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Effect of salt stress on accumulation of proline and soluble sugars in cladodes and roots of two *Opuntia* species existing in Algerian steppe

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Abstract

The accumulation of soluble salts in the soil diminishes significantly the value of the productivity of cultivated or not cultivated species. The introduction of plants tolerant to salinity is one of the techniques used for the valuation of marginal soils. Faced with this problem, the *Opuntia* has a great capacity to adapt to various weather conditions. The objective of the present study was the influence of sodium chloride (NaCl) at different levels (control, 200, 400, 600meq.l^{-1}) on synthesis of proline and soluble sugars in two species of Opuntia. The results show the variability of the proline accumulation and soluble sugars according to the organ, species and dose of salts. The accumulations of these compounds are much more in cladodes compared to the roots. In *O. engelmanii var. longuiformis* the content of proline will increase from 0.46 µg/100g FW in the control plants to 0.89 and 0.93 µg/100g FW in the stressed plants at 400 and 600 meq.l^{-1} of NaCl respectively in young cladode. At O. *streptacantha* Lem. the highest soluble sugars content in the aged cladode (98 µg/100g FW) was recorded with the stress at 600 meq.l^{-1}. This suggests that, both *Opuntia* species are able to accumulate these biochemical compounds, order to ensure the osmotic adjustment under stress conditions at varying proportions.

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Introduction

In arid and semi-arid areas, soil salinization is a major abiotic factor that reduced agricultural yields of many cultures (Zid and Grignon, 1991; Baatour *et al.*, 2004). The accumulation of soluble salts in the soil diminishes significantly the value of the productivity of cultivated or not cultivated lands (Pessarakli, 1999). The FAO (1996) estimates that from 237 millions of hectares of irrigated lands, around of 30 millions are gravely affected for the salt; other 80 millions of hectares have been affected in some grade and around 1.5 millions of hectares lost annually because of watering due to the flooding or the salinity. During the past 45 years, much attention has been given to the effect of NaCl on plant growth and production; however, the fundamental mechanisms determining the responses of plants to salinity are still not understood (Amzallag*et al.*, 1990). In Algeria, saline soils cover an area of 3.2 million ha (Hamdy, 1999). According to Le Houérou (2000), Algeria is among the most affected countries by salinization.

The introduction of plants tolerant to salinity is one of the techniques used for the valuation of marginal soils. Faced with this problem, theOpuntiahas agreat capacity to adaptto a variety of climatic conditions, and has been cultivated in the arid and semi-arid climates of many countries (Pimienta, 1994). Tolerance of cacti to salinity has been studied by (Berry and Nobel, 1985), (McCree and Richardson, 1987),(Silverman *et al.*,1988), (Gersani*et al.*,1993), (Le Houérou, 1996), (Nobel, 1998), (Le Houérou, 2002),(Murillo-Amador *et al.*, 2001), (Cony*et al.*, 2006), (Ochoa-Alfaro*et al.*, 2008), (Nieto–Garibay*et al.*, 2011). Among other *Opuntia quimilo* from Argentina is reported to be more tolerant to NaCl than other *Opuntia* species. Another cactus with high tolerance to salinity is *Cereus validus*(Nobel, 1998).

In this study, our objectiveis to study thebiochemical responsesof twoOpuntiaspeciesin relation tosalt stress.Order to valorization these species in the revegetation of degraded areas in arid and semi-arid. In this context these species could be a source of germplasm of great agricultural and ecological interests.

Materials and methods

Plant material and culture conditions

The plant material used in this study consists of cladodes and roots of two *Opuntia* species: *Opuntia engelmanii var. longuiformis* and *Opuntia streptacantha*Lem.grown in the region of M'sila (Algeria).

The cladodes are planted in 2012, in the form ofcompletelyrandomized designwith twentyreplications, the cladodes are planted in pots 2 liter volume, filled with sand and exposed to natural growing conditions. The cladodes are watered weekly by distilled water. Salt stress is applied after the second year of culture. Sodium chloride (NaCl) was chosen as a stress agent. Four concentrations of (NaCl) were used:0 (control), 200, 400 and 600meq.l^{-1}.

Studied traits

The studied traits included; proline and soluble sugars. After 60 daysof application to the salt stress, weperformed leviesfromplant material (youngcladodes, agedcladodes and roots) (Fig. 1). 20 samplesper species perconcentrationareintended for the analysis.

Methods of measuring traits

Proline is extracted and assayed according to the method of Troll and Lindsley (1954), streamlined and developed by Drier and Goring (1974). Soluble sugars were extracted and assayed according to the method of Schields and Burnett (1960). The contents are expressed by µg/100g Fresh Weight(FW)for proline and solubles sugars.

Statistical analysis

The results obtained were analyzed using the statistical software StatBox Pro. V. 6,4 and the mean of data were compared by Newman-Keuls test at 5 %.

Results

Effect of NaCl on accumulation of Proline

Fig. 2 indicatesthat, prolinecontent increasesin the differentplant organsaccording to theincreasing salinity. Note also that, young cladodes of *Opuntia engelmanii var. longuiformis* are rich to prolinecomparedto other organs, both in the control plants than those treated with different concentrations of salts.The contentof prolinewillincreasefrom 0.46 µg/100g FW in the control plants to 0.89 and 0.93 µg/100g FWin thestressed plants at 400 and 600 meq.l^{-1}of NaClrespectively.

Table 1. Effect of salt stress (NaCl) on proline content (µg/100g FW) in different organs of O.engelmaniivar. longuiformis and O.*streptacantha* Lem. Each value represents an average of 20 samples ± SD. Values bearing the same letter in each column are not significantly different at p <0.05.

	O.engelmanii var. longuiformis			*O. streptacantha*Lem.		
	Aged cladode	Young cladode	Root	Aged cladode	Young cladode	Root
Control	0,40±0.007^{c}	0,46±0.006^{d}	0,23±0.003^{c}	0.31±0.004^{d}	0.31±0.005^{d}	0,13±0,006^{c}
200 meq	0,75±0.004^{b}	0,84±0.003^{c}	0,46±0,002^{b}	0.68±0.005^{b}	0.60±0.006^{b}	0,34±0,003^{b}
400 meq	0,78±0.003^{a}	0,89±0.004^{b}	0,67±0,003^{a}	0.70±0.007^{a}	0,65±0.001^{a}	0,45±0,003^{a}
600 meq	0,79±0.004^{a}	0,93±0.003^{a}	0,67±0,001^{a}	0.62±0.003^{c}	0,53±0.001^{c}	0,44±0,004^{a}
Probability	0.0001	0.00008	0.00006	0.0001	0.0001	0.00011

We also observe that the accumulation of proline in the roots is about doubleandthree timeshigher in the treated plants to 400 and 600 meq.l^{-1} of salt (0.46 and 0.67 µg/100g FW, respectively), compared to the control plants, against to 0.23 µg/100g FW. In aged cladodes, the highest proline content is recorded in the plants treated with 600 meq.l^{-1}(0.79 µg/100g FW). Furthermorethe quantity of prolineaccumulatedis increasedin the direction of roots, agedcladodes and youngcladodes. The content of proline accumulated is increased in the control plants toward stressed plants with salts (NaCl) (table 1).

Table 2. Effect of salt stress (NaCl) on soluble sugars content (µg/100g FW) in different organs of O.engelmaniivar.longuiformisandO. *streptacantha*Lem. Each value represents an average of 20 samples ± SD. Values bearing the same letter in each column are not significantly different at p <0.05.

	O. engelmanii var. longuiformis			*O. streptacantha*Lem.		
	Aged cladode	Young cladode	Root	Aged cladode	Young cladode	Root
Control	54±0.28^{d}	89±0.35^{d}	24±0.07^{d}	52±0.21^{d}	23±0.49^{d}	17±0.49^{d}
200 meq	73±0.49^{c}	92±0.35^{c}	50±0.70^{c}	64±0.21^{c}	28.3±0.49^{c}	20±0.21^{c}
400 meq	77±0.42^{b}	95±0.49^{b}	58±0.49^{b}	71±0.14^{b}	55±0.28^{b}	50±0.35^{b}
600 meq	80±0.56^{a}	99±0.49^{a}	63±0.42^{a}	98±0.49^{a}	75±0.14^{a}	69±0.21^{a}

In*Opuntia streptacantha*Lem.the proline content is changing in all organs with increasing salt concentration, compared to control plants (Fig. 2). Prolinecontentsfluctuate withthe organ and thesalttreatment. In roots the accumulationis low with values: 0.13 µg/100g FW, 0.34µg/100g FW, 0.45 µg/100g FW and 0.44 µg/100g FW within control plant, 200, 400and 600 meq.l^{-1} respectively, comparedatagedcladode with values: 0.31 µg/100g FW, 0.68 µg/100g FW, 0.70 µg/100g FW and 0.62 µg/100g FW within control plant, 200, 400and 600 meq.l^{-1} respectively. The quantity of prolineaccumulatedis increasedin the direction of roots, youngcladodes and agedcladodes (table 1). Statistical analysis (Table 1) indicates that the root system is significantly less rich to proline,

comparatively with cladodes, for all treatments.

Effect of NaCl on the accumulation of soluble sugars

In *O. engelmaniivar. longuiformis*, the resultsanalysisindicates thatthe amounts ofsoluble sugarsarevariable, in root, aged cladode and young cladode (fig. 3).

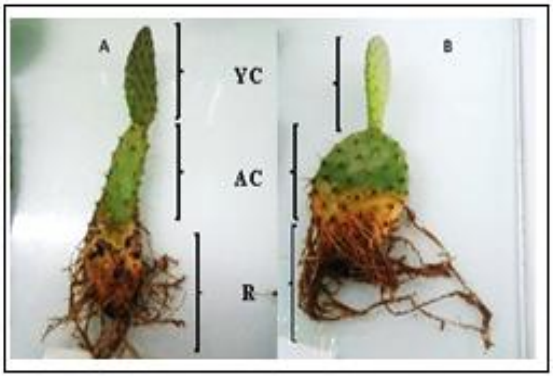

Fig. 1. Different organs of Opuntia plant(YC: Young cladode; AC: aged cladode; R: root):(A)O. engelmaniivar.longuiformis;(B)O. *streptacantha*Lem. intendedfor theanalysis.

Indeed, we note that the carbohydrate compounds are more concentrated in the cladodes than in the roots,as well in the control plants than those treated with different concentrations of salts. This accumulation is higher in young cladodes especially when plants are stressed at 600 and 400 meq.l^{-1} with values 99 µg/100g FW and 95 µg/100g FW respectively. The increase in salinity level, there was an increase; in soluble sugars content was noticed. A35.18%, 42.59% and 48.14% increaseinthestressed outplantswith 200, 400and 600 meq.l^{-1} respectively, compared to the control plants.Whereas in roots, soluble sugars amount is about 63 µg/100g FW with stressed outplantsby600 meq.l^{-1}(table 2).

AtO.*streptacantha*Lem.the quantity of soluble sugarsaccumulatedis increasedin the direction of roots, youngcladodes and agedcladodes. The highest soluble sugars content in the aged cladode (98 µg/100g FW) was recorded with the stress at 600meq.l^{-1}. Besides, the amount of soluble sugars in roots is lowest (69 µg/100g FW) at plants treated with 600meq.l^{-1}, in comparison with the controls (17 µg/100g FW)(fig. 3, table 2). When O.*streptacantha*Lem.was treated with NaCl with different concentration there was significant difference was noticed for soluble sugarscontent in roots, young cladodes and aged cladodes.

Discussion

The biochemical response, analyzed through the expression of the accumulation of proline and soluble sugars of both species of *Opuntia*: *O. engelmaniivar. longuiformis* and *O. streptacantha*Lem. in salt stress conditions with (NaCl) 200, 400 and 600 meq.l^{-1}, show that the plants accumulate these compounds in different organs. This accumulation varied from one organ to another, from one species to another, depending on the intensity of concentration salt stress.

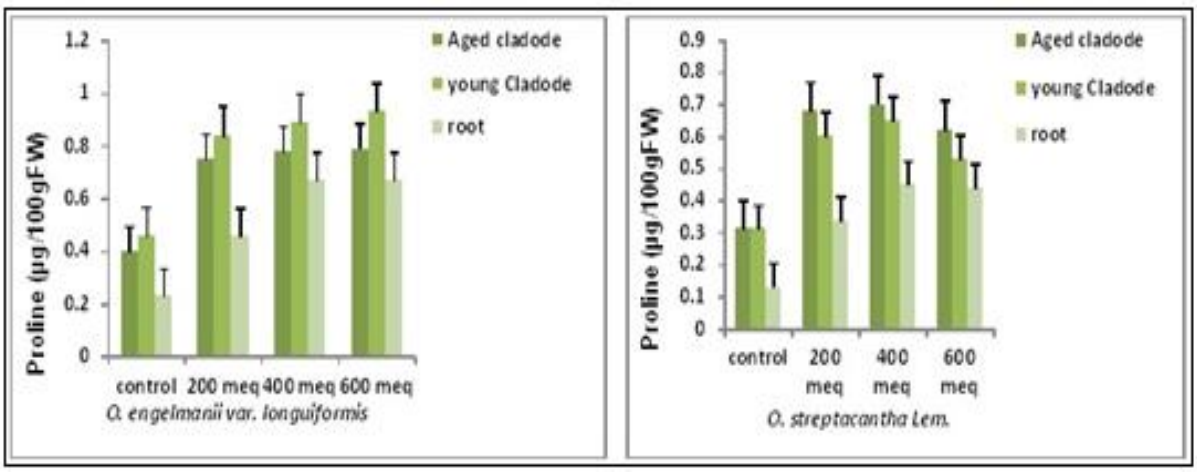

Fig. 2. Prolinecontentin different organsof Opuntia plants (O. engelmaniivar.longuiformisandO.*streptacantha*Lem.) stressed by NaCl for two months.

In*O. engelmanii var.longuiformis*, the accumulation of proline is in the directionroot, cladodes, as well in control plants toward stressed plants with different concentrations. In the cladodes, the prolinepreferentially concentrated in young cladodes at significantly higher levels when the salinity of NaCl increases. These results are in conformity with the study of Hadjadj*et al.* (2011) on Atriplex. Indeed, in the *O. engelmanii var. longuiformis* stressed by NaCl (200, 400 and 600 meq.l^{-1}) prolinecontent increasing much more in young cladodescomparatively to aged cladodes. Ould El Hadj-Khelil (2001) noticed that in tomato plants under stress of 200 mMNaCl, the accumulation of proline is greater in young leaves than in the basal ones, and application of a second salt treatment generates a further increase in the amino acid. This increase is more important when leaf tissues are young and salinity is high. As against Martinez *et al.* (2005) showed that exposure of plants *Atriplexhalimus* L. at 50 mMof NaCl induced accumulation of proline in aged leaves,relatively of the young leaves.

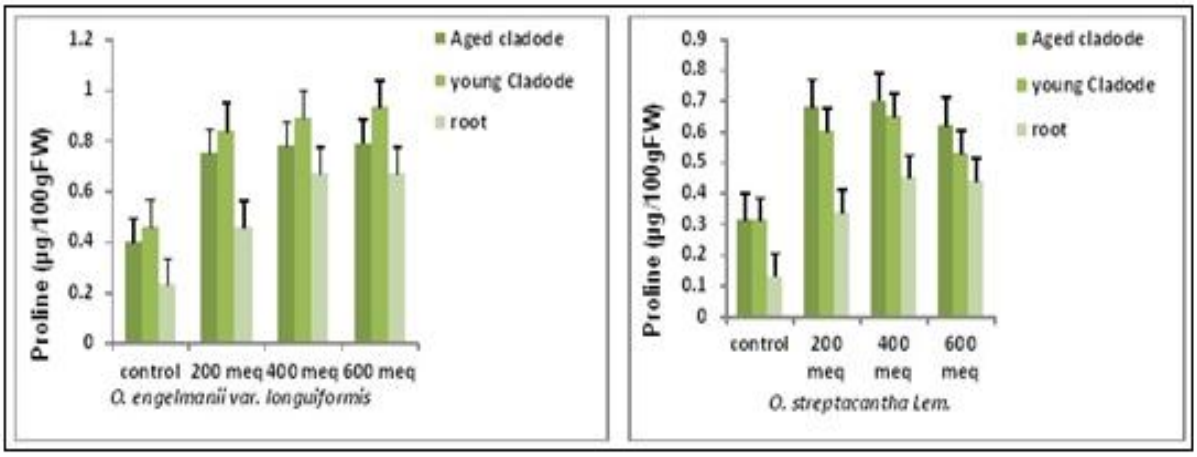

Fig. 3. Soluble sugars contentin different organsof Opuntia plants (O. engelmaniivar. LonguiformisandO. *streptacantha*Lem.) stressed by NaCl for two months.

For *O. streptacantha*Lem. in agedcladode the quantity of prolineaccumulatedis increased with 0.70 µg/100g FWat 400 meq.l^{-1}. In roots the accumulationis high with 0.45 µg/100g FWat 400 meq.l^{-1}comparatively to control plant with 0.13 µg/100g FW. These results are accordance with the work of (De-lacerda*et al.*, 2001) have found that exposure of two sorghum genotypes (*Sorghum bicolor* L. *Moench*) one sensitive and the other tolerant to 100 mMof NaCl, causes an increase in proline in all parts of the plants two genotypes and more particularly in the aged leaves: third and fourth leaf from the apex.

The accumulation of proline in young cladodes may result from the strong accumulation of salts in the latter. Ould el-Hajj khelil (2001) reports that older basal leaves are involved in the sequestration of excess Na^+, this is done to benefit young leaves growing, which appear to be protected while Na^+ in excess. Huber (1974) showed that the salt inhibits the catabolism of the amino acid at the proline dehydrogenase, an enzyme involved in the degradation of proline. It could also correspond to the stimulation of the synthesis. Indeed, (Hu *et al.*, 1992) assert that in *Vignaaconitifolia*, agenecoding forsynthetase pyrroline-5-carboxylate, is highly expressed in leaves and roots of plants treated with 200 mMof NaCl.

The accumulation of proline would be caused by the increased synthesis from glutamate and ornithine (Hare and Cress, 1997). Under stress conditions, the amino acid metabolism is greatly altered and protein synthesis increases as a result of these metabolic conditions, the synthesis of proline can be promoted by an increase in concentrations of metabolites accompanied by a production of precursors proline

and may be the main cause of the accumulation of proline in the tissues of plants exposed to stress conditions (Silveira*et al.*, 2001).

Also, our results show that the amount of accumulated proline in both species of Opuntia is fairly high, which suggests that this osmolyte is among the most effective biochemical parameters for this plant's resistance to extreme environmental conditions and expressingmore markedly in the cladodes. It would migrate from the various organs to these cladodes for purposes of protection and increased photosynthetic efficiency because some studies have reported that proline is synthesized in the leaves and transported to the resistance sites of aggressions (VezinaandPaquin, 1982; LaLiberteandPaquin, 1984) others report that the proline migrates to the leaves for stressed plants; such is the case for sorghum (Weimberg*et al.*, 1984), eggplant (Joshi, 1984), cotton (Boutelier, 1986), vines (ImamulHuqandLarher, 1984), beans (Belkhodja, 1996; BelkhodjaandBenkablia, 2000). Bellinger*et al.* (1989) reported that thisaccumulationis not asensibilityindicatorof plant to stress, but an indicatorof acquisition to tolerance for stresses.

Concerning soluble sugars, according to the results on accumulation of soluble sugars in the root and cladodes of *O. engelmanii var. longuiformis* and *O. streptacantha* Lem. it appears that these carbohydrate compounds have also increased depending to the increase in salt concentration for both species. The accumulation of soluble sugars occurs significantly in cladode than in roots under normal conditions or under salt stress in *Opuntia*. Belfakih*et al.* (2013), showed an increase in the content of soluble sugars in a banana variety (dwarf) in a salt stress situation. Ben Khaled*et al.* (2003) reported that exposure of plantlets to clover (Alexandria) at 2, 4, 6 and 8 $gl^{-1}$of NaCl, induced an accumulation of soluble sugars in the leaves. This increase was 37% to 4 gl^{-1} and 57% to 8 $gl^{-1}$of NaCl. These results reflectfirstly, the variability of biochemical metabolism of both *Opuntia*species under salt stress, and secondly, express their ability to synthesize and accumulate the soluble sugars in different organs. Sugarsplay an importantrole inosmotic adjustment, well as at of stabilization of some proteins.The accumulation ofsugarsseems to inducegelationof the cell contentsby saturatingthe intracellularenvironment, this phenomenon allows avoid the crystallization of contained molecules in cell, therefore limit the damage of cellular structures (Dubos, 2001). There seems to be some proportionality between the amounts of accumulated proline and those of soluble sugars. Therefore, the increase content of proline, under the effect of saline treatment, is proportional to the increase in quantity of soluble carbohydrates.These results explain the existence of a connection between the biosynthesis pathway of proline and the accumulation of soluble sugars; in general, they could provide the carbon precursor and the chemical energy required for the synthesis of proline. The α-ketoglutarate, intermediary respiratory tract during glucose oxidation, play a particularly important role such as acceptor amino group of glutamine, forglutamateconversion. This one;the main precursor of proline during a stress situation (Hopkins, 2003).

Conclusion

Accumulationsof proline andsoluble sugarsin both speciesof Opuntia would be involved in osmotic adjustment mechanisms and would also serve such as osmoprotecteurs. Thesebiochemical elements can be considered as «Biochemical markers» of degree of tolerance to salt stress and consequently can be used to early selection of salt tolerant species. However, quantitative variation depends upon the species, the organ and level of stress provoked. This variation is probably related to the role of this amino acid at the cellular level and its involvement in osmotic adjustment. According to our findings, the two *Opuntia* species are able to grow in the presence of different levels salinity ranging from 200to 600 $meq.l^{-1}$NaCl.

References

Amzallag GN, Lerner HR, Poljakoff-Mayber A. 1990. Exogenous ABA as a modulator of the response

of sorghum to high salinity. Journal of Experimental Botany**41,** 1529-1534.
http://dx.doi.org/10.1093/jxb/41.12.1529

Baatour O, M'rah S, Ben Brahim N, Boulesnem F, Lachaal M. 2004. Réponse physiologique de la gesse (*Lathyrussativus*) à la salinité du milieu. Revue des Régions Arides **1,** 346-358.

Belfakih M, Ibriz M, Zouahri A. 2013. Effet de la salinité sur les paramètres morphophysiologiques de deux variétés de bananier (*Musa acuminata* L).Journal of Applied Biosciences **70,** 5652-5662.
http://dx.doi.org/10.4314/jab.v70i1.98808

Belkhodja M. 1996. Action de la salinité sur les teneurs en proline des organes adultes de trois lignées de fève (*Vicia faba* (L.)) au cours de leur développement. ActaBotanicaGallica **143(1),** 21-28.
http://dx.doi.org/10.1080/12538078.1996.10515315

Belkhodja M, Benkablia M. 2000. Proline reponse of faba bean (*Viciafaba*L.) under salt stress. Egyptian Journal of Agricultural Research **78(1),** 185-195.

Bellinger Y, Bensaoud A, Larher F. 1989. Physiological significance of proline accumulation, a trait of use to breeding for tress tolerance.Colloque Physiology-Breeding of winter Cereals for Stressed Mediterranean Environments, Montpellier, France, 3-6 July 1989. Les colloques n°55. Paris, Ed. Inra. 1991.

Ben Khaled L, Morte Gömez A, Honrubia M, Oihabi A. 2003. Effet du stress salin en milieu hydroponique sur le trèfle inoculé par rhizobium. Agronomie, EDP Sciences **23(7),** 553-560.
https://hal.archives-ouvertes.fr/hal-00886208

Berry WL, Nobel PS. 1985. Influence of soil and mineral stresses on cacti. Journal of Plant Nutrition **8,** 679-696.

Boutelier E. 1986. Essai du chlorure de sodiumsur la physiologie du cotonnier (*Gossypiumhirsutum* L.). Son rôle dans l'acquisition de la résistance à la sécheresse. Thèse de Doc. Université de Paris 6, France, 142p.
http://www.sudoc.fr/174778058

Cony MA, Trione SO, Guevara JC. 2006. Macrophysiological responses of two forage *Opuntia Species* to salt stress. Journal of the Professional Association for Cactus Development **8,** 52-62.

De-Lacerda CF, Cambraia J, Oliva Cano MA, Ruiz HA.2001. Plant growth and solute accumulation and distribution in two sorghum genotypes, under NaCl stress.Revista Brasileira de Fisiologia Vegetal **13(3),** 270-284.
http://dx.doi.org/10.1590/S010331312001000300003

Dreier W, Göring M. 1974. Der EinflusshoherSalzkonzentration auf verschiedenephysiologische Parameters von Maiswurzeln. Wiss. Z. Humboldt Univ. Berlin, Reihe/Math. Naturwiss **23,**641-644.

Dubos C. 2001. Réponse moléculaire de jeunes plants de pin maritime soumis à un stress hydrique en milieu hydroponique. Thèse de Doc. Université Henri Poincaré, Nancy I,p. 54-55.

FAO. 1996. Alimentos para todos. CumbreMundial sobre la Alimentación. 13-17 noviembre, Roma, Italia, 64 p.

Gersani M, Graham AE, Nobel PS. 1993. Growth response of individual roots of *Opuntia ficus-indica* to salinity. Plant Cell and Environment **16,** 827-834.
http://dx.doi.org/10.1111/j.13653040.1993.tb00504.x

Hadjadj S, Djerroudi O, Bissati S. 2011.Etude comparative des mécanismes biochimiques de tolerance au stresse salin de deux espèces d'Atriplex: *Atriplexhalimus*L. et *Atriplexcanescens*(Purch) Nutt.Algerian journal of arid environment **1(2),** 3-10.
http://dspace.univouargla.dz/jspui/handle/123456789/6608

Hamdy A. 1999. Saline irrigation and management for a sustainable use.In: Advanced Short Course on Saline Irrigation Proceedings Agadir, Morocco, 152-227.

Hare PD, Cress WA. 1997. Metabolic implications of stress induced proline accumulation in plants. Plant Growth Regulation **21,** 79-102. http://dx.doi.org.ez.statsbiblioteket.dk:2048/10.1023/A:1005703923 3.47

Hopkins WG. 2003. Physiologie végétale. 2éme Ed. De Boeck, Bruscelles, 61-476.

Hu CA, Delauney AJ, Verma DP. 1992. Abifunctional enzyme (b1-pyrroline-5- carboxylate synthetase) catalyzes the first two steps in proline biosynthesis in plant. Proceeding of the National Academy of Sciences of the United States of America **89(19),** 9354-9358.

Huber W. 1974. Influence of NaCl and abscisic acid treatment on praline metabolism and some further enzymes of amino acid metabolism in seedlings of *Pennisetumtyphoides*. Planta. **121,** 379-394.

Imamul Huq SM, Larher F. 1984. Osmoregulation in Higher Plants:Effect of maintaining a constant Na: Ca ratio on the growth, ion balance and organic solute status of NaCl treated cowpea. *Vignasinensis*L.Pflanzenphysiologie **113,** 163-176. http://dx.doi.org/10.1016/S0044-328X(84)80051-3.

Joshi S. 1984. Effect of salinity stress on organic and mineral constituents in the leaves of *Pigeonpeacajanus* L. Plant and soil **82,** 69-76.

La Liberte G, Paquin R. 1984. Effets des basses températures avec ou sans apport d'acide glutamique et de saccharose sur la teneur en proline et la tolérance au gel du blé d'hiver. Physiologievégétale **22,** 305-315.

Le Houérou HN. 1996. The role of cacti (*Opuntia spp*) in erosion control, land reclamation, rehabilitation and agricultural development in the Mediterranean Basin. Journal of Arid Environments **33,** 135-159. http://dx.doi.org/10.1006/jare.1996.0053

Le Houerou HN. 2000. Use of fodder trees and shrubs (trubs) in the arid and semi-arid zones of West Asia and North Africa: history and perspectives. In: Gintzburger G, Bounejmate M, Nefzaoui A, Ed. Fodder Shrub Development in Arid and Semi-arid Zones. Proceedings of the Workshop on Native and Exotic Fodder Shrubs in Arid and Semi-arid Zones, 27 October-2 November 1996, Hammamet, Tunisia. ICARDA, Aleppo, Syria **1,** 9-53.

Le Houérou HN. 2002. Cacti (*Opuntia spp*) as a fodder crop for marginal lands in the Mediterranean Basin. Acta Horticulturae **581,** 21-46. http://agris.fao.org/aos/records/US201400.119918

Martinez JP, Kinet JM, Bajji M, Lutts S. 2005. NaCl alleviates polyethylene glycolinduced water stress in the halophyte species *Atriplexhalimus*L. Journal of Experimental Botany **56(419),** 2421-2431. http://dx.doi.org/10.1093/jxb/eri235

McCree KJ, Richardson SG. 1987. Salt increases the water use efficiency in water stressed plants. Crop Science **27,** 543-547. http://dx.doi.org/10.2135/cropsci1987.0011183X002700030024x

Murillo-Amador B, CorteÂs-Avila A, Troyo-DieÂguez E, Nieto-Garibay A, Jones HG. 2001. Effects of NaCl salinity on growth and production of young cladodes of *Opuntia ficus-indica*. Journal of Agronomy and Crop Science **187,** 269-279. http://dx.doi.org/10.1046/j.1439-037X.2001.00529.x

Nieto–Garibay A, Troyo–Diéguez E, Valdez–Cepeda RD, Rueda–Puente E, García–Hernández JL, Flores–Hernández A, Zamora–Salgado S, Orona–Castillo I, Murillo–Amador B. 2011. Plant height and

mineral content of *Opuntia taponag*rowing along the coasts of Baja California Sur, México. Journal of the Professional Association for Cactus Development **13,** 77-87.

Nobel PS. 1998. Los Incomparables Agaves y Cactos. Ed. Trillas, México, 211 p.

Ochoa-Alfaro AE, Silva-Ortega CO, Becerra-Flora A, Flores-Rivas J, Jiménez-Bremont JF. 2008.Effect of salt stress, proline, and polyamines on seed germination of *Opuntia streptacantha*. Journal of the Professional Association for Cactus Development **10,** 56-70.

Ould El, Hadj-Khelil A. 2001. Contribution à l'étude de réponses métaboliques de la tomate à la salinité. Thèse de doc. Université de Rennes I, France, 34-44 P.

Pimienta BE. 1994. Prickly pear (*Opuntia spp*.): a valuable fruit for the semi-arid land of Mexico. Journal of Arid Environments **28,** 1-11. http://dx.doi.org/10.1016/S0140-1963(05)80016-3

Pessarakli M. 1999. Response of green beans (*Phaseouls vulgaris* L.) to salt stress. In: Handbook of plant and crop stress. Pessarakli M. Ed. Marcel Dekker, Inc. U.S.A., 827-842 P.

Schields R, Burnett W. 1960. Determination of protein bound carbohydrate in serum by modifiedanthronemethod.Analytical Chemistry **32,** 885-886. http://dx.doi.org/10.1021/ac60163a053

Silveira JAG, Melo ARB, Viégas RA, Oliveira JTA. 2001. Salinity-induced effects on nitrogen assimilation related to growth in cowpea plants. Environmental and Experimental Botany **46(2),** 171-179.

Silverman FP, Young DR, Nobel PS. 1988. Effects of applied NaCl on *Opuntia hemifusa*. PhysiologiaPlantarum **72,** 343-348. http://dx.doi.org/10.1111/j.13993054.1988.tb05843.x

Vezina L, Paquin R. 1982. Effets des basses températures sur la distribution de la proline dans les plants de Luzerne. *Medicagomediapers*.Physiologie Végétale **20,** 101-109.

Weimberg R, Lerner HR, Poljakooff–Mayber A. 1984. Changes in growth and water-soluble solute concentrations in *Sorghum bicolor* stressed with sodium and potassium salts. PhysiologiaPlantarum **62(3),** 472-480. http://dx.doi.org/10.1111/j.13993054.1984.tb04605.x

Zid E, Grignon C. 1991. Les tests de sélection précoce pour la résistance des plantes aux stress. Cas des stress salin et hydrique. L'amélioration des plantes pour l'adaptation aux milieux arides, AUPELF-UREF. Jon LibbeyEurotext, Paris, 91-108 P.