work revealed that seed germination and early seedling growth of all tested *Portulaca* taxa are inhibited, in a concentration-dependent manner, in the presence of increasing concentrations of both, PEG (osmotic stress) and NaCl (salt stress). In general, all determined germination and seedling parameters (germination percentages and velocity, seedling fresh weight, seedling vigour index, and radicle, hypocotyl and cotyledon lengths) decreased in response to stress, although with quantitative differences between the selected species and cultivars. This is a common behaviour of plants and, in fact, these early phases of the life cycle are generally the most sensitive to stress (e.g., VICENTE & al. [23]; ALMANSOURI & al. [24]; REHMAN & al. [25]; OKCU [26]; FRANCO [11]). For all accessions, the salt solutions had a stronger negative effect on germination than the PEG solutions generating the same osmotic potentials; this can be easily explained by the 'ion toxicity' component of salt stress that, in addition to the osmotic component, has a number of deleterious effects in plants, inhibiting many enzymatic activities and cellular processes, directly inactivating proteins or interfering with mineral nutrition (MUNNS [5]; ZHU [27]).

Strong inhibition of germination and early seedling growth under high salinity conditions has also been observed in many halophytes, salt-tolerant plants that can withstand much higher salt concentrations in the adult stage (see VICENTE & al. [23]; CORDAZZO [28]; HOULE & al. [29], for a few examples). However, in some halophytic species, low or moderate salt concentrations may induce a certain *stimulation* of germination, increasing germination percentages and/or germination velocity; this has been observed, for example, in quinoa (PANUCCIO & al., [30]) and some *Limonium* species (AL HASSAN & al., [31]). In our experiments, we have also detected a significant increase of germination percentages in the commercial cultivars of *P. grandiflora* ('Sundial Mango', 'Double Pink' and 'Double White'), at PEG concentrations generating -0.25 MPa and -0.5 MPa osmotic potentials, or in the presence of the lowest NaCl concentration tested. It should be pointed out, however, that these taxa showed lower germination percentages in water, as compared to the wild accessions, *P. oleracea* L. ssp. *oleracea*, *P. grandiflora* Hook. and *P. halimoides* L.

It is difficult to establish the relative degree of tolerance to osmotic or salt stress of the analysed accessions if we consider the changes observed in all germination and seedling parameters since no uniform patterns were obtained in the different taxa. In practical terms, looking only at the final germination percentages and germination velocity under stress conditions, we could conclude that *P. halimoides* L. is the taxon most tolerant to water deficit, followed by *P. grandiflora* Hook and *P. oleracea* L. ssp. *oleracea*. The same three species appear to be the most tolerant to salt stress – as far as the salt concentration is not too high to cause a complete inhibition of seedling development after radicle emergence – although in this case *P. grandiflora* Hook shows the highest germination percentages. In any case, the commercial *P. grandiflora* cultivars seem to be slightly more stress-sensitive, probably because selection for other traits (such as their ornamental value) has reduced the efficiency of their stress responses. Further studies, using adult plants, will be required to establish which of these species are the most suitable to be commercially cultivated with reduced irrigation or using low-quality, saline water, in the context of climate change and a more sustainable agriculture.

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