

N ° d'ordre : 42333

N ° d'ordre : 1205

L'UNIVERSITÉ DE LILLE, SCIENCES ET TECHNOLOGIES

**ECOLE DOCTORALE DES SCIENCES DE LA MATIÈRE, DE RAYONNEMENT ET DE
L'ENVIRONNEMENT**

Pour obtenir le grade de
DOCTEUR

Spécialité : **GEOSCIENCES, ECOLOGY, PALEONTOLOGY, OCEANOGRAPHY**

Par

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**RELATIONS EAU ET CULTURES DE LEGUMES : EFFET DES SOLS SALINS ET
CONDITIONS DE SECHERESSE SUR LA CROISSANCE, COMPOSITION MINERAL
ET PHOTOSYNTHESE**

**EFFECT OF SALINE SOILS AND DROUGHT CONDITIONS ON GROWTH,
MINERAL COMPOSITION, PHOTOSYNTHESIS AND WATER RELATIONS OF
VEGETABLE CROPS**

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ACKNOWLEDGEMENTS

I am deeply thankful to my supervisor Professor Baghdad Ouddane (Université de Lille, Sciences et Technologies, Équipe Physico-Chimie de l'Environnement, LASIR UMR-CNRS 8516, Villeneuve d'Ascq Cedex, France) for his knowledge and support to improve my scientific research, for his assistance with all the documents for the Doctoral School in University Lille 1 (Co-Tutorial, inscription and others), the technical maintenance and assistance in providing several analyses in the LASIR laboratory, and finally the successful completion of my PhD thesis defense under his supervision.

I would like to express thanks to my first supervisor Professor Mukhamadkhon Khamidov, Doctor of Agricultural Science, Rector of Tashkent Institute of Irrigation and Melioration, Tashkent, Uzbekistan for teaching me when I was a student in his faculty, for advising me to expand my scientific career as a head of department, and for supporting me in finishing my PhD thesis at a European university under his co-supervision.

My special thanks to my first supervisor in Europe, Professor Jose Beltrao, CIEO (Research Centre for Spatial and Organizational Dynamics), University of Algarve, Portugal for his support in problem-solving, and his guidance and scientific advices as my supervisor during my study in Algarve (2009-2010). I am grateful for his critical comments that very much improved this work and his great contribution to finish the thesis. We have published five scientific articles in journals and conferences with co-authors related to my work.

I am also thankful to my second supervisor, Professor Maria Alcinda Neves (Department of Agronomy, Sciences and Technology, University of Algarve) for her useful advices, for organizing several experiments in the greenhouse with me, for helping to prolong my stay in Algarve to finish the experiment, and for her constant encouragement and eagerness during all stages of this work (2010 - 2011).

I am particularly appreciative to the Head of Department, Doctor Ilkhom Begmatov, and other professors of the department Agricultural Hydro Technical Melioration (AHTM), and the Vice-rector for Research, Professor Tohirjon Sultanov, Tashkent Institute of Irrigation and Melioration, Tashkent, Uzbekistan for advice and support during the studying period.

Additional gratitude to the University of Algarve. Professor Isabel Barrote for her assistance in analyzing biochemical parameters (pigments, chlorophyll and soluble sugar). Moreover, I am

extremely grateful to Camilo Portela (Laboratory Assistant) for his laboratory assistance in the Faculty of Engineering and Natural Resources (FENR), University of Algarve. I gratefully acknowledge the advices of Professor Carlos Guerrero, his assistance in helping me prepare to using SPSS Software for statistical analysis and Professor Clara Costa for her help on plant chemical analysis and soil chemical parameters for these experimental studies.

Thanks to Mrs. Catherine Ouddane for her assistance to providing analysis and Mrs. Nathalie Faure for helpful consultations during the experiment in the greenhouse at the University Lille 1, France.

I want to thanks with respect Professor Igor Khmelinskii (coordinator of ERASMUS MUNDUS ECW Lot 8 at the University of Algarve) and Doctor Mojca Maher Pirc (coordinator ERASMUS MUNDUS Euro - Asian CEA, University of Ljubljana, Faculty of Economics) for their support in the European Joint Doctorate Program to my PhD thesis.

I would like to give special thanks to my mother Jumagul Bekmirzaeva and to my wife Shakhzada Ibragimova, for her constant rearing to our children and support of my research during the long period.

Finally, I would like to thank everybody else who contributed in one way or another to the completion of this study.

Thanks
To all of you ...

ABSTRACT

Salinity is a global problem that retards plant growth and slows down productivity worldwide. Progressive salinization of soils and water is one of the most serious problems affecting today's agriculture and economy in many countries. The world scientists have conducted many experiments to solve salinization problems of agricultural areas and they have estimated that every minute the world is losing, on average, 10 ha of cultivable land, three of which are due to salinization and this is equivalent to a loss of about 1 500 000 ha per year. The world's food production will need to increase by up to 70% by 2050 to match the predicted population growth (Panta et al. 2014). To solve the above problems used of conventional techniques to control soil salinization process - soil leaching or fertilization enhancing - contribute highly to soil and aquifers contamination; on the other hand, the use of salt tolerant species will be very useful to the plants, but does not solve the problem of soil or groundwater contamination. Hence, the only way to control the salinization process to maintain the sustainability of landscapes and agricultural fields is to combat the salinization problems with environmentally safe and clean techniques. One of these new techniques is the use of salt removing species. In order to study the potential capacity to remove soil salts, two agronomic species *Portulaca oleracea* and *Tetragonia tetragonioides*, and the salt sensitive crop lettuce (*Lactuca sativa* L) were evaluated for their efficiency to remove salts from sandy soil. Plants were analysed relatively to total growth and mineral composition of the leaves and soil. According to the results, it was seen that *T. tetragonioides* is better as a salt removing species and complementary, it has other benefits, which are as follows: high biomass production; several harvests; high content of minerals; horticultural importance, as a leaf vegetable crop; easy multiplication; easy crop management; tolerance to drought conditions; *P. oleracea* is a high drought tolerant species, followed by *T. tetragonioides*. The purpose of planting salt sensitive crop lettuce in the pots (were grown salt removing crops on the high salinity condition) to identify which salt tolerant species were usefull as a salt removing species. On the other hand, lettuce is an irrigated crop, and therefore, is not adequate to clean the soil. As concluding remarks, it was shown that this new technique to control salinity is a powerful and environmental clean tool to maintain the sustainability of the landscape and of the irrigated areas.

Key words: salt tolerant species; salt sensitive crop; soil and water salinity; salinity and drought stress; control of salinity and soil erosion; leaf and soil mineral compositions.

RÉSUMÉ

La salinité est un problème mondial qui retarde la croissance des plantes et ralentit la productivité à cause de la salinisation progressive des sols et de l'eau, c'est l'un des problèmes les plus graves qui touchent l'agriculture d'aujourd'hui et de l'économie de nombreux pays. Les scientifiques ont mené de nombreuses expériences pour résoudre le problème de salinisation des zones agricoles et ils ont estimé une perte moyenne de 10 ha de terres cultivables par minute, le tiers est dus aux problèmes de salinisation et cela équivaut à une perte de plus de 1 500 000 ha par an.

Les techniques classiques utilisées pour contrôler les processus de salinisation des sols, comme le lessivage des sols ou l'amélioration de la fertilisation contribuent fortement à la contamination des sols et des aquifères; d'autre part, l'utilisation des espèces tolérantes au sel sera très utile mais ne résoudra pas le problème de la contamination des sols ou des eaux souterraines. Par conséquent, la seule façon de contrôler les processus de salinisation pour maintenir la durabilité des paysages et des champs agricoles est de lutter contre les problèmes de salinisation par des techniques plus respectueuses de l'environnement. Une de ces nouvelles techniques est l'utilisation d'espèces végétales pour enlever le sel. Afin d'étudier la capacité potentielle de certaines plantes pour éliminer les sels du sol, deux espèces *Portulaca oleracea* et *Tetragonia tetragonioides* et la laitue (*Lactuca Sativa* L) ont été étudiées pour évaluer leurs efficacités à éliminer les sels d'un sol sableux. Les plantes ont été analysées selon certains critères de croissance et de composition minérale des feuilles et du sol. D'après les résultats obtenus, on constate que la *T. tetragonioides* est le meilleur choix des espèces testés qui permet la réduction de la salinité. Il y a d'autres avantages pour l'utilisation de cette espèce comme : une production élevée de biomasse; plusieurs récoltes au cours de l'année (été et hiver); teneurs élevés en minéraux; l'importance de l'horticulture, en tant que culture de feuille végétale; multiplication facile des semences; tolérance à la sécheresse et des conditions de chaleurs; contrôle en raison de son excellente couverture du sol qui réduit les phénomènes d'érosion des sols. L'espèce *P. Oleracea* a une tolérance élevée pour la sécheresse suivie de l'espèce *T. tetragonioides*. D'autre part, la laitue est une culture irriguée et par conséquent, ne suffit pas à réduire la salinité du sol. Suite aux observations, il a été montré que cette nouvelle technique de culture permet de contrôler la salinité et peut être utilisée comme un bon outil de remédiation pour maintenir la durabilité du paysage et des zones irriguées.

Mots clés: tolérance au sel; sensibilité de culture au sel; salinité des sol; salinité de l'eau; érosion des sols; composition minérale; stress hydrique.

ABSTRAKT

Hosildor tuproqlar va suvning sho'rlanishi global muammo bo'lib, qishloq xo'jaligi ekinlarining o'sib rivojlanishi va hosildorligiga, ya'ni yer yuzidagi ko'plab mamlakatlarning qishloq xo'jaligi va iqtisodiyotiga katta zarar yetkizmoqda. Hosildor yerlarning sho'rlanish jarayonini oldini olish va uni bartaraf qilish maqsadida dunyo olimlari tomonidan ko'plab tajribalar olib borildi. Tahlillar natijasiga ko'ra, yer yuzida bir minutda 10 gektarga yaqin hosildor yerlar zararlanib, bu ko'rsatkich bir yilda qariyb 1.5 million gektarni tashkil qiladi. Dunyo oziq-ovqat ishlab chiqarish xajmini 2050 yilgacha bashorat qilinayotgan aholining o'sib borishiga mos ravishda 70% gacha ko'paytirish kerak bo'ladi (Panta va boshqalar, 2014). Yuqoridagi muammolarni hal qilish uchun an'anaviy usullarni qo'llash orqali tuproqning sho'rlanish jarayonini nazorat qilish - tuproqni sho'rini yuvish, o'g'it me'yorini oshirish - tuproq va yer osti suvlari tarkibiga katta ta'sir ko'rsatadi. Tuzga chidamli turlardan foydalanish sho'rlangan tuproqlarda juda foydali bo'lishi mumkin, lekin tuproq va yer osti suvlarining ifloslanish muammolarini to'liq bartaraf etmaydi. Binobarin, ekologik xavfsiz bo'lgan tuz o'zlashtiruvchi o'simliklarni qo'llash orqali faqatgina sho'rlanish jarayonini nazorat qilish, joyning landshaftini va qishloq xo'jaligi yerlarini barqaror saqlab qolishga erishiladi. Bunday zamonaviy va an'anaviy usullardan biri sho'rlangan yerlarda tuzga chidamli bo'lgan o'simliklardan foydalanishdir. Sho'rlangan tuproqlarni yaxshilashda tuz o'zlashtiruvchi o'simliklarning potensial imkoniyatlarini o'rganish maqsadida, tajriba uchun sho'rga chidamli ikkita agronomic turlar *Portulaca oleracea*, *Tetragonia tetragonioides* va sho'rga o'rtacha chidamli bo'lgan lettuce (*Lactuca sativa* L) tanlandi. Tajribalar davomida ularning samaradorligi tahlil qilinib, olingan natijalar *T. tetragonioides* eng yaxshi tuz o'lashtiruvchi tur ekanligini tasdiqladi va qo'shimcha quyidagi foydali tomonlari aniqlandi: yuqori biomassa to'plash; yil davomida bir necha hosil olish; minerallarga boy; sabzavot ekini sifatida ahamiyati; oson ko'payish (urug'idan ko'paytirish) va oson boshqarish; qurg'oqchilik va issiq sharoitlarga chidamli; tuproq eroziyasini oldini olish. *P. oleracea* ham qurg'oqchilik sharoitiga chidamli ekanligi natijalar orqali yana bir bora tasdiqlandi. Lettuce hosili tahlil qilinib, agronomic turlarning qaysi biri sho'rlangan tuproqlar uchun foydali ekanligi tasdiqlandi. Yakuniy xulosada tuz o'lashtiruvchi agronomik turlar - sho'rlanishni nazorat qilish va oldini olishda, joyning landshafti va sug'oriladigan yerlarning barqarorligini saqlab qolishda, tuproq eroziyasini oldini olishda eng yaxshi va ekologik toza vosita ekanligini ko'rsatdi.

Kalit so'zlar : tuz o'zlashtiruvchi turlar; tuproq va suvning sho'rlanishi; sho'rlanish va tuproq eroziyasini nazorat qilish; barg va tuproqning mineral tarkibi.

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List of symbols and abbreviations

EC _e	Electrical conductivity of soil solution at saturation point (dS m ⁻¹)
EC _w	Electrical conductivity of drainage water (dS m ⁻¹)
FAO	Food and Agriculture Organization of the United Nations
GWT	Groundwater table (dimensionless)
GVA	Good visual appearance
FW	Fresh weight
DW	Dry weight
DI	Drought index
θ _{wp}	Volumetric soil water content (m ³ m ⁻³)
θ _{fc}	Field capacity
RDI	Regulated deficit irrigation
ADB	Asian Development Bank
LR	Leaching requirement
AW	Depth of applied water (mm year ⁻¹)
ET	Total annual crop water demand (mm year ⁻¹)
Yr	Relative crop yield (%)
ICARDA	International Center for Agricultural Research in the Dry Areas
IWMI	International Water Management Institute
MAWR	Ministry of Agriculture and Water Resources
CRD	Completely randomized design

List of publications

Articles published

Bekmirzaev, G.T., Ouddane, B., Khamidov, M., Beltrao, J., 2016. Growing of halophytic species and salt-sensitive crops on saline soils. *International Journal of Agricultural Science and Research (IJASR)* 6(3): 71-82. (<https://www.academia.edu/27464914/>).

Bekmirzaev, G.T., Ramazanov, U., 2015. Effects of drought and salt conditions on the germination of salt removing species. *Journal of Irrigation and Melioration* № 01, 34-38. <http://tiim.uz/wp-content/uploads/2016/01/Irrigatsiya-va-melioratsiya-jurnali-1-sonPDF.pdf>.

Bekmirzaev G.T., Beltrao J., Neves M.A., 2012. Effects of salt removal species in lettuce rotation. *8th WSEAS International Conference on Energy, Environment, Ecosystems and sustainable development (EEESD '12)*, *WSEAS Journal*, 79-85. (<http://www.wseas.us/e-library/conferences/2012/Algarve/EEESD/EEESD-09.pdf>).

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Bekmirzaev, G.T., Khamidov, M., Beltrao, J., Neves, M.A., 2010. Salt removing crops in arid climates. 3rd *WSEAS International Conference on "Climate changes, global warming, biological problems and natural hazards"* *WSEAS Journal*, 36-41. (<http://www.wseas.us/e-library/conferences/2010/Faro/NAGB/NAGB-05.pdf>).

Articles submitted

Bekmirzaev, G.T., Ouddane B., Beltrao, J., Khamidov, M., 2017. Yield, Minerals and Photosynthetic Pigments of *Tetragonia tetragonioides* in Several Water Regimes. Submitted in the *Agronomy Journal*, ASA, 5585 Guilford Rd., Madison WI 53711.

Oral communications

Oral presentation in session of the PhD Welcome Lille Nord de France, December, 10 and 11, 2015. Mercury Hotel Marcq - on – Baroeul, Lille, France.

Salt removing vegetable crops. Under the project of BMBF "Reduction of the salinity of irrigation water through the use of macrophytes" (FKZ 01DK 12049) ZALF Munchenberg Institute Landscape Hydrology, 15 August and 01 September, 2013, Germany.

Oral presentation in the session of young scientists on the Innovation, Integration and Scientific Research Center under the Ministry of Agriculture and Water Resources (MAWR) of the Republic of Uzbekistan, 2012, Tashkent, Uzbekistan.

Salt removing crops in arid climates, 2010. *3rd WSEAS International Conference* on “Climate changes, global warming, biological problems and natural hazards” *WSEAS Conference*, 3-5 November, Faro, Portugal.

GENERAL INTRODUCTION

The total area of salt-affected soils in the world is 831 million hectares which includes 397 and 434 million hectares of saline and sodic soils, respectively (Munns and Tester, 2008). The agricultural land is decreasing constantly due to population pressure, adverse environmental conditions, continuously increasing natural calamities, and global climate change (Mahajan and Tuteja, 2005 and Munns, 2005). More than 45 million hectares of irrigated land are affected by salt which accounts for 20% of total land and 1.5 million ha of land are taken out of production each year owing to high salinity levels (Hasanuzzaman et al., 2012, Manchanda, 2008). If it continues this way, 50% of cultivable lands will be lost by the middle of the 21st century (Ashraf et al., 2005). In recent years, plants with the ability to remove salts from contaminated soils have been studied and identified by many researchers (Flowers, 2008). At the same time, halophytes can be exploited as significant and major plant species bearing the potential capability of desalination and restoration of saline soils and phytoremediation as well. Due to the rapid climate change, the saline area in the world is increasing day by day and currently there is an ample need to develop highly salt-tolerant crops to cope with the adverse situation. Halophytes are able to provide satisfactory yields under high salt conditions. An increasing world population and the development of urbanism created high competition on the limited water resources (Hamdy, 2002). In arid and semi-arid areas, such as the Mediterranean regions, supplies of good quality water allocated to agriculture are expected to decrease because most of available fresh/potable water resources have been already mobilized (Costa et al., 2011). According to FAO (2011), due to the shortage of water, there is an enlargement of saline land in agricultural areas in some of the developing countries. As a result, yields are decreasing, provoking an increasing cost of the agricultural products (Pretty et al., 2003; Rivera-Ferre, 2009; Seck et al., 2010; Bekmirzaev et al., 2011).

Agricultural losses caused by salinity are difficult to assess but estimated to be substantial and expected to increase with time. Secondary salinization of agricultural lands is particularly widespread in arid and semiarid environments where crop production requires irrigation chemes. At least 20% of all irrigated lands are salt-affected, with some estimates being as high as 50% (Pitman and Lauchil, 2002). Improved crop salt tolerance can contribute to the remediation of soil salt problems because it extends the choice of crops that can grow at each soil salinity level; allows irrigation using more saline water; increases soil organic matter and improves soil structure (Eynard et al., 2013). Water deficit and salinity are the major limiting factors in plant productivity, affecting more than 10 percent of arable land on our planet and

resulting in a yield reduction, on average of more than 50% for most major crop plants (Bartels and Sunkar, 2005). Crops grown in arid and semi-arid regions are often exposed to adverse environmental factors such as drought or high soil salinity (Tuna et al., 2008). One of the major effects of salt stress in plants is induced nutritional disorders. These disorders may result from the effect of salinity on nutrient availability, competitive uptake and transport or partitioning within the plant (Munns and Tester, 2008). Salinity stress is a key factor that limits crop production worldwide 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 (Hamidov et al., 2007). Recently, a new environmentally clean and safe technique known as phytoremediation has been introduced to address the salinity problem. This includes the introduction of salt (ion) removing species to control salinity and to maintain the sustainability of agricultural fields (Kilic et al., 2008 and Shaaban and El-Fouly, 2002). Generally, the growth of glycophytes decreases with increasing of salinity while halophytes improve. In the present study, the growth of New Zealand spinach increased under salt stress, agreeing with previous data reported on the halophytes *Salicornia europaea* and *Suaeda maritima* (Moghaieb et al., 2004). Salinity is one of the rising problems causing tremendous yield losses in many regions of the world, especially in arid and semiarid regions. The use of salt-tolerant crops does not remove the salt and hence halophytes that have the capacity to accumulate and exclude the salt can be an effective way (Hasanuzzaman, 2014). Salinity and drought do not only affect agriculture, but they also decry the economic situation of most developing as well as developed countries. Salt-spoiled soils worldwide consist of 20% of all irrigated lands, an area identical to France; comprehensive costs include \$27 billion in lost crop value/year. Salinity and drought stress show a high degree of similarity with respect to physiological, biochemical, molecular and genetic effects (Sairam and Tyagi, 2004). Many researches were carried out on saline soil or water with a high concentration of NaCl and many crops were tested: vegetable crops (Prasad and Chakravorty, 2015; Maksimovich and Ilin, 2012;), tomato (De Pascale et al., 2003), sunflower (Flagella et al., 2004), snap bean (Mori et al., 2012) and potato (Patel et al., 2001). High concentrations of soluble salts in the soil moisture of the root zone are always associated with saline soils. All plants are subject to the influence of high osmotic pressure of soluble salts, but sensitivity to high osmotic pressure varies widely among plant species and it puts various problems to the plants either at the population, organism or even at the molecular level. Physiologically and genetically salt tolerance is complex among the variety of plants with a wide range of adaptations in halophytes and less tolerant plants (Flowers, 2004). The ability of vegetation to survive under higher salinity conditions is

important for the distribution of plants and agriculture around the world. Enhancing the salt tolerance of plants is an important breeding objective in areas which are affected by soil salinity (Flowers and Flowers, 2005). A plant's ability to acclimate to salt stress includes alterations at the leaf level, associated with morphological, physiological and biochemical characteristics whereby many plants adjust to high salinity and the consequent low soil water availability (Ashraf, 2004). Salt stress also induces a decrease in stomatal conductance and transpiration. Under saline conditions, stomatal closure helps to maintain higher leaf water content; however, this leads to a decrease in the leaf CO₂ assimilation rate (Parida et al., 2004).

Salt treatments induced reduction in germination. Growth and chlorophyll contents can be improved by exogenous application of proline (Khalid Nawaz, 2010). Soil salinity is one of the limiting factors for crops grown in arid and semi-arid regions. Salinity is one of the major environmental factors that leads to a deterioration of agricultural land and reduction in crop productivity worldwide (Chinnusamy et al., 2005). Salinity is one of the major abiotic stresses that adversely affect crop productivity and quality (Ouda, 2008) with increasing socio-economic and health impacts, especially in the farming communities. More than half of the 2.32 million hectares of irrigated land in Uzbekistan is salt affected and built-up salinity is seriously threatening agricultural productivity (ICARDA, 2002). Around 202.3 ha of farmland in arid and semi-arid regions are lost every day to damage caused by salt, according to Qadir et al. (2014). This problem has already affected one-fifth of irrigated land in the world and will continue to worsen without new land and water management practices. Salinization the accumulation of salt in soil occurs in areas with little rainfall and poor drainage. Normal salt levels in soil are between 0-175 mg of salt per liter. When levels reach 3.5 mg/l, crop production plummets. Purslane has been rated as salt tolerant with a threshold value, given in terms of the electrical conductivity of saturated-soil extract, EC_e of 6.3 dS m⁻¹ and yield was reduced by 50% when EC_e reached 11.5 dS m⁻¹ (Kumamoto et al., 1990).

Growth retardation and fresh and dry weight loss of roots and shoots under salinity stress were revealed in previous studies (Lolaei 2012; Navarro et al., 2008; Villarino and Mattson, 2011). In addition, based on the fresh or dry weight, it was demonstrated in several studies that the root/shoot ratios of many plants increase under salinity stress (Maggio et al., 2007). Differential response of plants to salt and alkali stresses are largely due to high-pH associated stress (Munns, 2002; Li et al., 2012). Under general NaCl toxic conditions, plants absorb a higher amount of Na, which thus decreases the K/Na ratio (Ahmad and Prasad, 2012). Salinity stress involves changes in various physiological and metabolic processes, varies with stress severity and its duration and ultimately inhibits crop production (Munns, 2005; Rozema and

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Flowers, 2008). The effect of salinity stress arises as a result of the combination of the relationship between the morphological, physiological, and biochemical processes of plants (Parida and Das 2005). Salinity leads to significant changes in water potential, ion uptake, ion balance, ion toxicity and oxidative stress (Parida and Das 2005). Sodium and chloride can influence the uptake of nutrients by competing with nutrients or affecting the ion permeability of the membrane. In most plants, an increase in NaCl concentration in the plant leads to an increase in sodium (Na^+) and chloride (Cl^-) ions but may result in a decrease in N, P, K^+ , and Ca^{2+} (Karimi et al., 2005; Tuna et al., 2007; Navarro et al., 2008). One of the most damaging effects is accumulation of sodium and chloride ions in tissues of plants exposed to soils with high NaCl concentrations. Higher sodium blocks potassium uptake, results in lower productivity and may even lead to cell death (James et al., 2011). Plant adapting and tolerance to salinity stress includes complex physiological features, metabolic pathways and molecular or gene networks. Comprehensive knowledge of how plants respond to salinity stress at various levels and an integrated approach combining molecular, physiological, and biochemical techniques are imperative for developing salt-tolerant diversities in salt-affected areas (Ashraf, 2014).

This problem is intensified in coastal areas, due to sea water intrusion which results from the reduction of ground water levels as water demand exceeds the annual groundwater recharge (Ben Asher et al., 2002). As reported above, some of the emerging regions in risk of increasing levels of salinization of their soils are located in the Mediterranean Basin (Nedjimi et al., 2006, Beltrao et al., 2014), Australia (FAO, 2008), Central Asia (Hamidov et al., 2013), the Middle East (Ben Asher et al., 2002), and Northern Africa (Yensen, 2008). The saline water that may be used in halophyte crops irrigation can be, for example, seawater, salt contaminated phreatic sheets, brackish water, wastewater, or drainage water from other irrigated plantations (Ben Asher et al., 2012b). Water deficit and salinity are, therefore, the major limiting factors in plant productivity, affecting more than 10 % of arable land on our planet, resulting in a yield reduction of more than 50% for most major crop plants (Bartels and Sunkar, 2005).

Drought stress is considered to be a moderate loss of water, which leads to stomatal closure and limitation of gas exchange and induces a range of physiological and biochemical responses in plants. Plants respond and adapt to water deficit at both the cellular and molecular levels; for instance, by the accumulation of osmolytes and proteins specifically involved in stress tolerance (Shinozaki and Yamaguchi-Shinozaki, 2007). Drought has a major impact on plant growth and development, limiting crop production. The abiotic stresses usually noted that include a component of cellular water deficit, are salinity and low temperature stresses that can also

severely limit crop production (Ansari and Lin, 2010). Abiotic stresses, such as drought, salinity, extreme temperatures, chemical toxicity and oxidative stress are serious threats to agriculture and result in the deterioration of the environment. The abiotic stresses of drought, salinity and freezing are linked by the fact that they all decrease the availability of water to plant cells (Verslues et al., 2006). Abiotic stress is the primary cause of crop loss worldwide, reducing average yields for most major crop plants by more than 50% (Bray et al., 2000). Hence, our aims were to choose a salt removing crop, tolerant to salinity, while at the same time interesting as a food crop, and to test its drought tolerance through its response to several water regimes.

Tetragonia (*Tetragonia tetragonioides*) was the selected crop. It is known as New Zealand spinach, annual grown for its edible leaves like spinach; successful in autumn, winter and spring in a Mediterranean climate. For early crops, one should sow the seeds in boxes or pots and maintain a temperature of 15-16 °C, being transplant at 45-60 cm apart in rows 90-100 cm apart; for direct seeding sow in 6 mm deep drills and space the drills 90 cm apart; soak seed overnight before sowing; thin the seedlings 45-60 cm apart; pinch out the flower buds; known as trouble free; pick the leaves in bunches for marketing (Ben Asher et al., 2012a). New Zealand-spinach is propagated from seed, which can be purchased from garden stores, and planted any time of the year in a sunny or partially shaded spot (Steples and Herbst, 2005). Recently, a new environmentally safe and clean technique known as phytoremediation has been introduced to address the salinity problem. This includes the introduction of salt (ion) removing species to control salinity and to maintain the sustainability of agricultural fields (Cuartero and Shaaban, 2002). The increase of Na⁺ accumulation in New Zealand spinach was associated with reduced K⁺, Mg²⁺ and Ca²⁺, indicating a restriction in the uptake of these nutrients, as noted in other halophytic plants (Debez et al., 2004). In order to evaluate the wild plants' ability to remove salts from soil, several species from the Mediterranean coastal flora, living in saline environments were studied (Panagopoulos, 2006). New Zealand spinach (*Tetragonia tetragonioides*) is a member of Tetragoniaceae and is distributed widely from tropical and subtropical to temperate areas (Matraszek, 2008).

Purslane (*Portulaca oleracea* L. aggregate; Portulacaceae), known as a summer weed in the Mediterranean region, has long been consumed as a vegetable and used as a medicinal plant. In the last decade, considerable research has focused on its biological activity (Rashed, 2003). *Portulaca oleracea* is a cosmopolitan succulent species that includes more than 15 microspecies, which differ in several morphological characteristics and in their ploidy levels (Danin, 2008). Purslane (*Portulaca oleracea* L.), a common plant found throughout the United States, is a vigorous colonizer of disturbed, waste habitats from the yield response model of

Maas and Hoffman (1977). Soil salinity causes great losses to agriculture each year by lowering the yield of crops through the creation of conditions unfavorable for plant growth and development. Soil salinity is a major abiotic constraint affecting crop yield, much research has been conducted to develop plants with improved salinity tolerance. Salinity stress impacts many aspects of a plant's physiology (Negrao et al., 2017). As a significant source of omega-3 oils, *P. oleracea* could yield considerable health benefits to vegetarian and other diets where the consumption of fish oils is excluded. Scientific analysis of its chemical components has shown that this common weed has uncommon nutritional value, making it one of the potentially important foods for the future (Kaymak, 2013; Uddin et al., 2014).

Lettuce is the most important and useful horticultural crop in the world. Lettuce is considered to be a moderately salt-sensitive crop, with a threshold electrical conductivity of 1.3 dS m^{-1} , but a wide range of salt tolerance can be identified among its different cultivars: e.g. the „Romaine“ or „Cos“ lettuce cultivars (*Lactuca sativa* var. *longifolia*) are considered far more tolerant to salinity than the „Iceberg“ cultivars (*Lactuca sativa* var. *capitata*) (Eraslan et al., 2007; Kohler et al., 2009; Oh et al., 2010). Salinity levels of more than 2.0 and 2.6 dS m^{-1} reduce lettuce fresh yield and plant growth, respectively. It has been reported that lettuce has a threshold value of 1.1 dS m^{-1} and the relative yield decreases in slope after this threshold as 9.3% (Unlukara et al., 2008). Salts in the external medium are supposed to adversely affect growth and the metabolism of glycophytic higher plants (Younis et al., 2008). Effects of electrical conductivity of the nutrient solution on plant growth and yield have been previously demonstrated (Andriola et al., 2005). The obtained results demonstrated that the high NaCl concentrations ($> 60 \text{ mol m}^{-3}$) in nutrient solution strongly affected the germination rate and root elongation, seedling and mature vegetative growth of both spinach and lettuce, but especially in lettuce (Kaya et al., 2002; Khaydarova, 2006). Crop rotation, if properly designed is an efficient tool to keep soils more productive. Introducing species with a market value and high salt removal capacity into rotation programs is an environmentally friendly tool to evaluate the salt removal ability of the plants (Kilic et al., 2008). For commercial use an available form of lettuce is available in some supermarkets and markets as a fresh green vegetable (Vincent, 2009). The seeds are widely available for propagation from seed dealers and nurseries.

However, use of halophytes for soil reclamation is still in an exploratory stage and only a few field studies for bio-reclamation of saline soil using halophytes have been carried out so far and therefore, more research is needed to study the utilization of halophytes to remove excess salinity added by irrigation.

CHAPTER I: LITERATURE REVIEW

I.1 Salinity

More than 800 million hectares of land throughout the world are salt affected (FAO, 2008). This amount accounts for more than 6% of the world's total land area. Most of this salt-affected land has arisen from natural causes, from the accumulation of salts over long periods of time in arid and semiarid zones (Rengasamy, 2002). Apart from natural salinity, a significant proportion of recently cultivated agricultural land has become saline owing to land clearing or irrigation, both of which cause water tables to rise and concentrate the salts in the root zone. Of the 1500 million ha of land farmed by dryland agriculture, 32 million ha (2%) are affected by secondary salinity to varying degrees. Of the current 230 million ha of irrigated land, 45 million ha (20%) are salt affected. Salinity stress is a limitation to the productivity of agricultural crops worldwide. It has been estimated that almost 80 million hectares of arable lands worldwide is currently affected by salinity (FAO, 2008).

I.1.1 Soil salinity

Soil salinity is a major environmental constraint to crop production, affecting an estimated 45 million hectares of irrigated land, and is expected to increase due to global climate changes and as a consequence of many irrigation practices (Munns and Tester, 2008; Rengasamy, 2010). Salinity on about 80% of these lands is of natural origin (primary salinization), whereas on the remaining 20% it is a result of secondary soil salinization under conditions of poor drainage in irrigation. Between 9 and 25% of irrigated areas in Tunisia, the United States, India, China and South Africa are salt-affected (CISEAU, 2006).

Table I.1. Soil salinity classifications according to the FAO (<http://www.fao.org/soils-portal/soil-survey/soil-classification/en/>)

Salinity level	ECe (dS m ⁻¹)
Non saline	0 - 2
Low saline	2 - 4
Moderately saline	4 - 8
High salinity	8 - 16
Severely salinity	≥ 16

I.1.2 Water salinity

I.1.2.1 Irrigation water quality

Water for irrigation is a major limitation to agricultural production in many parts of the world. Use of waters with elevated levels of salinity is one likely option to meet the supply of increased demands. The sources of these waters include drainage water generated by irrigated agriculture, municipal wastewater, and poor quality groundwater (Letey et al., 2011). Salinity is a major abiotic stress limiting growth and productivity of plants in many areas of the world due to increasing use of poor quality of water for irrigation and soil salinization (Gupta, 2014). The scientists in the World has predicted water scarcity and the impact of climate change will necessitate the use of alternate available water resources in agriculture, such as saline water, to narrow the gap between demand and supply of freshwater. Bezborodov et al. (2010) suggested possible saline water, in combination with freshwater or alone used to irrigate cotton in Central Asia in summer when there are often severe freshwater shortages. But to use of saline water without appropriate management can result in the accumulation of salts in the root zone with associated negative impacts on crop productivity. Water used for irrigation can vary greatly in quality depending upon the type and quantity of dissolved salts. Salts are present in irrigation water in relatively small but significant amounts (Ayers and Westcot, 1994). Soil scientists use the following categories to describe irrigation water effects on crop production and soil quality: salinity hazard - total soluble salt content; sodium hazard - relative proportion of sodium to calcium and magnesium ions; pH - acid or basic; alkalinity - carbonate and bicarbonate; specific ions: chloride, sulfate, boron, and nitrate. Another potential irrigation water quality impairment that may affect suitability for cropping systems is microbial pathogens (Bauder et al., 2011).

The most common parameters used for determining the irrigation water quality, in relation with its salinity, are EC_w (Table I.2).

Table I.2. Salinity hazard (irrigation water quality) (Bauder et al., 2011)

Limitation for use	EC_w (dS m ⁻¹)*	Salinity hazard
None	≤ 0,75	Low
Some	0.76 – 1.5	Medium
Moderate ¹	1.51 – 3.00	High
Severe ²	≥ 3.00	Very high

*dS m⁻¹ at 25° C = mmhos/cm; ¹ leaching required at higher range; ² good drainage needed and sensitive plants may have difficulty at germination.

I.2 Saline soils in the World

Soil salinity is one of the most important problems affecting many areas of the world. Saline soils present in agricultural areas reduce the annual yields of most crops. The world is losing 2.000 ha of farm soil daily to salt-induced degradation. Salt-spoiled soils worldwide 20% of all irrigated lands-an area equal to France; extensive costs include \$27 billion in lost crop value/year (Qadir et al., 2014).

Well known salt-degraded land areas include:

- Aral Sea Basin, Central Asia
- Indo-Gangetic Basin, India
- Indus Basin, Pakistan
- Yellow River Basin, China
- Euphrates Basin, Syria and Iraq
- Murray-Darling Basin, Australia
- San Joaquin Valley, United States

Canada, Jordan, Pakistan and Sri Lanka, deal with crop productivity losses at farm, regional, and global scales, the cost of doing nothing, and the net economic costs of preventing and/or reversing land degradation (Qadir et al., 2014). The beginning of the 21st century is marked by global deficiency of water resources (especially, for drinking and irrigation), environmental pollution and increased salinization of soil and water. Saline soil is a huge problem for agriculture under irrigation. In the arid and semi-arid regions of the world, the soils are often saline with low agricultural potential. The increasing human population and reduction in available land for cultivation are two threats for agricultural sustainability (Shahbaz and Ashraf, 2013).

I.2.1 Salinity problems of Central Asia

Land degradation due to water logging and its influence on secondary soil salinization processes pose a major threat to the sustainability of irrigated agriculture in the semi-arid production ecologies of Central Asia (Devkota et al., 2015). Salt-induced land and water resources degradation in Central Asia are the consequence of both naturally occurring phenomena (primary salinity) and anthropogenic activities causing secondary salinity. Recent estimates suggest that more than 50% of irrigated soil are salt-affected and/or waterlogged in Central Asia (Qadir et al., 2009). The region has a total area of 3 994 400 km² (FAO, 2008a) and a population of over 55 million. The climate in the region is classified as arid or semi-arid, with mean annual precipitation varying between 600–800 mm in the mountainous zones to 80–150 mm in the

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desert regions that are predominantly distributed over the winter and spring. Nearly half of the region's irrigated lands are affected by varying levels of salinity (Kijne, 2005). Recent estimates reveal that more than 50% of irrigated soils in Central Asia are salt-affected and/or waterlogged (Dukhovny, 2005). Irrigated lands of the Republic of Uzbekistan are mostly prone to salinization. This is interlinked to the aridity of climate, geological and hydrogeological conditions of the irrigated territories. Salinization is one of the factors which reduce soil fertility and productivity of irrigated lands significantly. Depending on the degree of soil salinity, crop losses could be 15 to 80%. According to the land reclamation monitoring service of the MAWR, saline soil represents more than 50% of the total irrigated land (4.4 mln. ha), including 32% of slightly saline, 15% of medium saline and 3.5% of saline land (Shirokova and Morozov, 2014).

I.3 Crop response to salinity

I.3.1 Soil salinity effect on crop growth

Plant species differ greatly in their growth response to salinity. It is beneficial to study the physiological reactions of such halotolerant plants (mechanisms of salt tolerance) to saline conditions to characterize salinity responses (Koyro, 2006). Plants exposed to salt stress undergo changes in their environment. The ability of plants to tolerate salt is determined by multiple biochemical pathways that facilitate retention and/or acquisition of water, protect chloroplast functions, and maintain ion homeostasis (Parida and Das, 2005). Agricultural productivity is severely affected by soil salinity because salt levels that are harmful to plant growth affect large terrestrial areas of the world. The damaging effects of salt accumulation in agricultural soils have influenced ancient and modern civilizations (Yamaguchi and Blumward, 2005). Flowers (2004) suggested possible ways, which were appropriate at that time, to cultivate salt-tolerant crops: develop halophytes as alternative crops; use interspecific hybridization to raise the tolerance of current crops; use the variation already present in existing crops; generate variation within existing crops by using recurrent selection, mutagenesis or tissue culture, and breed for yield rather than tolerance. These all remain possible solutions to the problem. Although conventional forms of mutagenesis have not, in general, delivered salt-tolerant genotypes (Tester and Davenport, 2003).

I.3.2 Salts affect crop yield

Much research has been done to determine crop response to salinity by measuring crop yields at increasing salinity and relating yield reduction directly to soil salinity. This method permits to distinguish salt tolerant and salt sensitive crops and to choose a cropping pattern

corresponding with the expected soil salinity. The method is simple and practical, but it does not, however, explain the behavior of crops under saline conditions, nor why crops differ in salt tolerance (Katerji et al., 2003). Accumulation of excessive salt in irrigated soils can reduce crop yields, reduce the effectiveness of irrigation, ruin soil structure, and affect other soil properties (Horneck et al., 2007). Maas et al. (1986) exposed two sorghum cultivars to salinity and determined the reduction in dry matter and grain yield. Rehabilitation of abandoned salt-affected soils through crop-based interventions is an attractive low-cost opportunity that can be undertaken by farmers (Qadir and Oster, 2004; Qadir et al., 2007). The introduction of halophyte species licorice (*Glycyrrhiza glabra* L.) as a bioremediation crop for the reclamation of saline soils and subsequent restoration of irrigated cropping systems has been studied by Kushiev et al. (2005) on abandoned land in the Hungry Steppes of Uzbekistan. The average yields of wheat and cotton for the Hungary Steppes of Central Asia are 1.75 and 1.5 t ha⁻¹, respectively.

I.3.3 Salts affect mineral nutrients

I.3.3.1 Nutrient imbalance

The high frequency of dry seasons in many regions of the world and the problems associated with salinity in irrigated areas often result in the consecutive occurrence of drought and salinity on cultivated land. Nutrient disturbances under drought and also salinity reduce plant growth by affecting the availability, transport, and partitioning of nutrients. However, drought and salinity can differentially affect the mineral nutrition of plants. Salinity may cause nutrient deficiencies or imbalances, due to the competition of Na⁺ and Cl⁻ with nutrients such as K⁺, Ca²⁺ and NO₃⁻ (Hu and Schmidhalter, 2005). Under salinity stress, changes in the nutritional balance of NaCl result in higher levels of Na⁺/Ca²⁺, Na⁺/K⁺, Na⁺/Mg²⁺, Cl⁻/NO₃⁻ and Cl⁻/H₂PO₄⁻, thus causing plant growth retardation (Grattan and Grieve 1999). Plants take in nutrients through the root system. Ion regulation is important under normal conditions and is also vital under saline conditions for plant growth (Aşık et al., 2009). The problem of low productivity of saline soils may be ascribed not only to their salt toxicity or damage caused by excess amounts of soluble salts but also arising from the lack of organic matter and available mineral nutrients especially N, P, and K⁺ (Lakhdar et al., 2009). Potassium homeostasis is disrupted, possibly due to the ability of Na⁺ to compete for K⁺ binding sites. A high ratio of Na⁺ to K⁺ and high concentrations of all ions inactivate enzymes and inhibit protein (Bartels and Sunkar, 2005).

I.3.4 Salt tolerant crop

Salinity tolerance is too complex to be easily amenable to improvement through selection as a trait in itself, but traits that are hypothesized to contribute to salinity tolerance are more genetically tractable and genes underlying these can be discovered using molecular genetics tools and genomics. Alterations in crops can then be made using both marker-assisted selection and genetic modification, and the relevance of such traits on whole plant salinity tolerance can then be tested, as measured by yield maintenance in saline conditions (Roy et al, 2014). Soil salinity is a major environmental constraint to crop productivity worldwide. The “biological” approach to this problem focuses on the management, exploitation, or development of plants able to thrive on salt-affected soils (Ashraf et al., 2008). Slow growth in low-salt medium is a general feature of halophytic species and a potential problem of transgenic salt-tolerant crops in an agricultural setting. Salt tolerance may require more selective ion acquisition and transport; this selectivity, recognition and exclusion of toxic elements might cost energy and require altered sink–source relations (Volkov et al., 2004).

I.4 Techniques to control salinization

I.4.1 Conventional techniques

Conventional techniques to combat the salinization process can be characterized by four generations: 1) Problem of root zone salinization by soil leaching - two options can occur - when there is an impermeable layer, salts will be concentrated above this layer; on the other hand, when there is no impermeable layer, aquifers contamination can be observed (Ben Mechlia, 2002; Schubert et al., 2009); 2) Use of subsurface trickle irrigation - economy of water, and therefore less additional salts; however the problem of groundwater contamination due to natural rain or artificial leaching remains (Malash et al., 2008); 3) Enhanced fertilization increases tolerance to salinity, but sensitivity increases as well (Beltrao et al., 2002; Silberbush and Ben Asher, 2002; Munns and James, 2003; Pathania, 2015), while the contamination will be increased by other hazardous chemicals such as nitrate (Beltrao and Ben Asher, 1997); 4) Use of salt tolerant species - this technique will be very useful to the plants, but it does not solve the problem of soil or groundwater contamination (Hamdy, 2000; Geissler et al., 2009). When using saline water, the only way to control the salinization process and to maintain the sustainability of landscape and agricultural fields is to combat the salinization problems by environmentally safe and clean techniques, as follows: use of salt (ions) removing species (Cuartero et al., 2002; Shaaban and El Fouly, 2002; Neves et al., 2005; Corwin et al., 2007;

Beltrao et al., 2009); use of drought tolerant crops species, because less water is applied and, therefore, less salts are infiltrated (Glenn et al., 1999); reduction of salt application by deficit irrigation (Nasr and Ben Mechlia, 2002; application of minimal levels of water enough to obtain a good visual appearance GVA of the landscape (Beltrao et al., 1999).

I.4.2 Environmentally useful techniques

I.4.2.1 Salt removing species

Phytoremediation – salt removing species have become the best technique to remediate saline soils and decontaminate the environment (Anac et al., 2005). Most of the literature and research studies conducted by different scientists have shown that phytoremediation is the most economical and social approach through which salt affected wasteland can be successfully utilized for crop production. Toxic ions like Na^+ and Cl^- are removed by the salt tolerant species used for phytoremediation. Qadir and Oster (2004) conducted 14 experiments to compare the remediation of salt-affected soils by chemicals and through vegetation and reported that soil amendment with gypsum reduced 62% of sodicity levels while it was 52% by phytoremediation. Shekhawat et al. (2006) conducted experiments with salt tolerant crops viz. *Salsola baryosma*, *Haloxylon recurvum* and *Suaeda nudiflora*. *Haloxylon recurvum* removed the highest Na^+ . Akhter et al. (2003) reported that cultivation of Kallar grass on salt-affected soils significantly reduces the soil Na^+ content. The reduction in the soil Na^+ is 71% after five - year cultivation with the species when compared with uncultivated control areas. Many salt tolerant species have also been recommended by Geieve and Suarez (1997) *Portulaca oleracea*, Glenn et al. (1996) *Salicornia bigelovii*, and Kushiev et al. (2005) *Glycyrrhiza globra*. However, the main concern of their research were ontogenetic aspects of the wild species. In general, this new technique to mitigate salinity is a powerful and environmentally clean tool to maintain the sustainability of the agricultural areas and landscapes (Neves et al., 2005).

I.4.2.2 Drought tolerant species

Drought is the most important environmental stress affecting agriculture worldwide (Cattivelli et al, 2008). Exploiting yield potential and maintaining yield stability of crops in water-limited environments are urgent tasks that must be undertaken in order to guarantee food supply for the increasing world population (Yang et al., 2010). Cattivelli et al. (2008) suggested that drought is the most significant environmental stress in agriculture worldwide and improving yield under drought is a major goal of plant breeding. A review of breeding progress pointed out that selection for high yield in stress-free conditions has, to a certain extent, indirectly improved

yield in many water-limiting conditions. Climate change is predicted to increase the incidence and severity of droughts in ecosystems worldwide (Sheffield and Wood 2008). Pooter and Markesteijn (2008) had studied the 62 species which were ranked along independent axes of drought- and shade-tolerance. They used a drought index (DI) as an ecological indicator of a species drought-tolerance. Drought and high salinity are common stress conditions that adversely affect plant growth and crop production (Xiong et al., 2002).

I.4.3 Using remote sensing techniques

Remote sensing data and techniques have been progressively applied to monitor and map soil salinity since 1960s when black-and-white and color aerial photographs are used to delineate salt-affected soils. Advantages of using remote sensing technology include saving time, wide coverage (satellite remote sensing provides the only source when data is required over large areas or regions), are faster than ground methods, and facilitate long term monitoring (Allbed and Kumar, 2013). Remote sensing has been widely used to detect and map salt-affected areas, since thousands of medium to high-resolution imageries from the earth surface are available (Farifteh et al., 2006). Remote sensing data have been increasingly used in soil-salinity studies as they are not only quicker but are also useful for making realistic predictions (Shrestha, 2006). Soil salinity can be identified from remote sensing data obtained by different sensors by way of direct indicators that refer to salt features that are visible at the soil surface as well as indirect indicators such as the presence of halophytic plants and assessing the performance level of salt-tolerant crops (Allbed and Kumar, 2013). Soil salinity can be detected directly from remotely sensed data through salt features that are visible at the soil surface, such as bare soil with white salt crusts on the surface (Matinfar et al., 2013) or indirectly from indicators such as the presence of halophytic plants and the performance level of salt-tolerant crops (Aldakheel, 2011). Remote sensing has been widely used to detect and map salt-affected areas, since thousands of medium to high-resolution imageries from the earth surface are available (Farifteh et al., 2006).

CHAPTER II: TESTING THE EFFECTS OF SALT AND DROUGHT ON VEGETABLE CROPS

II.1 Effect of salts on growth, biomass production (yield) and mineral composition of salt removing species *Tetragonia tetragonioides* and *Portulaca oleracea*

Abstract: Conventional techniques used to control the soil salinization process - soil leaching or enhanced fertilization - contribute highly to soil and aquifers contamination; on the other hand, the use of salt tolerant plant species will be very useful to the plants, but it does not solve the problem of soil or groundwater contamination. Hence, the only way to control the salinization process and to maintain the sustainability of landscapes and agricultural fields is to combat the salinization problems by environmentally safe and clean techniques. One of these techniques is the use of salt removing species. In order to study the effects on the potential capacity to remove soil salts, two horticultural leaf species (*Tetragonia tetragonioides* and *Portulaca oleracea*) were planted. The total growth and the leaf's mineral composition of these species were studied. According to the results of plant growth and leaf analysis, it was seen that *Tetragonia tetragonioides* is the best salt removing species; on the other hand, *Portulaca oleracea* was the most tolerant species to soil and water salinity. As final remarks, it is concluded that in arid climates, the clean and environmental safe procedures to control salinity could be associated with the conventional techniques, combining environmental, economic and social aspects, contributing, therefore, to increase the sustainability of the environment and plant growth.

II.1.1 Materials and methods

II.1.1.1 Experimental procedure

To conduct the experimental study, we selected two horticultural species (salt tolerant) which are *Tetragonia tetragonioides* (Fig. II.1.1) and *Portulaca oleracea* (Fig. II.1.2) (the characteristics of the species are given in II.1.1.2 Characteristics of species).

Similar previous studies on the horticultural species *P. oleracea* and *T. tetragonioides* were carry out under different conditions. The former experiment studies differed in purpose, salinity levels (saline treatments) and other factors.

Before starting, we have chosen the design for the experimental study that is shown in Figure II.1.3. The species were submitted to 3 salinity treatments (T) which are salinity levels of irrigation water – 1 dS m⁻¹ (T0), 10 dS m⁻¹ (T1) and 20 dS m⁻¹ (T2). In order to obtain these electrical conductivity values of the irrigation water EC_w, NaCl was added to the tap water as

follows: 0 (T0) is without salt as a control group, 5,78 g/L (T1) is 100 mM NaCl and 11.7 g/L (T2) is 200 mM NaCl. The number of plants per treatment was four. The number of replications was four.

The experimental study was carried out in an investigation greenhouse, in the Horto of the University of Algarve, Campus de Gambelas, Faro, Portugal in 2009, during May and June, with randomized potted plants. Four leaves plants were transplanted to 7 litter soil randomized pots in a greenhouse during the beginning of May, 5. A nitrogen fertigation was applied daily with concentrations of 2 mM NO₃⁻ and 2 mM NH₄⁺. Irrigation water amounts were minimal, but still sufficient for the plant's survival (0.2 L/pot). The plants were irrigated every three days until water drained from the bottom of the pot. The salinity treatments were started with nitrogen concentration 16 days after transplanting salt tolerance species to the randomised pots. The analyses of plants' germination (stem length, number of nodes and number of big leaves) were obtained each three days before irrigation of the species. The analyses of electrical conductivity (EC_w) and pH of drainage water were obtained every ten days after irrigation of the species. The soil electrical conductivity (EC_s) and pH were analysed after completely finishing the experimental study. Plants were analysed on total growth and mineral compositions of the leaves.

At 33 days (June, 23), four plants 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 a forced drought oven at 70 °C for 48 hours and the dry weight (DW) was measured.

II.1.1.2 Characteristics of species

Tetragonia tetragonioides

Family: Aizoaceae

Distribution: Widespread in coastal areas and inland salt marshes. Also occurs in New Zealand.

Common name: New Zealand spinach

Genus: *Tetragonia*

Species: *T. tetragonioides*

Tetragonia tetragonioides (Fig. II.1.1) is found scattered throughout Australia and has become naturalized in many parts of the world. It is considered an agricultural weed in in parts of Queensland. It is a prostrate, sprawling plant with soft stems and foliage and can spread to around 2 meters. Leaves are oval or diamond-shaped and about 75 - 100 mm long. The small, greenish yellow flowers appear at the leaf bases throughout most of the year.

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Fig. II.1.1 - *Tetragonia tetragonioides* (in the greenhouse at the University of Algarve, Portugal)

New Zealand spinach or *Tetragonia tetragonioides* is a bushy plant originating from New Zealand and rumored to have been “discovered” by Captain Cook, used by the seafarer to escape scurvy and then brought back to England where it was quite popular in the 19th century. The seeds of this plant are big and weird (almost evil looking) and should be soaked overnight before planting. The plant is easily grown in moist, reasonably drained soils in sun or partial shade. It is, however, short lived and needs to be regularly propagated. Although edible (and, like common garden spinach), the leaves contain a high level of oxalic acid which must be leached out by blanching before eating. This can be done by plunging the leaves into boiling water for a minute or so. The water should be discarded.

Portulaca oleracea

Family: Portulacaceae

Genus: *Portulaca*

Species: *P. oleracea*

Portulaca oleracea (Fig. II.1.2) (Common Purslane, also known as Verdolaga, Pigweed, Little Hogweed or Pusley), is an annual succulent of the family Portulacaceae which can reach 40 cm in height. About 40 varieties are currently cultivated. It has an extensive old-world distribution extending from North Africa through the Middle East and the Indian Subcontinent

to Malesia and Australasia. The species' status in the New World is uncertain: it is generally considered an exotic weed; however, there is evidence that the species was in Crawford Lake deposits (Ontario) in 1430-89 AD, suggesting that it reached North America in the pre-Columbian era. It is naturalized elsewhere and in some regions it is considered an invasive weed. It has smooth, reddish, mostly prostrate stems and alternate leaves clustered at stem joints and ends. The yellow flowers have five regular parts and are up to 6 mm wide. The flowers appear depending upon rainfall and may occur year round. The flowers open singly at the center of the leaf cluster for only a few hours on sunny mornings. Seeds are formed in a tiny pod, which opens when the seeds are ready. Purslane has a taproot with fibrous secondary roots and is able to tolerate poor, compacted soils and drought.



Fig. II.1.2 - *Portulaca oleracea* (in the greenhouse at the University of Algarve, Portugal)

II.1.1.3 Experimental design

The experimental design was a completely randomized design (CRD) with four replications per treatment: T0 (non saline) – control group, T1 (100 mM NaCl) and T2 (200 mM NaCl) – salinity treatments (Fig. II.1.3).

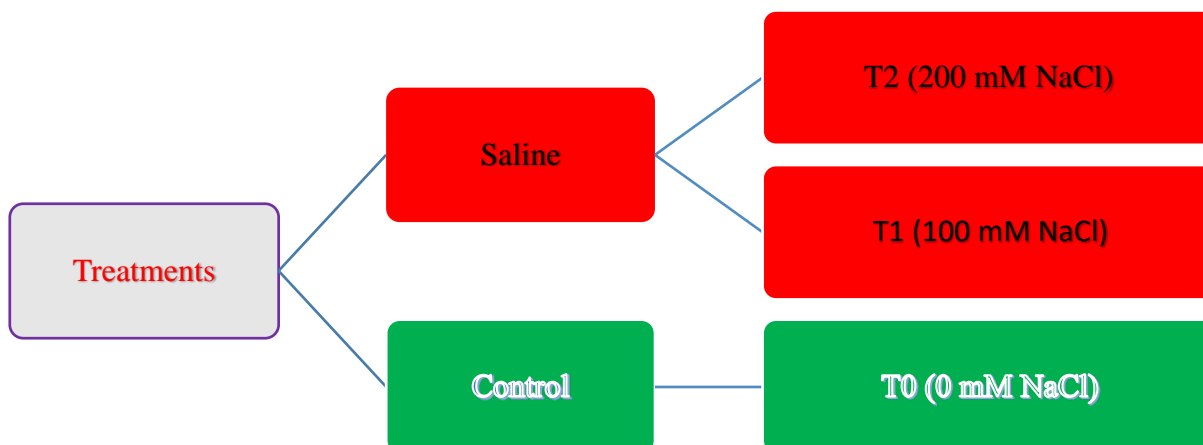


Fig. II.1.3 – Experimental design

II.1.1.4 Climate condition in greenhouse

The average climatic data of the greenhouse during the experimental period:

- Maximal relative humidity – 91.2 %
- Minimal relative humidity – 16.2 %
- Maximal temperature – 45.1 °C
- Minimal temperature – 12.1 °C

During the experimental period, the relative humidity of the greenhouse was increased, and the temperature was decreased (Fig. II.4).

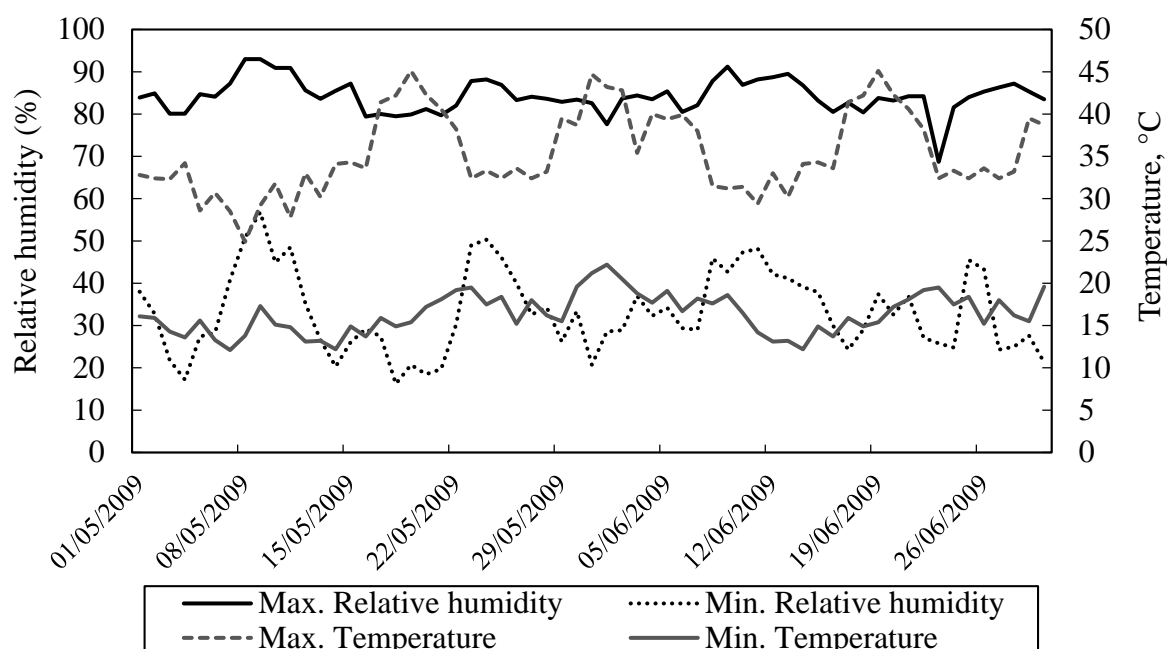


Fig. II.1.4 – Relative humidity (%) and temperature (°C) of the greenhouse during the experiment period, May and June, 2009

II.1.1.5 Soils

Table II.1.1 demonstrates the data of physical and chemical parameters which were taken before the experiment. The soil for the experiment was selected from the soil layer at 0-35 and 30-70 cm depths. The soil characteristics in Algarve (Portugal) were similar to some observed saline areas in the Mirzachul (Hungry Steppe), Syr-Darya Region of Uzbekistan (ADB project, 2008). The chemical reaction is slightly alkaline (pH 8.5) and the soil salinity at the saturation point is moderate (0.3 dS m⁻¹). There was significantly increase of calcium (Ca) content in soil. The soil texture is heavy – silt loam and silt clay loam. The soil structure is moderate.

Table II.1.1. The data of soil physical and chemical parameters were taken before the experimental study

Soil physical parameters	Soil parameters					
	Soil layers (cm)	0 - 30 cm		30 - 70 cm		
Volumetric fc, (%)	23.8	Sand (%)		57.8	49.7	
Volumetric wp, (%)	11.9	Lime (%)		17.7	15.4	
Bulk density, (g cm ⁻³)	1.41	Clay (%)		24.5	34.9	
		Classification		Sandy Clay Loam	Sandy Clay	
Soil chemical parameters		(ppm g sample ⁻¹)				
pH (H ₂ O)	8.5	P ₂ O ₅	K ₂ O	Fe	Mn	Zn
EC _s (dS m ⁻¹)	0.3	31	189	118	40	4.5
Total calcareous (%)	41.2	(% g sample ⁻¹)				
		Ca	Mg	Na	K	N
		27.08	6.56	1.2	1.17	0.11

Note: fc - field capacity; wp - wilting point;

II.1.1.6 Chemical analyses

Dried leaves and stems were finally grounded and analyzed by using the dry-ash method. The levels of sodium (Na) and potassium (K) were determined by a flame photometer and the remaining cations (Na, K, Ca, Mg and Fe) were assessed by atomic absorption spectrometry. Chloride ions were determined in the aqueous extract by titration with silver nitrate according to Radojevic and Bashkin (1999). Plant nitrogen was determined by the Kjeldhal method.

Phosphorus was determined by the colorimetric method according to the vanadate – molybdate method. All mineral analyses were only performed on the leaves.

II.1.1.7 Statistical analyses

Data (n = 4) were examined by one – way ANOVA analysis of variance. Multiple comparisons of the means of data between different salinity treatments within the plants were performed using the Duncan’s test at the P<0.05 significance level (all tests were performed with SPSS Version 17.0 for Windows 7 program).

II.1.2 Results and discussions

II.1.2.1 Electrical conductivity (EC_w) of drainage water of the species *Tetragonia tetragonioides* and *Portulaca oleracea*

The electrical conductivity (EC_w) of drainage water for different species and the high salt concentration (NaCl) treatments (T0, T1 and T2) are shown in the Figs. II.1.5 and II.1.6. There was increased of both species (*Tetragonia tetragonioides* and *Portulaca oleracea*) with the enhancement of the salinity treatments of irrigation water during the vegetation period. It may be seen that the electrical conductivity EC_w of drainage water was higher in both species, which shows that these species are more efficient as salt removal species.

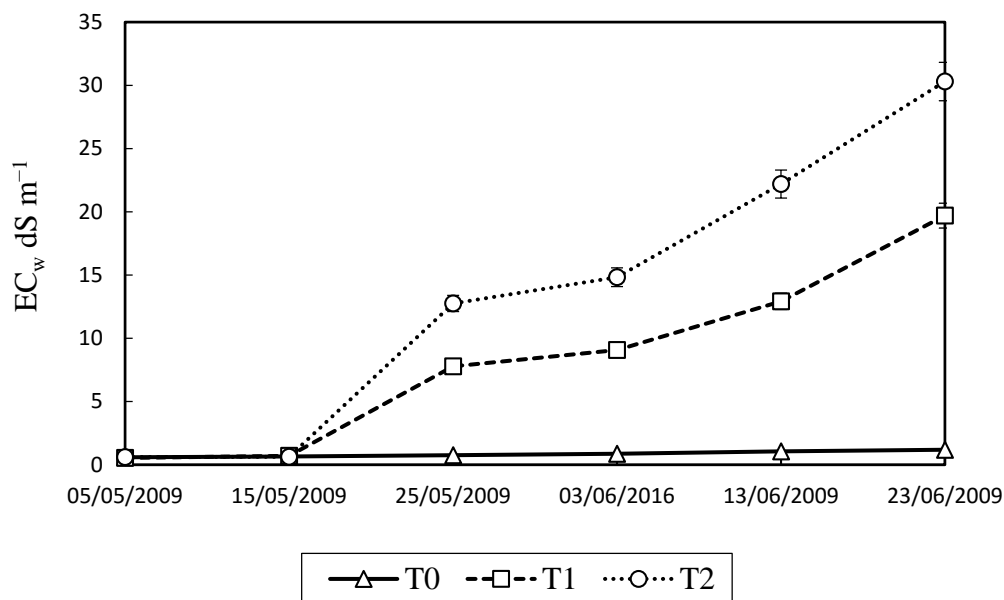


Fig II.1.5. The electrical conductivity (EC_w) of the drainage water of *T. tetragonioides*

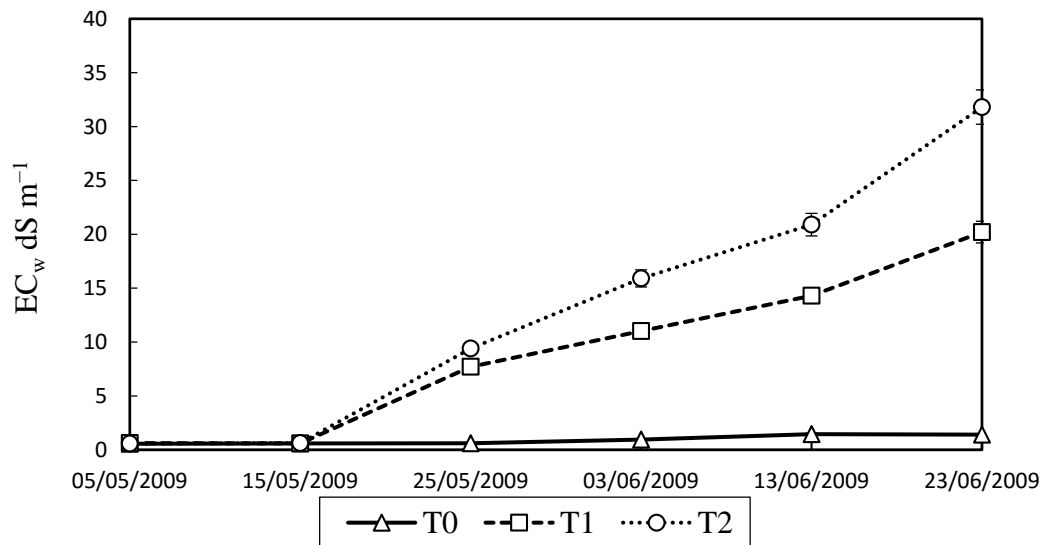


Fig II.1.6. The electrical conductivity (EC_w) of the drainage water of *P. oleracea*

II.1.2.2 Effect of salt treatments on germination of the species

The salinity treatments had a significant effect on the stem length (Fig. II.1.7) 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 Neves (2006) and Yousif et al. (2010). On the other hand, there was a great increase of the stem length in the T0 treatment. The stem length of all treatments (where it was grown *P. Oleracea*) was more different among them than the stem length of *T. Tetragonioides*. The number of big leaves of all treatments (where it was grown *P. Oleracea*) was slightly different than the number of big leaves of *T. tetragonioides* (Fig. II.1.8).

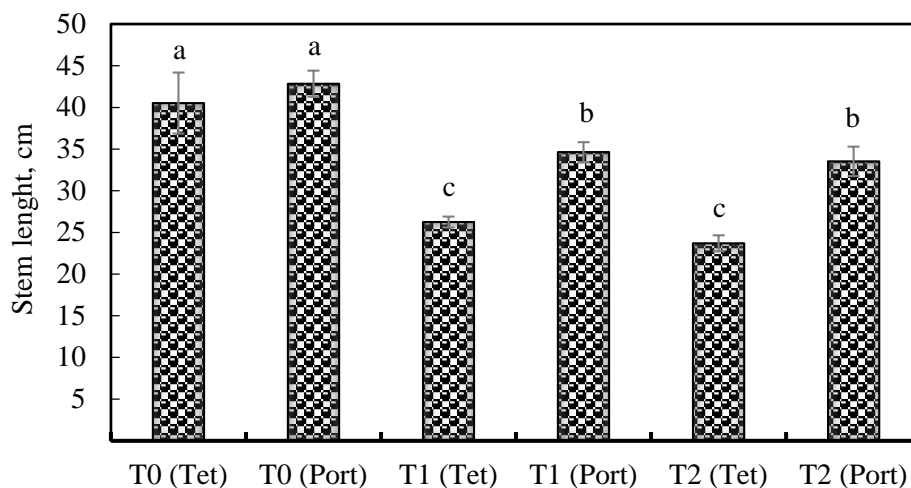


Fig. II.1.7 - Effect of salt treatments (NaCl) on each salt concentration of stem length *Tetragonia tetragonioides* and *Portulaca oleracea*

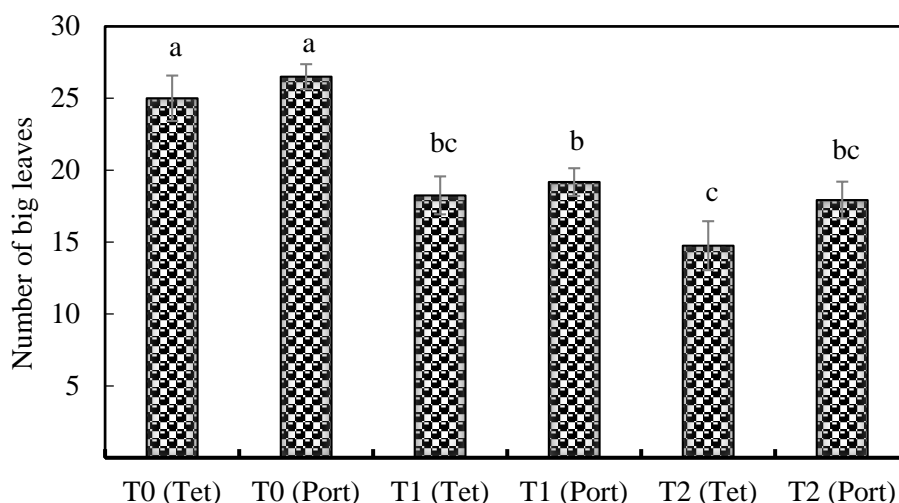


Fig. II.1.8 - Effect of salt treatments (NaCl) on each salt concentration of number big leaves *Tetragonia tetragonioides* and *Portulaca oleracea*

II.1.2.3 pH and electrical conductivity (EC_s) of soil

The soil analyses of pH showed that the soil which was used for the experiment decreased in comparison to the soil without treatment. This result shows that species were influenced by the soil during the experiment period.

Electrical conductivity (EC_s) of the soil saturated with distilled water (dS m⁻¹) for the different species and treatments (T1, T2 and T3) are shown in Table II.1.2. For each treatment, it may be seen that the electrical conductivity of the soil amplified with the increasing salinity of the irrigation water. It may be also seen that the soil electrical conductivity was larger in case of *T. tetragonioides*, which shows that these species have higher potential capacity to remove salts from the soil than *P. oleracea* species.

Table II.1.2. pH and electrical conductivity (EC_s) of the soil was obtained at the end of the experiment

Treatment	<i>Tetragonia tetragonioides</i>		<i>Portulaca oleracea</i>	
	EC _s , dS m ⁻¹	pH	EC _s , dS m ⁻¹	pH
T0	0.13	7.87	1.94	7.89
T1	0.61	7.78	2.20	7.76
T3	1.09	7.69	2.63	7.70

Note: the electrical conductivity (EC_s) of soil was 0.3 dS m⁻¹ at the beginning of the experiment and the pH of the soil samples (used for the experiment) was 8.5.

II.1.2.5 Fresh and dry weight of the species

The results related to *Tetragonia tetragonioides* and *Portulaca oleracea* have revealed that the fresh weights (FW) consistently decline from 526 g plant⁻¹ to 226 g plant⁻¹ (*T. tetragonioides*) and 76.9 FW g plant⁻¹ to 31.5 FW g plant⁻¹ (*P. oleracea*) as they were affected by enhanced salinity. The fresh weight of all treatments (where *T. tetragonioides* were grown) was different ($p < 0.05$) than the fresh weight of *P. oleracea* (Fig. II.1.9). The biomass production of stem (DW g plant⁻¹) (where *T. Tetragonioides* was grown) was significantly different between treatments than the biomass production of stem (DW g plant⁻¹) of *P. oleracea* (Fig. II.1.10). The dry weight of leaves (DW g plant⁻¹) (where *T. Tetragonioides* was grown) was considerably different between treatments than the dry weight of leaves of (DW g plant⁻¹) *P. oleracea* (Fig. II.1.11). The dry weight of seed (DW g plant⁻¹) (where *T. Tetragonioides* was grown) was substantially different between treatments than the dry weight of seeds (DW g plant⁻¹) of *P. oleracea* (Fig. II.1.12).

In the study two agronomic species (*T. tetragonioides* and *P. oleracea*,) were exposed to salt stress by increasing the NaCl concentration (0, 100 and 200 mM NaCl) of irrigation water. When the salinity increased, the plants' dry weight of both species markedly abated; however, the dry weight of *T. tetragonioides* decreased under salt conditions. The dry weight of *T. tetragonioides* diminished 1.2 and 1.4 times and the dry weight of *P. oleracea* decreased 1.2 times on 100 and 200 mM NaCl treatment, respectively compared with the control group. Generally, the growth of species decreases with increasing salinity, while that of halophytes improves. In the study, the growth of the species *T. tetragonioides* amplified under salt stress, according to the previous data reported on the halophytes *Salicornia europaea* and *Suaeda maritima* (Moghaieb et al., 2004) and *Alhagi pseudoalhagi* (Kurban et al., 1999), in which salt treatment improved plant growth at low levels. 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 more dry matter than *P. oleracea* and illustrated, therefore, great capacity for ions accumulation.

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 other similar present studies of many researchers (Anac et al., 2005; Neves, 2006), the fresh and dry weight of *T. tetragonioides* significantly increased on 50 mM NaCl treatment and remained unchanged on 100 and 200 mM NaCl treatment relative to the control group. This is confirmed by the results obtained by Neves et al. (2008) who found an increased yield of salt absorbing species *T. tetragonioides*. The effect of different concentrations of NaCl on growth, increased salt stress and decreased the dry matter biomass length of shoot. This is affirmed by the results obtained by Zayed (2006).

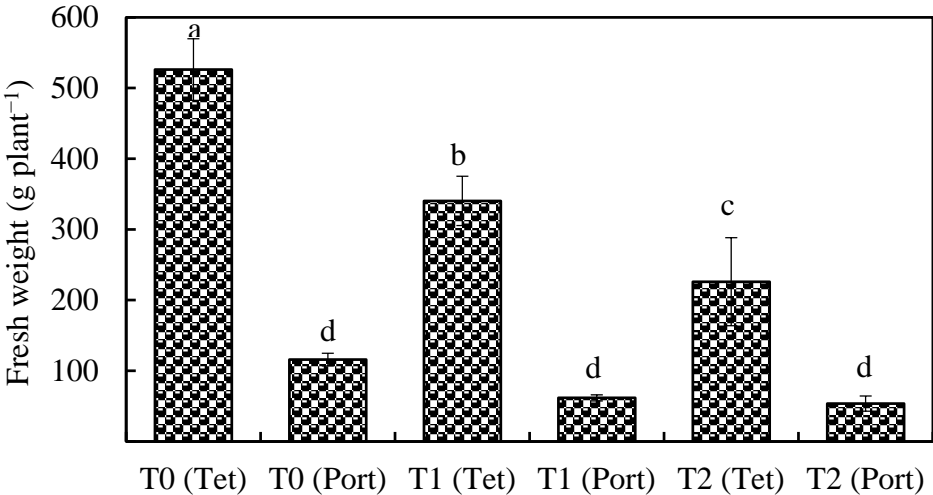


Fig. II.1.9 - Effect of salt treatments (NaCl) on each salt concentration of biomass production in the fresh weight (FW g plant⁻¹) of stem, leaves and seeds of *Tetragonia tetragonioides* and *Portulaca oleracea*

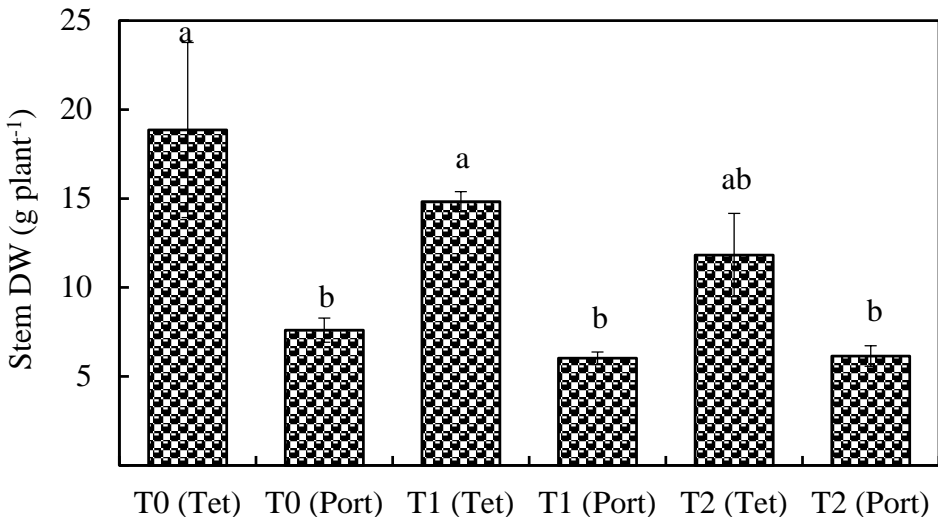


Fig. II.1.10 - Effect of salt treatments (NaCl) on each salt concentration of biomass production in the dry weight of stem (DW g plant⁻¹) of *Tetragonia tetragonioides* and *Portulaca oleracea*

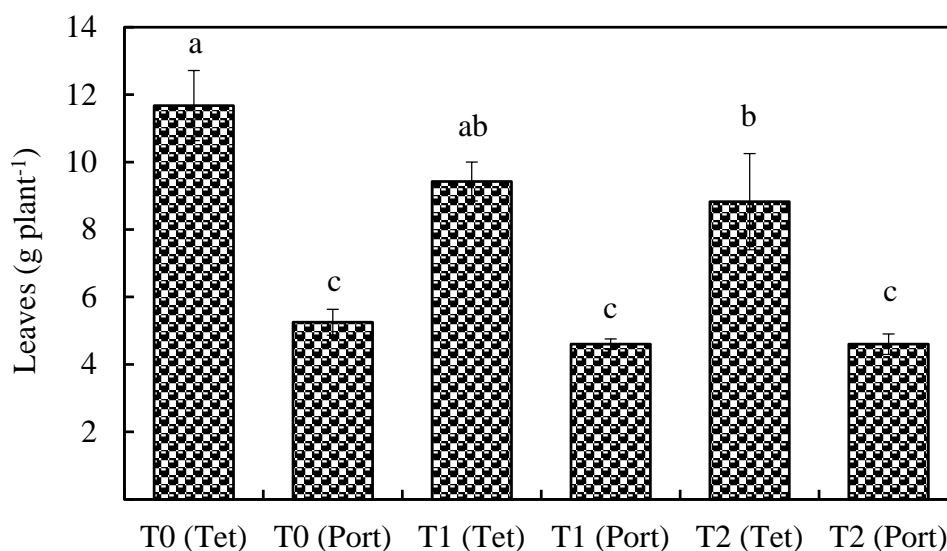


Fig. II.1.11 - Effect of salt treatments (NaCl) on each salt concentration of biomass production in the dry weight of leaves (DW g plant⁻¹) of *Tetragonia tetragonioides* and *Portulaca oleracea*

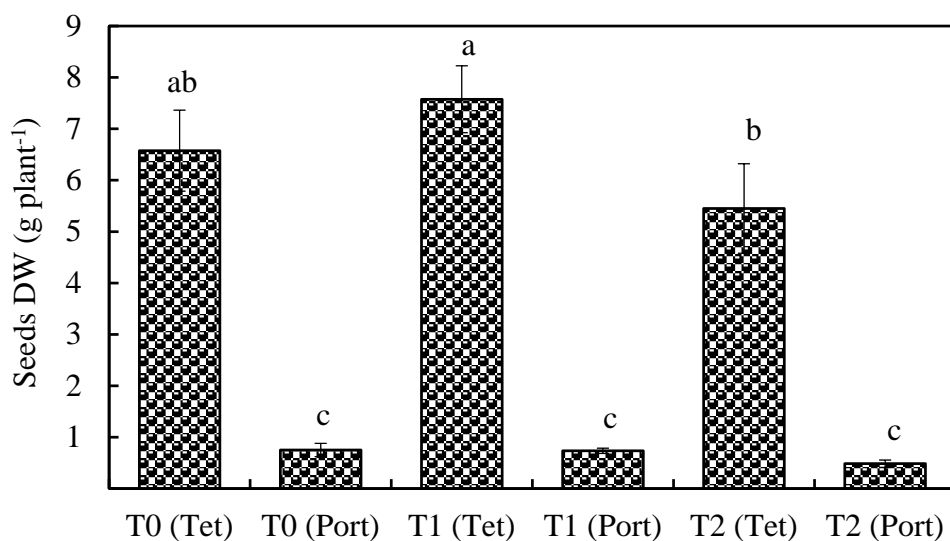


Fig. II.1.12 - Effect of salt treatments (NaCl) on each salt concentration of biomass production in the dry weight of seeds (DW g plant⁻¹ B, C, D) of *Tetragonia tetragonioides* and *Portulaca oleracea*

II.1.2.6 Mineral compositions in leaf content

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 (Table II.1.3): 1) the total nitrogen leaf content highlighted low variations among treatments; 2) there was an immense increase of sodium and chloride concentrations, with the increase of salinity

concentration; 3) a low increase of potassium leaf content was shown, with the saline water increment; 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 the iron content of the leaf. Similarly in a previous study conducted by Anac et al. (2005), the mineral content of *P. oleracea* which are Mg, P, N decreased, and Na, K, Ca, Fe and Cl increased when salt treatments increased.

The salinity had a significant effect on the leaf mineral composition of *Tetragonia tetragonioides* for the majority of analysed mineral elements, as follows (Table II.1.4): 1) there was a great increase of sodium concentrations when water salinity increased; 2) chloride concentrations of the leaves were different between treatments; 3) the total nitrogen leaf content of species showed low variations among treatments, apparently not related with the salinity; 4) there was a general decrease of potassium, calcium, and magnesium leaf content of the crop; 5) the salinity of irrigation water affected phosphorus and iron leaf content. In similar previous studies (Anac et al., 2005; Neves et al., 2008), the mineral contents of *T. tetragonioides* which are Mg, P, N decreased, and Na, K, Ca (Munns, 2002), Fe and Cl increased when salt treatments were increased.

On the other hand, the Na concentration of plants *T. tetragonioides* increased from 3,5% to 6.3% (% g sample⁻¹ of leaf) and *P. oleracea* increased from 1.41% to 2.87% (% g sample⁻¹ of leaf) in accordance with increased salinity levels. This is confirmed by the results obtained by Anac et al. (2005).

Table II.1.3. Effect of salt treatments on the mineral compositions of leaves on each salt concentration of *Portulaca oleracea* (% g sample⁻¹)

<i>Portulaca oleracea</i>				
Treatment	Na	Cl	Mg	Fe
T0	1.41 ± 0.249 c	0.17 ± 0.006 c	0.20 ± 0.001 a	0.0002 ± 0.0003 c
T1	2.87 ± 0.113 b	0.26 ± 0.005 b	0.19 ± 0.004 a	0.0003 ± 0.0001 bc
T2	2.81 ± 0.710 b	0.34 ± 0.003 a	0.19 ± 0.002 a	0.0002 ± 0.0004 c
	N	K	P	Ca
T0	0.39 ± 0.005 ab	1.93 ± 0.244 ab	0.12 ± 0.029 a	0.0027 ± 0.0002 a
T1	0.37 ± 0.008 ab	2.44 ± 0.512 a	0.04 ± 0.005 bc	0.0023 ± 0.0002 ab
T2	0.38 ± 0.009 ab	2.66 ± 0.164 a	0.09 ± 0.014 ab	0.0020 ± 0.0002 ab

Table II.1.4. Effect of salt treatments on the mineral compositions of leaves on each salt concentration of *Tetragonia tetragonioides* (% g sample⁻¹)

<i>Tetragonia tetragonioides</i>				
Treatment	Na	Cl	Mg	Fe
T0	3.49 ± 0.250 b	0.13 ± 0.009 d	0.15 ± 0.016 b	0.0003 ± 0.0002 ab
T1	6.42 ± 0.114 a	0.30 ± 0.034 ab	0.10 ± 0.004 bc	0.0003 ± 0.0001 ab
T2	6.33 ± 0.111 a	0.35 ± 0.015 a	0.08 ± 0.001 c	0.0004 ± 0.0003 a
	N	K	P	Ca
T0	0.33 ± 0.029 b	1.58 ± 0.649 ab	0.06 ± 0.004 bc	0.0018 ± 0.001 ab
T1	0.35 ± 0.017 b	0.62 ± 0.426 b	0.07 ± 0.011 bc	0.0015 ± 0.0004 b
T2	0.43 ± 0.053 a	0.99 ± 0.449 b	0.02 ± 0.003 c	0.0008 ± 0.0002 c

II.1.2.7 Yield of crop

The species leaf, stem and seed dry matter yield was reduced significantly by saline irrigation water (Tables II.1.5 and II.1.6). The yield of the species *Tetragonia tetragonioides* and *Portulaca oleracea* were calculated and the results of salinity treatments (T1 and T2) was compared to the control group (T0). Despite the fact that the species were irrigated in the high salt concentration (NaCl) of irrigation water, the dry matter (DM) was not much lower than that of the control (T0) treatment. The high biomass production of the species *T. tetragonioides* averaging from 4452 to 3132 DM kg ha⁻¹ was obtained (Annex II.1.1). The obtained results show that the dry matter of the species had low variation between salinity treatments. The dry matter of the species in the salinity treatments significantly increased by 1.42 – 1.22 times, compared to T0. The depressing effects of salinity on plant growth were in accordance with our previous findings (Irshad et al., 2002b and Wilson et al., 2000).

The biomass production of the species *P. oleracea* averaging from 2720 to 2240 DM kg ha⁻¹ was obtained (Annex II.1.2). Plants grown at the low levels of NaCl (control) reaches relatively to the higher total dry weights and did not exhibit the toxicity symptoms. However, the total dry weight was significantly reduced at higher levels of salinity (10 - 20 dS m⁻¹), in which the symptoms of salt toxicity were revealed as growth depression. The salinity concentration of 10 - 20 dS m⁻¹ NaCl slightly increased the dry matter of the species by 1.21 – 1.02 times, compared to the control group. These results are confirmed by previous research implemented by Ehni, (1997); Cros et al. (2007) and Hamidov (2007).

Table II.1.5. Yield of species *Tetragonia tetragonioides*

<i>Tetragonia tetragonioides</i>					
Treatment	FM (g plant ⁻¹)	DM (g plant ⁻¹)	Yield %	FM kg ha ⁻¹	DM kg ha ⁻¹
T0	526 ± 0.26 a	37 ± 0.19 a	7 ± 0.17 b	63132 a	4452 a
T1	340.5 ± 0.13 b	32 ± 0.93 a	9.5 ± 0.56 ab	40842 b	3819 a
T2	226 ± 1.07 b	26.3 ± 0.71 a	13 ± 0.83 a	27120 b	3132 a

Table II.1.6. Yield of species *Portulaca oleracea*

<i>Portulaca oleracea</i>					
Treatment	FM (g plant ⁻¹)	DM (g plant ⁻¹)	Yield %	FM kg ha ⁻¹	DM kg ha ⁻¹
T0	121 ± 0.48 a	16.5 ± 1.13 a	13.5 ± 0.21 b	24300 a	2720 a
T1	62 ± 0.27 b	13.5 ± 0.34 b	21.8 ± 0.36 a	12340 b	2280 a
T2	53.5 ± 0.63 b	13.5 ± 0.42 b	27.5 ± 0.64 a	10660 b	2240 a

II.1.3 Conclusion

Tetragonia tetragonioides proved to be the most potential salts (ions) removal species. Moreover, at the end of the experiment, *T. tetragonioides* was the sole species that had produced significant amounts of dry matter. The reasons were: fast growth rate, higher biomass production (if properly managed), easy cropping (as winter or summer crop). Moreover, it can be suggested that *T. tetragonioides* is a very interesting species because:

- The higher biomass production potential: besides the growth rate, this specie can produce several yields during the year (summer and winter).
- Easy multiplication (seed propagation) and easy crop management.
- Species tolerant to drought and salts.
- Protection from soil erosion due to excellent soil covering.
- High mineral composition of leaves.

The potential of *Portulaca oleracea* as salts removal plant was also high, but lower than *T. tetragonioides*. This was explained by the larger biomass production of *T. tetragonioides*. On the other hand, for very arid climates, *P. oleracea* may substitute *T. tetragonioides* successfully, once that *P. oleracea* is much more tolerant to drought and salt conditions. Moreover, these species can be planted, as ornamentals, in saline soils, even without irrigation.

These new clean techniques to control salinity showed that agricultural production can be maintained through the reduction of salts application due to the decrease of irrigation amounts, reducing the leaching. On the other hand, the applied salts through the irrigation can be eliminated by using the salt (ions) removing species. As final remark, it is concluded that in arid climates, the clean and environmental safe procedures to control salinity could be associated with the conventional techniques, combining environmental, economic and social aspects. Hence, these two salt removing species may contribute to increase the soil sustainability of irrigated areas under climatic changes, and also may be used as food crops.

II.2 Effect of saline soil (where grown on the high salinity conditions (NaCl) of the species) on growth, mineral compositions and yield of lettuce

Abstract: The purpose of the experimental study was to planting salt sensitive crop lettuce (*Lactuca sativa* L) in the pots (were grown salt removing crops on the high salinity condition) to identify which salt tolerant crops were usefull as a salt removing species. In several experiments on lettuce we conducted several conditions. The former experiments of scientists differed in purpose, salinity levels (saline treatments) and other factors. Due to the fact that lettuce 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 yield reduction and reduced crop quality. Hence, the experimental results showed that *Tetragonia tetragonioides* and *Portulaca oleracea* have high potential as soil salt removing species, and, therefore, they are recommended to start crop rotation, in order to reduce soil salinity. The salt removal crops above are a good contribution, mainly the *T. tetragonioides* species, to the quality and yield of the lettuce, a moderate salt sensitive crop. Hence, it is demonstrated that this technique is a clean and environmental safe tool to avoid salinization and to maintain the sustainability of agricultural systems.

II.2.1 Materials and methods

II.2.1.1 Experimental procedure

The experimental studies were conducted at the University of Algarve, Campus de Gambelas in the greenhouse from September, 25 to November, 10, 2009, randomized with potted plants. Plants were sowed in soil pots where just before *Tetragonia tetragonioides* and *Portulaca oleracea* species were grown. Plants were irrigated each three days with tap water until October, 20. The total number of plants was 24, distributed according to the above salt treatments (3), every treatment was applied to four pots with four plants. Lettuce was irrigated with nitrogen N treatments during the last twenty days (October, 20 to November,10) with concentrations of 2 mM NO₃⁻ and 2 mM NH₄⁺. Electrical conductivity (EC_w), pH of the drainage water and stem length of the crop were measured periodically and the mineral composition in leaf content was determined at the end of the experiment.

Plants were harvested at 20 days (November, 10, 2009) after treatments, and washed with distil water for a few minutes, wiped with paper and the fresh matter (FM) was measured. The fresh samples were dried in a forced drought oven et 70° C for 48 hours and the dry matter (DM) was measured and plant materials for chemicals and organic analyses were collected.

Lettuce (*Lactuca sativa* L)

Family: Asteraceae

Genus: *Lactuca*

Species: Lettuce (Fig. II.2.1) is a temperate annual or biennial plant of the daisy family Asteraceae. It is most often grown as a leaf vegetable. It is eaten either raw, notably in salads, sandwiches, hamburgers, tacos, and many other dishes, or cooked, as in Chinese cuisine in which the stem becomes just as important as the leaf. Both the English name and the Latin name of the genus are ultimately derived from *lac*, the Latin word for “milk”, referring to the plant’s milky juice. Mild in flavor, it has been described over the centuries as a cooling counterbalance to other ingredients in a salad. The Food and Agriculture Organization of the United Nations reports that the world production of lettuce and chicory for the calendar year 2007 was 23.55 million tons, primarily coming from China (51 %), the United States (22 %) and Spain (5 %).



Fig. II.2.1 - Lettuce (*Lactuca sativa* L) (in the greenhouse at the University of Algarve, Portugal)

II.2.1.2 Chemical analyses

Dried leaves and stems were finally grounded and analyzed by using the dry-ash method. The levels of sodium (Na) were determined by a flame photometer and the remaining cations were assessed by atomic absorption spectrometry. Chloride (Cl) ions were determined in the aqueous extract by titration with silver nitrate according to Radojevic and Bashkin (1999). All mineral analyses were only performed in the leaves (Fig. II.2.2).

Bekmirzaev, G./ Effect of saline soils and drought conditions on growth, mineral composition, photosynthesis and water relations of vegetable crops – Thesis (2017)



Fig. II.2.2 - Analysis process in the laboratory at the University of Algarve, Portugal

II.2.1.3 Statistical analyses

Data ($n = 4$) were examined by one – way ANOVA analysis of variance. Multiple comparisons of the means of data between different salinity treatments within the plants were performed using the Duncan's test at the $P < 0.05$ significance level (all tests were performed with SPSS Version 17.0 for Windows program).

II.2.1.4 Soil

The soil was used for the experimental study just after the previous salt removing species *T. tetragonioides* and *P. oleracea*. The characteristics of the pH and electrical conductivity of the saturated soil ECs is given in the Table II.1.2 (II.1.2 Results and discussions - II.1.2.3 pH and electrical conductivity (EC_s) of soil).

II.2.2 Results and discussions

II.2.2.1 Height leaves of lettuce

Height leaves of lettuce (grown in the pots of *T. tetragonioides* and *P. oleracea*) are shown in Fig 1. The total grown height leaves of lettuce showed low variations among treatments. There was a great increase of the height leaves in treatment T0. There was a general decrease in treatment T1 and T2. These effects were generally very clear when compared to the control T0.

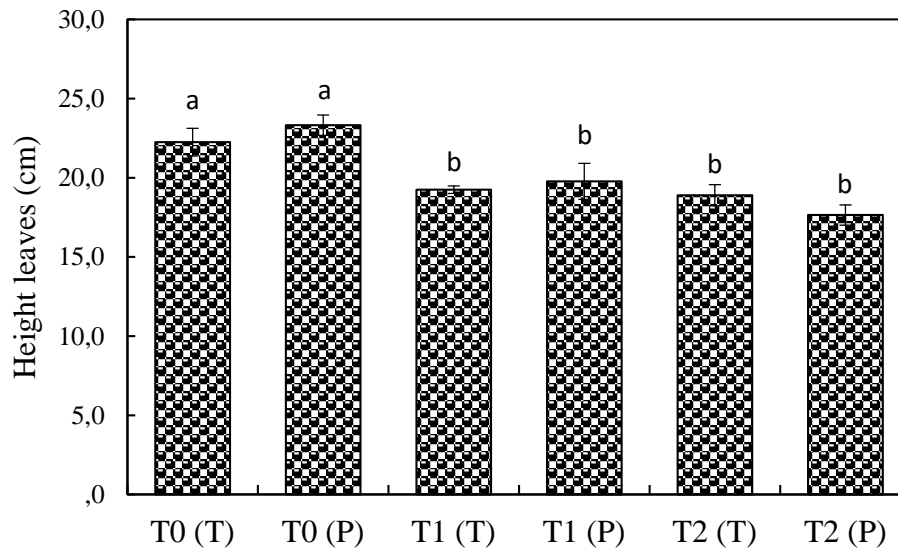


Fig. 1 - Height leaves of lettuce during of the experiment (grown in the pots of *T. tetragonioides* and *P. oleracea*) related to treatments of Duncan test. Means \pm S.E., $n = 4$.

Bars with different letters are significantly different at $P < 0.05$

II.2.2.1 Effect of electrical conductivity (EC_w) of drainage water to lettuce leaf height

Growth decreasing the leaves' height of lettuce was significantly affected by an increase in salinity levels. The leaves' height of lettuce (grown in the pots of *T. tetragonioides*) in the treatment T2 (high salinity regime) were reduced by 1.02 times compared to the treatment T1 (middle salinity regime) (Fig. II.2.3) and the height of lettuce's leaves (grown in the pots of *P. oleracea*) in the treatment T2 (high salinity regime) reduced by 1,12 times compared to the treatment T1 (middle salinity regime) (Fig. II.2.4). The presented results confirm that the height of leaves was highly affected by high salinity (Cemek et al., 2011). Decrease in the height of leaves indicates an inverse relationship between salinity and biomass production. Change in electrical conductivity levels in treatments is the most important.

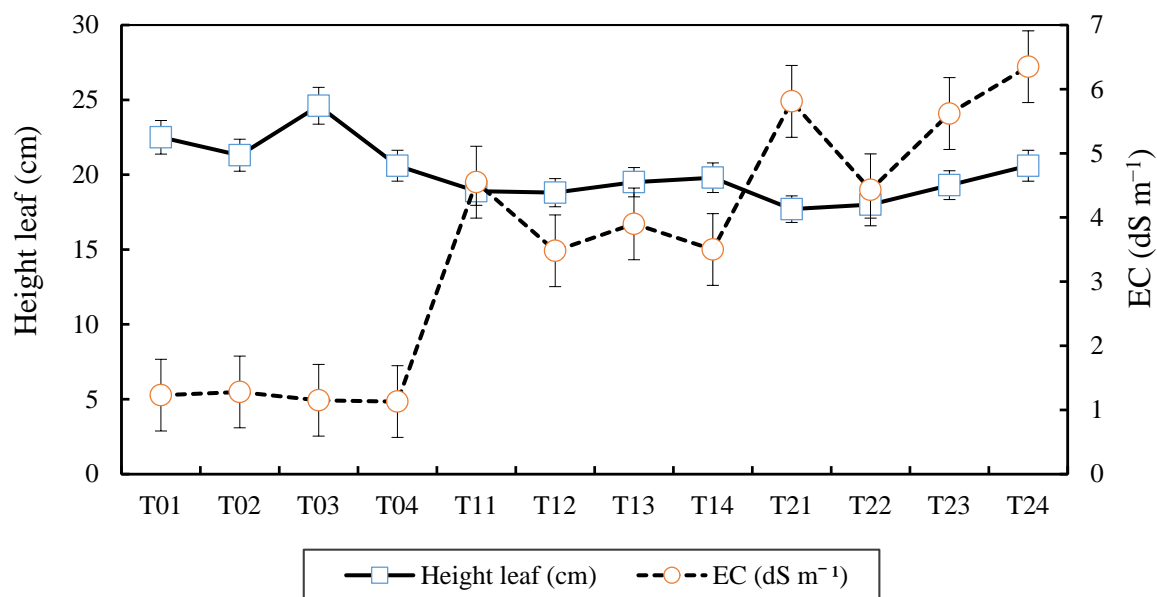


Fig. II.2.3 - Electrical conductivity (EC_w) of the drainage water comparing lettuce's height of leaves (grown in the pots of *Tetragonia tetragonioides*). Means \pm S.E., $n = 4$. Bars with different letters are significantly different at $P < 0.05$.

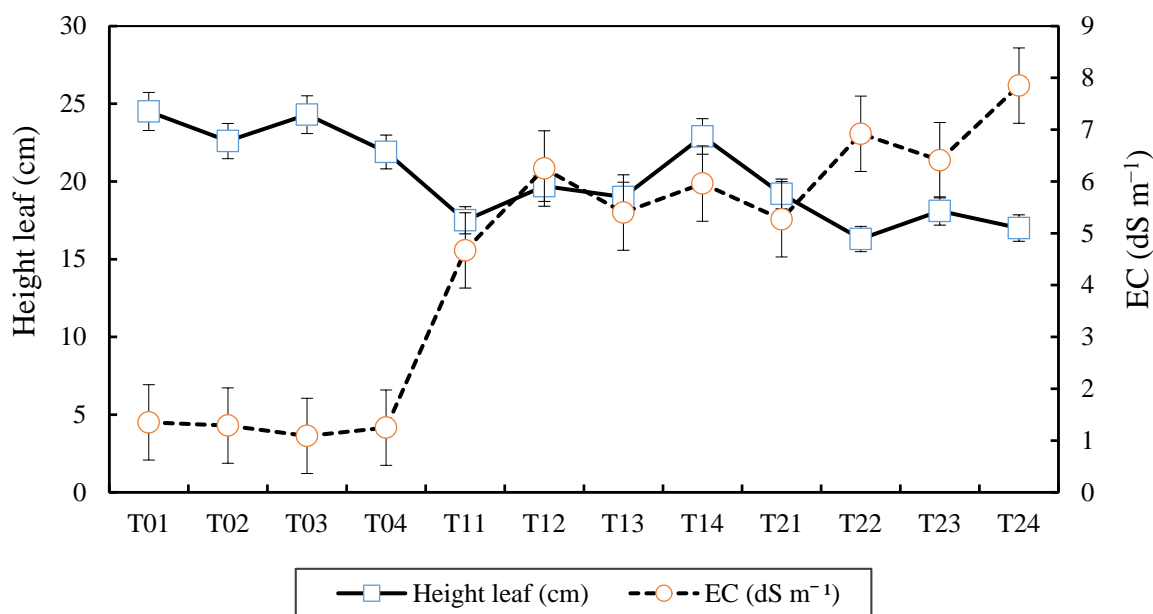


Fig. II.2.4 - Electrical conductivity (EC_w) of the drainage water comparing lettuce's leaves height grown in the pots of *Portulaca oleracea*. Means \pm S.E., $n = 4$. Bars with different letters are significantly different at $P < 0.05$.

II.2.2.2 Fresh (FW) and dry (DW) weight of plant

Fresh weight of lettuce (grown in the pots of *T. tetragonioides* and *P. oleracea*) showed no difference in each treatment (Fig. II.2.5). The fresh weight of lettuce ($225.6 \text{ g plant}^{-1}$) (grown in the pots of *T. tetragonioides*) was lower than the fresh weight of lettuce ($255.4 \text{ g plant}^{-1}$)

(grown in the pots of *P. oleracea*) in the treatment T0, but there was shown that the fresh weight is higher on the saline soil in the treatments T1 (186.8/178.2 g plant⁻¹) and T2 (167.8/154.9 g plant⁻¹). It was indicated that the soil in which *T. tetragonioides* was grown had less salinity than the soil in which *P. oleracea* was grown (Fig. II.2.6). The dry weight of lettuce (grown in the pots of *T. tetragonioides*) showed no variations among treatments. There was a great increase of dry matter in treatment T0. The dry weight of lettuce (grown in the pots of *P. oleracea*) demonstrated low variations among treatments. There was a great increase of dry matter in the treatment T0.

In support of the previous observations implemented by Puttanna et al. (2001), we demonstrated that foliar application of fertilizer significantly enhanced the growth and yield of citronella plants. High rates of fertilizer can lead to high salinity which can damage plants and reduce growth and yield. Miceli et al. (2003) 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 the number of leaves per plant. Recently, studying the effects of urea fertilization on cluster bean plants subjected to water stress, revealed that water stress significantly decreased a shoot water potential, fresh and dry mass and maintained a reduction of water content according to similar researches (Samarakoon, 2006; Al-Maskri et al., 2010; 1996; Younis et al., 2009).

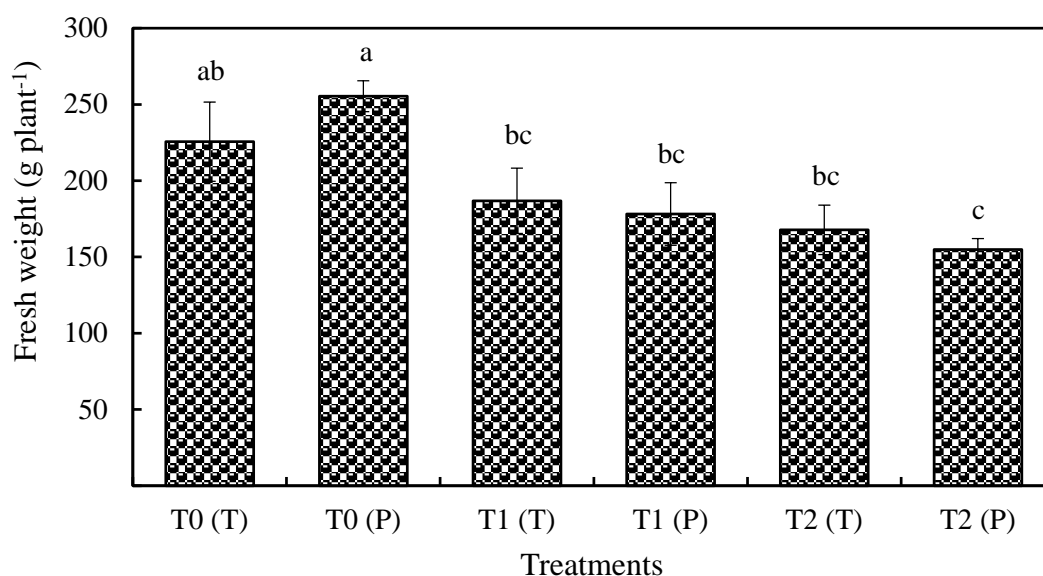


Fig. II.2.5 - Fresh weight of lettuce (grown in the pots of *Tetragonia tetragonioides* and *Portulaca oleracea*) on the different treatments of nitrogen concentrations. Means \pm S.E., n = 4. Bars with different letters are significantly different at P < 0.05.

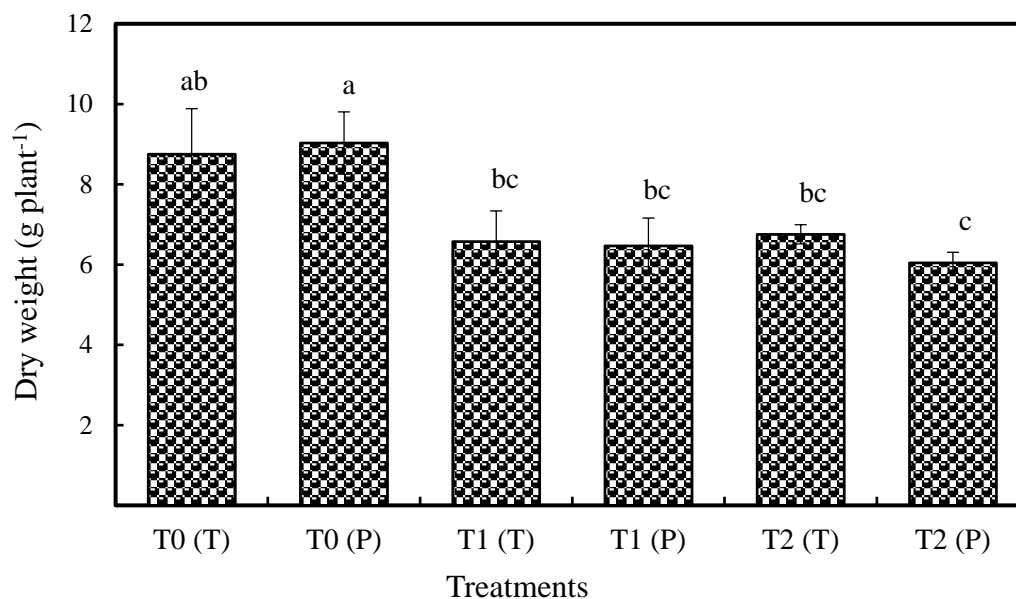


Fig. II.2.6 - Dry weight of lettuce (grown in the pots of *Tetragonia tetragonioides* and *Portulaca oleracea*) on the different treatments of nitrogen concentrations. Means \pm S.E., n = 4. Bars with different letters are significantly different at P < 0.05.

II.2.2.3 Mineral compositions in leaf content

The significant effect was in the mineral composition of lettuce's leaf content (grown in the pots of *T. tetragonioides* and *P. oleracea*) analysed for the majority of mineral elements. 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 compared to treatments (Fig. II.2.7) There was an increased percentage of high salt concentration soil in the treatments T1 and T2, approximately in average 0.49 – 0.59 % g sample⁻¹ (Annex II.2.3). These results confirmed similar research by Coudela and Petricova (2008) and Santos et al. (2003).

Chloride (Cl) content of the lettuce leaves grown in the pots of *T. tetragonioides* was significantly different between treatments (Fig. II.2.8). There was slightly increased chloride in leaf content of lettuce grown in the pots of *Tetragonia tetragonioides* in the salt concentration treatments (T1 and T2) compared to the lettuce grown in the pots of *Portulaca oleracea*.

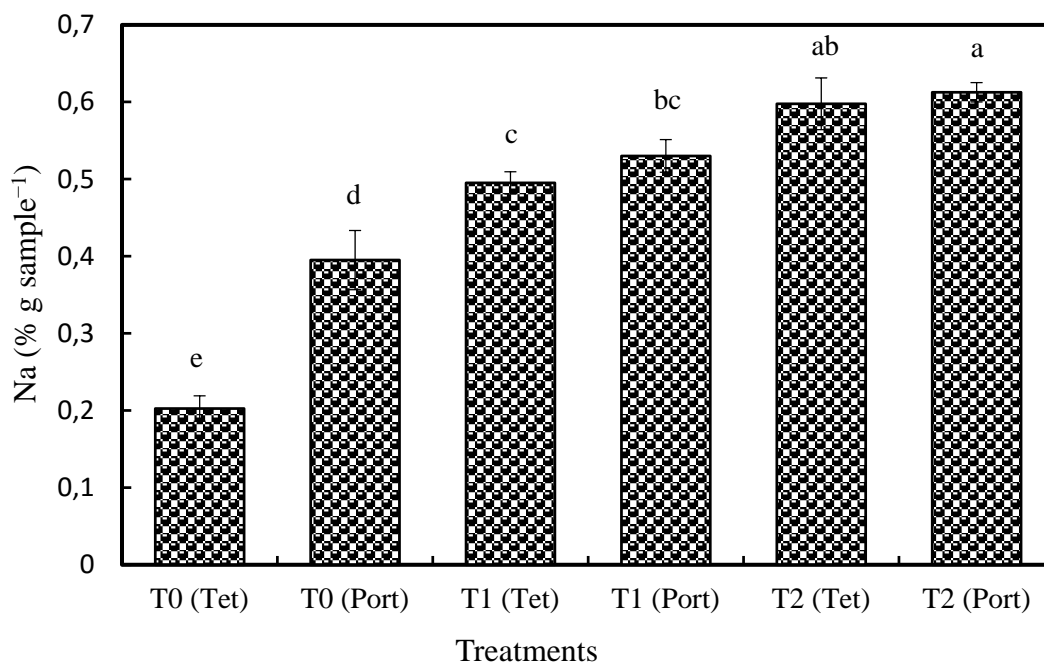


Fig. II.2.7 - Mineral composition (Na) of lettuce leaves (grown in the pots of *Tetragonia tetragonioides* and *Portulaca oleracea*) on each salt concentration. Means \pm S.E., n = 4. Bars with different letters are significantly different at P < 0.05.

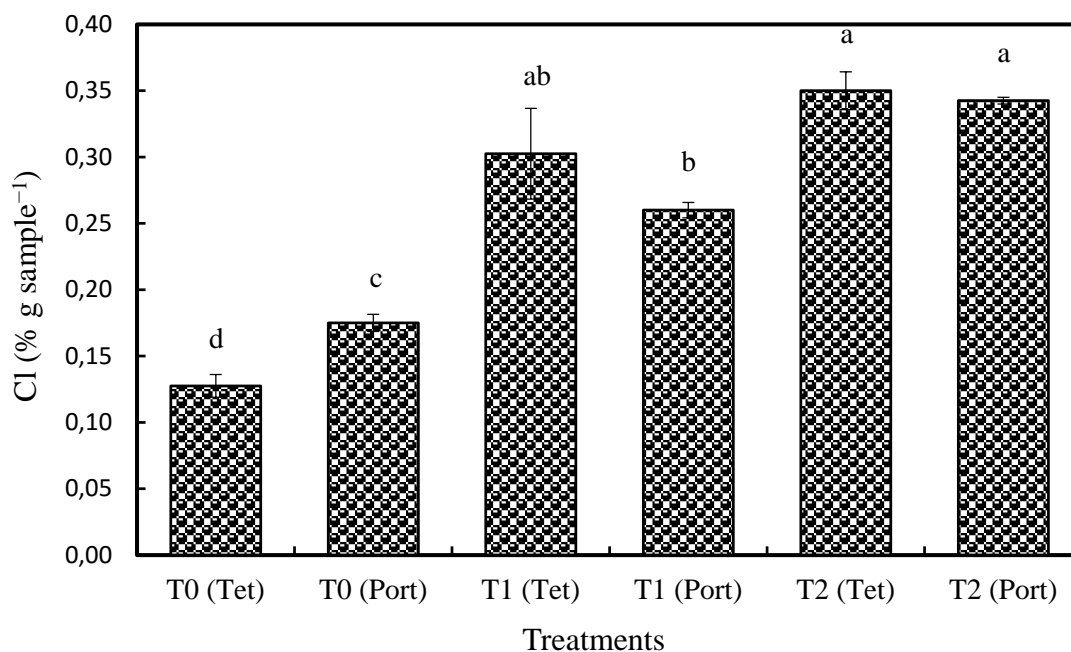


Fig. II.2.8 - Mineral composition (Cl) of lettuce leaves (grown in the pots of *Tetragonia tetragonioides* and *Portulaca oleracea*) on each salt concentration. Means \pm S.E., n = 4. Bars with different letters are significantly different at P < 0.05.

II.2.2.4 Yield of lettuce

The calculation of lettuce (*Lactuca sativa* L) yield (grown in the pots of *Tetragonia tetragonioides* and *Portulaca oleracea*) was significantly different between treatments in soil of the species' previous experiment (Tables II.2.1 and II.2.2). The dry weight of lettuce grown in the pots of *T. tetragonioides* in the treatments of T2 (4.02) increased slightly more than the dry weight of lettuce grown in the pots of *P. oleracea* in the treatments of T2 (3.90). The dry weight of lettuce (*T. tetragonioides*) was 788 – 608 kg ha⁻¹ (Annex II.2.1) and the dry weight of lettuce of (*P. oleracea*) was 813 – 544 kg ha⁻¹ (Annex II.2.2). Both species' dry weight greatly increased in comparison to the control group (T). The lettuce's dry weight (*T. tetragonioides*) in the saline soil significantly increased compared to the dry weight of lettuce (*P. oleracea*). Lettuce yield response to the three treatments was similar with the findings of Karam et al. (2005). Unlukara et al. (2008) and Yasgan et al. (2008) also reported that salinity soil reduced the yield of lettuce in a constant manner. The previous experiment confirmed that *T. tetragonioides* is the best salt removing crop.

Table II.2.1. Lettuce yield (grown in the pots of *Tetragonia tetragonioides*)

Treatment	Lettuce yield				
	FM (g plant ⁻¹)	DM (g plant ⁻¹)	Yield %	FM kg ha ⁻¹	DM kg ha ⁻¹
T0	225.6 ± 26 ab	8.7 ± 1.1 ab	4.0 ± 0.00 a	20307.5 ± 2338.2 a	788 ± 100.9 a
T1	186.8 ± 21.4 bc	6.6 ± 0.8 bc	3.8 ± 0.25 a	16813.3 ± 1927.7 b	91.8 ± 68.6 b
T2	167.8 ± 16.2 bc	6.8 ± 0.2 bc	4.0 ± 0.41 a	15105.3 ± 1459.2 b	608 ± 21.6 b

Table II.2.2. Lettuce yield (grown in the pots of *Portulaca oleracea*)

Treatment	Lettuce yield				
	FM (g plant ⁻¹)	DM (g plant ⁻¹)	Yield %	FM kg ha ⁻¹	DM kg ha ⁻¹
T0	255.4 ± 10.1 a	9.0 ± 0.8 a	3.5 ± 0.28 a	22987.8 ± 910.6 a	812.8 ± 69.9 a
T1	178.2 ± 20.5 bc	6.5 ± 0.7 bc	3.8 ± 0.25 a	16037.5 ± 1849.5 b	581.8 ± 62.8 b
T2	154.9 ± 7.2 c	6.1 ± 0.3 c	4.0 ± 0.00 a	13938.3 ± 643.8 b	544.3 ± 23.8 b

II.2.3 Conclusions

The obtained results highlight that lettuce (*Lactuca sativa*, L) is not tolerant to dry and hot conditions and it needs large amounts of irrigation water. This irrigation water contains a certain quantity of soluble salts which will be accumulated in the soil and cause reduction of crop yield and quality. The positive contribution was indicated for the above salt removal crops, mainly

T. tetragonioides and *Portulaca oleracea* halophytic species to the quality and yield of the lettuce (a moderate salt - sensitive crop). Therefore, it is demonstrated that this technique is a clean and environmentally safe tool to avoid salinization 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, economic and social aspects of problem solving. Hence, these two salt removing species may contribute to the increase of the soil sustainability of irrigated areas under climatic changes and may also be used as food crops.

II.3 Effect of several irrigation water regimes on yield, leaf minerals and photosynthetic pigments of the species *Tetragonia tetragonioides*

Abstract: The main purpose of this experiment was to study the effect of several irrigation water regimes on *Tetragonia tetragonioides* in semi-arid regions. We measured its effects on growth, biomass production (fresh and dry mass), yield, mineral composition and photosynthetic pigments (chlorophyll, carotenoid and soluble carbohydrates content in leaves). The experiments were conducted in the greenhouse at the University of Algarve (Portugal). The study ran from February to April 2010. Three irrigation treatments were based on replenishing the 0.25 m deep (depth of the pots) to field capacity when the soil water level dropped to 70 % (T1, wet treatment), 50% (T2, medium treatment), and 30% (dry treatment) of the available water capacity. The experimental results showed that the leaf mineral composition of chloride and sodium, the main responsible ions for the soil salinization and alkalization in arid and semi-arid regions, enhanced with the decrease of the soil water content. However, the minimum amount of chlorophyll, carotenoids and soluble carbohydrates in leaf content were obtained in the medium and driest treatment. On the other hand, growth differences among the several irrigation regimes were very low, and the crop yield increased in the dry treatment (T3) compared to the medium treatment (T2). Thus, the high capacity of salt removing species as *T. tetragonioides* suggests an advantage of its cultivation under dry conditions. This research recommends the use of *T. tetragonioides* in arid and semi-arid regions.

II.3.1 Material and methods

II.3.1.1 Experimental procedure

The experimental work was conducted in the greenhouse of Horto at the University of Algarve, Faro, Portugal (37°02'37.1N 7°58'30.8W), from February to April 2010. The salt removing species *T. tetragonioides* were the selected species. Four leaf plants were transplanted to 7 L capacity and 0.25 m depth randomized pots on February, 10. The number of plants per pot was eight, with three replications. The species were irrigated with tap water every three days until the beginning of the treatments (February, 1 – March, 8). A nitrogen fertigation treatment was started on March, 8 (daily applied with concentrations of 2 mM NO₃⁻ and 2 mM NH₄⁺, being the cumulative amount of NO₃NH₄ (g plant⁻¹) to the end of the experimental studies (April, 22). The electrical conductivity (EC_w) of irrigation water was 0.6 dS m⁻¹ and pH 7.



Fig. II.3.1 *Tetragonia tetragonioides* (in the greenhouse at the University of Algarve, Portugal)

The treatments consisted of three irrigation regimes in a randomized complete block design with three replicated treatments based on replenishing the 0,25 m deep (depth of the pots) to field capacity when the soil water level dropped to 70 % (T1, wet treatment), 50% (T2, medium treatment), and 30% (T3, dry treatment) of the available water capacity (aw). This concept has been developed by Veihemeyer and Hendrickson (1931), where ‘aw’ is the range of available water that can be stored in soil and is available for growing crops. It was assumed by the same authors that the soil available water content readily to plants (θ_{aw}) is the difference between the volume of water content at field capacity (θ_{fc}) and at the permanent wilting point (θ_{wp}), as follows:

$$\theta_{aw} = \theta_{fc} - \theta_{wp} \quad (1)$$

The watering volume was estimated to replenish of the soil profile to field capacity at a depth of 0.25 m. The volumetric soil water content - m^3 water / m^3 soil ($m^3 m^{-3}$) – was determined just before the water application (see Table II.3.2).

To control soil water along the soil profile, the irrigation frequency and water amounts, the pots were weighted every day. The soil water content was monitored periodically, gravimetrically measured for a 0.00–0.25 m depth.

The plants were harvested after the treatments (April, 26), washed with distil water for a few minutes and wiped with paper. Then the fresh weight (FW) was measured. The fresh samples

were dried in a forced drought oven at 70° C for 48 hours and the dry weight (DW) was measured. Plant materials were collected for chemical analyses. The soil electrical conductivity (EC_s) and pH were measured before and after the experiment.

II.3.1.2 Growth and chemical analysis

During the vegetation period we measured the stem length, number of nodes and number of leaves of *T. tetragonioides* every seven days.

The plants' leaves were analysed on total growth and mineral compositions (Na, Cl, N, K, P, Ca and Mg). Dried leaves and stems were finally grounded and analyzed by using the dry-ash method. The levels of Na and K were determined by a flame photometer and the remaining cations (Na, K, Ca and Mg) were assessed by atomic absorption spectrometry. Chloride ions were determined in the aqueous extract by titration with silver nitrate according to the method of Radojevic and Bashkin (1999). Plant nitrogen (N) content was determined by the Kjeldhal method. Phosphorus was determined by the colorimetric method according to the vanadate – molybdate method. All mineral analyses were only performed on the leaves.

The analysis of pigments was done on four discs with a size of 0.66 cm and a total area of 1.37 cm². For sugars there were ten discs, with the size of a disc of 0.66 cm and a total area of 3.42 cm². The amount of photosynthetic pigments (chlorophyll a, b total and carotenoids) was determined according to the method of Lichtenthaler (1987). Shoot samples (0.25 g) were homogenized in acetone (80%). The extract was centrifuged at 3.000 g and absorbance was recorded at wavelengths of 646.8 and 663.2 nm for chlorophyll assay and 470 nm for carotenoids assay by the Varian Cary 50 UV–Vis spectrophotometer. Chla, Chlb, ChlT and carotenoids were calculated. Soluble sugars (glucose) in leaves were extracted as described by Dubois et al. (1956). The change in absorbance was continuously followed at 340 nm using an Anthos hat II microtiter-plate reader (Anthos Labtec Instrument, Hanau).

II.3.1.3 Statistical analyses

Data (n = 4) were examined by a one – way ANOVA analysis of variance. Multiple comparisons of the means of data between different salinity treatments within the plants were performed using the Duncan's test at the P<0.05 significance level (all tests were performed with SPSS Version 17.0 for Windows program).

II.3.1.4 Soil

Table 1 shows the soil texture and soil parameters before the experiment. According to FAO, based on the USDA particle-size classification, the soil texture was sandy clay loam. The soil

parameters show that the range in the soil's pH value is slightly alkaline and that the electrical conductivity (ECs) is 1.1 dS m^{-1} (non - saline soil) at $25 \text{ }^\circ\text{C}$.

Table II.3.1. Soil parameters before the experiment

Soil texture		Soil parameters			
Sand (%)	58.9	Field capacity θ_{fc} ($\text{m}^3 \text{ m}^{-3}$)	0.238	pH (H ₂ O)	7.7
Silt (%)	18	Wilting point θ_{wp} ($\text{m}^3 \text{ m}^{-3}$)	0.119	EC _e * (dS m ⁻¹)	1.1
Clay (%)	24.1	Available soil water θ_{aw} ($\text{m}^3 \text{ m}^{-3}$)	0.119		
Classification:					
Sandy Clay Loam		Bulk density (g cm ⁻³)	1.41		

EC_e* - Electrical conductivity of the extract of a saturated soil-paste (dS m^{-1})

Table 2 shows the volumetric soil water content - $\text{m}^3 \text{ water} / \text{m}^3 \text{ soil}$ ($\text{m}^3 \text{ m}^{-3}$) - just before the water application. The volumetric soil water content in soil ranges between $0.202 - 0.155 \text{ m}^3 \text{ m}^{-3}$.

Table II.3.2. Volumetric soil water content - $\text{m}^3 \text{ water} / \text{m}^3 \text{ soil}$ ($\text{m}^3 \text{ m}^{-3}$) - just before the water application

Treatment	Determination	Θ ($\text{m}^3 \text{ m}^{-3}$)
T1	$\theta_{wp} + 0.70 \cdot \theta_{aw}$	$\Theta_1 = 0.202$
T2	$\theta_{wp} + 0.50 \cdot \theta_{aw}$	$\Theta_2 = 0.178$
T3	$\theta_{wp} + 0.30 \cdot \theta_{aw}$	$\Theta_3 = 0.155$

II.3.1.5 Climate condition in greenhouse

The average climatic data during the experimental period in the greenhouse were: maximal relative humidity – 88.4 %, minimal relative humidity – 11.3 %, maximal temperature – $45.8 \text{ }^\circ\text{C}$ and minimal temperature – $11.4 \text{ }^\circ\text{C}$.

During the experimental period, the relative humidity of the greenhouse was increased, and the maximal temperature decreased.

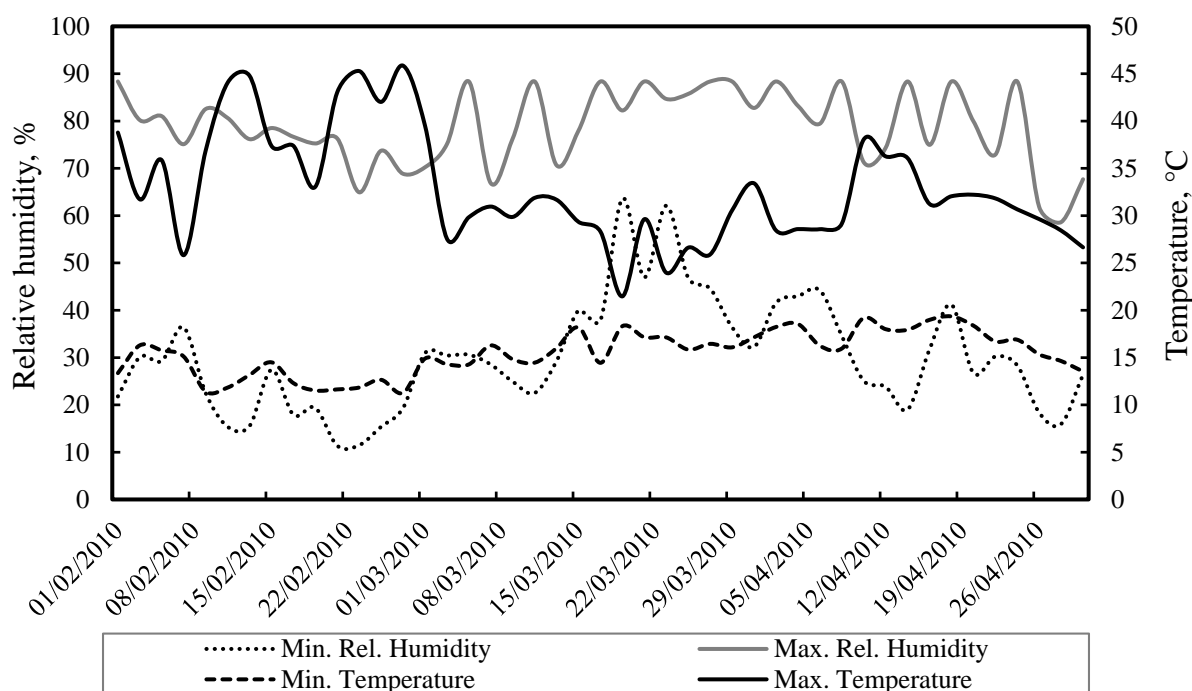


Fig. II.3.2 - Relative humidity (%) and temperature (°C) in greenhouse, February - April, 2010

II.3.2 Results and discussions

II.3.2.1 Effect of irrigation water regimes on plant growth

Table 3 shows the irrigation water regimes' effects on the *T. tetragonioides*' growth (stem length, number of nodes and number of leaves). A significant effect on the stem length can be seen. In the beginning of the experiment, the stem length of the crop showed very low variations between T1 and T2 treatments. During the last three weeks of the experimental period, the stem length increased showing equal differences between each two treatments – T1 and T2, and T2 and T3 (Δ stem length \sim .5 cm). The number of nodes and number of leaves was also higher in treatment T1.

Table II.3.3. Effect of irrigation water regimes on the stem length, nodes and number of leaves of the species

Treatment	<i>Tetragonia tetragonioides</i>		
	Stem length, cm	Number of node	Number of leaf
T1	38.8 ± 1.9 a	22.5 ± 0.6 a	9.9 ± 0.58 a
T2	34.2 ± 0.5 b	18.1 ± 0.6 b	8.1 ± 0.37 b
T3	29.3 ± 1.5 c	19.2 ± 0.8 b	9.5 ± 0.22 a

II.3.2.2 Fresh (FW) and dry (DW) weight of crop

The fresh weight (FW) of *T. tetragonioides* species shows low variations among treatments (Fig. II.3.3). There was a low increase of the fresh weight of stem, leaves and seeds in treatment T1. Surprisingly the obtained results in treatment T3 were slightly higher than in treatment T2.

The obtained results of dry matter show that the stem, leaves and seeds of treatment T1 were slightly higher than in the other treatments. There were very low variations of dry matter between T2 and T3 treatments (Fig. II.3.4).

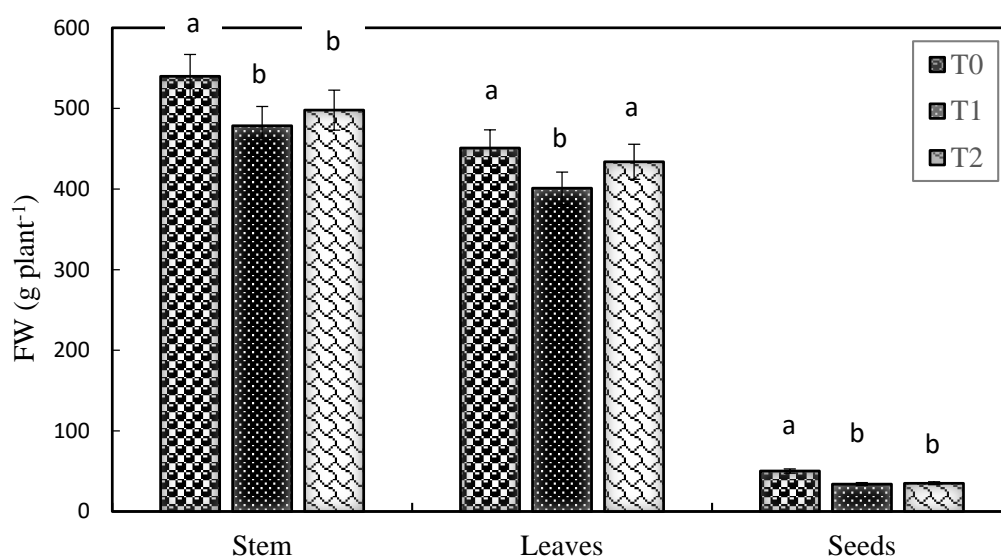


Fig. II.3.3 - Fresh weight response of *Tetragonia tetragonioides* to the different irrigation treatments

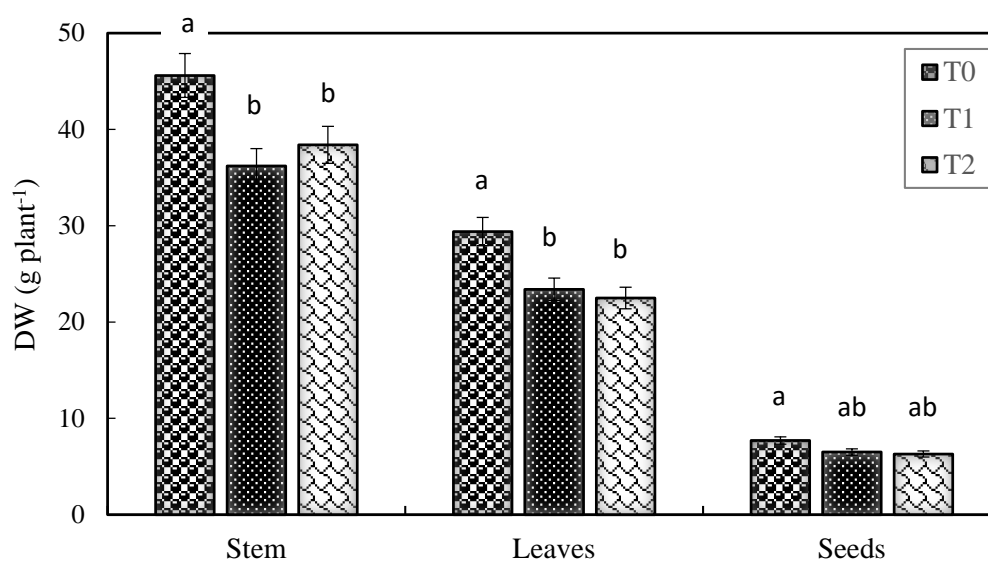


Fig. II.3.4 - Dry weight response of *Tetragonia tetragonioides* to the different irrigation treatments

II.3.2.3 Effect of irrigation water regimes on mineral composition in plant leaves

Table II.3.4 shows the effects of water application treatments on the mineral composition of *T. tetragonioides*' leaves, as follows: the total nitrogen leaf content of the species showed low variations among treatments. There was an enhancement of chloride and sodium concentration with the decrease of water content. There was a general decrease of phosphorus, calcium, potassium, iron and magnesium in the leaf content under drought conditions (Annex II.3.2).

Table II.3.4. Effect of irrigation water regimes on the leaf mineral composition

Treatment	Leaf mineral composition, (%)			
	Na	Cl	Mg	Ca
T1	3.4 ± 0.19 c	1.3 ± 0.07 c	0.43 ± 0.03 a	0.0006 ± 0.04 a
T2	4.3 ± 0.25 b	3.0 ± 0.22 b	0.36 ± 0.02 b	0.0004 ± 0.02 b
T3	4.4 ± 0.42 a	3.5 ± 0.09 a	0.35 ± 0.01 b	0.0004 ± 0.03 b
	N	K	P	Fe
T1	0.34 ± 0.02 b	4.2 ± 0.23 a	3.1 ± 0.02 a	0.0001 ± 0.02 b
T2	0.34 ± 0.01 b	3.7 ± 0.17 b	2.7 ± 0.04 b	0.0002 ± 0.03 a
T3	0.37 ± 0.01 a	4.1 ± 0.14 ab	2.7 ± 0.02 b	0.0001 ± 0.01 b

II.3.2.4. Effect of irrigation water regimes on chlorophyll content

The response of chlorophyll content in leaves of *T. tetragonioides* to the different water regimes is shown in Table II.3.5. The results show that the chlorophyll content was higher in treatment T2 and lower in treatments T1 and T3. These results are in agreement with the findings obtained by Pirzad et al. (2011) where the minimum amount of chlorophyll a, chlorophyll b and total chlorophyll were obtained from the wettest and driest treatment in *Matricaria chamomilla* L. potted plants. Similar results were obtained by Bradford and Hsiao (1982) and Chartzoulakis et al. (1993).

Table II.3.5. Leaf chlorophyll content in leaf

Treatment	Chlorophyll content					
	C _a , mg m ⁻²	C _b , mg m ⁻²	C _{a+b} , mg m ⁻²	C _a , mg g ⁻¹ (DM)	C _b , mg g ⁻¹ (DM)	C _{a+b} mg g ⁻¹ (DM)
T1	232.1 ± 12.1 b	80.9 ± 5.8 b	312.9 ± 17.6 b	26.5 ± 1.4 b	9.2 ± 0.7 a	35.8 ± 1.9 ab
T2	289.9 ± 7.3 a	110.1 ± 4.5 a	400.1 ± 11.4 a	32.3 ± 1.3 a	12.2 ± 0.5 a	44.5 ± 1.7 a
T3	253.9 ± 13.2 ab	91.6 ± 5.6 ab	345.6 ± 18.7 ab	29.8 ± 1.6 ab	10.7 ± 0.6 a	40.5 ± 2.1 ab

II.3.2.5 Effect of irrigation water regimes on carotenoid content in leaves

Carotenoids in all higher plants are synthesized and located in the chloroplast along with the chlorophyll. Table II.3.6 shows the carotenoid content of the leaves of *T. tetragonioides* under different irrigation water regimes. The maximum leaf carotenoid content was 8.44 mg g⁻¹ DW in treatment T2. On the other hand, for wetter and drier treatments (T1 and T3) the carotenoid content was lower, respectively, 7.2 and 7.9 mg g⁻¹ DW. Lower carotenoid content was also obtained for stress water regimes of some fenugreek varieties (Hussein and Zaki, 2013). Moreover, a decrease of leaf carotenoid content in green beans was attributed to water stress and the Vegetation Index (NDVI) showed the highest correlations with the chlorophyll (a, b and total) and carotene content of leaves (Koksal et al., 2010).

Table II.3.6. Carotenoid leaf content of the species

Treatment	Leaf carotenoid content	
	Car, mg m ⁻²	Car, mg g ⁻¹ (DW)
T1	62.9 ± 3.2 b	7.2 ± 0.3 b
T2	75.8 ± 2.2 a	8.4 ± 0.4 a
T3	67.6 ± 4.1 ab	7.9 ± 0.5 ab

II.3.2.6 Effect of irrigation water regimes on soluble carbohydrates content in leaves

The irrigation water regimes had a slight effect on the soluble carbohydrates' content in leaves of the species *T. tetragonioides*. The glucose and soluble carbohydrates content in leaves increased in the wet (T1) and dry (T3) treatments, respectively glucose – 0.58 and 0.57 mg ml⁻¹ and soluble carbohydrates – 1.71 and 1.67 mg. These results are confirmed by Redillas et al. (2012). There was decreased glucose (0.54 mg ml⁻¹) and soluble carbohydrates (1.59 g) content in leaves in the medium (T2) treatment (Table II.3.7).

Table II.3.7. Soluble carbohydrates content in leaves

Treatment	Soluble carbohydrates				
	Glucose, mg ml ⁻¹	Area, cm ²	Solub. Carb, mg	DW, cm ²	Solub. Carb, g
T1	0.57 ± 0.05 a	3.42 ± 0.0 a	1.67 ± 0.14 ab	0.001 ± 0.0 a	1.9 ± 0.15 a
T2	0.54 ± 0.03 a	3.42 ± 0.0 a	1.59 ± 0.08 b	0.001 ± 0.0 a	1.8 ± 0.09 ab
T3	0.58 ± 0.03 a	3.42 ± 0.0 a	1.71 ± 0.09 a	0.001 ± 0.0 a	2.0 ± 0.12 a

II.3.2.7 Yield of species

T. tetragonioides produced significant amounts of dry matter, which ranged from 82.7 to 66.1 g plant⁻¹. The partition of the plant dry matter to plant organs was changed by the effect of the irrigation water regimes (Table II.3.8). The fact that the species was irrigated during the vegetation period T1 (70 % - wet treatment), significantly increased the dry biomass of the species at the harvest time averaging 6616 kg ha⁻¹. The dry matter of the species decreased when the soil water decreased in treatments T2 (50% - medium treatments) and T3 (30% - dry treatment). There was no significant difference between treatments. The obtained results confirmed that the species *T. tetragonioides* is tolerant to drought condition. The yield of the crop shows that the drought had less effect than the salinity (6616 – 5288 kg DM ha⁻¹). These results are confirmed by the previous study of Bekmirzaev et al. (2011).

Table II.3.8. Effect of irrigation water regimes on yield of the species

<i>Tetragonia tetragonioides</i>					
Treatment	FW (g plant ⁻¹)	DW (g plant ⁻¹)	Yield, %	FM kg ha ⁻¹	DM kg ha ⁻¹
T0	1041.2 ± 12.1 a	82.7 ± 4.0 a	7.8 ± 0.3 a	83284.2 ± 967 a	6609 ± 329 a
T1	913.7 ± 22.5 b	66 ± 3.1 b	7.2 ± 0.5 a	73094.7 ± 1805 b	5289 ± 248 b
T2	966.2 ± 22.3 b	67.3 ± 3.1 b	6.8 ± 0.3 a	77300.3 ± 1787 b	5377 ± 242 b

II.3.3 Conclusions

The experimental results showed several effects of the water irrigation regimes on the growth, mineral composition and photosynthetic pigments of *T. tetragonioides*, as follows:

- Plant growth (stem, leaves and seeds) increased slightly with enhancement of the water level (near the field capacity), the growth difference between the drier water regimes is very low. This increase is probably due to the increase of stomatal conductance, and consequently, transpiration and CO₂ fixation are higher. Hence, it is not surprising that experimental results in which the only variable was water application agree quite well on this supposed theory.
- Leaf mineral composition of chloride and sodium, respectively, the main responsible ions for the soil salinization and alkalization in arid and semi-arid regions, enhanced with the decrease of the soil water content. These contents were very high in relation to other plants, showing their high capacity as salt removing species.

- There was a general low decrease of phosphorus, calcium, potassium, iron and magnesium in leaf content under drought conditions, probably due to the chloride and potassium competition.
- The total nitrogen leaf content of species showed a very low variation probably due to the same fertigation for all irrigation treatments.
- The minimum carotenoids' amount of chlorophyll a, chlorophyll b and total chlorophyll were obtained from the wettest and driest treatment in *T. tetragonioides* plants, probably due to higher plant senescence provoked by these regimes.
- The glucose and soluble carbohydrates content in leaves increased in the driest treatments and enhanced tolerance to drought condition.
- The yield of the species increased in the wettest and driest treatments.

As concluding remarks, it can be suggested that the *T. tetragonioides* are species tolerant to drought conditions. Its capacity as a halophyte and salt removing species when the soil water content decreased has been shown, suggesting its use in arid and semi-arid regions. Moreover, growth and yield differences in the several irrigation regimes were very low, which suggests the other important advantage of these species - its cultivation under dry conditions, when used as a leafy vegetable for human consumption or for animal feeding. Nevertheless, more research is needed in order to test the plant development under drier conditions, in arid and semi-arid climates.

II.4 The high salinity conditions (NaCl) effect on growth, photosynthesis, root length and mineral nutrients of species *Tetragonia tetragonioides*

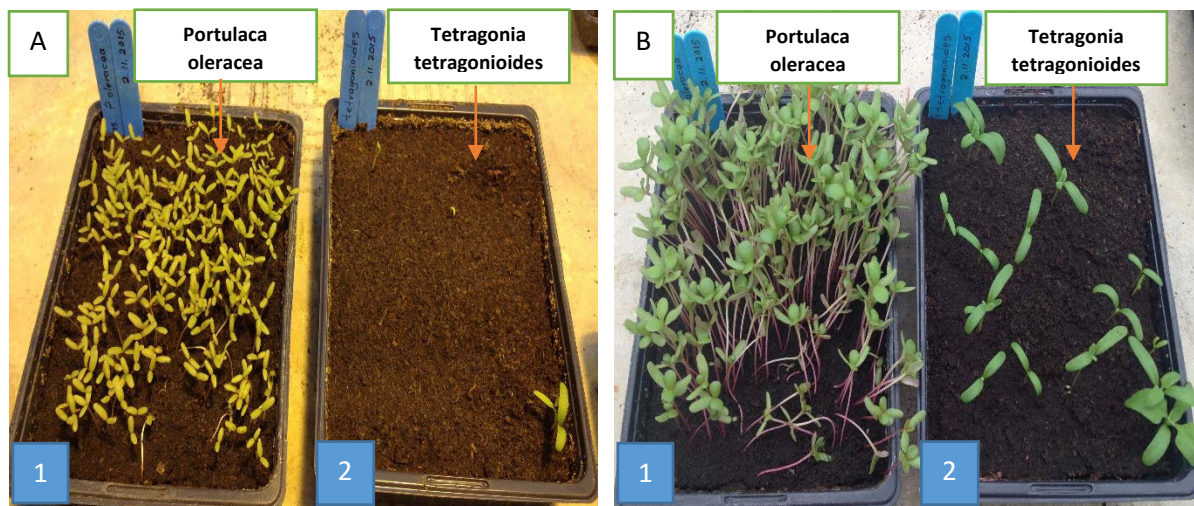
Abstract: The purpose of the experiment was to study the effect of high salinity concentration (NaCl) on growth, biomass production (total yield), photosynthesis process (chlorophyll) during the vegetation period and mineral compositions (macro – micronutrients in leaf and soil content), and the root length of the species *Tetragonia tetragonioides*. The experimental work was conducted in the greenhouse at the University of Lille 1, France, from November 2015 to January, 2016. Four treatments, in these three salinity treatments (T1, T2, T3) and a control treatment (T0) were applied. Analysis of the results showed that the total yield of the species had low variation between salinity treatments compared to the control group. The high salt concentration had effect on the macro and micronutrient contents in leaf and soil. The root length of the species showed the great effect of soil erosion in saline lands. As concluding remarks, the higher capacity of *T. tetragonioides* as salt removing species was shown. On the other hand, growth and chlorophyll content differences in the treatments were very low, which suggests the benefit of its growing under salinity conditions – the main important finding of this research.

II.4.1 Materials and methods

II.4.1.1 Experimental procedure

The experimental study was conducted in the greenhouse at the University Lille 1, France (50°36'31.8"N 3°08'43.2"E). The seeds of the species *Tetragonia tetragonioides* were sowed to soil for planting on November 2, 2015. *T. tetragonioides* seeds were germinated on November 9, seven days after seedling (Fig. II.4.1). Six leaves plants were transplanted to the three litter soil randomized pots (each pot 1300 g⁻¹ of soil) on November, 26, 2015 (Fig. II.4.2).

The species were irrigated with tap water until the beginning of the salinity treatments. The salinity treatments were the following: T0 (1-4) 0mM NaCl (control), T1 (5-8) 50 mM NaCl, T2 (9-12) 100 mM NaCl and T3 (13-16) 200 mM NaCl. The number of plants per treatment was four. The crops were irrigated salinity water in a minimal amount, enough for the plant's survival (0,25 L/pot – in the beginning of the experiment). The salinity treatments were applied with water amounts of 0.50 L/pot on December, 17. Measurement of plant germination: two days after transplantation to the randomised pots. Stem length and number of nodes were analysed each ten days during the vegetation period of the species.



Figs. II.4.1 - *T. tetragonioides* seeds were germinated (A (2) – November, 9 – after seven days and B (2) – November, 16 – after two weeks of seedling)

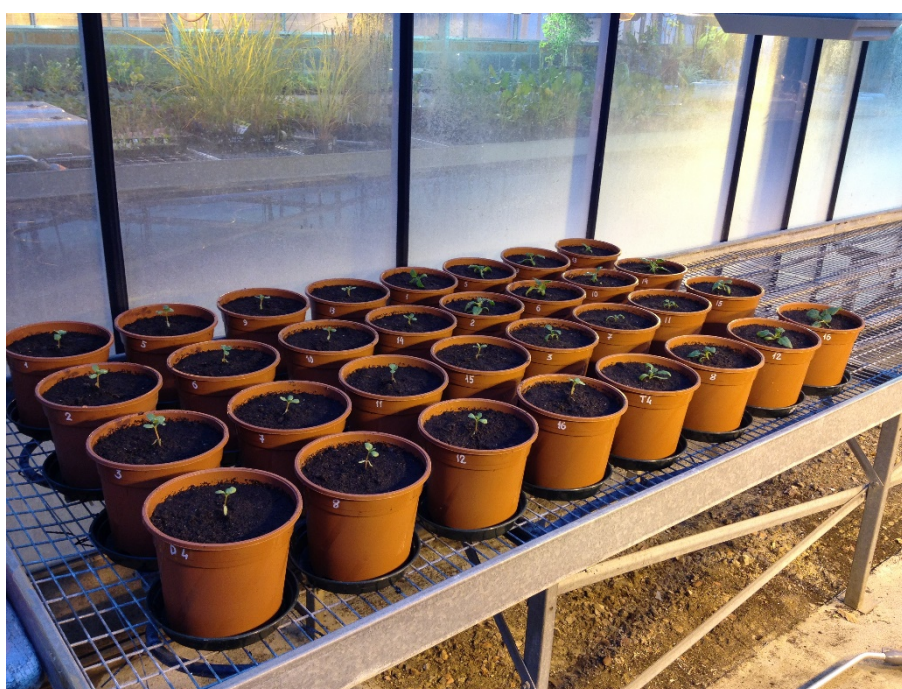


Fig. II.4.2 - *Tetragonia tetragonioides* (beginning (November, 26, 2015) of the experiment in greenhouse at the University of Lille 1, France)

At 60 days (January, 25, 2016) after salinity treatments, four plants of each treatment were harvested and washed with distil water for a few minutes, wiped with paper and the fresh weight (FW) was measured. The fresh samples were dried in a forced drought oven et 65° C for 72 hours and the dry weight (DW) was measured and plant materials for chemicals and organic analyses were collected.

The analyses of electrical conductivity (EC_w) and pH of drainage water was obtained every ten days after irrigation of plants (the analyses began on November 26) during the period of the experiment.

The soil electrical conductivity (EC_s) and pH were analysed before and after the experimental study.

The root length of the crops was determined after the experimental study.

II.4.1.2 Chemical analyses

The dried samples of leaves were used to analyze the ion concentrations. The dry materials were ground and were digested via the dry digestion method. The concentrations of copper (Cu^{2+}), manganese (Mn^{2+}), iron (Fe^{2+}), zinc (Zn^{2+}), calcium (Ca^{2+}), magnesium (Mg^{2+}), potassium (K^+), phosphorus (P) and sodium (Na^+) were determined by inductively coupled plasma-atomic emission spectrometry (ICP-AES) (Torun et al. 2013). After determining the ion concentrations, the K^+/Na^+ and Ca^{2+}/Na^+ ratios were calculated.

Dry soil and leaves were analyzed (Na^+ , K^+ , P, Ca^{2+} , Mg^{2+} , Zn^{2+} and Fe^{2+}) by using the ICP- AES method. One of the principal methods of analyzing samples that shows up frequently in the National Geochemistry Data Base is inductively coupled plasma-atomic emission spectrometry (ICP-AES). This method provides a rapid and precise means of monitoring up to 50 elements simultaneously for minor and trace levels. The ICP-AES technique is widely regarded as the most versatile analytical technique in the chemistry laboratory.

Chloride (Cl) ions were determined in the aqueous extract by titration with silver nitrate according to Radojevic and Bashkin (1999). Plant nitrogen (N) was determined by the Kjeldhal method. All mineral analyses were performed with soil and leaves.

II.4.1.3 Photosynthesis analysis

The chlorophyll content of leaves was determined by the Hansatech method (CL-01 Chlorophyll content meter). The measurement of chlorophyll content provides an indicator of photosynthetic activity related to the salt concentration of the sample. The field-portable, hand-held device determines relative chlorophyll content using dual wavelength optical absorbance (620 and 940 nm wavelength) measurements from leaf samples. Relative chlorophyll content is displayed in the range 0 – 2000 units.

II.4.1.4 Statistical analysis

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.

II.4.1.5 Climate conditions in the greenhouse

The average of climatic data during the experimental period in the greenhouse was the following: maximal temperature – 22.18 °C; minimal temperature -18.97 °C.

Along the experimental period, the temperature of the greenhouse decreased two times in the middle (November, 30) and at the end of the experiment (January, 20). The minimal temperatures were 18.9 °C and 19.2 °C on November, 30 and January, 20 respectively. The maximum temperature was 22.2 °C (November, 10) at the start of the experiment (Fig. II.4.3).

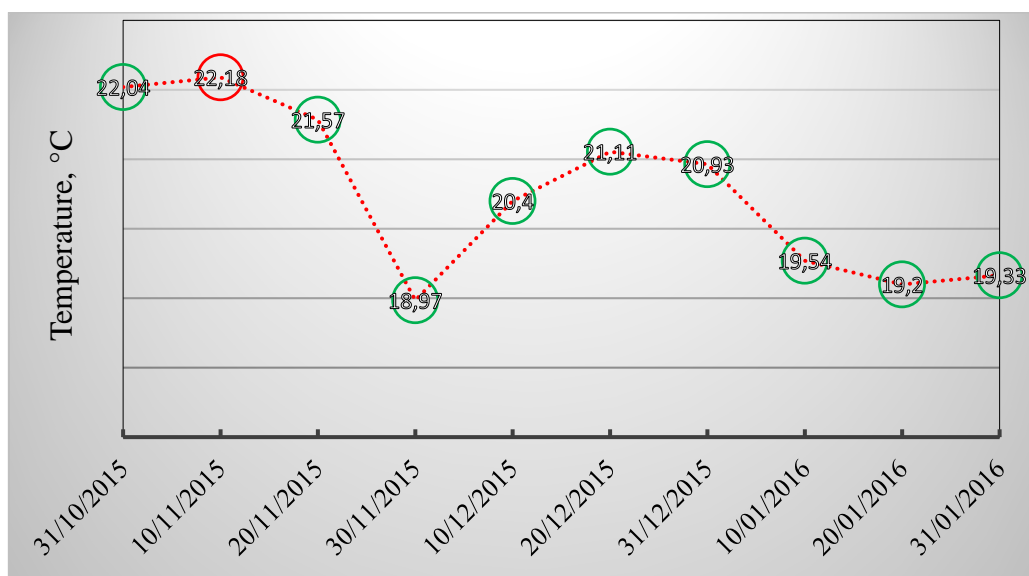


Fig. II.4.3 - Climate condition in the greenhouse

II.4.1.6 Soil

The soil for the experiment we used is NEUHAUS, which is used generally for horticultural crops in greenhouses. The soil analyses were conducted at the special laboratory of the LASIR, before the beginning of the experimental study (Table II.4.1). The pH 6 and EC_s 0.3 ($dS\ m^{-1}$) of the soil was analyzed. Table 1 shows that the mineral composition (macro – micro nutrients) in the soil contents were generally low ($\% g\ soil^{-1}$). The macronutrients' (N, P, K) percental volumes are low in the soil content. The micronutrients' volumes are very low.

Table II.4.1. Soil parameters and mineral elements (macro and micronutrients)

Soil parameters		Macronutrients (%)					
pH	EC _s (dS m ⁻¹)	N	P	K	Ca	Mg	S
6	0.3	0.014	0.032	0.166	0.880	0.212	0.142
Micronutrients (%)							
Fe	Al	Sr	Zn	Cu	Pb	Na	Cl
0.344	0.18	0.002	0.002	0.001	0.001	0.029	0.010

II.4.2 Results and discussions

II.4.2.1 pH and EC_w of drainage water

The pH and EC_w of drainage water were determined every ten days after crop irrigation during the vegetation period. Fig. II.4.4 shows that the pH of drainage water was not different between treatments (T1, T2, T3) when the species were irrigated with tap water (November, 27) and fertility dissolved water (December, 7). When they were irrigated with salinity treatments, the pH of drainage water decreased.

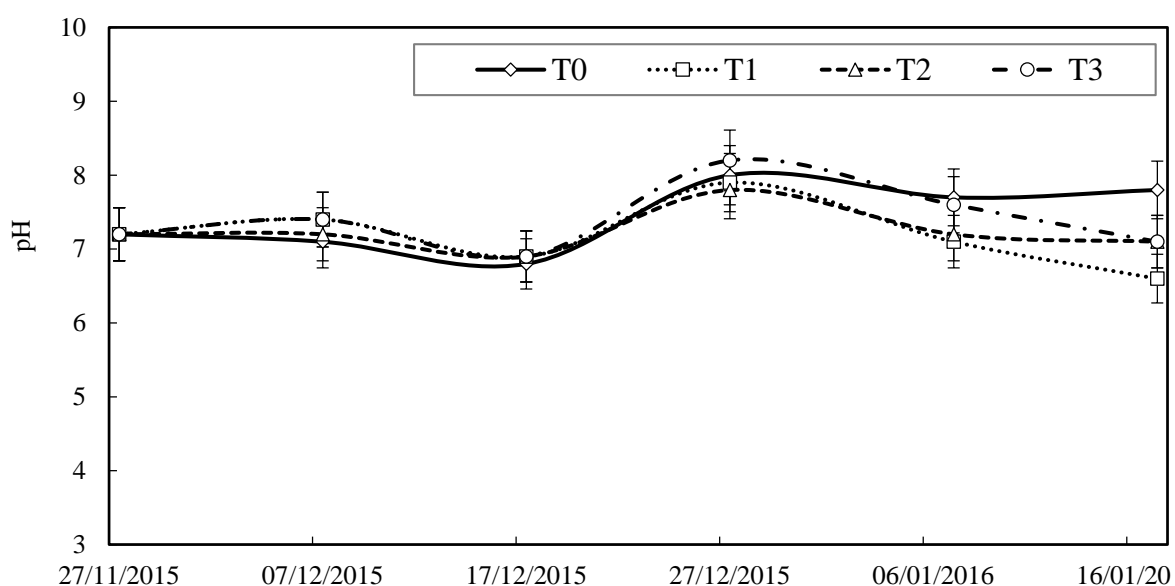


Fig. II.4.4 – pH of the drainage water

The electrical conductivity (EC_w) of drainage water was analyzed for each treatment of the vegetation period. The EC_w of drainage water slightly increased till the next irrigation of the species on the salinity treatments (Fig. II.4.5) and T0 (control) decreased. They were constant during all of the treatments in the last two irrigations of the vegetation period.

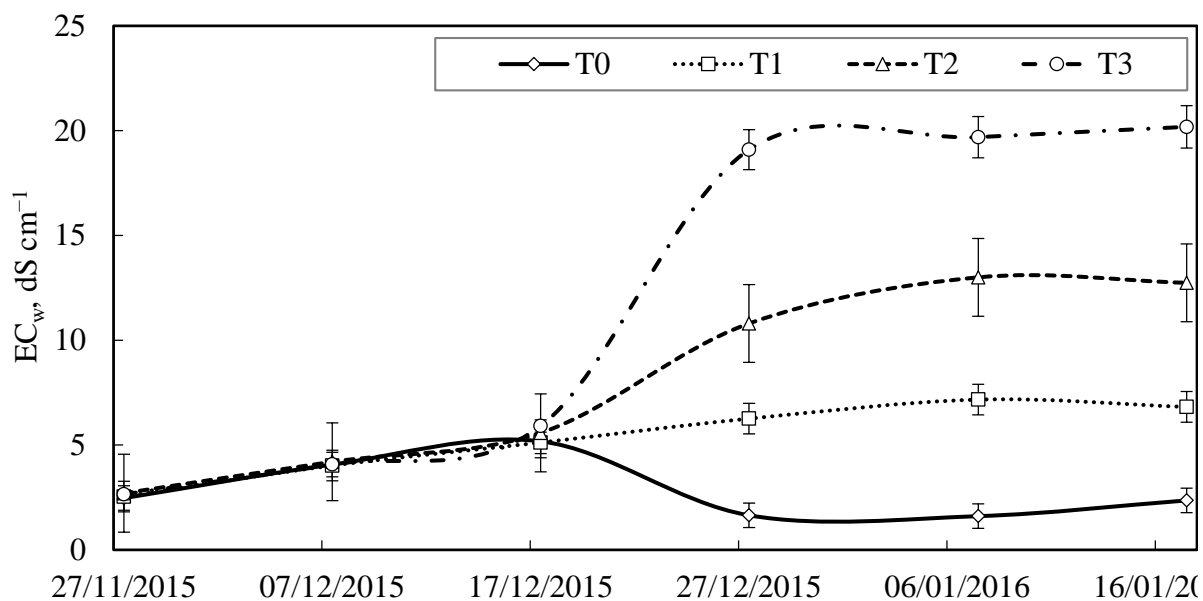


Fig. II.4.5 – Electrical conductivity (EC_w) of the drainage water

II.4.2.2 Growth rate of species

Plant growth (stem length, number of nodes and leaves of the species) was analyzed every ten days during the vegetation period (30 days). Fig. II.4.6 shows a crop developing periodically until the start of salinity treatments on December, 17. In the research study, the species New Zealand spinach was exposed to salt stress by increasing the NaCl concentration (0, 50, 100 and 200 mM NaCl) of irrigation water. The salinity treatments had a significant effect on the growth rate of the species (01.01 - 16.01.2016). The growth rate of the species in the salinity treatment slightly decreased 1.1, 1.2 and 1.3 times on 50, 100 and 200 mM NaCl treatment, compared to the control treatment (T0). Generally, the growth rate decreases with increasing salinity, while that of halophytes improves and the treatments (T1 and T2) showed no significant differences between. Thus, with an increase of salinity treatments, the species' growth decreased and this result corresponds with Maas and Hoffmann (1997). In the present study, the growth of New Zealand spinach increased under salt stress, agreeing with previous data reported on the halophytes *Salicornia europaea* and *Suaeda maritima* (Moghaieb et al., 2004) and *Alhagi pseudoalhagi* (Kurban et al., 1999), in which salt treatment at low levels improved plant growth. These results indicated that New Zealand spinach is a halophyte and so the salt tolerance of this species.

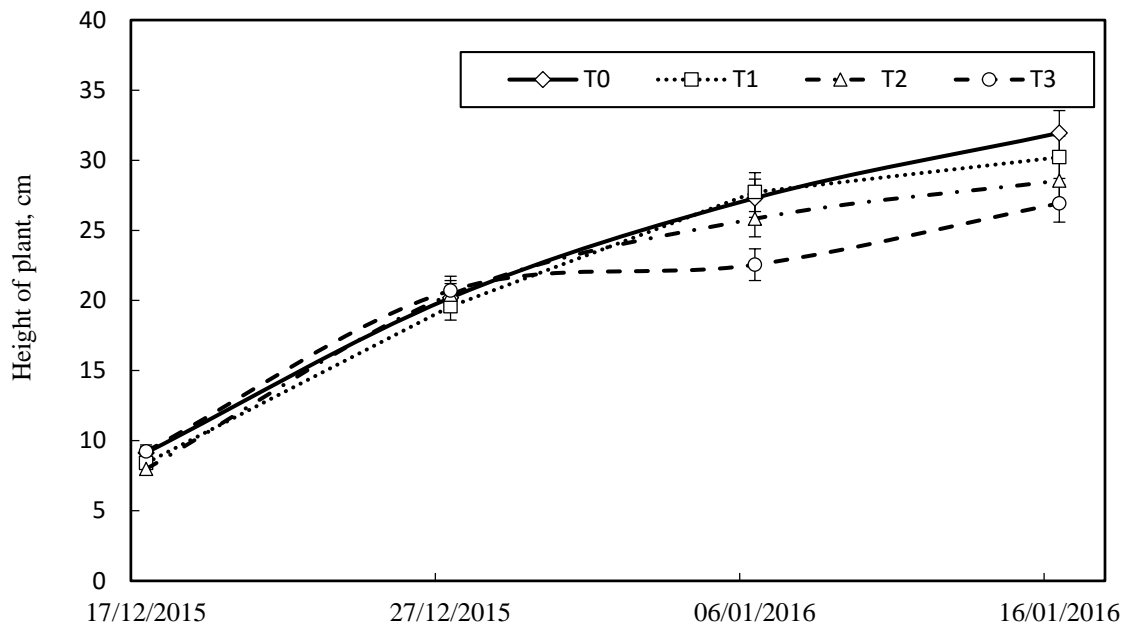


Fig. II.4.6 - Growth rate of species

II.4.2.3 Fresh and dry mass of the crop

The results related to *Tetragonia tetragonioides* have revealed that the fresh weights consistently did not decline compared to the control group from 261 g plant⁻¹ to 326 g plant⁻¹, 315 and 283 g plant⁻¹ as they were affected by increased salinity (Fig. II.4.7). The fresh weight of all treatments was different. The biomass production of stem (DW g plant⁻¹) and dry weight of leaves (DW g plant⁻¹) (where it was grown *T. Tetragonioides*) were not different between salinity treatments (Fig. II.4.8) and higher than the control treatment. The dry weight of seed (DW g plant⁻¹) was lower between salinity treatments than the dry weight of seeds (DW g plant⁻¹) of the control treatment (T0) (Annex II.4.1).

It was shown that the partition among plant organs was affected by the medium salt concentration of the species, as follows: there was an increase of the percentage of dry matter of the stems and leaves in saline conditions and a decrease in seeds. In other similar present studies of many researchers (Anac et al., 2005; Neves, 2006), the fresh and dry weight of *T. tetragonioides* significantly increased on 50 mM NaCl treatment and remained unchanged on 100 and 200 mM NaCl treatments relative to the control group. This is confirmed by the results obtained by Neves et al. (2008), who found an increased yield of salt absorbing species (*T. tetragonioides*).

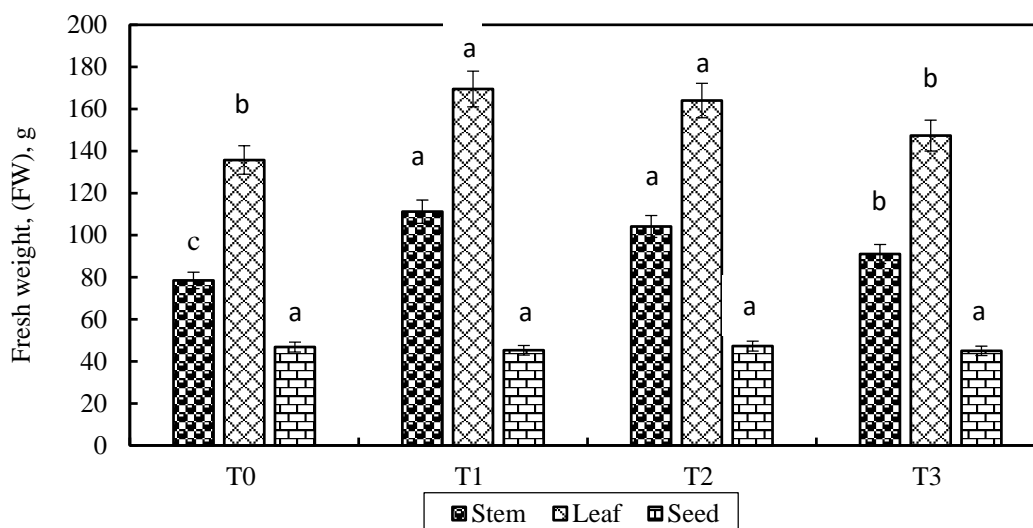


Fig. II.4.7 - Fresh weight of the species

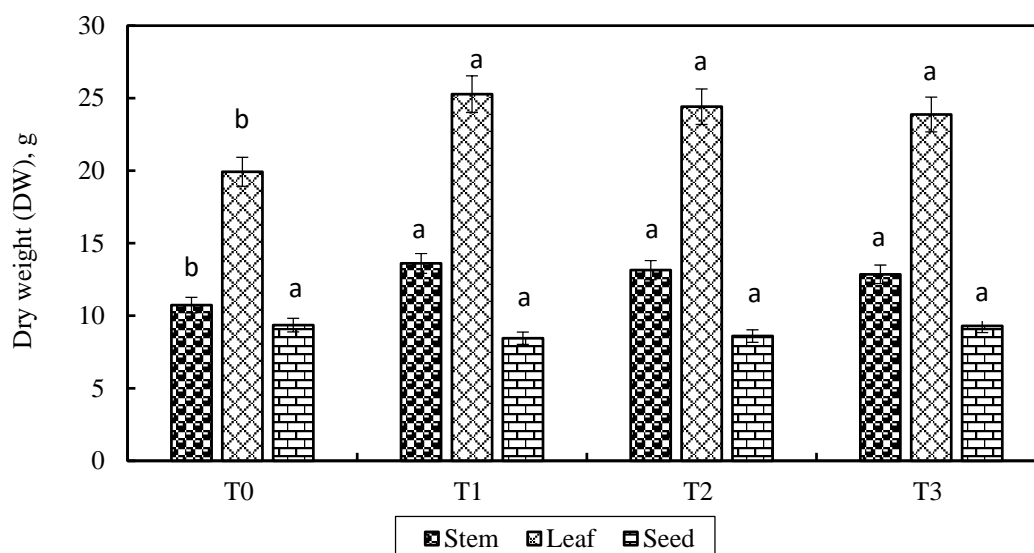


Fig. II.4.8 - Dry weight of the species

II.4.2.4 Leaf area

The leaf area is an important part of the plant to analyze the growth and predict the yield. The grid count method and gravimetric method are generally used for leaf area measurement. But these methods are laborious and time consuming when applied to a large number of leaves.

To calculate the surface of the leaf (small, medium, large and very large) (cm²) after the salinity treatments (the number of plants per treatment was four and the number of replications were four) its average values are calculated.

The leaf area (Fig. II.4.9) of the plant was slightly different in salinity treatments. The high salt concentration of irrigation water affected the leaf area in salt treatments T3. There was very low effect on the leaf area in treatments T1 and T2 compared to treatment T0.

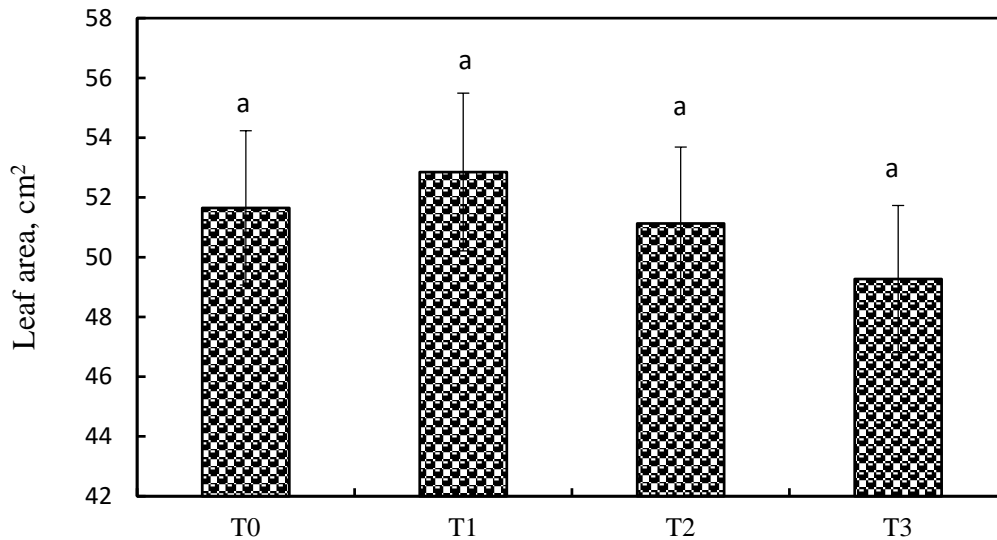


Fig. II.4.9 – Leaf area of species

II.4.2.5 Length of root

At the end of the experiment, the root length of the species was measured by a plastic ruler and compared between salinity treatments (Fig. II.4.11). The results are as follows: the root length of the species in the control treatment (T0) has an average of 49.1 cm; the root length of the species in the salinity treatments were T1 (37.3 cm) and T2 (37.1 cm). They slightly increased in the treatments T3 (42 cm) compared to the other salinity treatments. The root length of the species demonstrated that it can be grown in high salinity. In addition, the root layer of the species shows the high achievement of soil erosion in saline lands (Fig. II.4.10).



Fig. II.4.10 – Root of the species is an excellent soil covering (574 g/soil)

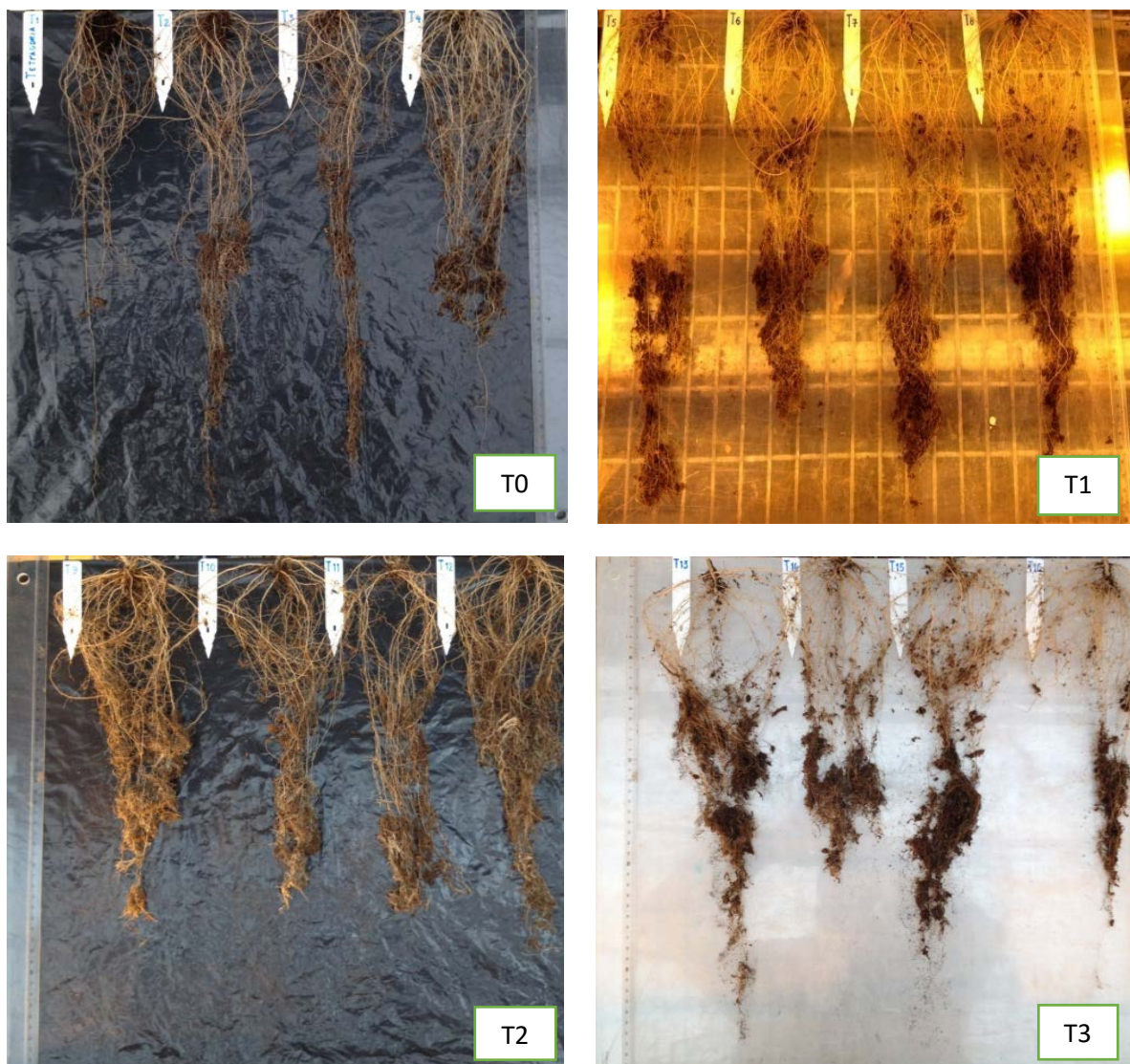


Fig. II.4.11 – Root length. The root length – 38.3 cm, fresh/dry weight of the root – 13.1/2.8 g

II.4.2.6 pH and EC_s of soil after the experiment

The pH and electrical conductivity (EC_s) in the soil was analysed by the EC meter in a dissolved soil sample at the end of the experiment. The table II.4.2 shows that the pH of the soil was decreasing when salinity treatments enhanced. It may be seen that the soil content will be alkaline. The EC_s of the soil increased with the enhanced salinity treatments.

Table II.4.2 pH and EC_s in the soil at the end of the experiment

Treatment	pH	EC _s (dS m ⁻¹)
T0	5.5 ± 0.29 a	1.0 ± 0.00 c
T1	5.0 ± 0.00 ab	2.75 ± 0.48 bc
T2	4.75 ± 0.25 b	3.75 ± 0.63 b
T3	5.0 ± 0.00 ab	7.25 ± 1.11 a

II.4.2.7 Macro and micronutrients in the leaves of the halophytic species (*Tetragonia tetragonioides*)

The relationship between the salinity and mineral nutrition of plants is extremely complex. Plant growth adversely affected by salinity induces nutritional disorders, which may result from the effect of salinity on nutrient availability, competitive uptake, transport, or partitioning within plant organs (Greenway & Munns, 1980). The macronutrients in leaf nitrogen (N), phosphorus (P) and sulphur (S) were lower in each treatment (Fig. II.4.12). In the experiment study, salt stress reduced K^+ uptake and the Na^+/K^+ ratio increased. The Ca^{2+} and Mg^{2+} concentrations in leaves of *Tetragonia tetragonioides* species decreased on salt treatment (Annex II.4.2). The increase of Na^+ in the species was associated with reduced K^+ , Mg^{2+} and Ca^{2+} , indicating a restriction in the uptake of these macro nutrients, as noted in other halophytic plants (Maggio et al., 2000; Debez et al., 2004).

Fig. II.4.13 shows that the micronutrients (Fe, Al, Sr, Zn and Cu) in leaves of the species had a very low concentration. Na^+ ions in the leaves, as observed in many reports, whereby halophytic plants accumulate and compartmentalize large amounts of Na^+ in vacuoles to lower the osmotic potential (Cheeseman, 1988; Munns, 2002; Ashraf, 2004). Surprisingly, chloride (Cl^-) in leaf content was of low concentration and there was no variation between salinity treatments.

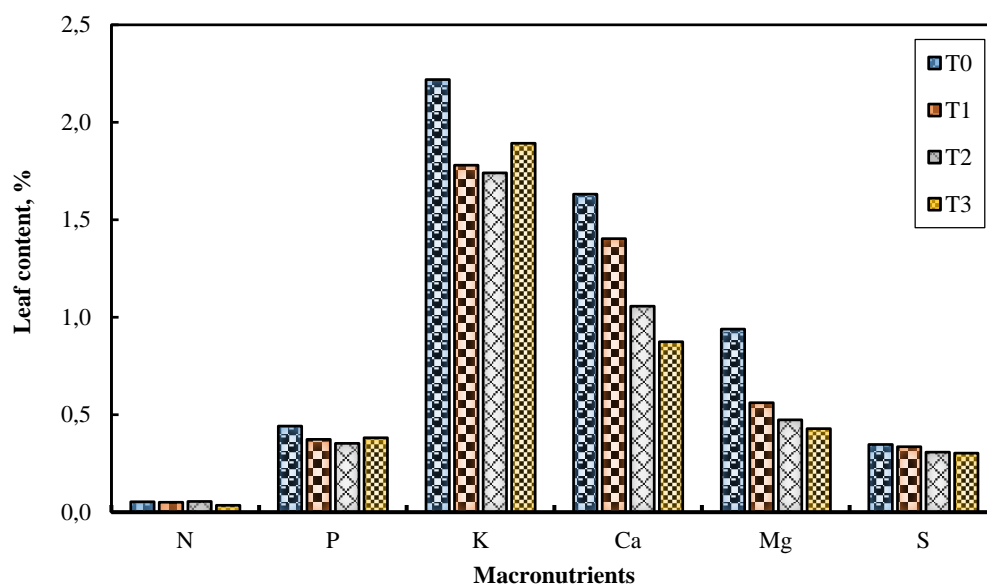


Fig. II.4.12 – Macronutrients in leaf content

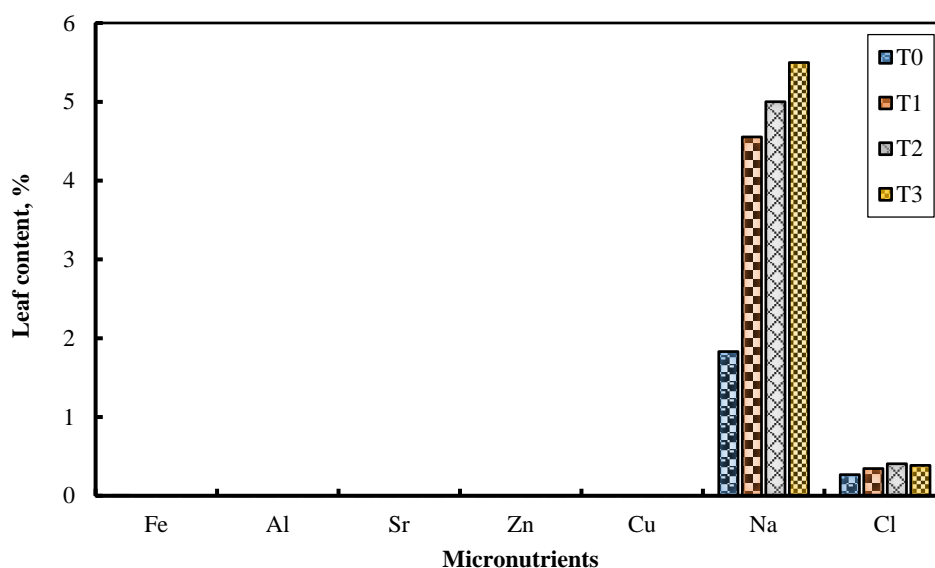


Fig. II.4.13 – Micronutrients in leaf content

II.4.2.8. Macro and micronutrients content in soil (*Tetragonia tetragonioides*)

Among the plant macronutrients, potassium plays an important role to tolerate/mitigate ill effects of high salt contents in soils. It also helps to conserve water within the plant itself. Its adequate levels in plants also enable roots to absorb/extract water from soils even under low moisture conditions. The macronutrients (N P K) are the essential elements for plant growth in soil shown (Fig. II.4.14) very low concentration (0.03 – 0.07 %) in all treatments to compare Epstein, (1965). The potassium (K) fertilizer application increased K movement from soil to root-surface. Phosphorus, like K^+ and nitrogen (N), is an important macronutrient involved in many essential functions in plant life especially in energy storage and transfer. The salinity phosphorus (P) interaction in plant nutrition is highly complex and sometimes confusing depending on the plant species, growth stage, salt types, degree of salinity and P content of growth media (Zhukovskaya, 1973). Calcium (Ca^+) has a large share in soil compared to other macronutrient contents. Magnesium (Mg^{2+}) and sulfur (S) are 0.41 – 0.47 % between treatments in soil content (Annex II.4.3).

The micronutrients in soil content (Fig. II.4.15) (were *Tetragonia tetragonioides* species was grown) were different between salinity treatments. There were low concentrations of iron (Fe) and aluminum (Al) in the soil content (0.34 – 0.72 %) for each treatment. The macronutrients Ba, Sr, Zn, Cr, Cu and Pb in the soil content were of very low concentrations (0.001 – 0.01 %). The high salt treatments had a clear effect on the micronutrients of sodium (Na) and chloride (Cl) in soil content.

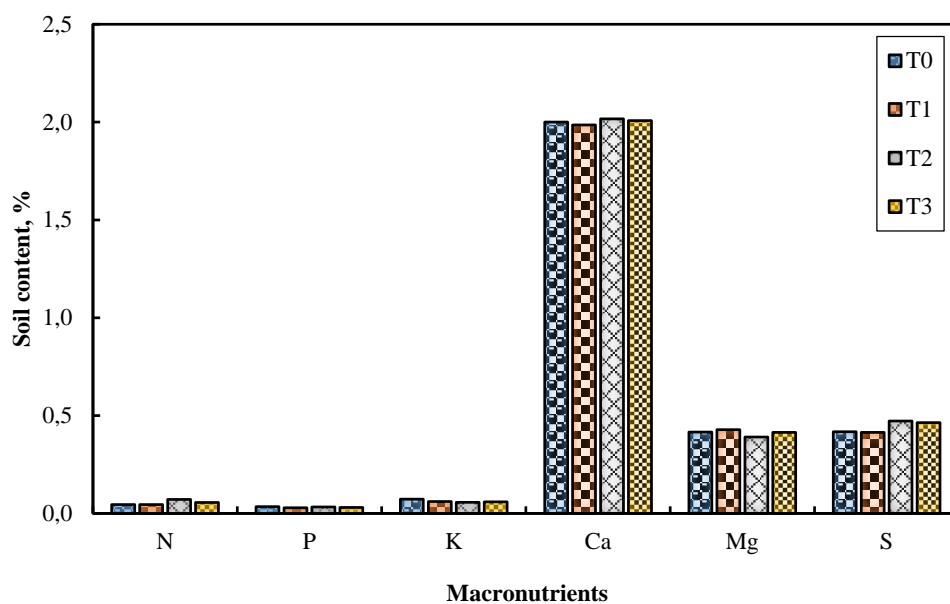


Fig. II.4.14 - Macronutrients in soil content

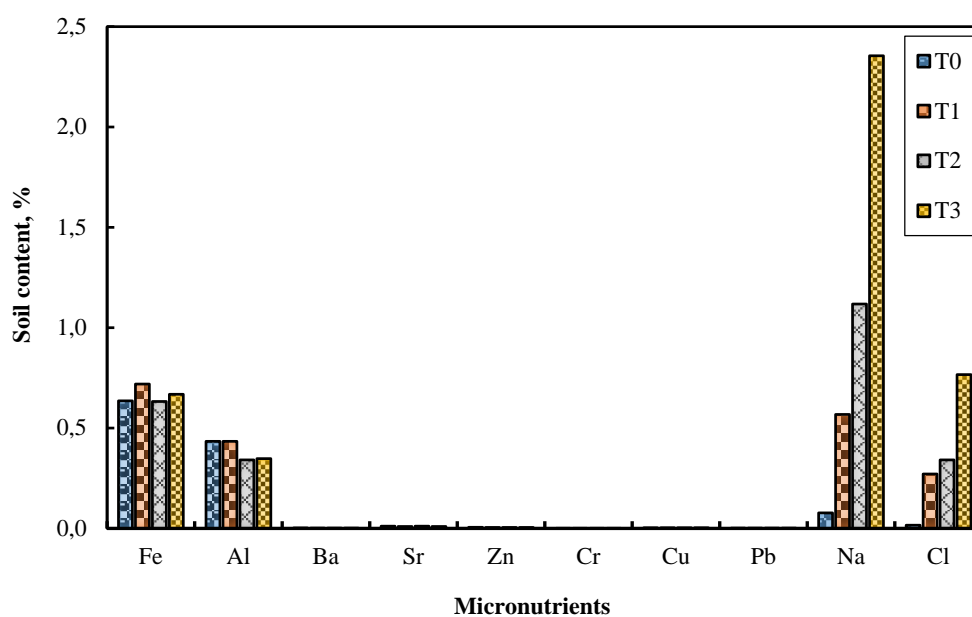


Fig. II.4.15 – Micronutrients in soil content

II.4.2.9 Yield of the species

The fresh leaves and stems are the consumed parts; therefore, the fresh weight is an important component of yields as well as plant height which was positively correlated to fresh yield. The average fresh weight of the crop ranged from 325.9 FW g plant⁻¹ (T1), 315.4 FW g plant⁻¹ (T2) and 283.4 FW g plant⁻¹ (T3) in the salinity treatments and 261 g plant⁻¹ (T0) in the control group (Table II.4.3).

T. tetragonioides produced significant amounts of dry matter, which ranged from 47,3 to 40 g plant⁻¹ and 4733 to 4000 DW kg ha⁻¹. The partition of the plant's dry matter to plant organs changed as an effect of the high salinity concentration (NaCl). Surprisingly, there was an increase of the dry matter of the species in saline conditions (T1, T2, T3) and a decrease of dry matter in the control group (T0).

Table II.4.3. Yield of species

Treatment	DW				
	FW (g plant ⁻¹)	(g plant ⁻¹)	Yield %	FM kg ha ⁻¹	DM kg ha ⁻¹
T0	261 ± 6.2 b	40 ± 1.5 b	15.3 ± 0.48 ab	31329 ± 748.4 b	4800 ± 177.5 b
T1	325.8 ± 12.1 a	47.3 ± 2.4 a	14.8 ± 0.25 ab	39114 ± 1454.2 a	5679 ± 286.9 a
T2	315.5 ± 8.3 a	46.3 ± 2.7 ab	14.5 ± 0.65 b	37851 ± 1008.9 a	5538 ± 315.9 ab
T3	283.5 ± 6.4 b	46 ± 1.4 ab	16.3 ± 0.47 a	34008 ± 751.2 b	5523 ± 149.3 ab

II.4.3 Conclusions

It can be suggested that the *Tetragonia tetragonioides* are species tolerant to high salinity conditions. The species proved to be the most potential salt (ions) removal species (Annex II.4.4). It was shown that the growth and yield differences on the several salinity treatments were low, which suggests the other important advantage of these species - its cultivation under salinity conditions, when used as a salt remover from soil and as animal feeding. In addition, it can be used for protection from soil erosion due to excellent soil covering. In conclusion, the growth of the species *T. tetragonioides* was promoted under saline conditions, indicating that the species is a halophytic. As final remarks, it is concluded that in salinity conditions, the clean and environmental safe procedures to control salinity could be associated to the conventional techniques, combining environmental and social aspects. Hence, the species may contribute to increase the soil sustainability of irrigated areas and may also be used as food crop.

II.5 Effect of saline soil (where grown just before the species *Tetragonia tetragonioides*) on growth, leaf chlorophyll content, micro-macronutrients and yield of species

Abstract: The experiment was carried out in the greenhouse at the University Lille 1, France (February, 8 to April, 26, 2016). The main purpose of the study was to confirm that the species (which was grown just before the species *Tetragonia tetragonioides*) can be used as a salt removing crop in saline soil. The species was irrigated with tap water among 0,25 – 0,50 L/pot and analysed of plant growth and leaf chlorophyll content during the vegetation period. The crop yield, macro and micronutrients were analysed at the end of the research study by using the ICP - AES method. To obtained results of the experiment is following: The results of the experiment confirmed previous conclusions.

II.5.1 Material and methods

II.5.1.1 Experimental procedure

Before planting, the seeds were soaked for three hours in warm water. They were planted 7 mm deep in the soil pots on January, 19. The seeds of the species were germinated 12 days after being planted, on January, 30.

Four leaves plants were transplanted to the three litter soil randomized pots (each pot 1300 g/soil) on February 8, and were grown just before the species *T. tetragonioides* in the several salinity treatments.

The plants were irrigated every six days with tap water of minimal amounts enough to the plant's survival (0,25 L/pot – in the beginning of the experiment). The measurement of plant germination: at four days after transplantation to the randomised pots. Height shoot, electrical conductivity (EC_w) and pH of the drainage water was analysed every six days during the vegetation period.

Plants were harvested (April, 26) after treatments, and washed with distil water for a few minutes, wiped with paper and the fresh weight (FW) was measured. The fresh samples were dried in forced drought in an oven at 65° C for 72 hours and the dry weight (DW) was measured. For chemicals analyses plant materials were collected. The soil electrical conductivity (EC_s) and pH were analysed before and after the experimental study.

II.5.1.2. Chemical analyses

Dry soil and leaves were analyzed (Na, K, P, Ca, Mg, Zn and Fe) by using the ICP - AES method. One of the principal methods of analyzing samples that shows up frequently in the National Geochemistry Data Base is inductively coupled plasma-atomic emission spectrometry

(ICP - AES). This method provides a rapid and precise means of monitoring up to 50 elements simultaneously for minor and trace levels. The ICP - AES technique is widely regarded as the most versatile analytical technique in the chemistry laboratory. Chloride (Cl) ions were determined in the aqueous extract by titration with silver nitrate according to Radojevic and Bashkin (1999). Plant nitrogen (N) was determined by the Kjeldhal method. All mineral analyses were performed in soil and leaves materials.

II.5.1.3. Photosynthesis analysis

The chlorophyll meter is a simple, portable diagnostic tool that measures the greenness or relative chlorophyll content of leaves. The measurement of chlorophyll content provides an indicator of photosynthetic activity relating to the salt concentration of the sample. Five leaf SPAD readings from each plant were taken and then averaged to have the mean SPAD reading for each replicate. The NaCl - induced effects on the total chlorophyll content were calculated and compared to the untreated control treatment value.

The root length of the crops was determined after the experimental study.

II.5.1.4 Statistical analysis

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.

II.5.1.5. Climate condition in greenhouse

Climatic conditions were monitored with an automated system at hourly intervals. Daily air temperatures ranged from 18.1 to 26.7 °C (average, 19.4 °C) in February, 18.3 to 28.4 °C (average, 20.2 °C) in March and 24.2 to 28 °C (average, 21.7 °C) in April. Night temperatures ranged from 16.3 to 22.7 °C (average, 19.5 °C) during the experiment (Fig. II.5.1).

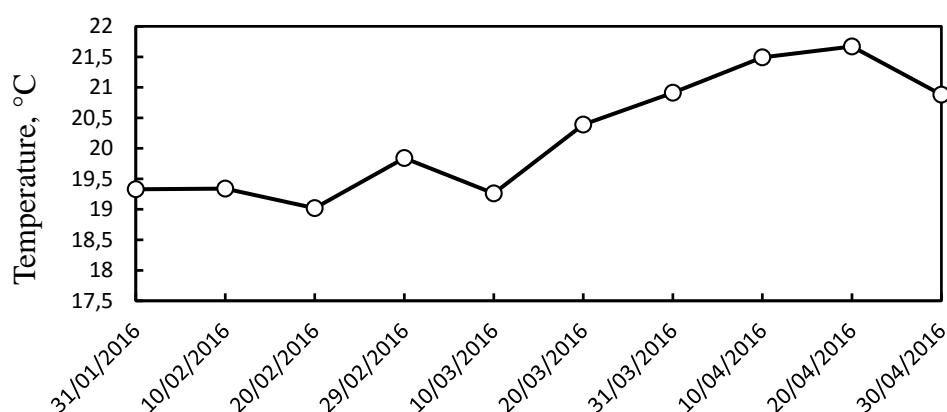


Fig. II.5.1 - Climate condition in greenhouse

II.5.1.6. Soil

The analyses of soil parameters, macronutrients (Table II.5.1) and micronutrients (Table II.5.2) of the previous experiment, were grown just before *Tetragonia tetragonioides* species. The soil chemical parameters were taken before the experiment. The pH of soil decreased and there was a slight difference between the salinity treatments. The chemical reaction is very strongly acidic up to between pH 4.6 – 5.5. The soil salinity at the saturation point is not saline treatment T0 – 0.8 dS m⁻¹, slightly saline (T1 – 2.6 dS m⁻¹ and T2 – 3.9 dS m⁻¹) and moderately saline (T3 – 7.5 dS m⁻¹). There was great increase between control and salinity treatments.

The macronutrients in soil content, nitrogen (N), phosphorus (P) and potassium (K) were of a very low concentration and there was no variation between salinity treatments. The calcium (Ca) in soil content slightly increased between macronutrients. The magnesium (Mg) and sulphur (S) had low variation between salinity treatments. It will be seen that the salinity treatments in irrigation water affected Ca in soil content.

The micronutrients iron (Fe) and aluminum (Al) were low in soil content and there was no variation between salinity treatments. Ba, Sr, Zn, Cr, Cu and Pb were very low in soil content and there was no effect of salinity treatments. The sodium (Na) in soil content increased (two times in each treatment) in salinity treatments. The chloride (Cl) in soil content had a slight variation and increased with salinity irrigation water between treatments.

Table II.5.1. Soil chemical parameters and macronutrients

Treatment	Soil parameters		Soil macronutrients (%)					
	pH	EC _s dS m ⁻¹	N	P	K	Ca	Mg	S
T0	5.5	0.8	0.04	0.03	0.07	2.00	0.42	0.42
T1	5.1	2.6	0.05	0.03	0.06	1.99	0.43	0.41
T2	4.6	3.9	0.07	0.03	0.06	2.02	0.39	0.47
T3	4.8	7.5	0.06	0.03	0.06	2.01	0.41	0.46

Table II.5.2. Soil micronutrients

Treatment	Soil micronutrients (%)									
	Fe	Al	Ba	Sr	Zn	Cr	Cu	Pb	Na	Cl
T0	0.64	0.43	0.002	0.01	0.005	0.001	0.003	0.001	0.08	0.02
T1	0.72	0.43	0.001	0.01	0.005	0.001	0.003	0.001	0.57	0.27
T2	0.63	0.34	0.001	0.01	0.004	0.000	0.003	0.001	1.12	0.34
T3	0.67	0.35	0.001	0.01	0.004	0.000	0.003	0.001	2.35	0.77

II.5.2 Results and discussions

II.5.2.1 pH and EC_w of drainage water

The results of electrical conductivity (EC_w) and pH in the soil saturated with distilled water (dS m⁻¹) are shown in Figs. II.5.2 and II.5.3. It slightly increased when the electrical conductivity (EC_w) in soil decreased. The electrical conductivity in drainage water generally decreased between treatments (T1, T2, T3) compared to the start of the experiment. There was a great decrease in treatment T3. The electrical conductivity in soil in the treatment T0 (control) did not change during the experiment. The electrical conductivity (EC_w) in saline soil decreased at the end of the experiment as follows: treatments in saline soil T1 – 1.7 times; T2 – 2.8 times and T3 – 3.4 times. These results confirm that the species of *Tetragonia tetragonioides* are the most potential salt (ions) removal species.

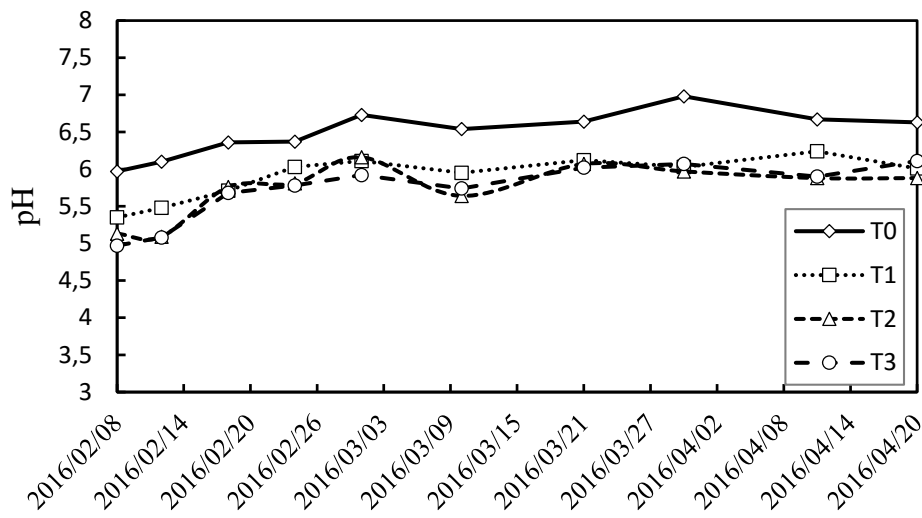


Fig. II.5.2 - pH of drainage water

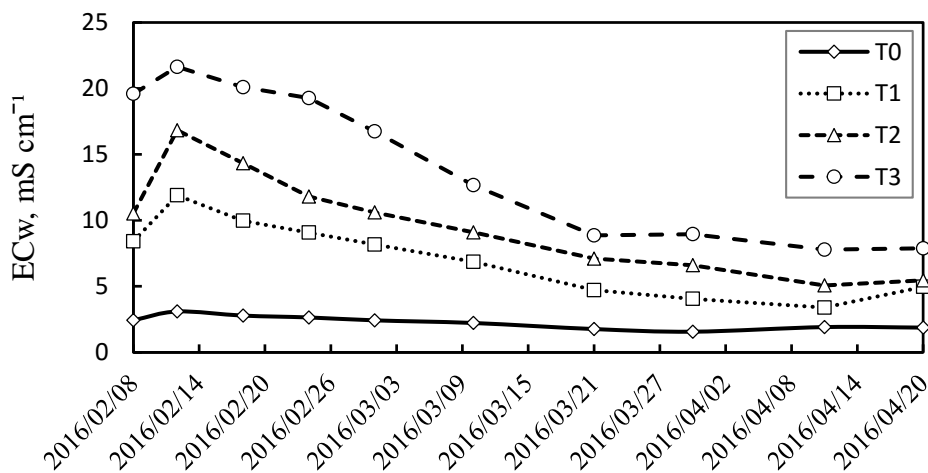


Fig. II.5.3 – Electrical conductivity (EC_w) of drainage water

II.5.2.2 Growth rate of species

The salinity soil had a significant effect on the stem length (Fig. II.5.4) of *Tetragonia tetragonioides*. The stem length of the species showed low variations between T1 and T2 treatments. This is confirmed by the results obtained by Neves (2006) and Yousif et al. (2010). On the other hand, there was a slight increase of the stem length on the T0 treatment. The stem length was increased periodically till March, 31 and there was an unexpected decrease of plant growth in the last two decades. The reason for this may be related to the vegetation period of the species.

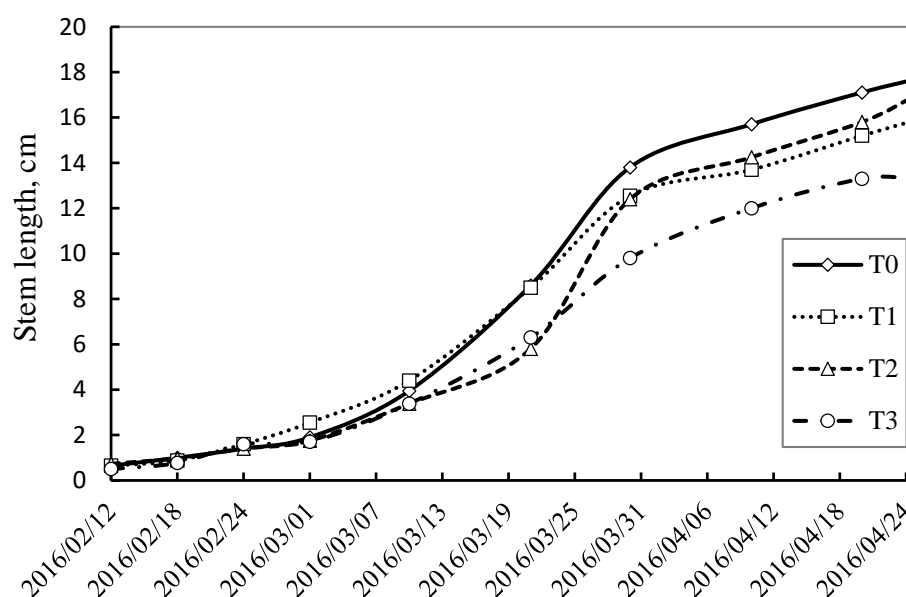


Fig. II.5.4 – Growth rate of species

II.5.2.3 Fresh (FW) and dry weight (DW) of crop

The species were grown with irrigated tap water (without fertilizer and salinity treatments) where the wild crop demonstrated to be the most potential salt (ions) removal species and the sole species that had produced significant amounts of fresh mass. Fig. II.5.5 shows that the fresh biomass production in the leaf (T0 – 30.6 g plant⁻¹) of the control treatment decreased more than the leaf (T1 – 47.4 g plant⁻¹; T2 – 38.7 g plant⁻¹; T3 – 41.1 g plant⁻¹) of saline soil. The fresh weight of the stem (T0 – 30.3 g plant⁻¹) and seeds (T0 – 23.6 g plant⁻¹) of the control treatment was lower than the fresh weight of the stem (T1 – 32.6 g plant⁻¹) and seeds (T1 – 26.6 g plant⁻¹) in salinity treatment of the species *Tetragonia tetragonioides* (Annex II.5.1).

Despite the low dry matter production (dry weight per plant at the end of the experiment (13.2 g plant⁻¹), *Tetragonia tetragonioides* grew without fertility and salinity treatments and plant yield was not greatly affected by salinity soil. In addition, plant tissues accumulated large

amounts of sodium and chloride in saline soils. Saline soils (which were grown just before *Tetragonia tetragonioides*) in several salinity treatments (Fig. II.5.6)) had produced higher dry weight per plant (17.3 g plant⁻¹) and showed, therefore, great capacity for ions accumulation. It is the only plant suitable for temporary use of agricultural crops for soil desalinization. The dry matter of stem, leaf and seeds (T0) was of lower production than the saline soil (T1). Its partition among plant organs was affected by the medium salt concentration. There was an increase of the percentage of dry matter of the leaves and seeds in saline conditions. The percentage of dry matter of stems was not of big difference in all treatments.

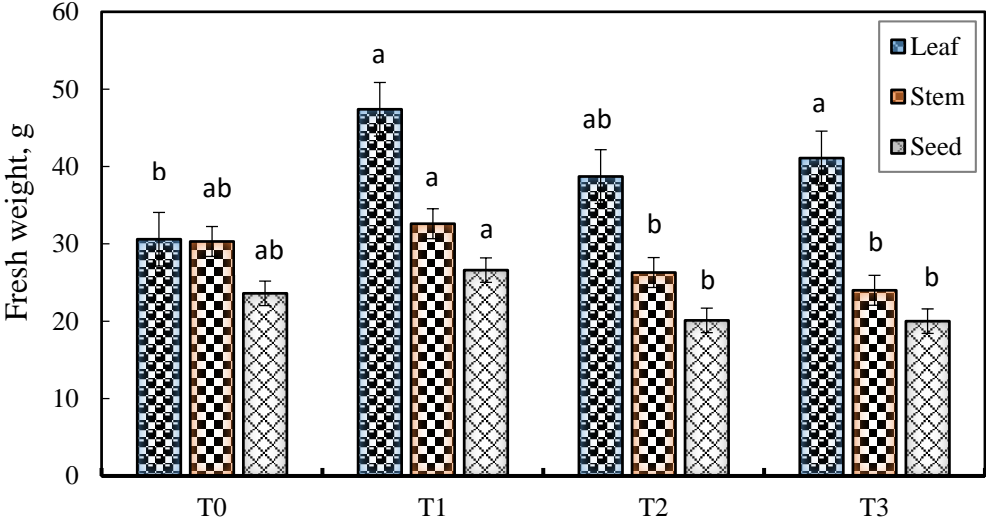


Fig. II.5.5 - Fresh weight of crop

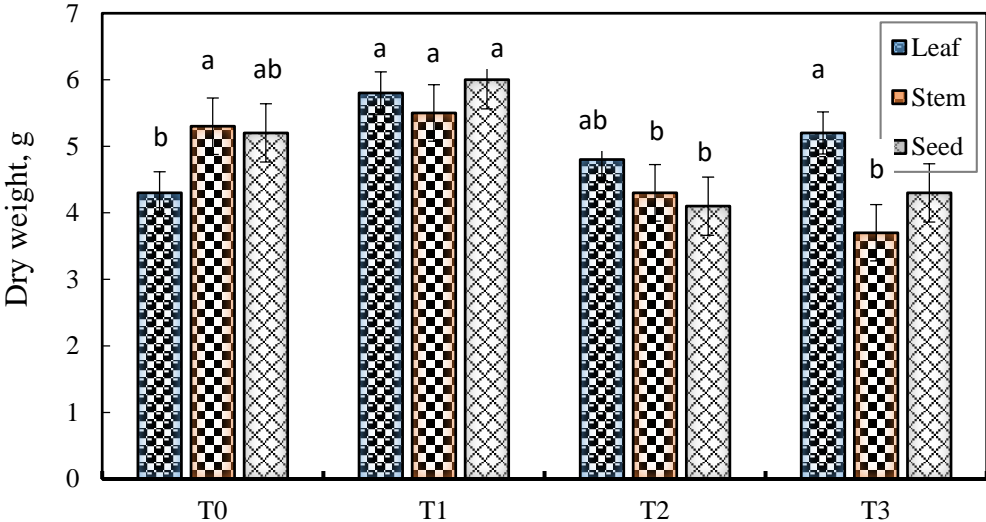


Fig. II.5.6 - Dry weight of crop

II.5.2.4 Chlorophyll content in leaf

The measurement of chlorophyll in leaf content provides an indicator of photosynthetic activity relating to the salt concentration of the sample. Despite being irrigated every six days, the chlorophyll content in leaves decreased periodically. There was a slight difference of chlorophyll content in leaves between salinity soil (which were grown just before of the species in the previous experiment). The chlorophyll content in leaves was lost.

The chlorophyll leaf contents were determined by using the device of the Hansatech each seven days. To analyse the chlorophyll leaf contents, we selected three parts of the plant (lower, middle and upper part). The analysis showed (Fig. II.5.7) that the chlorophyll leaf contents sharply decreased at the bottom of the plant in the next 7 days. This was more than double the height compared to the upper part of the plant. In the saline soil the chlorophyll leaf content decreased as follows: T0 – 1.17 times; T1 – 1.15 times; T2, T3 and 1.19 times – 1.27 times. This figure after 2 weeks is as follows: T0 – 2.57 times; T1 – 1.91 times; T2 and T3 – 3.7 times.

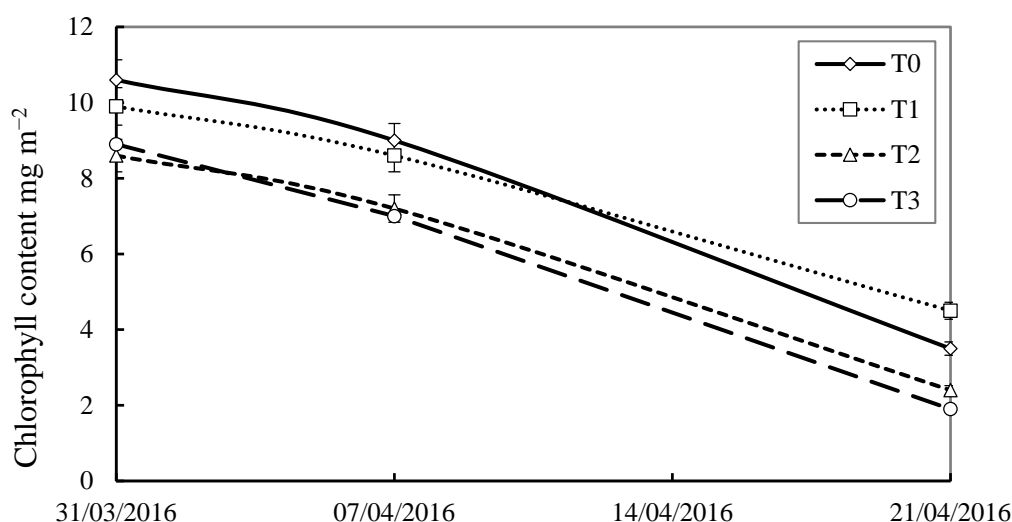


Fig. II.5.7 – Chlorophyll content in leaf

II.5.2.5 Leaf area

The leaf area (Fig. II.5.8) of the plant showed low variation between treatments. The high electrical conductivity (EC_s) of soil slightly affected the leaf area in the treatments T2 and T3. Saline soil did not affect the leaf area in treatment (T1) compared to T0 - control.

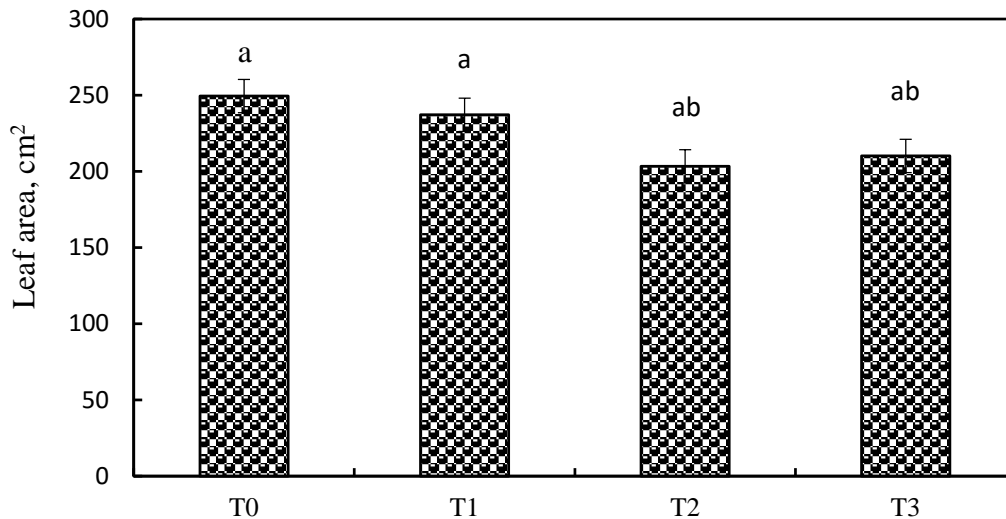


Fig. II.5.8 – Leaf area of crop

II.5.2.6 Root length of crop

The root length of the species *Tetragonia tetragonioides* was measured at the end of the experiment (Fig. II.5.9). The results are as follows: the root length of the species in the control treatment (T0) had an average of 53.9 cm; the treatments in saline soil T1 (43.3 cm); T2 (49.7 cm) and T3 (42.9 cm). There was a slight increase in the treatment T2 in saline soil compared to the other treatments in saline soil (T2 and T3). The root length of the species demonstrated that it can be grown in saline soil.

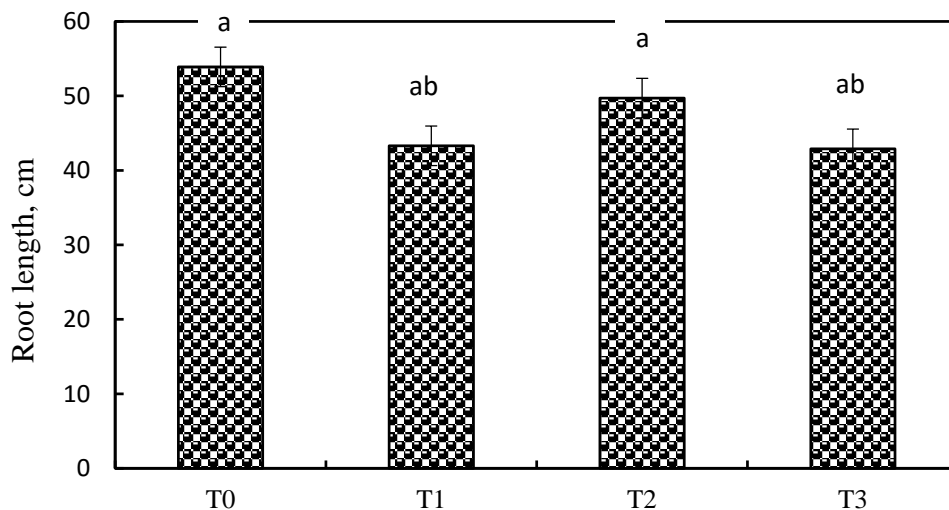


Fig. II.5.9 – Root length

II.5.2.7 Macro and micronutrients content in leaf

The salinity of irrigation water had a significant effect on the macro and micro nutrients of *Tetragonia tetragonioides*' leaf content for the majority of the analysed mineral elements, as

follows (Figs. II.5.10 and II.5.11): 1) the total nitrogen leaf content was not analysed because of very low concentration among treatments; 2) a slight increase of potassium (K) and calcium (Ca) leaf content was shown, with the saline water increment; 3) a low decrease of magnesium (Mg) and a very low decrease of sulphur (S) leaf content was verified when the salt concentration of irrigation water was greater; 4) the salinity did not affect the micronutrients' (Fe, Al, Sr, Zn and Cu) content of leaf. Similarly, in a previous study conducted by Anac et al. (2005); 5) there was a great increase of sodium concentrations, with the increase of salinity concentration; 6) the salinity did not affect the chloride content of the leaf (Annex II.5.2).

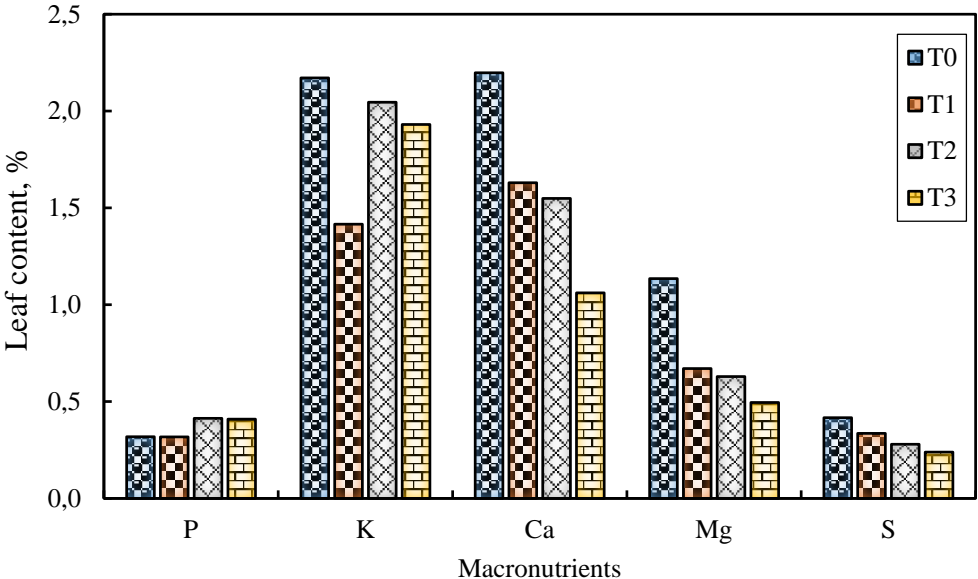


Fig. II.5.10 – Macronutrients content in leaf

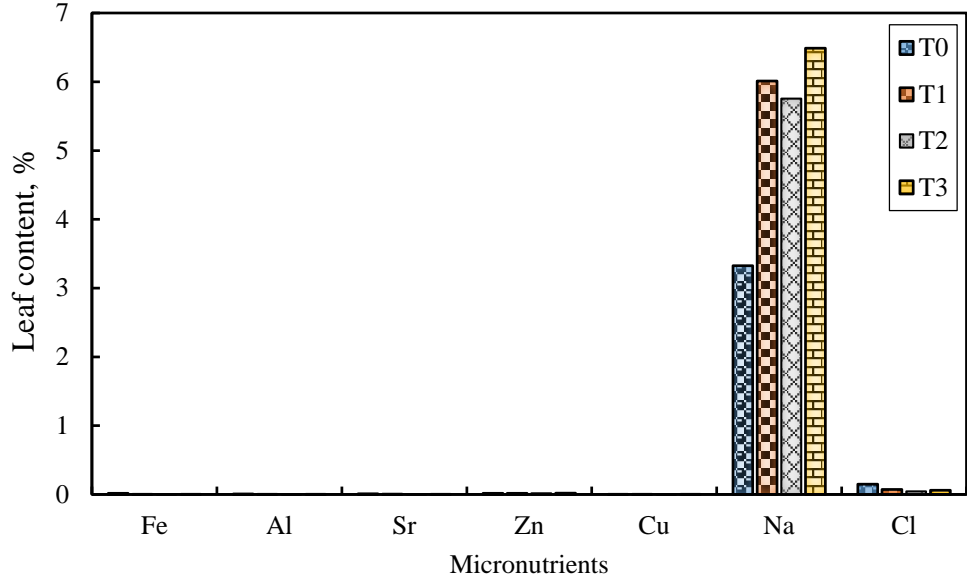


Fig. II.5.11 – Micronutrients content in leaf

II.5.2.8 Macro and micronutrients content in soil

The salinity had a significant effect on the soil's macro and micronutrients of *Tetragonia tetragonioides* for the majority of the analysed mineral elements, as follows (Figs. II.5.12 and II.5.13): 1) the total nitrogen (N) and phosphorus (P) soil content of species showed very low variations among treatments, apparently not related to the salinity; 2) the potassium (K) soil content showed low variations between treatments, 3) there was a slightly decrease of magnesium (Mg) and sulphur (S) soil content of crop; 4) the calcium (Ca), 1.9 – 2.3 % (g sample⁻¹ of soil) soil content slightly increased between macronutrients in treatments; 5) there was a slightly increase of iron (Fe) and aluminium (Al) soil content in each treatment. It will be confirmed when soil pH decreased from 6.5, the soil has become more acidic, the solubility of the elements was increased; 6) The other micronutrients (Ba, Sr, Zn, Cu, Pb) were of very low concentration in the treatments; 7) there was a great increase of sodium concentrations when water salinity increased; 8) chloride concentrations of the soil were of low concentration, but different between treatments; 9) the salinity of irrigation water affected phosphorus and iron leaf content (Annex II.5.3). In similar previous studies (Neves et al., 2008), the mineral contents of *T. tetragonioides* which are Mg, P, N decreased, and Na, K, Ca (Munns, 2002).

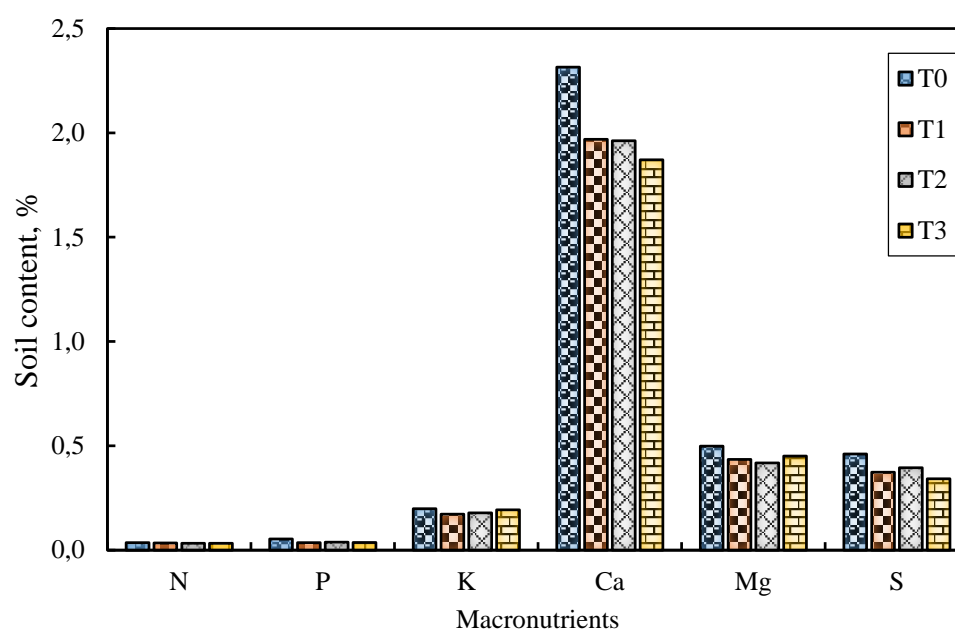


Fig. II.5.12 – Macronutrients content in soil

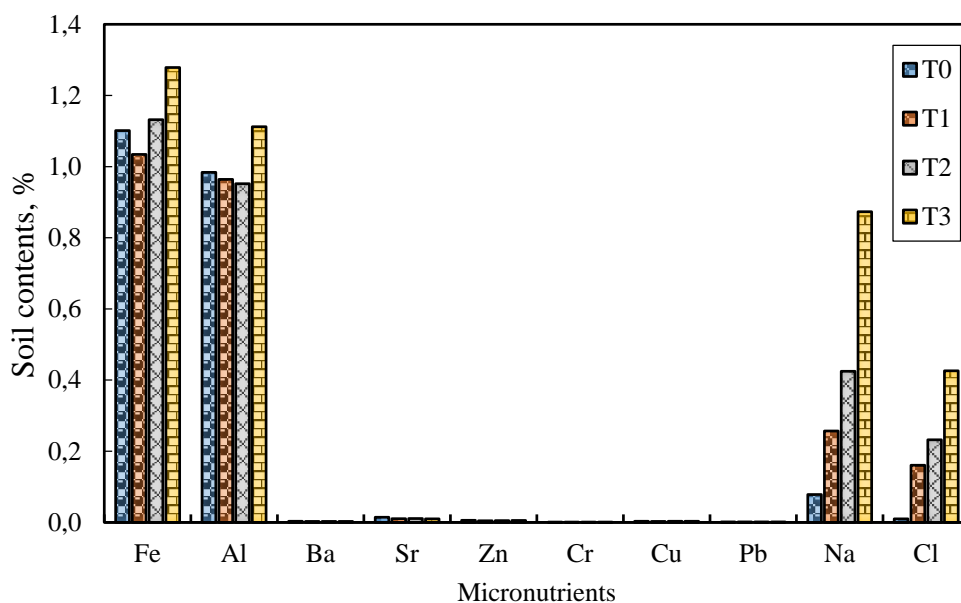


Fig. II.5.13 - Micronutrients content in soil

II.5.2.9 Yield of species

The yields of crop in saline soil on the treatments (T1, T2, T3) were slightly different compared to the non-saline soil in the control group (T0). The fresh weight averaged from 106.7 (12798 kg ha⁻¹) – 84.6 g plant⁻¹ (10150 kg ha⁻¹). The dry weight of the species averaged from 1582 – 1770 DW kg ha⁻¹. The fresh and dry weights of species significantly increased between treatments. These values show that the species *Tetragonia tetragonioides* is salt tolerant and that it can remove salts from soil in the previous experiment (Table II.5.3).

Table II.5.3. Yield of species

Treatment	<i>Tetragonia tetragonioides</i>				
	FW (g plant ⁻¹)	DW (g plant ⁻¹)	Yield, %	FM kg ha ⁻¹	DM kg ha ⁻¹
T0	84.5 ± 5.2 b	15.0 ± 2.6 ab	17.3 ± 0.5 a	10149 ± 642.5 b	1770.3 ± 158.5 ab
T1	106.5 ± 6.1 a	17.3 ± 1.1 a	16.3 ± 0.6 a	12798 ± 717.1 a	2072.8 ± 138.6 a
T2	85.3 ± 17.4 b	13.5 ± 2.7 c	15.3 ± 0.5 ab	10200 ± 2067.5 b	1580.8 ± 322.9 b
T3	85.0 ± 18.6 b	13.3 ± 3.3 c	14.8 ± 1.3 b	10206 ± 2232.6 b	1591.0 ± 403.0 b

II.5.3 Conclusions

T. tetragonioides showed to be a valuable salt (ions) removal species. In the experiments, *T. tetragonioides* produced significant amounts of dry matter. The reasons were: fast rate growth (0.31 – 0.52 cm per day), deep root length (42.9 – 53.9 cm), high biomass production (12798 FW kg ha⁻¹), easy cropping (as winter or summer crop), and absence of diseases and pests of

major importance, as well as good acceptance by local consumers as a leafy vegetable. The obtained results suggest that plants can be grown in high salinity levels (NaCl) in soil. It was shown that this new technique to control salinity is a powerful and environmental clean tool to maintain the sustainability of the agricultural areas and landscape; however, additional research is needed. Taken into account the values of sodium and chloride content observed in *T. tetragonioides*, it could be extrapolated potential soil removal of these mineral elements by the species on different saline conditions. From the data presented, it could be expected that in very saline soils, that species could extract just to 200 - 450 kg ha⁻¹ of sodium and 300 - 500 kg ha⁻¹ of chloride. *T. tetragonioides* partition among plant organs was affected by the medium salt concentration. There was an increase of the percentage of dry matter of the leaves in saline conditions and a decrease in seeds. The reduction of biomass production by salinity, from a horticultural point of view, could be counterbalanced by the relative increase of the marketable part of the crop (leaves). The reduction of the seeds' production of plants in saline conditions showed that the reproductive system was more susceptible to salinity than the vegetative system.

II.6 Salinity (NaCl) effects on growth, photosynthesis, root length and macro-micronutrients of the species *Portulaca oleracea*

Abstract: The purpose of the experiment was to study the effect of high salinity concentration (NaCl) on growth, total yield during the vegetation period and mineral compositions (macro – micronutrients content in leaf and soil), and the root length of the species *Portulaca oleracea* at the end of the experiment. The experimental work was conducted in the greenhouse at the University of Lille 1, France, January to March, 2016. Four treatments were adopted for the study, of which three salinity treatments NaCl (T1 – 50 mMol, T2 – 100 mMol, T3 – 200 mMol) and a control group (T0). Analysis of the results showed that the total yield of the species had low variation between salinity treatments compared to the control group. The high salt concentration affected the macro and micro - nutrients in leaf and soil contents. As concluding remarks, an increase of the higher capacity as salt removing species of *P. oleracea* with the high salinity treatments was shown.

II.6.1 Materials and methods

II.6.1.1 Experimental procedure

The experimental study was conducted in the greenhouse at the University of Lille 1, France, January to March, 2016. The experiment carried out salinity treatments on the selected halophytic species of *Portulaca oleracea* (Fig. II.6.1).

Seeds of the crop were sowed in soil to obtain seedling on January, 10, 2016. The plant shoots four days after sowing (January, 14, 2016).

Four leaves plants were transplanted to three litter soil randomized pots (each pot 1300 g/soil) 12 days after the plants' shooting on January, 26, 2016. At the start of the experiment plants were irrigated with tap water of minimal amounts enough to the plant's survival (0,25 L/pot). The number of plants per treatment was four. The treatments were the following: T0 (1-4) 0 mM NaCl (control), T1 (5-8) 50 mM NaCl, T2 (9-12) 100 mM NaCl and T3 (13-16) 200 mM NaCl. Plants were irrigated with salinity water with amounts of 0.50 L/pot on February, 9. During the vegetation period with the development of the plant water demand had increased.

Analysis of plant germination began two days after transplantation of the species to randomised pots. Stem length and number of nodes of the species were analysed each five days during the vegetation period in the experiment.



Fig. II.6.1 – *Portulaca oleracea* (the greenhouse experiment at the University Lille 1, France)

At 50 days (Mart, 15, 2016) after salinity treatments, four plants one of each treatment were harvested and washed with distilled water for a few minutes, wiped with paper and the fresh weight (FW) was measured. The fresh samples were dried in a forced draught oven at 70° C for 48 hours and the dry weight (DW) was measured and plant materials for chemical and organic analyses were collected.

The electrical conductivity (EC_w) and pH of the drainage water were obtained every ten days (the analyses began on January 28) in the period of the experiment.

The soil electrical conductivity (EC_s) and pH were analysed before and after the experimental study.

II.6.1.2 Chemical analysis

Mineral elements (Na, K, P, Ca and Mg) of the soil (before and after the experiment) and plant leaves (only after the experiment) were analysed by the ICP (Inductively Coupled Plasma and Optical Emission Spectrometer: ICP - OES). Chloride ions were determined in the aqueous extract by titration with silver nitrate according to Radojevic and Bashkin (1999). Plant leaves and soil nitrogen (N) content were determined by the Kjeldhal method. All mineral analyses were performed on soil and leaves.

II.6.1.3 Statistical analysis

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.

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II.6.1.4 Climate conditions in greenhouse

Climatic conditions of the greenhouse were monitored with an automated system at hourly intervals (every 12 minutes). Daily air temperatures ranged from 20.1 to 24.2 °C (average, 22.2 °C) and night temperatures from 16.8 to 19 °C (average, 17.9 °C) during the experiment period (Fig. II.6.2).

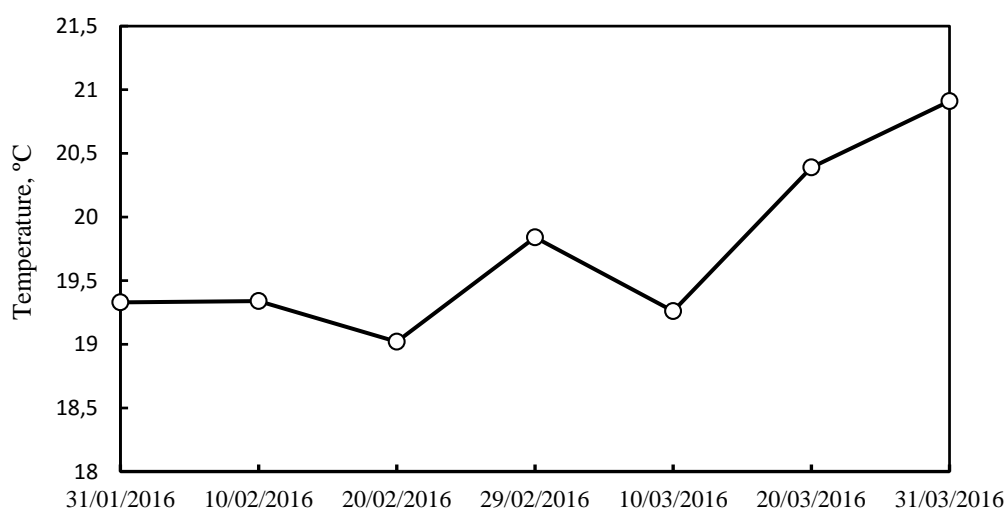


Fig. II.6.2 - Climate conditions in greenhouse

II.6.2 Results and discussions

II.6.2.1 pH and electrical conductivity (EC_w) of drainage water

The pH of drainage water showed no differences between treatments. Fig. II.6.2 shows that the pH of drainage water decreased when the electrical conductivity (EC_w) of irrigation water in the salinity treatments increased.

The electrical conductivity (EC_w) of drainage water was increased at the start of the salinity treatments (two weeks after transplantation of plants, February, 9). There was general increase with each salinity treatment (Fig. II.6.3). The electrical conductivity (EC_w) of drainage water in the salinity treatment T3 greatly increased on March, 7. The treatment (T0) periodically decreased during the vegetation period of the species *Portulaca oleracea*.

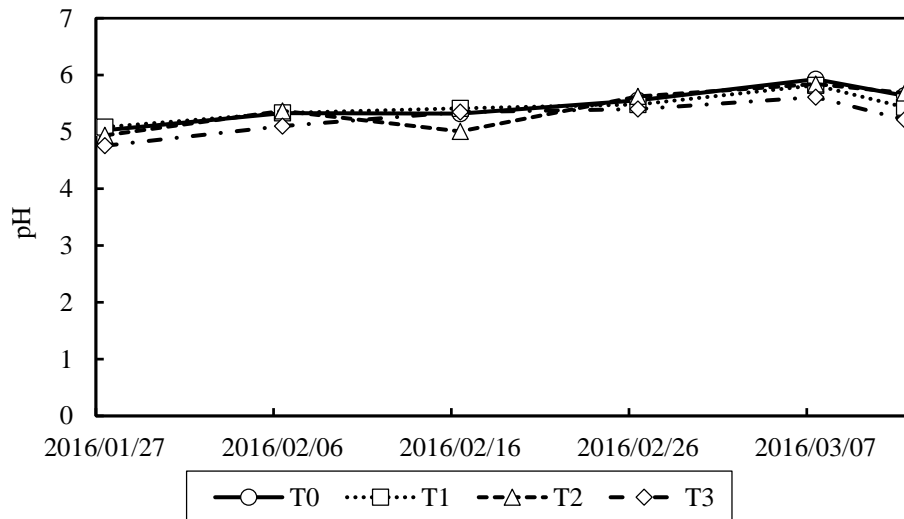


Fig. II.6.2 – pH of drainage water

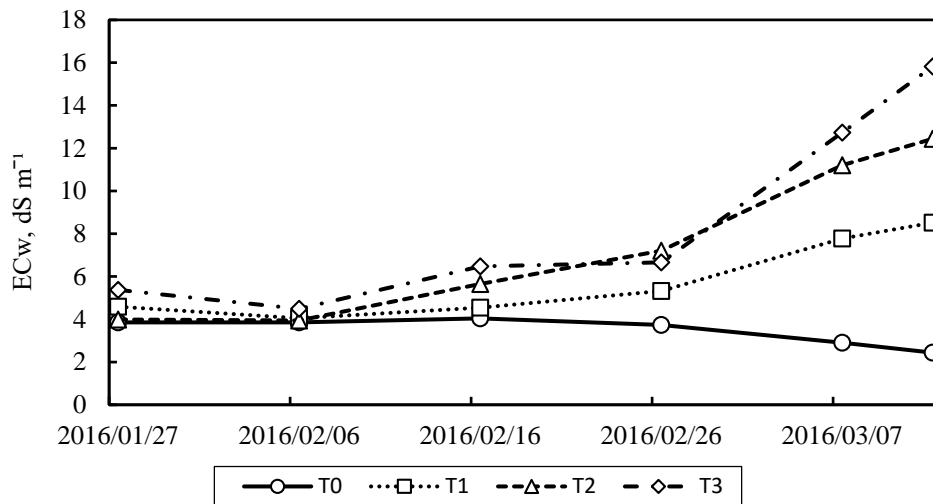


Fig. II.6.3 – Electrical conductivity (EC_w) of drainage water

II.6.2.2 Stem length of crop

Fig. II.6.4 shows that the irrigation water with salinity treatments affected the growth of stem length of the species *Portulaca oleracea*. In the beginning of the experiment the stem length of the species was 2.5 – 2.9 cm. The crop was irrigated each treatment with tap water 200 - 250 ml pot⁻¹ (26.01 - 9.02.2016) until the start of salinity treatments (salinity concentration NaCl (T0 (control) – 0 mM, T1 – 50 mM, T2 - 100 mM and T3 - 200 mM)). The species were irrigated (500 ml pot⁻¹ until February, 21) with several concentrations of sodium and chloride (NaCl) on February, 9. The electrical conductivity (EC_w) of saline water significantly affected all plant growth of the species. In the beginning of the experiment, stem length of the crop showed very low variations between all treatments. The species was flowered on February, 24, one-month after plant transplantation to the pots. On the other hand, during the last three weeks in the

vegetation period of the species, stem length showed differences between treatments – T0 and T1, T2 and T3 (Δ stem length \sim 6 - 8 cm).

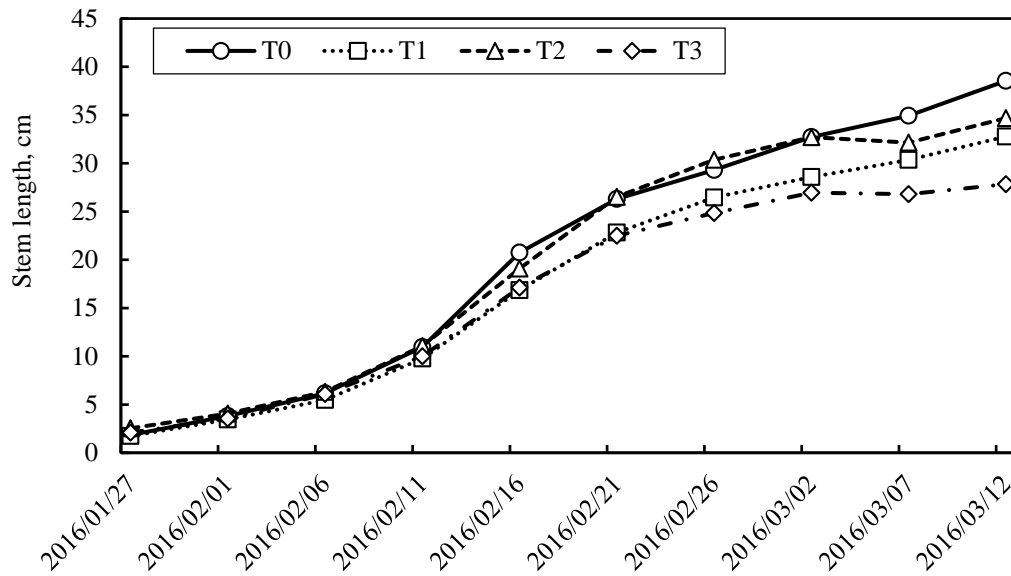


Fig. II.6.4 – Stem length

II.6.2.3 Leaf area

To calculate the leaf area of the species (small, medium, large and very large) in the treatments (number of plant per treatment was four and number replications were four), the surfaces was identified (cm^2) and the obtained results (sum) were calculated to average values. The leaf area (Fig. II.6.5) of the species was slightly different in the salinity treatments. The high salt concentration of salinity treatments affected the leaf area. There was a slight decrease of leaf area in salinity treatment T3 (13-16). There was very low effect to leaf areas in treatments T1 (5-8) and T2 (9-12) compared to the control treatment T0 (1-4).

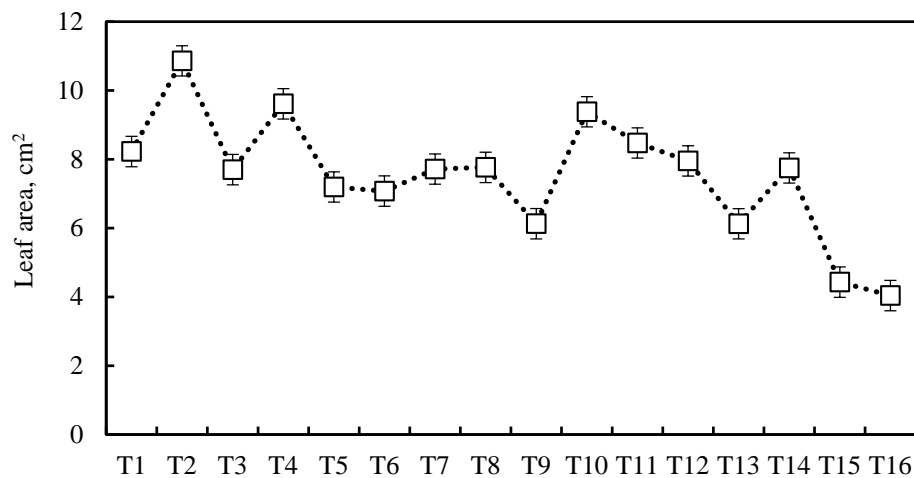


Fig. II.6.5 - Leaf area of species

II.6.2.4 Fresh and dry weight of crop

Fresh weight (FW) changes related to the roots and shoots of the species *Portulaca oleracea* under salinity stress is presented in Fig. II.6.6. The lowest value of stem, leaf and seeds of species FW were found at 200 mM NaCl (T3); the other NaCl treatments (T1 and T2) had no effect on the leaf and seeds FW of species. However, with NaCl concentrations of more than 200 mM, the crop FW decreased by approximately 33,6 % compared to the control group (Annex II.6.1). A change in the weight of plants due to salinity stress was also observed in the ratio of shoot FW. In the present study, the ratio of stem and leaf FW increased under salinity stress. Increases in the ratio of stem and leaf FW in the 50 and 100 mM NaCl treatments were significant in comparison to the control group and the other treatments. *P. oleracea* produced low amounts of dry matter, which ranged from 1.5 to 0.53 g plant⁻¹ (Fig. II.6.7). There was an increase of the percentage of dry matter of the stems and leaves in saline conditions and a decrease of dry matter allocated to the seeds with the increase of salinity treatments (100 – 200 mM NaCl). Growth retardation and fresh and dry weight loss of stem and leaves under salinity stress were revealed in previous studies (Hamidov, 2007; Lolaei 2012; Navarro et al., 2008; Villarino and Mattson 2011). In addition, based on the fresh or dry weight, it was demonstrated in several studies that the shoot ratios of many plants increase under salinity stress (Debouba et al., 2006; Maggio et al., 2007).

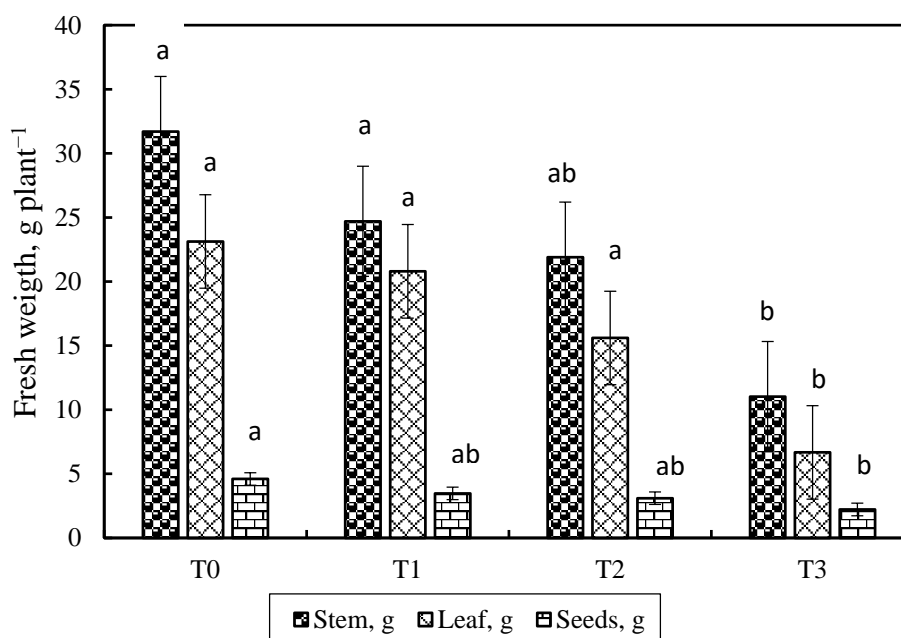


Fig. II.6.6 – Fresh weight of crop

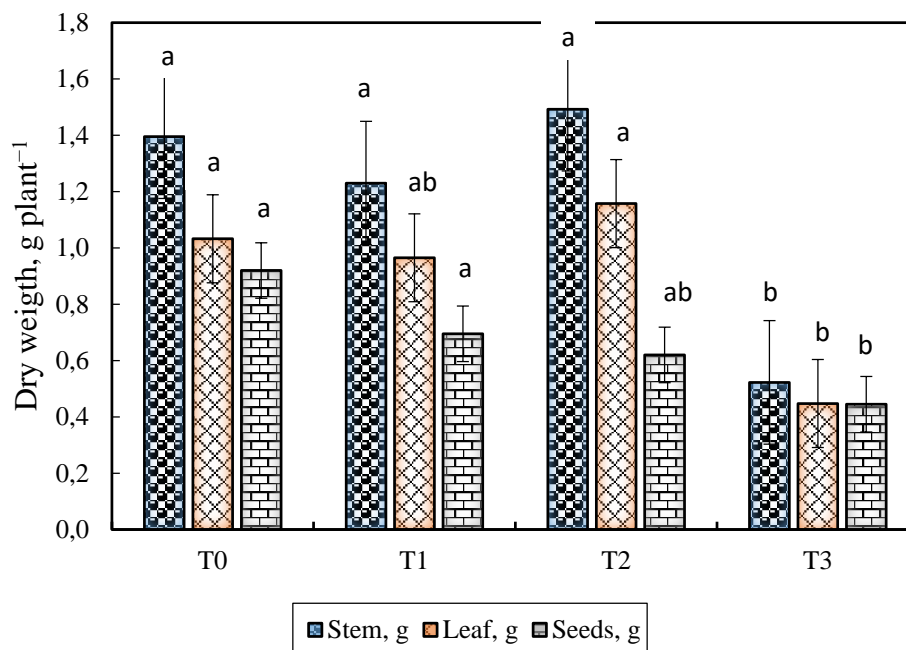


Fig. II.6.7 – Dry weight of crop

II.6.2.5 Root length of crop

The root length of the species showed low variation between treatments (Table. II.6.1). There was a decrease of the root length of crop in the high salinity treatment T3. Approximately 1.4 times decreases in the ratios of root length of species were observed in the 200 mM NaCl (T3) treatment compared to the T1 (50 mM NaCl), T2 (100 mM NaCl) and control (T0) treatments. The species *P. oleracea* was demonstrated to be the most tolerant to salinity. These aspects have already been studied in several crops, where the development was adversely affected with a significant decrease in production due to shallow GWT (Torres and Hanks, 1989; Beltrao et al., 1996).

Table. II.6.1 Root length of the species *Portulaca olerace*

Treatment	Root length, cm
T0	25.0 ± 4.88 a
T1	25.5 ± 3.18 a
T2	25.0 ± 1.73 a
T3	18.5 ± 1.32 ab

II.6.2.6 Macro and micronutrients in leaf content

In this study, salinity stress affected the micronutrient and macronutrient content in leaf uptake (Figs. II.6.8 and II.6.9). The analysis revealed a significant difference between salinity levels, in terms of macronutrients' accumulation in the plants. The salinity levels significantly affect all macronutrients in leaf content. The salinity levels significantly caused changes in performances of all treatments. All treatments showed increased potassium K^+ in leaf content. This was slightly significant in terms of calcium content (Ca^{2+}). Moreover, the calcium (Ca^{2+}) accumulation increased under salt treatment. Interestingly, the macronutrients of phosphorus (P), magnesium (Mg^{2+}) and sulphur (S) in leaf content were constant in all treatments (Fig. II.6.8).

The salinity levels significantly affected all treatments in terms of Na^+ and Cl^- content, while there were significant differences among treatments (Fig. II.6.9). There was an increase of sodium leaf content of the species with high salt concentration. Among the treatments, the highest Na^+ accumulation (2.76 %) occurred in the highest salinity level T3. In the high salinity level, chloride Cl^- in leaf content showed significantly less Na^+ . The micronutrients Fe, Al, Ba, Sr, Zn and Cu in leaf content were very low in all treatments (Annex II.6.2).

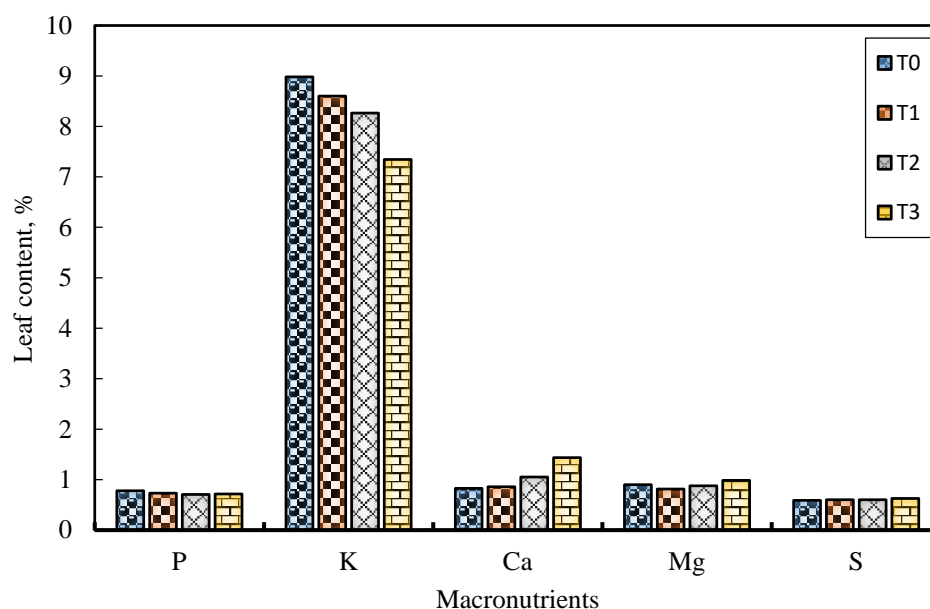


Fig. II.6.8 – Macronutrients content in leaf

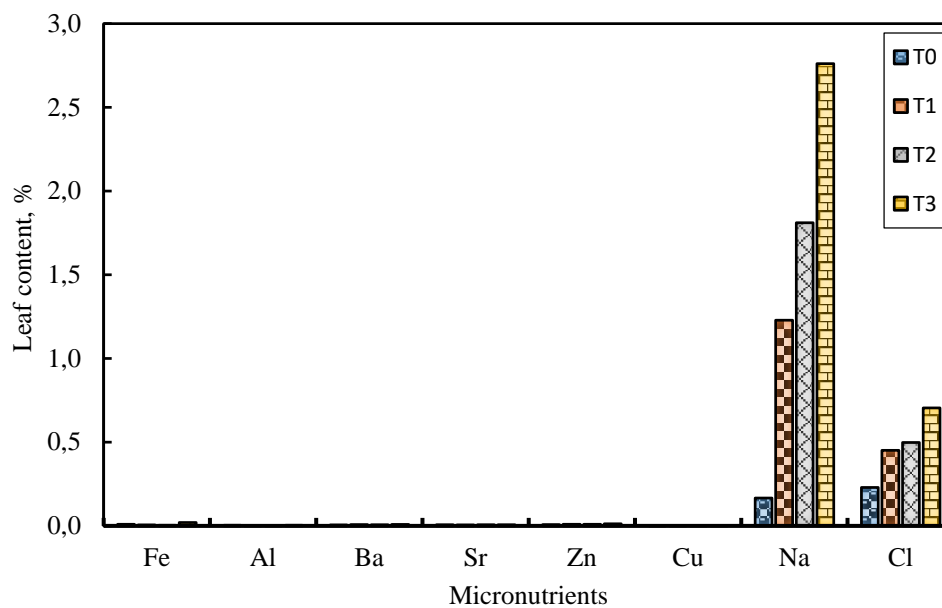


Fig. II.6.9 – Micronutrients content in leaf

II.6.2.7 Macro and micronutrients content in soil

The high salinity treatments were significantly affect macro and micronutrients in soil content. The low concentration of the macronutrients N, P, K in soil content of the species showed not variation between treatments (Fig. II.6.10). There was an increase of calcium (Ca^{2+}) in soil content in all treatments. However, non-saline soil had an amount of Ca in the species according to the control group (Kaymak, 2013). Magnesium (Mg^{2+}) and sulphur (S) showed low variation between treatments.

Macronutrients in soil content iron (Fe) and aluminum (Al) slightly increased in the salinity treatments (T2) compared to T0 (Fig. II.6.11). There was low variation between treatments of aluminum (Al) soil content. There was very low Ba, Sr, Zn, Cu and Pb in soil content in all treatments. Salinity levels of irrigation water had a significant effect on sodium Na^+ in soil content. There was an increase of sodium Na^+ in soil content when salinity concentration was high. There was variation in all treatments. Chloride (Cl^-) in soil content showed low variation between salinity treatments (Annex II.6.3).

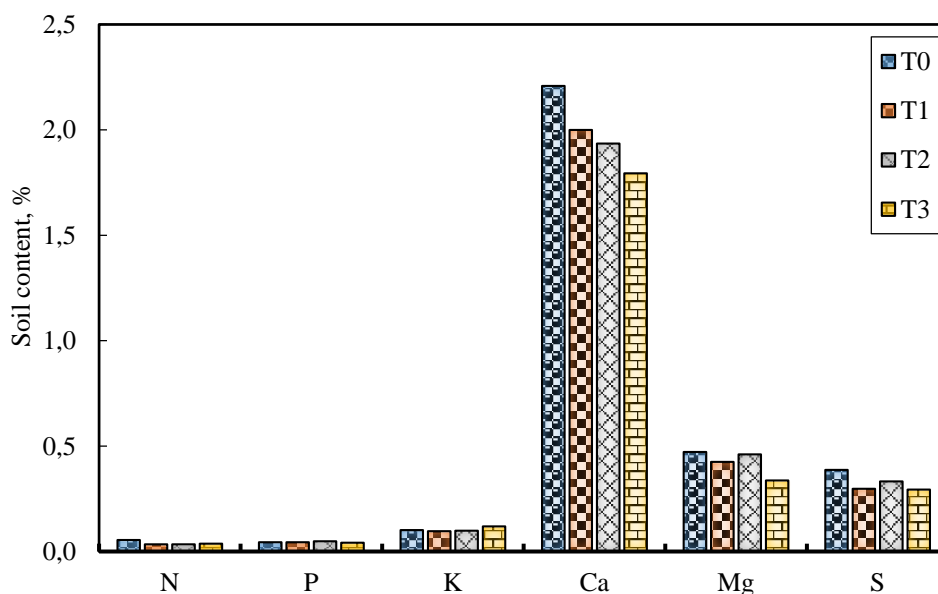


Fig. II.6.10 – Macronutrients content in soil

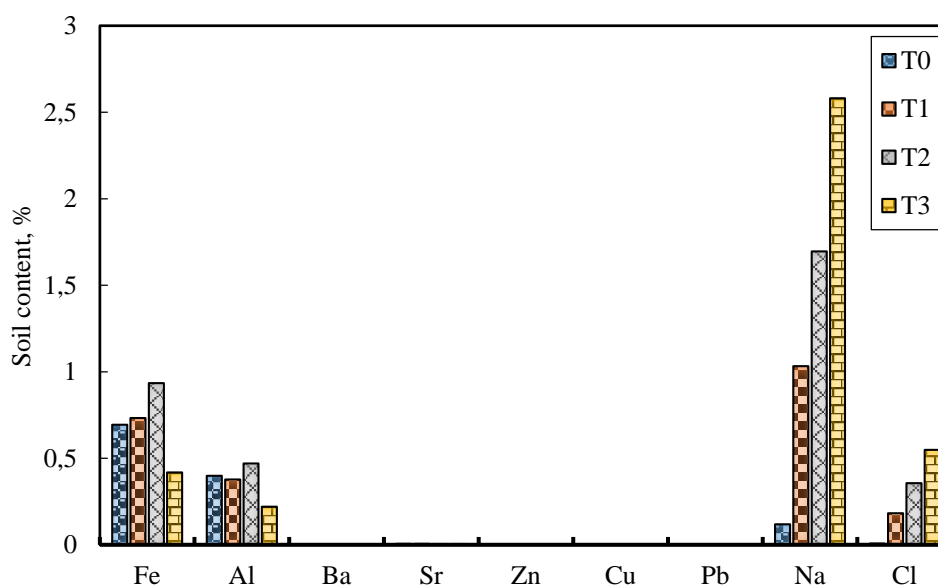


Fig. II.6.11 – Micronutrients content in soil

II.6.2.8 Yields of species *Portulaca oleracea*

The several salinity levels of salts NaCl affected the yield of *P. oleracea*. The value of the total fresh weight (FW) and dry weight (DW) of species were significantly different between treatments (Table II.6.2). The highest yield of the species was 1206 DW kg ha⁻¹ (21312 FW kg ha⁻¹) in the control group. A similar result was obtained by Vural et al. (2000) with a yield of cultivated purslane ranging between 30 - t ha⁻¹ and 50 - t ha⁻¹. In addition, the yield of cultivated purslane was affected by the seed quality; planting time, growing conditions and plant care practices (Vural et al., 2000). For example, Ehni et al. (1997) report the yield of purslane varied

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to the years and variety, and the highest yields were obtained from *Portulaca sativa* (70003 kg ha⁻¹) and *Egyptian* (37130 kg ha⁻¹). The lowest yield values were obtained in the highest salinity concentration with irrigated water treatment (T3 – 200 mMol NaCl) about 511 DW kg ha⁻¹. Plants grown at low levels of NaCl (control) reaches relatively to the higher total dry weight and did not exhibit the toxicity symptoms. However, the total dry weight was significantly reduced at higher levels of salinity treatments (5 - 20 dS m⁻¹), in which the symptoms of salt toxicity were revealed as growth depression. The plants' growth commenced to fall during the day on December, 17 and January, 20 by applying salinity. The concentration of 20 dS m⁻¹ NaCl significantly reduced the dry weight by 42.4 %, compared to the control group. These results are confirmed by previous research implemented by Hamidov (2007).

Table II.6.2. Yield of species *Portulaca oleracea*

<i>Portulaca oleracea</i>					
Treatment	FW (g plant ⁻¹)	DW (g plant ⁻¹)	Yield, %	FM kg ha ⁻¹	DM kg ha ⁻¹
T0	59,25 ± 6.1 a	3,25 ± 0.48 a	5,5 ± 0.28 ab	14806,3± 1520.8 a	837,3±97.8 a
T1	49 ± 9.7 a	2,75 ± 0.75 ab	5,5 ± 0.28 ab	12243,8± 2415.7 a	722,8±181.7 a
T2	40,75 ± 5.2 a	3,25 ± 1.03 a	7,25 ± 1.60 a	10150± 1264.9 a	817,8±269 ab
T3	20 ± 2.7 b	1,5 ± 0.29 b	7 ± 0.41 a	4981,3±691.3 b	353,7±46.4 b

II.6.3 Conclusion

Consequently, the results of this study suggest that cultivated species had increased the growth characteristics such as yield with the application of different salinity levels. The species *Portulaca oleracea* showed to be relatively tolerant to high saline conditions. The plant salt extraction analysis showed (Annex) that the species' tissues accumulated the largest amounts of sodium in the study (Figs. II.6.10 and II.6.12). In the current study, salt concentrations higher than 100 mM NaCl significantly increased the dry weights of the whole plant (total biomass), while high salt levels (200 mM NaCl) significantly decreased the dry weight DW ratio. Therefore, salinity stress was determined to have a significant impact in terms of micronutrient and macronutrient uptake. Despite the increasing salinity stress, an increase in Ca²⁺ in soil content to an increase in the plant's tolerance to stress. Under saline conditions, the Ca²⁺, K⁺, and Na⁺ uptakes of species were important parameters in terms of revealing the effects of stress.

II.7 Saline soil (where grown just before on the high salinity condition of the species) effects on growth, photosynthesis, root length and mineral nutrients of the species

Abstract: Salinity is one of most significant environmental stresses. *Portulaca oleracea* is high tolerant to salinity stress. Therefore, in this study, the fresh and dry weight of stem, leaf and seed ratio on a high concentration of saline soil and micronutrient and macronutrient concentrations in leaf and soil content of species were determined under salinity stress. The soil for the study was used of the previous study, saline soil (four salinity treatments (0, 50, 100, and 200 mM NaCl – were grown just before of *P. oleracea*) affected the biomass production of species *Portulaca oleracea*. The experiment was carried out in the greenhouse at the University Lille 1, France (April - June, 2016). The main purpose of the study was to confirm that the species is halophyte and can be used as a salt removing crops in the high salt concentration of saline soil. The species was irrigated with tap water among 0,25 – 0,50 L/pot and plant growth and chlorophyll content were analysed during the vegetation period.

II.7.1 Materials and methods

II.7.1.1 Experimental procedure

Seeds of the crop were sowed directly to the saline soil in three litter soil randomized pots (each pot 1400 g/soil) to obtain seedling on April, 7. The plant shoots seven days after sowing (April, 14, 2016).

The number of plants per treatment was four. In the beginning of the experiment plants were irrigated with water amounts minimal enough to the plant's survival (0,25 L/pot). The treatments are the following: T0 (1-4), T1 (5-8), T2 (9-12) and T3 (13-16). Analysis of plant germination began 12 days after plant shooting in the randomised pots. Stem length and number of nodes were analysed each seven days during the vegetation period in the experiment. Plants were irrigated with tap water amounts of 0,50 L/pot on May, 19. With the development of the plant, water demand increased.

Plants were harvested at 50 days (May, 15) after treatments, and washed with distil water for a few minutes, wiped with paper and the fresh weight (FW) was measured. The fresh samples were dried in forced drought in an oven at 65° C for 72 hours and the dry weight (DW) was measured.

The soil electrical conductivity (EC_s) and pH were analysed before and at the end of the experiment study.

II.7.1.2 Chemical analysis

Mineral elements macronutrients (N, P, K, Ca, Mg and S) and micronutrients (Fe, Al, Ba, Sr, Zn, Na and Cl) of the soil (before and after the experiment) and plant leaves (only after the experiment) were analysed by the ICP (Inductively Coupled Plasma and Optical Emission Spectrometer: ICP - OES). Chloride ions were determined in the aqueous extract by titration with silver nitrate according to Radojevic and Bashkin, (1999). Plant leaves and soil nitrogen (N) content were determined by the Kjeldhal method. All mineral analyses were performed soil and leaves.

The root length of the crops was determined at the end of the experimental study.

II.7.1.3 Statistical analysis

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.

II.7.1.4 Climate conditions in greenhouse

Climatic conditions were monitored with an automated system at hourly intervals. Daily air temperatures ranged from 20.7 to 29.1 °C (average, 24.9 °C) and night temperatures from 19.3 to 22.6 °C (average, 20.9 °C) during the experiment (Fig. II.7.1).

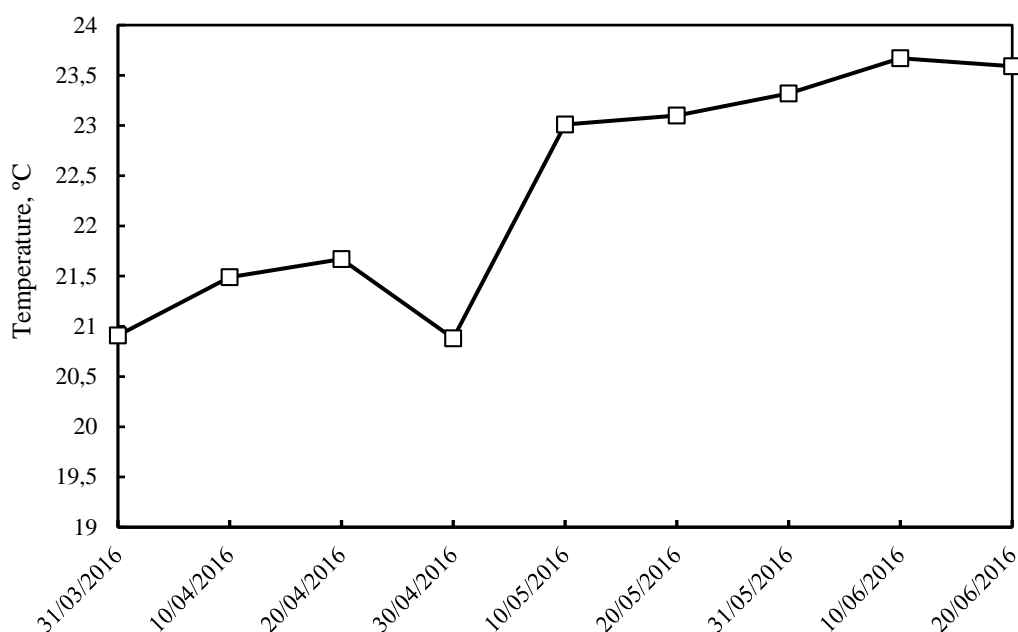


Fig. II.7.1 – Climate in greenhouse

II.7.2 Results and discussions

II.7.2.1 Growth rate of crop

The saline soil (several salt concentrations – were grown just before *Portulaca oleracea*) had a significant effect on the stem length of species *P. oleracea* (Fig. II.7.2). The total stem length of the species showed low variations among treatments, apparently not related with salinity. There was slightly increase of treatment T1 and T2 of crop. There was general decrease of treatments T0 and T3 of crop. The saline soil did affect the stem length of the crop. The salinity effect was generally very clear between all treatments, with a higher stem length increase in saline conditions of the species.

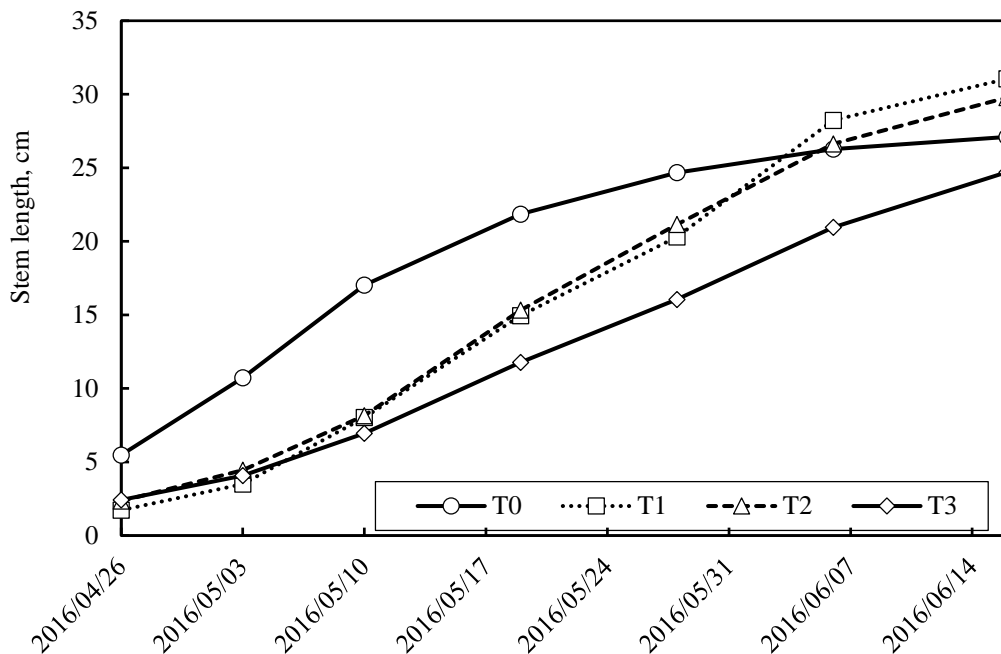


Fig. II.7.2 – Stem length of crop

II.7.2.2 Fresh and dry weight of plant biomass

The species *Portulaca oleracea* were grown in the same condition as a previous experiment (were grown just before the species *Tetragonia tetragonioides* in saline soil) (Fig. II.7.3). The fresh biomass production in leaf of the control treatment T0 (5.4 g plant^{-1}) decreased more than the leaf FW T1 ($12.5 \text{ g plant}^{-1}$); T2 ($15.9 \text{ g plant}^{-1}$); T3 ($19.7 \text{ g plant}^{-1}$) and the fresh weight of stem T0 ($10.8 \text{ g plant}^{-1}$) of the control treatment was lower than the fresh weight of stem T1 ($18.5 \text{ g plant}^{-1}$); T2 ($21.7 \text{ g plant}^{-1}$); T3 ($19.7 \text{ g plant}^{-1}$) on the saline soil of the species.

The dry weight of the species significantly decreased in the control group T0 compared to the treatments in saline soils (T1, T2 and T3) (Fig. II.7.4). It decreased 2.27 times more than all the saline soil treatments (Annex II.7.1).

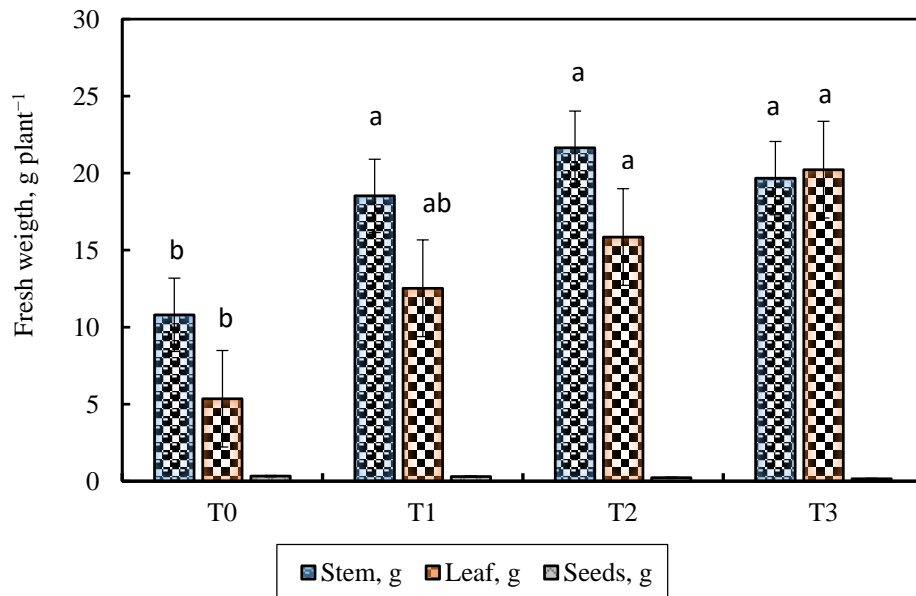


Fig. II.7.3 – Fresh weight of crop

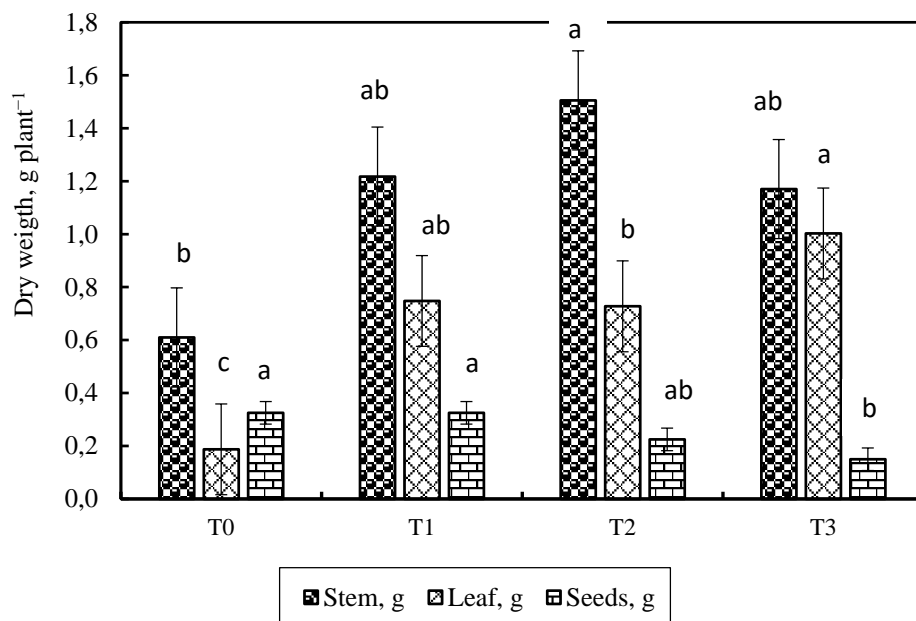


Fig. II.7.4 – Dry weight of crop

III.7.2.3 Root length of crop

The high salinity levels NaCl had a significant effect on the root length of species in the treatments T1 (19.7 cm) and T2 (20.3 cm) (Fig. II.7.5). There was low variation between

treatments. The results of root length show that the treatment T3 in high salt concentration was not different compared to T0. It will be seen that the species is halophyte.

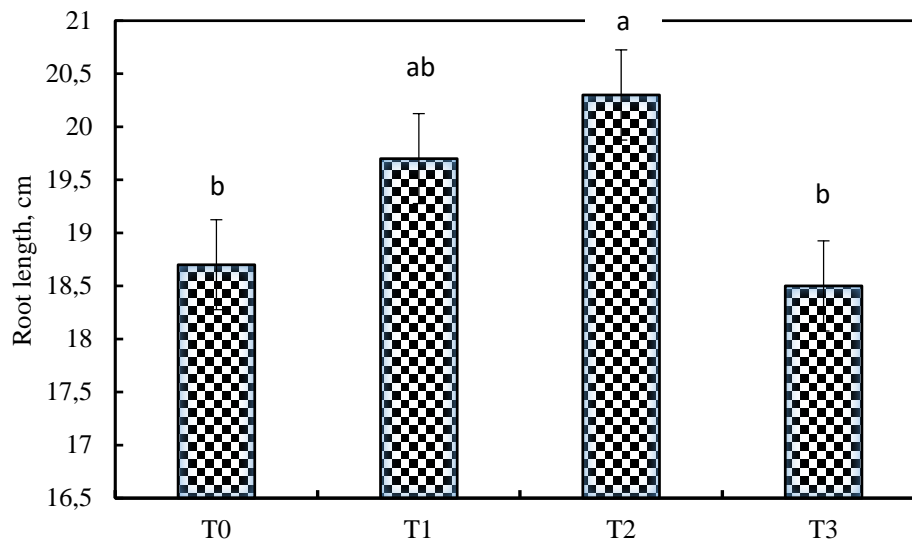


Fig. II.7.5 – Root length

II.7.2.4 Macro and micronutrients content in leaf

The species *Portulaca oleracea* demonstrated to be the most potential salt (ions) removal crop. High accumulation of Na^+ and Cl^- in soil increased macronutrients of potassium K^+ in leaf content in all treatments (Fig. II.7.6). The results indicated that the macronutrients of phosphorus P, calcium Ca^{2+} , magnesium Mg^{2+} and sulphur S in leaf content decreased in all treatments of saline soil compared to the control group.

The micronutrients Fe, Al, Ba, Sr, Zn and Cu were very low in all treatments. The sodium Na^+ in leaf content significantly increased with the increase of salinity levels in saline soil. There was increase of Chloride Cl^- in leaf content on the treatments (T1, T2, T3) of high salt concentration of soil compared to the control group (Fig. II.7.7). The effects of salinity stress on microelement uptake have been investigated in various studies (Villora et al., 2000; Lao and Plaza, 2013). However, the relationship between salinity and microelement uptake is complex. An increase or decrease may be observed in microelement uptake, or salinity may not have an effect on the microelement concentration of the plant. These differences result from factors such as plant species, plant tissues, level of salinity stress and composition, microelement concentration in the growth medium, growth conditions and stress duration (Grattan and Grieve 1999). Eom et al. (2007) suggested that salinity stress does not affect the Fe^{2+} or Zn^{2+} uptake of six different types of ground cover plants but reduces the concentration of Cu^{2+} (Annex II.7.2).

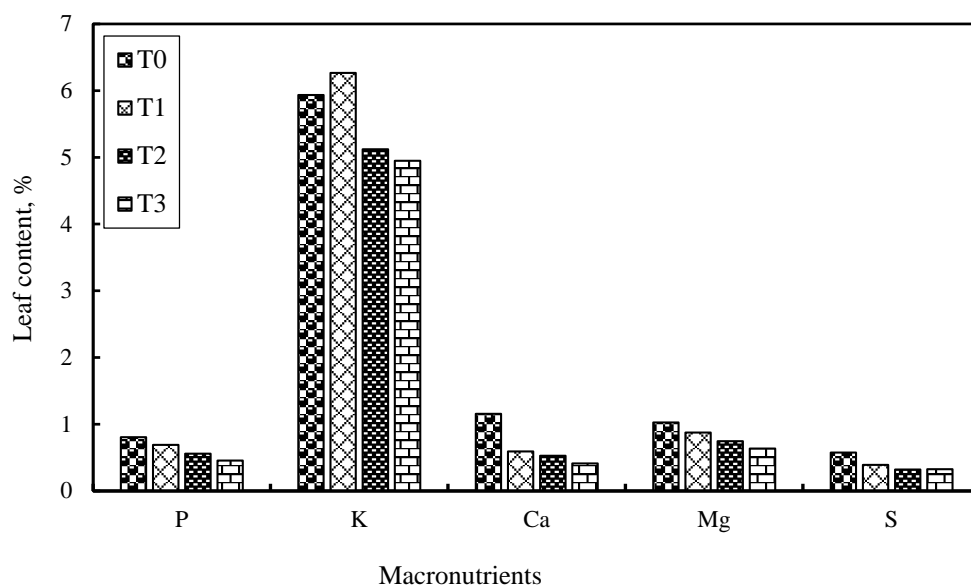


Fig. II.7.6 – Macronutrients content in leaf

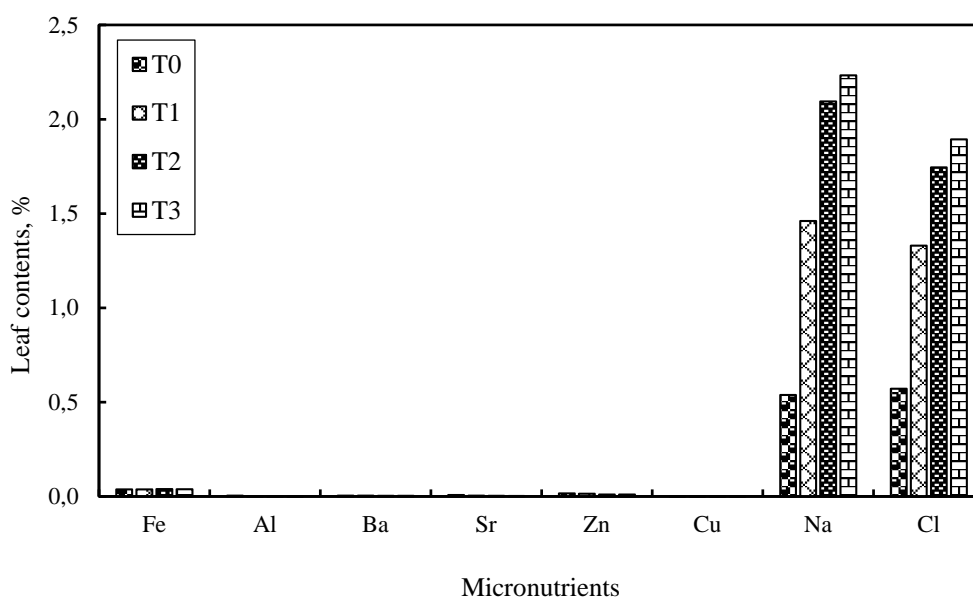


Fig. II.7.7 – Micronutrients content in leaf

II.7.2.5 Macro and micronutrients content in soil

The essential elements (P and K) in soil content were very low and there was no variation between treatments. Salinity soil had a significant effect on the macronutrient Ca^{2+} and the Mg^{2+} and S in soil content was not significantly affected (Fig. II.7.8).

The micronutrients Fe and Al concentrations were low in saline soil in all treatments. The saline soil stress had no effect on the micronutrients Ba, Sr, Zn and Cu the Cu^{2+} . Sodium Na in soil content was significantly increased in high salt concentration compared to the control

group. There was great increase of chloride Cl in soil content in all treatments T1 (1.13 %), T2 (4.9 %) and T3 (9.2 %) in saline soil (Fig. II.7.9) (Annex II.7.3).

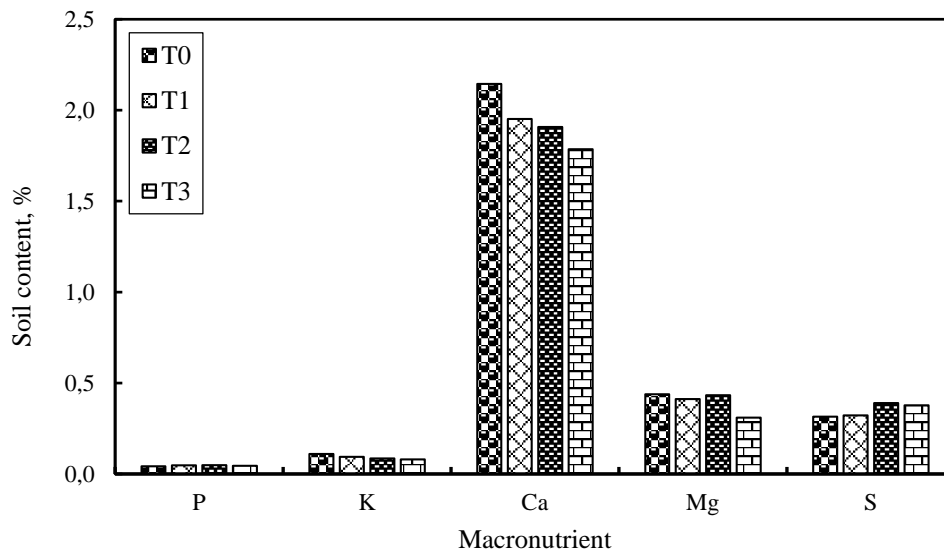


Fig. II.7.8 – Macronutrient content in soil

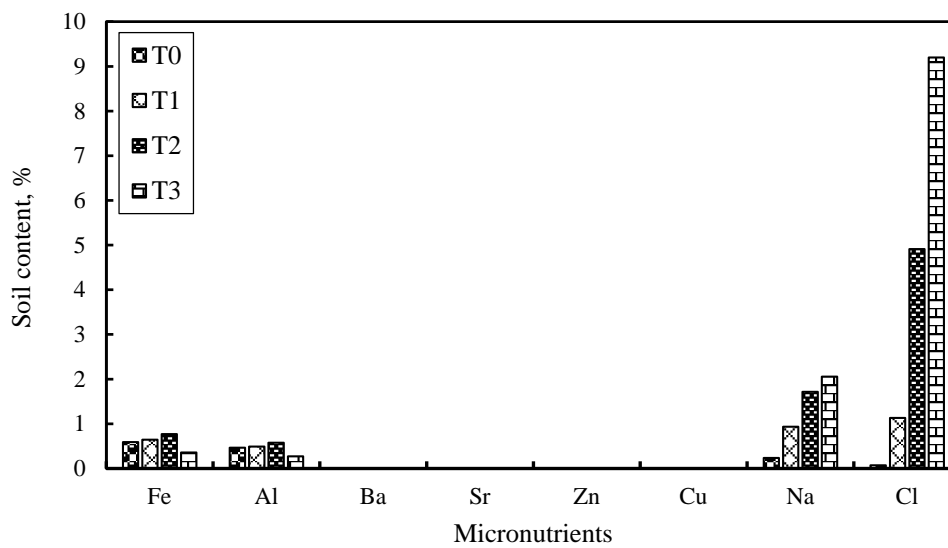


Fig. II.7.9 – Micronutrients content in soil

II.7.2.6 Macro and micronutrients in drainage water content

Plants that are irrigated by a large amount of water, may suffer from a loss of minerals (macro and micronutrients) in soil content. On the other hand, they may be helpful to remove salts from soil.

The drainage water was analyzed after each plant irrigation during the experiment study. Macro and micronutrients analysis of the drainage water was determined with the ICP method in the laboratory. The obtained results show that the macronutrients calcium Ca^{2+} and sulphur

S in drainage water increased from 150 mg/L to 260 mg/L (Annex II.7.4). Phosphorus P, potassium K^+ and magnesium Mg^{2+} in the drainage water decreased in all treatments (Fig. II.7.10).

The drainage water removed a large amount of micronutrients of sodium Na^+ and chloride Cl^- from the soil during the last irrigation of the species (Fig. 11). Na^+ in the drainage water increased between treatments T1 (870 mg/L), T2 (1214 mg/L) and T3 (1586 mg/L) in the high salinity soil compared to T0 (189 mg/L). Cl^- in the drainage water increased between treatments T1 (593 mg/L), T2 (1549 mg/L) and T3 (1984 mg/L) in the high salinity soil compared to T0 (163 mg/L). The results above confirm that the irrigation water removed a large amount of macro and micronutrients from the soil content (Fig. II.7.11).

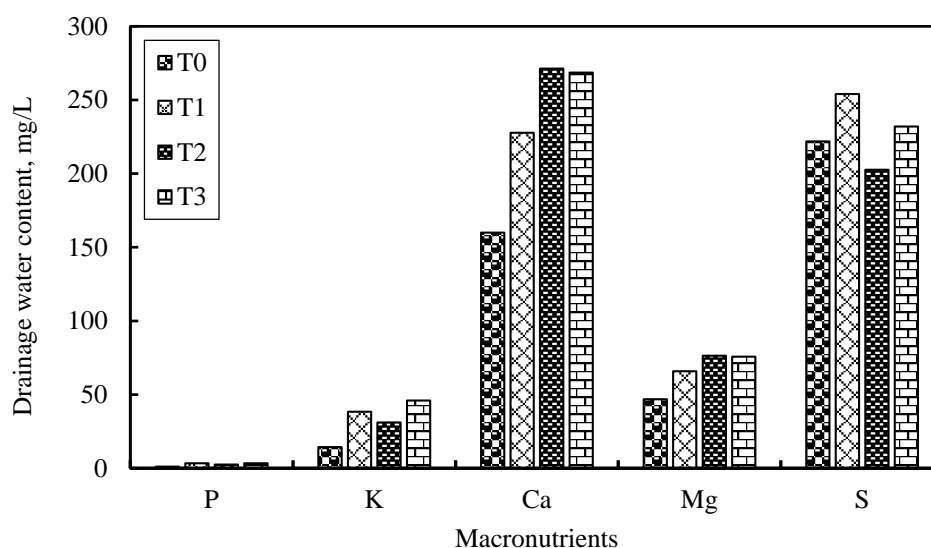


Fig. II.7.10 – Macronutrient content in drainage water

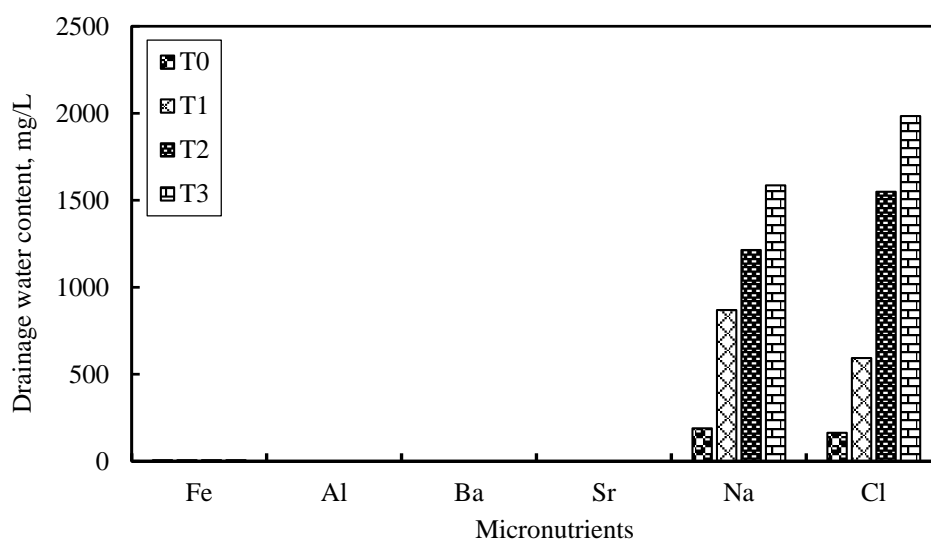


Fig. II.7.11 – Micronutrients content in drainage water

II.7.2.7 Yield of species

High significant variation was observed for fresh weights between treatments. The highest fresh weight (40.1 g) recorded in treatment T3 was in a high level of salinity soil and the lowest (16.5 g) was found in the control group (Table II.7.1). The obtained results show that saline soil has no effect on the yield of species *Portulaca oleracea*. The maximum amount of grain yield was measured in treatment T3 with an average of 14436 FW kg ha⁻¹ and the minimum amount of yield it was obtained in T0 (5940 FW kg ha⁻¹). Despite the saline soil effect on yield of species in the treatments T1, T2, T3 increased (1.9 – 2.4 times in the fresh weight and in dry weight 2 – 2.2 times) in comparison to the control group.

The driest matter was obtained in treatment T2 (886 kg ha⁻¹) and the least in the control group. The analysis of yield of the species *P. oleracea* confirmed that the species is halophyte.

Table II.7.1. Yield of crop

Treatment	<i>Portulaca oleracea</i>				
	FW (g plant ⁻¹)	DW (g plant ⁻¹)	Yield, %	FM kg ha ⁻¹	DM kg ha ⁻¹
T0	16,5 ± 2.99 b	1 ± 0.41 ab	6,5 ± 0.65 ab	4118,8 ± 770.3 b	281 ± 60.2 b
T1	31,75 ± 4.23 ab	2,3 ± 0.48 a	7 ± 1.08 a	7843,7 ± 1065.7 ab	572,7 ± 127.3 ab
T2	37,5 ± 11.55 ab	2,3 ± 0.75 a	6,5 ± 0.29 ab	9431,3 ± 2857.7 ab	614,7 ± 182.2 a
T3	40,25 ± 4.99 a	2,5 ± 0.64 a	5,75 ± 0.85 b	10012,5 ± 1280.1 a	580,7 ± 157.3 ab

II.7.3 Conclusion

Saline soil and water salinity are directly related to plant growth stress and decrease of the yield. Considering this global problem, in this study we tested agronomic species for biomass yield loss and physiological characteristics on saline soil stress. Analysis results revealed that although there had been significant variation among all treatments for measured parameters, commonly it has been proved as a high salt-tolerant crop plant capable of producing a satisfactory amount of dry matter content which is a great demand for any salt-tolerant plant species. In conclusion, our results indicated that the species *Portulaca oleracea* has been able to grow in high salinity soil. This is confirmed by the obtained dry matter in high saline conditions (grown just after *P. oleracea* in several concentrations of NaCl with salinity water).

CHAPTER III. GENERAL CONCLUSION

The main purposes of several experiments were carried out: effect of salt on growth and high biomass production during the vegetation period of the species (*Tetragonia tetragonioides* and *Portulaca oleracea*); effect of saline soil on growth of the moderately salt sensitive crop lettuce (*Lactuca sativa* L); effect of drought conditions and water relations on growth and photosynthesis of the species *T. tetragonioides*; high salt concentration effect on macro-micronutrients contents in leaf and soil and root length of the species (*T. tetragonioides* and *P. oleracea*); high biomass productions (dry matter) to compare ion extraction of the species (*T. tetragonioides* and *P. oleracea*) in high and moderate salinity levels of soil.

Based on the analysis of the results of the experiment, we can draw the following conclusions:

The first experiment measured the effect of salt on growth and biomass production of the two horticultural crops. The agronomic species were cultivated with the salinity treatments during the vegetation period (with salt concentration NaCl in irrigated water T1 (100 mM) and T2 (200 mM)). The obtained results confirmed that the both agronomic species are halophyte.

Tetragonia tetragonioides:

- The higher biomass production potential: apart the growth rate, this specie can produce several yields during the year (summer and winter).
- Species tolerant to high concentration of salts.
- High biomass production of the species (*T. tetragonioides*) averaging from 5220 to 7420 DM kg ha⁻¹.
- High mineral composition of leaves.

Portulaca oleracea:

- Higher tolerance to salinity and thirst
- Lower biomass and lower salt removal (the potential of *P. oleracea* as salts removal plant was also high (averaging from 2800 to 3400 kg DM ha⁻¹).

The obtained results highlights that lettuce (*Lactuca sativa*, L) is not tolerant to dry and hot conditions and it needs large amounts of irrigation water. 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. The positive contribution was indicated for the above salt removal crops, mainly *T. tetragonioides* and *P. oleracea* halophytic species to the quality and yield of the lettuce (a moderate salt - sensitive crop). Therefore, it has been demonstrated that this technique is a clean and environmentally safe tool to avoid salinization 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 combine environmental, economic and social aspects of problem solving. Hence, these two salt removing species may contribute to the increase of the soil sustainability of irrigated areas under climatic changes and may also be used as food crops.

As concluding remarks, it can be suggested that the *T. tetragonioides* is a species tolerant to drought conditions. The fresh weight FW of the species was not significantly different between treatments. The irrigation water regimes were (70 % (T1, wet treatment), 50% (T2, medium treatment), and 30% (dry treatment) of the available water capacity) effected on the process of photosynthesis (mineral composition, chlorophyll and carotenoid content in leaf) and yield of the species. Moreover, it was shown that the increase of its higher capacity as halophyte and salt removing species when the soil water content decreased, suggesting its use in arid and semi-arid regions. On the other hand, growth and yield differences on several irrigation regimes were very low, which suggests the other important advantage of these species - its cultivation under dry conditions, when used as a leafy vegetable for human consumption or for animal feeding. Nevertheless, more research is needed in order to test the plant development under drier conditions, in arid and semi-arid climates.

In conclusion of observation, it can be suggested that the *Tetragonia tetragonioides* are species tolerant to high salinity conditions. The high salt concentration affected macro-micronutrients contents in leaf and soil and root length of the species (*T. tetragonioides* and *P. oleracea*). In the experiments, *T. tetragonioides* produced significant amounts of dry matter. The reasons were: fast growth rate (0.31 – 0.52 cm/day), deep root length (42.9 – 53.9 cm), high biomass production (17000 – 21300 FW kg ha⁻¹). In addition, it can be used for protection from soil erosion due to excellent soil covering. As final remarks, it is concluded that in salinity conditions, the clean and environmental safe procedures to control salinity could be associated to the conventional techniques, combining environmental and social aspects. Hence, the species may contribute to increase the soil sustainability of irrigated areas and also may be used as food crops.

T. tetragonioides showed to be the most potential salt (ions) removal species. Moreover, at the end of the experiment, *T. tetragonioides* was the sole species that had produced significant amounts of dry matter. The reasons were: fast growth rate, high biomass production (if properly managed), easy cropping (as winter or summer crop), and absence of diseases and pests of major importance, as well as good acceptance by local consumers as a leafy vegetable. Moreover, it can be suggested that *T. tetragonioides* is very interesting species because:

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- The higher biomass production potential.
- High salt accumulation in leaf.
- Easy multiplication (seed propagation) and easy crop management.
- High macro and micronutrients content in leaf.

These new clean techniques to control salinity showed that agricultural production can be maintained through the reduction of salts application due to the decrease of irrigation amounts, reducing the leaching. On the other hand, the applied salts through the irrigation can be eliminated by using the salt (ions) removing species. As final remarks, it is concluded that in arid climates and under global warming, the clean and environmental safe procedures to control salinity could be associated to the conventional techniques, combining environmental, economic and social aspects, contributing, therefore, to increase the sustainability of irrigated areas and plant growth.

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 species can be planted, as ornamentals, in saline soils, even without irrigation.

These new clean techniques to control salinity showed that agricultural production can be maintained through the reduction of salts application due to the decrease of irrigation amounts, reducing the leaching.

As final remarks, it is concluded that in arid climates, the clean and environmental safe procedures to control salinity could be associated to the conventional techniques, combining environmental, economic and social aspects.

CHAPTER IV: PERSPECTIVES AND RECOMMENDATIONS

Based on the results during the years of the research studies, we can say that these are the agronomic species that can be cultivated in a high level of saline soils (10 - 20 dS m⁻¹).

The obtained results achieved the following successes:

- larger biomass accumulation during the vegetation period; (63132 FW kg ha⁻¹ and 4452 DM kg ha⁻¹) (*Tetragonia tetragonioides*);
- species can produce several yields during the year (summer and winter);
- easy multiplication (seed propagation) and easy crop management;

Tetragonia tetragonioides showed to be the most potential salts (ions) removal species. The potential of *Portulaca oleracea* as salts removal plant was also high, but lower than *T. tetragonioides*. The species (*Tetragonia tetragonioides*) removed up to 500 kg ha⁻¹ ions.

Previous investigations in the Mediterranean area have shown that annual *Tetragonia tetragonioides* crops produced the highest biomass and were efficient crops to remove ions from salt-affected soils. For instance, Beltrao et al. (2006) found that *Tetragonia tetragonioides* produced 4200 DM kg ha⁻¹ and removed up to 700 kg ha⁻¹ NaCl in Portugal. In another study, Hamidov (2007) obtained that *Portulaca oleracea* produced about 3950 DM kg ha⁻¹ and removed about 500 kg ha⁻¹ ions. Therefore, in future investigations, we need to assess other crops, such as *Tetragonia tetragonioides* for their efficiencies to remove ions from the saline soils.

The obtained results highlight that lettuce (*Lactuca sativa*, L) is not tolerant to dry and hot conditions and it needs large amounts of irrigation water. 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. The positive contribution was indicated for the above salt removal crops, mainly *T. tetragonioides* and *Portulaca oleracea* halophytic species to the quality and yield of the lettuce (a moderate salt - sensitive crop). Therefore, it has been demonstrated that this technique is a clean and environmentally safe tool to avoid salinization and maintain the sustainability of agricultural systems.

In arid areas, drought and salinity are the key factors that responsible for limiting crop productivity. Experimental results showed several effects of the water irrigation regimes on the growth, mineral composition and photosynthetic pigments of *Tetragonia tetragonioides*. Plant growth (stem, leaves and seeds) increased slightly with enhanced water levels (near the field capacity), with a very low growth difference between the drier water regimes. The obtained

results confirmed that the species can be a large biomass producer under arid and semi-arid conditions. Cultivation of these plants on saline soil and in arid climates can be considered as clean and environmentally safe techniques which combines environmental, economic 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 also be used as food crops. It can be suggested that the *T. tetragonioides* are species tolerant to drought conditions. Moreover, it was shown that the increase of its higher capacity as halophyte and salt removing species when the soil water content decreased, suggesting its use in arid and semi-arid regions. On the other hand, growth and yield differences on the several irrigation regimes were very low, which suggests the other important advantage of these species - its cultivation under dry conditions, when used as a leafy vegetable for human consumption or for animal feeding.

In general, the ideal multipurpose plant species should have a combination of the following features: ability to remove high levels of ions from the soil; high biomass production potential; a short vegetation period; low water consumption; good acceptance by local consumers as a leafy vegetable; tolerance to drought and hot conditions; and easy crop management.

The author recommends to obtained results of several experiments on the species of *Tetragonia tetragonioides* and *Portulaca oleracea* can use as a salt removing crops in high salinity levels on saline soil (abandoned land). The species were their root profiles because, it should be stated that the *Portulaca oleracea* can remove the salts only from 5-10 cm of the topsoil layer because of its low root length; and on the other hand, *Tetragonia tetragonioides* can remove the salts from around 20 - 25 cm of the topsoil layer.

However, the species can be planted in arid and semi-arid areas as drought tolerant. Moreover, both species can be planted, as ornamentals, in saline soils, even without irrigation.

We hope our findings will benefit readers, consumers, and producers to cultivate *Tetragonia tetragonioides* vegetables in any type of soils especially salinity affected areas.

In addition, the species *Tetragonia tetragonioides* can be a protection from soil erosion due to excellent soil covering.

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Annexes

Annex II.1.1 Fresh (FW) and dry weight (DW) of the species (*Tetragonia tetragonioides*), g plant⁻¹

Treatment	Total mass				Total mass			
	FW g plant ⁻¹	Leaves	Stems	Seeds	DW g plant ⁻¹	Leaves	Stems	Seeds
T01	601.4	309,7	261,5	30,2	49,8	10,5	33,5	5,8
T02	550.4	423,1	104,6	22,7	30,4	12,3	12,9	5,2
T03	400.0	288,7	101,2	10,1	29,4	9,6	13,3	6,5
T04	552.6	405,0	119,5	28,1	38,8	14,3	15,7	8,8
meanT0	526.1	356,6	146,7	22,8	37,1	11,7	18,9	6,6
T11	239.5	144,9	75,9	18,7	29,8	8,4	14,0	7,4
T12	359.2	229,8	102,8	26,6	35,5	10,7	16,2	8,6
T13	400.8	270,0	98,5	32,3	33,9	10,1	15,3	8,5
T14	361.9	265,8	80,4	15,7	28,1	8,5	13,8	5,8
mean T1	340.4	227,6	89,4	23,3	31,8	9,4	14,8	7,6
T21	78.9	53,3	21,9	3,7	13,8	5,1	5,4	3,3
T22	167.7	106,9	49,7	11,1	24,5	8,2	11,5	4,8
T23	313.2	207,5	82,0	23,7	31,4	10,4	14,4	6,6
T24	344.2	239,0	81,1	24,1	34,7	11,6	16,0	7,1
mean T2	226.0	151,7	58,7	15,7	26,1	8,8	11,8	5,5

Annex II.1.2 Fresh (FW) and dry weight (DW) of the species (*Portulaca oleracea*), g plant⁻¹

Treatment	Total mass				Total mass			
	FW g plant ⁻¹	Leaves	Stems	Seeds	DW g plant ⁻¹	Leaves	Stems	Seeds
T01	106,70	60,00	41,60	5,10	10,72	5,60	4,10	1,02
T02	130,00	89,80	37,50	2,70	14,44	8,20	5,70	0,54
T03	117,40	73,50	39,30	4,60	14,62	8,00	5,70	0,92
T04	131,90	84,40	44,90	2,60	14,62	8,60	5,50	0,52
meanT0	121,50	76,93	40,83	3,75	13,60	7,60	5,25	0,75
T11	56,10	32,40	20,50	3,20	10,74	5,40	4,70	0,64
T12	73,70	41,50	27,80	4,40	12,88	7,00	5,00	0,88
T13	61,50	32,60	25,40	3,50	10,80	5,70	4,40	0,70
T14	55,50	31,50	20,40	3,60	11,02	6,00	4,30	0,72
mean T1	61,70	34,50	23,53	3,68	11,36	6,03	4,60	0,74
T21	34,80	19,70	12,90	2,20	9,44	5,10	3,90	0,44
T22	75,90	43,70	28,70	3,50	13,10	7,20	5,20	0,70
T23	67,80	44,20	21,50	2,10	12,52	7,10	5,00	0,42
T24	34,50	18,40	14,20	1,90	9,88	5,20	4,30	0,38
mean T2	53,25	31,50	19,33	2,43	11,24	6,15	4,60	0,49

Annex II.1.3 Area, plant density, biomass production and ion extraction from the soil of the species *Tetragonia tetragonioides*

Treatment	<i>Tetragonia tetragonioides</i>											
							Na			Cl		
	Area, m ²	Plant density	FY g plant ⁻¹	DY, g plant ⁻¹	FY kg ha ⁻¹	DY, kg ha ⁻¹	ion conc., mg g ⁻¹ pl.	Ion extr, kg ha ⁻¹	ion extr, mg pie ⁻¹	ion conc., mg g ⁻¹ pl.	Ion extr, kg ha ⁻¹	ion extr, mg piece ⁻¹
T01	1	12	601,4	49,8	72168	5976	7,49	144,78	31,1	30,82	184,20	127,92
T02	1	12	550,4	30,4	66048	3648	13,43	148,99	34,02	70,53	257,30	178,68
T03	1	12	400	29,4	48000	3528	17,26	160,88	42,28	64,56	227,75	158,16
T04	1	12	552,6	38,8	66312	4656	10,07	146,89	32,56	45,20	210,47	146,16
T11	1	12	239,5	29,8	28740	3576	26,22	393,77	65,12	162,56	581,30	403,68
T12	1	12	359,2	35,5	43104	4260	22,51	395,88	66,58	101,33	431,65	299,76
T13	1	12	400,8	33,9	48096	4068	21,68	388,19	61,24	103,26	420,08	291,72
T14	1	12	361,9	28,1	43428	3372	27,19	391,68	63,67	194,84	656,99	456,24
T21	1	12	78,9	13,8	9468	1656	55,37	591,68	63,67	369,18	611,37	424,56
T22	1	12	167,7	24,5	20124	2940	30,71	590,27	62,69	216,29	635,90	441,6
T23	1	12	313,2	31,4	37584	3768	25,26	595,18	66,1	140,33	528,77	367,2
T24	1	12	344,2	34,7	41304	4164	21,01	587,48	60,75	152,67	635,73	441,48

Annex II.1.4 Area, plant density, biomass production and ion extraction from the soil of the species *Portulaca oleracea*

Treatment	<i>Portulaca oleracea</i>											
							Na			Cl		
	Area, m ²	Plant density	FY* g plant ⁻¹	DY, g plant ⁻¹	FY kg ha ⁻¹	DY* kg ha ⁻¹	ion conc., mg g ⁻¹ p	Ion extr., kg ha ⁻¹	ion extr., mg piece ⁻¹	ion conc., mg g ⁻¹ p	Ion extr., kg ha ⁻¹	ion extr., mg piece ⁻¹
T01	1	20	106,7	10,72	21340	2144	34,46	73,88	18,47	603,73	129,4	323,6
T02	1	20	130	14,44	26000	2888	23,56	68,04	17,01	459,28	132,4	331,6
T03	1	20	117,4	14,62	23480	2924	9,97	29,16	7,29	512,72	149,2	374,8
T04	1	20	131,9	14,62	26380	2924	18,62	54,44	13,61	498,77	145,4	364,6
T11	1	20	56,1	10,74	11220	2148	55,21	118,6	29,65	991,81	230,4	532,6
T12	1	20	73,7	12,88	14740	2576	43,77	112,7	28,19	843,48	272,8	543,2
T13	1	20	61,5	10,8	12300	2160	57,59	124,4	31,1	916,67	198,0	495
T14	1	20	55,5	11,02	11100	2204	46,75	103,04	25,76	921,96	203,2	508
T21	1	20	34,8	9,44	6960	1888	87,52	165,24	41,31	1475,00	378,4	696,2
T22	1	20	75,9	13,1	15180	2620	23,01	260,28	15,07	1049,47	374,9	687,4
T23	1	20	67,8	12,52	13560	2504	26,39	266,08	16,52	1086,26	372,0	680
T24	1	20	34,5	9,88	6900	1976	79,70	157,48	39,37	1361,54	369,4	672,6

Note: FY* - fresh yield; DY* - dry yield

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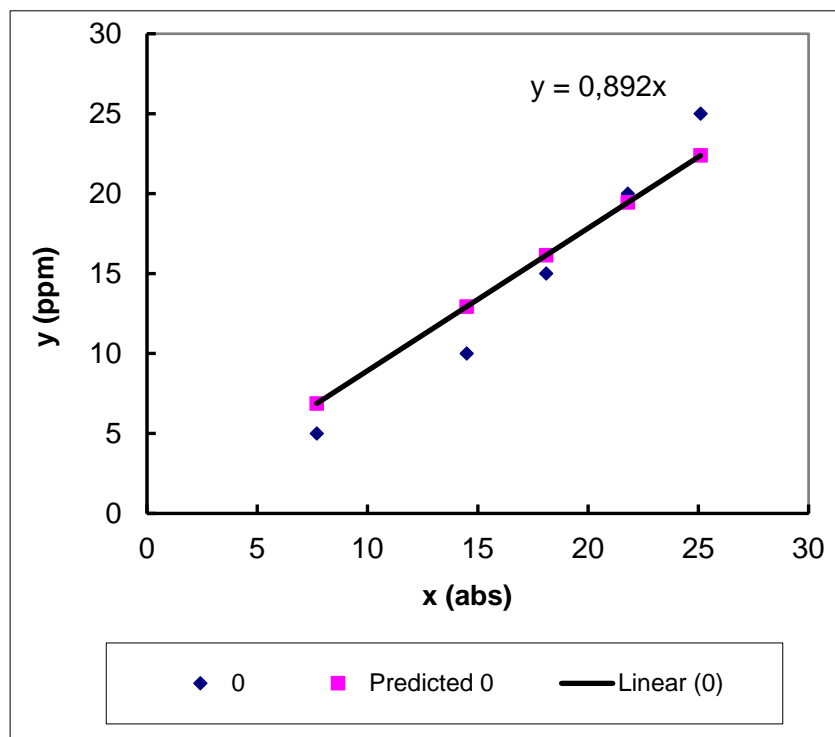
Annex II.2.1 Fresh (FM) and dry matter (DM) of lettuce (were grown just before *Tetragonia tetragonioides*), g plant⁻¹

Treatment	Fresh weight, plans mass g				Dry weight, plans mass			
	FM	m ²	Kg m ⁻²	Kg ha ⁻¹	DM	g m ⁻²	kg m ⁻²	kg ha ⁻¹
T01	214,66	1931,94	1,93	19319	8,32	74,88	0,07	749
T02	193,7	1743,30	1,74	17433	7,94	71,46	0,07	715
T03	302,04	2718,36	2,72	27184	12	108,00	0,11	1080
T04	192,16	1729,44	1,73	17294	6,75	60,75	0,06	608
mean T0	225,64	2030,76	2,03	20308	8,75	78,77	0,08	788
T11	162,65	1463,85	1,46	14639	5,37	48,33	0,05	483
T12	150,06	1350,54	1,35	13505	5,49	49,41	0,05	494
T13	187,99	1691,91	1,69	16919	6,8	61,20	0,06	612
T14	246,55	2218,95	2,22	22190	8,64	77,76	0,08	778
mean T1	186,8125	1681,31	1,68	16813	6,57	59,18	0,06	592
T21	165,75	1491,75	1,49	14918	7,25	65,25	0,07	653
T22	139,3	1253,70	1,25	12537	6,44	57,96	0,06	580
T23	152,59	1373,31	1,37	13733	6,26	56,34	0,06	563
T24	213,7	1923,30	1,92	19233	7,07	63,63	0,06	636
mean T2	167,835	1510,52	1,51	15105	6,75	60,80	0,06	608

Annex II.2.2 Fresh (FM) and dry matter (DM) of lettuce (were grown just before *Portulaca oleracea*), g plant⁻¹

Treatment	Fresh matter, plans mass g				Dry matter, plans mass			
	FM	m ²	kg m ⁻²	kg ha ⁻¹	DM	g m ⁻²	kg m ⁻²	kg ha ⁻¹
T01	244,73	2202,57	2,20	22026	7,65	68,85	0,07	689
T02	264,33	2378,97	2,38	23790	8,27	74,43	0,07	744
T03	279,03	2511,27	2,51	25113	11,21	100,89	0,10	1009
T04	233,58	2102,22	2,10	21022	8,99	80,91	0,08	809
meanT0	255,41	2298,76	2,30	22988	9,03	81,27	0,08	813
T11	138,21	1243,89	1,24	12439	5	45,00	0,05	450
T12	196,91	1772,19	1,77	17722	6,37	57,33	0,06	573
T13	150,8	1357,20	1,36	13572	6,13	55,17	0,06	552
T14	226,85	2041,65	2,04	20417	8,36	75,24	0,08	752
mean T1	178,19	1603,73	1,60	16037	6,46	58,19	0,06	582
T21	149,97	1349,73	1,35	13497	6,31	56,79	0,06	568
T22	149,9	1349,10	1,35	13491	5,33	47,97	0,05	480
T23	175,88	1582,92	1,58	15829	6,55	58,95	0,06	590
T24	143,73	1293,57	1,29	12936	5,99	53,91	0,05	539
mean T2	154,87	1393,83	1,39	13938	6,04	54,41	0,05	544

Annex II.2.3 Sodium (Na) analysis of lettuce leaf content (were grown just before of *T. tetragonioides* and *P. oleracea*)



Sodium (Na) in leaf content of lettuce (*T. tetragonioides*)

Treatment	mg/l	mg / g sample	g / g sample	%
T01	13,558	67,79	0,0678	6,78
T02	10,615	53,07	0,0531	5,31
T03	12,042	60,21	0,0602	6,02
T04	11,150	55,75	0,0558	5,58
T11	28,901	144,50	0,1445	14,45
T12	30,863	154,32	0,1543	15,43
T13	27,652	138,26	0,1383	13,83
T14	32,380	161,90	0,1619	16,19
T21	31,934	159,67	0,1597	15,97
T22	36,126	180,63	0,1806	18,06
T23	35,769	178,85	0,1788	17,88
T24	42,638	213,19	0,2132	21,32

Sodium (Na) in leaf content of lettuce (*P. oleracea*)

Treatment	mg/l	mg / g sample	g / g sample	%
T01	22,032	110,16	0,1102	11,02
T02	20,070	100,35	0,1004	10,04
T03	21,051	105,26	0,1053	10,53
T04	16,234	81,17	0,0812	8,12
T11	33,361	166,80	0,1668	16,68
T12	36,661	183,31	0,1833	18,33
T13	30,863	154,32	0,1543	15,43
T14	28,812	144,06	0,1441	14,41
T21	37,375	186,87	0,1869	18,69
T22	35,145	175,72	0,1757	17,57
T23	39,426	197,13	0,1971	19,71
T24	38,624	193,12	0,1931	19,31

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Annex II.3.1 Fresh FM and dry matter DM of the species (*Tetragonia tetragonioides*), g plant⁻¹

Groups	Stem	Leaves	Seed	Total mass FM	Stem	Leaves	Seeds	Total mass DM
T11	508,9	453,59	98,94	1061,43	55,03	35,78	9,68	100,49
T12	530,09	462,21	43,59	1035,89	45,44	33,39	8,16	86,99
T13	551,16	402,14	36,26	989,56	41,43	27,18	7,19	75,8
T14	558,07	474,26	32,25	1064,58	41,69	25,91	6,27	73,87
T15	522,96	461,35	44,95	1029,26	46,85	28,73	7,29	82,87
T16	568,26	452,11	45,23	1065,6	43,04	25,18	7,43	75,65
mean T0	539,9	450,9	50,2	1041,1	45,6	29,4	7,7	82,6
T21	473,28	403,19	28,88	905,35	33,73	24,81	5,79	64,33
T22	426,46	391,85	28,22	846,53	28,19	19,52	5,31	53,02
T23	452,65	409,73	37,97	900,35	40,55	21,98	7,09	69,62
T24	556,04	421,64	26,47	1004,15	36,82	23,02	4,53	64,37
T25	464,88	378,39	35,53	878,8	41,84	26,49	6,92	75,25
T26	498	401,47	47,46	946,93	36,28	24,37	9,43	70,08
mean T1	478,6	401,0	34,1	913,7	36,2	23,4	6,5	66,1
T31	507,31	458,74	44,98	1011,03	36,76	23,83	7,79	68,38
T32	444,02	401,83	22,85	868,7	37,45	18,26	4,85	60,56
T33	513,64	415,22	35,08	963,94	32,63	20,58	5,64	58,85
T34	517,8	485,54	20,12	1023,46	40,18	22,18	3,94	66,3
T35	506,41	426,62	41,26	974,29	45,2	26,45	7,93	79,58
T36	498,27	411,36	46,48	956,11	38,05	23,89	7,63	69,57
mean T2	497,9	433,2	35,1	966,3	38,4	22,5	6,3	67,2

Annex II.3.2 Mineral analysis in leaf and soil content of *Tetragonia tetragonioides*, (% g sample⁻¹)

Treatment	Mineral content in leaves, % g sample ⁻¹							Mineral content in soil, % g sample ⁻¹	
	K	P	Na	Ca	Mg	Fe	Cl	Na	Cl
T1.1	0,063	0,001	0,075	0,0006	0,435	0,0002	0,86	0,051	0,250
T1.2	4,040	0,030	0,077	0,0005	0,424	0,0002	0,76	0,040	0,220
T1.3	4,602	0,030	0,098	0,0005	0,431	0,0002	0,51	0,037	0,210
T1.4	4,551	0,030	0,093	0,0007	0,433	0,0002	0,98	0,039	0,230
T1.5	4,551	0,030	0,078	0,0005	0,420	0,0003	0,78	0,032	0,226
T1.6	4,244	0,031	0,074	0,0007	0,448	0,0003	0,81	0,029	0,210
T2.1	3,068	0,026	0,131	0,0004	0,358	0,0005	0,93	0,059	0,240
T2.2	3,886	0,029	0,130	0,0004	0,364	0,0003	0,99	0,048	0,260
T2.3	3,682	0,028	0,125	0,0004	0,368	0,0002	1,02	0,058	0,280
T2.4	3,989	0,028	0,138	0,0004	0,361	0,0002	0,82	0,061	0,260
T2.5	3,273	0,027	0,126	0,0003	0,353	0,0002	0,86	0,053	0,239
T2.6	4,142	0,028	0,127	0,0004	0,366	0,0003	0,95	0,057	0,275
T3.1	3,528	0,027	0,148	0,0003	0,324	0,0003	1,19	0,089	0,360
T3.2	4,091	0,027	0,135	0,0005	0,422	0,0003	0,93	0,108	0,270
T3.3	4,346	0,027	0,134	0,0003	0,339	0,0002	1,83	0,100	0,280
T3.4	4,193	0,028	0,149	0,0003	0,336	0,0002	1,10	0,093	0,270
T3.5	4,346	0,027	0,133	0,0004	0,355	0,0002	1,26	0,087	0,289
T3.6	3,733	0,027	0,148	0,0003	0,331	0,0002	1,36	0,104	0,307

Annex II.4.1 Fresh and dry mass of species (*Tetragonia tetragonioides*), g plant⁻¹

Treatment	Fresh mass, g plant ⁻¹				Dry mass, g plant ⁻¹			
	Stem	Leaf	Seed	Total mass	Stem	Leaf	Seed	Total mass
T1	87,68	135,72	37,60	261,00	10,26	19,05	6,70	36,00
T2	78,13	142,17	53,10	273,40	11,34	21,06	10,70	43,10
T3	70,32	126,88	46,80	244,00	10,50	19,50	10,10	40,10
T4	77,93	138,27	49,70	265,90	10,82	20,09	9,90	40,80
mean T0	78,52	135,76	46,80	261,08	10,73	19,92	9,35	40,00
T5	105,00	165,20	47,50	317,70	13,06	24,25	9,10	46,40
T6	111,26	173,84	49,20	334,30	14,11	26,20	9,80	50,10
T7	108,15	154,65	34,60	297,40	12,29	22,82	5,90	41,00
T8	120,31	184,29	49,80	354,40	14,98	27,82	9,00	51,80
mean T1	111,18	169,49	45,28	325,95	13,61	25,27	8,45	47,33
T9	100,72	171,18	57,30	329,20	13,97	25,94	11,60	51,50
T10	117,71	169,99	39,20	326,90	13,93	25,87	7,00	46,80
T11	104,15	152,15	36,30	292,60	11,62	21,58	5,70	38,90
T12	94,04	162,76	56,20	313,00	13,06	24,25	10,10	47,40
mean T2	104,15	164,02	47,25	315,43	13,14	24,41	8,60	46,15
T13	80,90	137,90	46,40	265,20	12,29	22,82	9,50	44,60
T14	96,62	151,68	43,40	291,70	14,11	26,20	9,40	49,70
T15	92,95	148,25	43,90	285,10	12,78	23,73	8,90	45,40
T16	93,77	151,63	46,20	291,60	12,25	22,75	9,40	44,40
mean T3	91,06	147,37	44,98	283,40	12,85	23,87	9,30	46,03

Annex II.4.2 ICP leaves analyses of the species (*Tetragonia tetragonoides*), % g plant⁻¹

Treatment	Fe	Al	Ba	Sr	S	Zn	Cu	Ca	Mg	Na	P	K
T1	0,010	0,005	0,000	0,005	0,382	0,006	0,000	1,831	1,058	2,009	0,485	2,739
T2	0,006	0,003	0,000	0,004	0,379	0,004	0,000	1,531	0,965	1,667	0,465	1,978
T3	0,004	0,002	0,000	0,004	0,298	0,004	0,000	1,526	0,804	1,843	0,369	2,018
T4	0,004	0,002	0,000	0,004	0,333	0,004	0,000	1,638	0,930	1,804	0,451	2,144
T5	0,004	0,002	0,000	0,003	0,377	0,006	0,000	1,582	0,599	4,618	0,362	1,864
T6	0,004	0,002	0,000	0,003	0,324	0,004	0,000	1,237	0,586	4,572	0,357	1,728
T7	0,004	0,002	0,000	0,004	0,337	0,006	0,000	1,722	0,571	4,488	0,402	1,819
T8	0,004	0,002	0,000	0,002	0,307	0,004	0,000	1,073	0,490	4,543	0,372	1,708
T9	0,003	0,001	0,000	0,002	0,286	0,003	0,000	0,836	0,444	4,758	0,329	1,403
T10	0,002	0,001	0,000	0,002	0,307	0,005	0,000	1,089	0,452	5,178	0,348	1,630
T11	0,003	0,001	0,000	0,003	0,356	0,008	0,000	1,427	0,535	5,114	0,371	1,980
T12	0,004	0,002	0,000	0,002	0,281	0,004	0,000	0,874	0,463	4,954	0,366	1,950
T13	0,002	0,001	0,000	0,002	0,265	0,005	0,000	0,753	0,348	5,339	0,363	1,826
T14	0,004	0,002	0,000	0,002	0,306	0,005	0,000	0,836	0,431	5,233	0,366	1,730
T15	0,005	0,002	0,000	0,002	0,314	0,005	0,000	0,938	0,452	5,635	0,394	1,892
T16	0,004	0,002	0,000	0,002	0,326	0,005	0,000	0,973	0,484	5,787	0,403	2,121

Annex II.4.3 ICP soil analyses of the species (*Tetragonia tetragonioides*), % g plant⁻¹

Treatment	Fe	Al	Ba	Sr	S	Zn	Cr	Cu	Pb	Ca	Mg	Na	P	K
T1	1,158	0,560	0,002	0,010	0,493	0,006	0,001	0,003	0,002	2,050	0,456	0,127	0,041	0,089
T2	0,427	0,428	0,002	0,009	0,398	0,005	0,001	0,002	0,001	1,768	0,383	0,076	0,034	0,078
T3	0,516	0,382	0,002	0,013	0,385	0,007	0,000	0,003	0,001	2,225	0,435	0,064	0,034	0,075
T4	0,440	0,363	0,001	0,010	0,394	0,004	0,000	0,003	0,001	1,957	0,388	0,043	0,030	0,051
T5	0,441	0,289	0,001	0,008	0,417	0,004	0,000	0,002	0,001	1,858	0,355	0,793	0,028	0,054
T6	0,390	0,297	0,001	0,009	0,503	0,004	0,000	0,002	0,001	1,962	0,382	0,521	0,025	0,049
T7	1,170	0,632	0,001	0,010	0,321	0,005	0,001	0,003	0,001	2,108	0,523	0,461	0,031	0,071
T8	0,875	0,515	0,001	0,010	0,414	0,005	0,001	0,003	0,001	2,014	0,454	0,496	0,031	0,068
T9	0,921	0,487	0,001	0,010	0,567	0,005	0,001	0,003	0,001	2,127	0,477	1,271	0,031	0,059
T10	0,528	0,278	0,001	0,016	0,498	0,004	0,000	0,003	0,001	2,171	0,386	1,233	0,040	0,058
T11	0,495	0,268	0,001	0,009	0,305	0,004	0,000	0,002	0,001	1,831	0,322	0,889	0,029	0,049
T12	0,583	0,330	0,001	0,008	0,519	0,004	0,000	0,003	0,001	1,936	0,376	1,079	0,030	0,062
T13	0,791	0,428	0,001	0,010	0,520	0,005	0,001	0,003	0,001	2,114	0,470	3,150	0,033	0,065
T14	0,532	0,281	0,001	0,008	0,427	0,004	0,000	0,002	0,001	1,865	0,344	1,523	0,027	0,040
T15	0,744	0,390	0,001	0,008	0,333	0,005	0,001	0,003	0,001	1,861	0,389	1,587	0,029	0,065
T16	0,603	0,291	0,001	0,010	0,577	0,004	0,000	0,002	0,001	2,191	0,454	3,159	0,031	0,066

Annex II.4.4 Biomass production, and ion extraction from the soil, *Tetragonia tetragonioides*, (Na) kg ha⁻¹

Treatment	Area, m ²	Plant density	FY*, (g plant ⁻¹)	DY*, (g plant ⁻¹)	FY, kg ha ⁻¹	DY, (kg ha ⁻¹)	mg g ⁻¹	Ion extr. mg plant ⁻¹	g m ⁻²	Ion extract. (kg ha ⁻¹)
T01	1	12	261	36	31320	4320	20,09	723,4	8,68	37,50
T02	1	12	273,4	43,1	32808	5172	16,67	718,5	8,62	44,59
T03	1	12	244	40,1	29280	4812	18,43	739,0	8,87	42,68
T04	1	12	265,9	40,8	31908	4896	18,04	736,2	8,83	43,25
T11	1	12	317,7	46,4	38124	5568	46,18	2142,8	25,71	143,17
T12	1	12	334,3	50,1	40116	6012	45,72	2290,4	27,48	165,24
T13	1	12	297,4	41	35688	4920	44,88	1839,9	22,08	108,63
T14	1	12	354,4	51,8	42528	6216	45,43	2353,0	28,24	175,52
T21	1	12	329,2	51,5	39504	6180	47,58	2450,3	29,40	181,72
T22	1	12	326,9	46,8	39228	5616	51,78	2423,5	29,08	163,32
T23	1	12	292,6	38,9	35112	4668	51,14	1989,5	23,87	111,44
T24	1	12	313	47,4	37560	5688	49,54	2348,3	28,18	160,28
T31	1	12	265,2	44,6	31824	5352	53,39	2381,4	28,58	152,94
T32	1	12	291,7	49,7	35004	5964	52,33	2600,8	31,21	186,14
T33	1	12	285,1	45,4	34212	5448	56,35	2558,3	30,70	167,25
T34	1	12	291,6	44,4	34992	5328	57,87	2569,6	30,84	164,29

*FY – fresh yield; *DY – dry yield;

Annex II.5.1 Fresh and dry biomass (matter) of the species (*Tetragonia tetragonioides*), g plant⁻¹

Treatment	Fresh mass, g plant ⁻¹				Dry mass, g plant ⁻¹			
	Stem	Leaf	Seed	Total mass	Stem	Leaf	Seed	Total mass
T1	28,30	34,00	23,70	86,00	3,96	6,34	5,27	15,57
T2	31,10	36,00	31,30	98,40	4,78	6,15	6,99	17,92
T3	29,40	22,10	21,30	72,80	3,84	3,46	4,38	11,68
T4	33,70	29,20	18,20	81,10	4,68	5,07	4,09	13,84
mean T0	30,63	30,33	23,63	84,58	4,32	5,26	5,18	14,75
T5	42,60	28,00	27,80	98,40	4,45	4,73	6,29	15,47
T6	54,90	41,50	27,60	124,00	7,20	7,20	6,04	20,44
T7	42,40	27,80	28,90	99,10	5,55	4,81	7,17	17,53
T8	49,80	33,10	22,20	105,10	6,09	5,12	4,44	15,65
mean T1	47,43	32,60	26,63	106,63	5,82	5,47	5,99	17,27
T9	58,70	37,70	27,10	123,50	7,23	6,12	5,36	18,71
T10	35,80	25,80	20,90	82,50	4,75	4,32	4,54	13,61
T11	41,20	29,10	23,40	93,70	4,88	4,76	4,92	14,56
T12	19,10	12,40	8,80	40,30	2,39	1,85	1,58	5,82
mean T2	38,70	26,25	20,05	85,00	4,81	4,26	4,10	13,18
T13	42,90	23,60	20,90	87,40	5,68	3,82	4,49	13,99
T14	39,50	25,70	27,80	93,00	5,32	4,30	6,35	15,97
T15	60,90	38,10	25,80	124,80	7,61	6,20	5,52	19,33
T16	21,00	8,50	5,50	35,00	2,28	0,63	0,83	3,74
mean T3	41,08	23,98	20,00	85,05	5,22	3,74	4,30	13,26

Annex II.5.2 ICP leaves analyses of the species (*Tetragonia tetragonioides*), % g plant⁻¹

Treatment	Fe	Al	Ba	Sr	S	Zn	Cr	Cu	Ca	Mg	Na	P	K
T1	0,03	0,01	0,00	0,01	0,37	0,02	0,00	0,00	2,23	1,28	3,40	0,29	2,10
T2	0,02	0,01	0,00	0,01	0,44	0,01	0,00	0,00	1,81	1,07	3,39	0,23	1,85
T3	0,02	0,01	0,00	0,02	0,54	0,02	0,00	0,00	2,87	1,30	3,12	0,39	2,57
T4	0,00	0,00	0,00	0,01	0,32	0,01	0,00	0,00	1,88	0,89	3,39	0,36	2,16
T5	0,01	0,00	0,00	0,01	0,38	0,02	0,00	0,00	1,47	0,64	6,37	0,28	1,28
T6	0,00	0,00	0,00	0,01	0,29	0,02	0,00	0,00	1,63	0,69	5,36	0,32	1,54
T7	0,00	0,00	0,00	0,01	0,36	0,02	0,00	0,00	1,78	0,69	6,38	0,28	0,91
T8	0,00	0,00	0,00	0,01	0,31	0,02	0,00	0,00	1,64	0,66	5,93	0,39	1,93
T9	0,00	0,00	0,00	0,00	0,23	0,01	0,00	0,00	1,34	0,56	5,52	0,39	1,74
T10	0,00	0,00	0,00	0,01	0,29	0,02	0,00	0,00	1,68	0,68	5,78	0,30	1,77
T11	0,01	0,00	0,00	0,00	0,27	0,02	0,00	0,00	1,47	0,58	6,04	0,33	1,85
T12	0,01	0,00	0,00	0,01	0,32	0,02	0,00	0,00	1,71	0,70	5,68	0,63	2,82
T13	0,00	0,00	0,00	0,00	0,27	0,01	0,00	0,00	1,03	0,45	6,92	0,35	1,49
T14	0,00	0,00	0,00	0,00	0,29	0,02	0,00	0,00	1,05	0,51	7,06	0,35	1,32
T15	0,01	0,00	0,00	0,00	0,19	0,03	0,00	0,00	0,92	0,42	6,22	0,29	1,65
T16	0,00	0,00	0,00	0,00	0,20	0,01	0,00	0,00	1,25	0,60	5,75	0,65	3,26

Bekmirzaev, G./ Effect of saline soils and drought conditions on growth, mineral composition, photosynthesis and water relations of vegetable crops – Thesis (2017)

Annex II.5.3 ICP soil analyses of the species (*Tetragonia tetragonioides*), % g plant⁻¹

Treatment	Fe	Al	Ba	Sr	S	Zn	Cr	Cu	Pb	Ca	Mg	Na	P	K
T1	1,34	0,98	0,00	0,01	0,43	0,01	0,00	0,00	0,00	2,24	0,45	0,14	0,06	0,21
T2	0,82	0,85	0,00	0,02	0,66	0,01	0,00	0,00	0,00	2,52	0,49	0,08	0,06	0,19
T3	1,27	1,15	0,00	0,01	0,39	0,01	0,00	0,00	0,00	2,23	0,57	0,06	0,05	0,19
T4	0,97	0,96	0,00	0,01	0,37	0,01	0,00	0,00	0,00	2,27	0,48	0,04	0,04	0,20
T5	1,37	1,19	0,00	0,01	0,37	0,01	0,00	0,00	0,00	1,88	0,49	0,36	0,04	0,21
T6	1,18	1,09	0,00	0,01	0,39	0,00	0,00	0,00	0,00	1,94	0,45	0,19	0,04	0,20
T7	0,83	0,80	0,00	0,01	0,38	0,00	0,00	0,00	0,00	1,95	0,39	0,21	0,03	0,14
T8	0,76	0,78	0,00	0,01	0,36	0,00	0,00	0,00	0,00	2,10	0,40	0,27	0,03	0,14
T9	1,47	0,95	0,00	0,01	0,41	0,01	0,00	0,00	0,00	1,95	0,41	0,48	0,04	0,19
T10	1,31	1,14	0,00	0,01	0,52	0,01	0,00	0,00	0,00	2,01	0,46	0,37	0,04	0,20
T11	0,85	0,84	0,00	0,01	0,31	0,00	0,00	0,00	0,00	1,91	0,39	0,44	0,04	0,16
T12	0,90	0,88	0,00	0,01	0,34	0,00	0,00	0,00	0,00	1,98	0,41	0,41	0,04	0,16
T13	1,56	1,21	0,00	0,01	0,31	0,01	0,00	0,00	0,00	1,76	0,48	1,04	0,04	0,20
T14	1,11	1,04	0,00	0,01	0,35	0,00	0,00	0,00	0,00	1,91	0,42	0,88	0,03	0,19
T15	1,09	1,05	0,00	0,01	0,35	0,00	0,00	0,00	0,00	1,90	0,43	0,74	0,04	0,21
T16	1,35	1,14	0,00	0,01	0,36	0,01	0,00	0,00	0,00	1,91	0,48	0,83	0,04	0,18

Annex II.6.1 Fresh and dry biomass (matter) of the species (*Portulaca oleracea*), g plant⁻¹

Treatment	Fresh mass, g plant ⁻¹				Dry mass, g plant ⁻¹			
	Stem	Leaf	Seed	Total mass	Stem	Leaf	Seed	Total mass
T1	39,80	26,20	4,60	69,80	1,67	1,12	0,92	3,71
T2	34,70	28,40	5,80	68,90	1,64	1,22	1,16	4,02
T3	29,40	17,40	6,40	53,20	1,29	0,86	1,28	3,43
T4	22,90	20,50	1,60	45,00	0,98	0,93	0,32	2,23
mean T0	31,70	23,13	4,60	59,23	1,40	1,03	0,92	3,35
T5	19,00	17,00	2,00	38,00	0,73	0,76	0,40	1,89
T6	13,50	12,60	1,80	27,90	0,61	0,47	0,36	1,44
T7	32,00	23,50	4,90	60,40	1,64	1,15	0,98	3,77
T8	34,30	30,10	5,20	69,60	1,94	1,48	1,04	4,46
mean T1	24,70	20,80	3,48	48,98	1,23	0,97	0,70	2,89
T9	12,80	11,70	2,60	27,10	0,50	0,43	0,52	1,45
T10	29,70	18,20	3,70	51,60	3,18	2,47	0,74	6,39
T11	21,90	17,70	2,90	42,50	1,05	0,95	0,58	2,58
T12	23,20	14,80	3,20	41,20	1,24	0,78	0,64	2,66
mean T2	21,90	15,60	3,10	40,60	1,49	1,16	0,62	3,27
T13	7,40	4,30	1,10	12,80	0,49	0,31	0,22	1,02
T14	14,90	7,40	3,80	26,10	0,68	0,42	0,76	1,86
T15	11,20	8,10	2,20	21,50	0,54	0,58	0,44	1,56
T16	10,60	6,90	1,80	19,30	0,38	0,48	0,36	1,22
mean T3	11,03	6,68	2,23	19,93	0,52	0,45	0,45	1,42

Annex II.6.2 ICP analyses of leaves, % g plant⁻¹ (*Portulaca oleracea*)

Treatment	Fe	Al	Ba	Sr	S	Zn	Cr	Cu	Ca	Mg	Na	P	K
T1	0,010	0,003	0,005	0,006	0,557	0,006	0,000	0,001	0,808	0,747	0,152	0,787	9,325
T2	0,005	0,002	0,005	0,007	0,602	0,006	0,000	0,001	0,872	0,798	0,167	0,811	8,967
T3	0,010	0,003	0,009	0,007	0,617	0,009	0,000	0,001	0,873	1,212	0,209	0,806	7,832
T4	0,012	0,004	0,004	0,006	0,587	0,006	0,000	0,001	0,759	0,837	0,137	0,709	9,804
T5	0,007	0,002	0,008	0,005	0,615	0,009	0,000	0,001	0,892	0,710	1,012	0,636	8,935
T6	0,007	0,002	0,006	0,006	0,625	0,009	0,000	0,001	0,896	0,834	1,351	0,728	8,659
T7	0,005	0,002	0,006	0,006	0,562	0,009	0,000	0,001	0,829	0,713	1,371	0,738	8,638
T8	0,002	0,001	0,007	0,005	0,612	0,011	0,000	0,001	0,813	1,004	1,180	0,827	8,173
T9	0,004	0,002	0,005	0,005	0,583	0,009	0,000	0,001	0,985	0,801	1,524	0,610	8,334
T10	0,002	0,002	0,007	0,007	0,633	0,009	0,000	0,001	1,130	1,178	1,829	0,797	8,270
T11	0,006	0,002	0,007	0,006	0,605	0,009	0,001	0,001	1,120	0,770	2,081	0,751	8,357
T12	0,003	0,002	0,008	0,005	0,584	0,008	0,000	0,001	0,974	0,760	1,805	0,665	8,095
T13	0,032	0,007	0,011	0,008	0,567	0,014	0,003	0,001	2,027	1,325	2,823	0,809	5,133
T14	0,020	0,003	0,007	0,005	0,645	0,009	0,003	0,001	1,132	0,822	2,781	0,637	8,472
T15	0,014	0,002	0,007	0,006	0,625	0,012	0,002	0,001	1,321	0,952	2,592	0,708	7,779
T16	0,014	0,003	0,009	0,005	0,680	0,013	0,003	0,001	1,262	0,845	2,846	0,709	7,998

Annex II.6.3 ICP analyses of soil, % g plant⁻¹ (*Portulaca oleracea*)

Treatment	Fe	Al	Ba	Sr	S	Zn	Cr	Cu	Pb	Ca	Mg	Na	P	K
T1	0,855	0,513	0,001	0,006	0,439	0,004	0,001	0,003	0,001	2,257	0,530	0,139	0,050	0,142
T2	0,848	0,456	0,001	0,007	0,318	0,004	0,001	0,003	0,001	2,335	0,523	0,095	0,047	0,088
T3	0,151	0,118	0,001	0,005	0,430	0,003	0,000	0,002	0,001	2,037	0,334	0,157	0,027	0,066
T4	0,922	0,508	0,001	0,006	0,362	0,004	0,001	0,003	0,001	2,204	0,503	0,084	0,053	0,112
T5	1,049	0,557	0,001	0,006	0,279	0,004	0,001	0,003	0,001	1,984	0,470	0,841	0,055	0,163
T6	0,917	0,478	0,001	0,006	0,328	0,004	0,001	0,003	0,001	2,190	0,521	1,015	0,050	0,110
T7	0,712	0,357	0,001	0,007	0,346	0,004	0,000	0,003	0,001	2,096	0,434	1,325	0,044	0,063
T8	0,254	0,120	0,001	0,004	0,238	0,002	0,000	0,002	0,001	1,729	0,276	0,951	0,026	0,051
T9	1,199	0,621	0,001	0,005	0,368	0,004	0,001	0,003	0,001	1,928	0,524	1,685	0,059	0,137
T10	0,790	0,410	0,001	0,006	0,355	0,003	0,001	0,002	0,001	2,111	0,474	1,836	0,046	0,065
T11	0,945	0,472	0,001	0,005	0,319	0,004	0,001	0,003	0,001	1,911	0,462	1,659	0,048	0,105
T12	0,805	0,380	0,001	0,005	0,290	0,003	0,001	0,002	0,001	1,791	0,385	1,604	0,042	0,089
T13	0,102	0,083	0,001	0,004	0,238	0,003	0,000	0,001	0,001	1,701	0,256	2,210	0,030	0,094
T14	0,523	0,264	0,001	0,005	0,338	0,003	0,000	0,002	0,001	1,889	0,372	2,673	0,047	0,134
T15	0,919	0,446	0,001	0,005	0,331	0,004	0,001	0,002	0,001	1,900	0,456	2,784	0,060	0,153
T16	0,129	0,091	0,001	0,004	0,268	0,002	0,000	0,001	0,001	1,687	0,265	2,658	0,032	0,097

Annex II.6.4 Biomass production, and ion extraction from the soil, *Portulaca oleracea*, (Na), kg ha⁻¹

Treatment	Area, m ²	Plant density	FY, (g plant ⁻¹)	DY, (g plant ⁻¹)	FY, kg ha ⁻¹	DY, (kg ha ⁻¹)	Ion extr.		Ion extract.	
							mg g ⁻¹	mg plant ⁻¹	g m ⁻²	(kg ha ⁻¹)
T01	1	20	69,8	3,71	17450	927,5	1,52	5,6	112,56	104,40
T02	1	20	68,9	4,02	17225	1005	1,67	6,7	134,18	134,85
T03	1	20	53,2	3,43	13300	857,5	2,09	7,2	143,21	122,80
T04	1	20	45	2,23	11250	557,5	1,37	3,1	61,16	34,10
T11	1	20	38	1,89	9500	472,5	10,12	19,1	382,62	180,79
T12	1	20	27,9	1,44	6975	360	13,51	19,5	389,10	140,07
T13	1	20	60,4	2,77	15100	942,5	13,71	38,0	759,30	715,64
T14	1	20	69,6	2,46	17400	1115	11,80	29,0	580,36	647,10
T21	1	20	27,1	1,45	6775	362,5	15,24	22,1	442,10	160,26
T22	1	20	51,6	2,39	12900	597,5	18,29	43,7	874,27	522,37
T23	1	20	42,5	2,58	10625	645	20,81	53,7	1073,73	692,56
T24	1	20	41,2	2,66	10300	665	18,05	48,0	960,29	638,59
T31	1	20	12,8	1,02	3200	255	28,23	28,8	575,92	146,86
T32	1	20	26,1	1,86	6525	465	27,81	51,7	1034,49	481,04
T33	1	20	21,5	1,56	5375	390	25,92	40,4	808,63	315,37
T34	1	20	19,3	1,22	4825	305	28,46	34,7	694,38	211,79

Annex II.7.1 Fresh and dry biomass (matter) of the species (*Portulaca oleracea*), g plant⁻¹

Treatment	Fresh mass, g plant ⁻¹				Dry mass, g plant ⁻¹			
	Stem	Leaf	Seed	Total mass	Stem	Leaf	Seed	Total mass
T1	13,70	5,10	0,50	19,30	0,86	0,15	0,50	1,51
T2	14,70	8,10	0,30	23,10	0,80	0,37	0,30	1,47
T3	9,60	4,80	0,30	14,70	0,55	0,18	0,30	1,03
T4	5,20	3,40	0,20	8,80	0,23	0,05	0,20	0,48
mean T0	10,80	5,35	0,33	16,48	0,61	0,19	0,33	1,12
T5	11,30	17,40	0,00	28,70	0,48	0,81	0,00	1,29
T6	11,80	8,30	0,40	20,50	0,65	0,56	0,40	1,61
T7	26,40	11,90	0,50	38,80	1,98	1,00	0,50	3,48
T8	24,60	12,50	0,40	37,50	1,76	0,62	0,40	2,78
mean T1	18,53	12,53	0,33	31,38	1,22	0,75	0,33	2,29
T9	34,30	28,30	0,20	62,80	2,53	1,38	0,20	4,11
T10	9,60	6,60	0,20	16,40	0,59	0,23	0,20	1,02
T11	14,80	5,20	0,40	20,40	0,91	0,16	0,40	1,47
T12	27,90	23,30	0,10	51,30	1,99	1,14	0,10	3,23
mean T2	21,65	15,85	0,23	37,73	1,51	0,73	0,23	2,46
T13	27,60	23,00	0,50	51,10	1,86	1,58	0,50	3,94
T14	12,30	14,20	0,00	26,50	0,51	0,46	0,00	0,97
T15	19,90	23,00	0,10	43,00	1,34	1,12	0,10	2,56
T16	18,90	20,70	0,00	39,60	0,97	0,85	0,00	1,82
mean T3	19,68	20,23	0,15	40,05	1,17	1,00	0,15	2,32

Annex II.7.2 ICP analyses of leaves, % g plant⁻¹ (*Portulaca oleracea*)

Treatment	Fe	Al	Ba	Sr	P	S	Zn	Cu	Ca	K	Mg	Na
T1	0,034	0,005	0,005	0,008	0,876	0,459	0,019	0,000	1,006	6,667	1,163	0,758
T2	0,037	0,005	0,005	0,011	0,785	0,977	0,013	0,000	1,527	6,536	1,027	0,438
T3	0,039	0,004	0,004	0,007	0,893	0,566	0,021	0,000	1,200	5,408	1,144	0,594
T4	0,040	0,005	0,004	0,005	0,669	0,303	0,013	0,000	0,891	5,132	0,769	0,361
T5	0,039	0,002	0,003	0,004	0,683	0,486	0,012	0,001	0,522	7,506	0,903	1,354
T6	0,037	0,002	0,005	0,005	0,800	0,439	0,016	0,001	0,720	6,215	0,928	1,806
T7	0,038	0,002	0,003	0,004	0,596	0,317	0,012	0,000	0,571	5,415	0,789	1,422
T8	0,035	0,002	0,005	0,004	0,680	0,317	0,018	0,000	0,553	5,926	0,879	1,261
T9	0,040	0,002	0,003	0,003	0,525	0,310	0,010	0,000	0,418	6,299	0,718	1,571
T10	0,045	0,001	0,003	0,004	0,626	0,290	0,010	0,000	0,575	4,475	0,793	2,516
T11	0,036	0,002	0,005	0,004	0,646	0,386	0,012	0,001	0,768	4,919	0,864	2,645
T12	0,036	0,002	0,003	0,003	0,441	0,295	0,009	0,000	0,352	4,790	0,612	1,647
T13	0,034	0,001	0,004	0,002	0,444	0,251	0,013	0,000	0,478	4,478	0,731	2,328
T14	0,036	0,001	0,003	0,002	0,481	0,423	0,009	0,001	0,400	5,207	0,635	2,296
T15	0,039	0,001	0,003	0,003	0,419	0,336	0,008	0,000	0,419	4,671	0,606	2,376
T16	0,045	0,001	0,003	0,002	0,472	0,291	0,010	0,000	0,351	5,441	0,572	1,934

Annex II.7.3 ICP analyses of soil, % g plant⁻¹ (*Portulaca oleracea*)

Treatment	Fe	Al	Ba	Sr	P	S	Zn	Cu	Pb	Ca	K	Mg	Na
T1	0,685	0,535	0,001	0,006	0,047	0,289	0,004	0,002	0,001	2,144	0,160	0,463	0,283
T2	0,773	0,605	0,001	0,006	0,046	0,359	0,004	0,002	0,001	2,137	0,100	0,466	0,213
T3	0,242	0,176	0,001	0,005	0,030	0,285	0,003	0,001	0,000	2,033	0,045	0,343	0,224
T4	0,660	0,540	0,001	0,006	0,046	0,328	0,004	0,002	0,001	2,265	0,135	0,479	0,218
T5	0,863	0,661	0,001	0,006	0,067	0,355	0,004	0,002	0,001	1,978	0,184	0,452	0,917
T6	0,685	0,507	0,001	0,005	0,047	0,264	0,004	0,002	0,001	2,048	0,106	0,441	0,839
T7	0,803	0,599	0,001	0,006	0,044	0,322	0,004	0,002	0,001	1,912	0,072	0,444	1,035
T8	0,230	0,189	0,001	0,005	0,032	0,347	0,003	0,001	0,000	1,867	0,017	0,312	0,948
T9	0,592	0,451	0,001	0,005	0,045	0,309	0,003	0,002	0,001	1,906	0,079	0,401	1,429
T10	0,642	0,477	0,001	0,005	0,047	0,427	0,003	0,002	0,001	2,043	0,082	0,432	2,079
T11	0,800	0,594	0,001	0,005	0,051	0,390	0,003	0,002	0,001	1,923	0,108	0,444	1,859
T12	1,040	0,779	0,001	0,005	0,052	0,434	0,004	0,002	0,000	1,760	0,074	0,454	1,489
T13	0,125	0,107	0,001	0,004	0,031	0,326	0,002	0,001	0,000	1,746	0,027	0,255	2,002
T14	0,499	0,372	0,001	0,006	0,061	0,423	0,003	0,002	0,000	1,833	0,144	0,372	2,242
T15	0,386	0,288	0,001	0,005	0,045	0,399	0,002	0,001	0,000	1,774	0,088	0,312	2,164
T16	0,428	0,318	0,001	0,005	0,042	0,361	0,003	0,001	0,000	1,790	0,063	0,298	1,820

Annex II.7.4 ICP analyses of drainage water, mg L⁻¹ (*Portulaca oleracea*)

Treatment	Fe	Al	Ba	Sr	P	S	Ca	K	Mg	Na
T1	5,873	0,166	0,034	0,536	1,759	236,087	177,318	17,017	50,972	251,838
T2	5,838	0,084	0,026	0,502	0,690	242,626	170,620	1,978	46,786	194,318
T3	5,804	0,040	0,035	0,367	0,654	227,449	149,017	8,374	48,957	167,685
T4	5,805	0,036	0,032	0,559	1,026	181,264	142,646	29,901	40,712	141,613
T5	5,842	0,144	0,039	0,657	1,653	182,229	166,763	80,920	51,076	666,813
T6	5,803	0,058	0,039	0,521	4,339	226,393	196,531	48,900	52,164	790,757
T7	5,803	0,030	0,045	0,701	2,403	274,656	262,319	17,271	73,199	1016,800
T8	5,811	0,055	0,061	0,567	4,909	332,674	285,431	6,281	87,045	1003,700
T9	5,792	0,027	0,044	0,837	0,487	174,438	211,357	22,352	62,451	864,408
T10	5,819	0,027	0,059	0,985	4,015	218,468	345,441	27,200	98,620	1426,490
T11	5,807	0,023	0,049	0,820	2,173	183,893	252,109	44,320	70,181	1166,700
T12	5,814	0,035	0,051	0,764	3,084	233,928	276,387	30,500	74,228	1400,200
T13	5,807	0,034	0,072	0,649	3,460	282,193	323,079	19,941	92,608	1765,660
T14	5,800	0,023	0,056	0,804	2,369	192,346	259,711	74,261	74,230	1490,190
T15	5,799	0,020	0,050	0,778	2,225	207,216	254,670	50,934	68,348	1463,990
T16	5,801	0,026	0,061	0,498	5,478	246,208	237,066	38,842	67,886	1622,610

Yield, Minerals and Photosynthetic Pigments of *Tetragonia tetragonioides* in Several Water Regimes

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Abstract

The main purpose of this experiment was to study the effect of several irrigation water regimes on *Tetragonia tetragonioides* in semi-arid regions. We measured its effects on growth, biomass production (fresh and dry mass), yield, mineral composition and photosynthetic pigments (chlorophyll, carotenoid and soluble carbohydrates content in leaves). The experiments were conducted in the greenhouse at the University of Algarve (Portugal). The study ran from February to April 2010. Three irrigation treatments were based on replenishing the 0,25 m deep (depth of the pots) to field capacity when the soil water level dropped to 70 % (T1, wet treatment), 50% (T2, medium treatment), and 30% (dry treatment) of the available water capacity. The experimental results showed that the leaf mineral composition of chloride and sodium, the main responsible ions for the soil salinization and alkalization in arid and semi-arid regions, enhanced with the decrease of the soil water content. However, the minimum amount of chlorophyll, carotenoids and soluble carbohydrates in leaf content were obtained in the medium and driest treatment. On the other hand, growth differences among the several irrigation regimes were very low, and the crop yield increased in the dry treatment (T3) compared to the medium treatment (T2). Thus, the high capacity of salt removing species as *T. tetragonioides* suggests an advantage of its cultivation under dry conditions. This research recommends the use of *T. tetragonioides* in arid and semi-arid regions.

Keywords: arid and semi-arid regions; soil water content; irrigation water regimes; crop yield; dry conditions.

INTRODUCTION

In arid and semi-arid regions, such as the Mediterranean regions, supplies of good quality water allocated to agriculture are expected to decrease because most of available fresh / potable water resources have already been mobilized (Costa et al., 2011). According to FAO (2011), due to the shortage of water, there is an enlargement of saline land in agricultural areas in some of the developing countries. As a result, yield is decreasing provoking an increasing cost of the agricultural products (Bekmirzaev et al., 2011).

Soil salinization has been recognized worldwide as being among the most important problems for crop production in arid and semi-arid regions (Szabolcs, 1994). Water deficit and salinity are the major limiting factors for plant productivity, affecting more than 10 % of arable land on our planet, resulting in a yield reduction of more than 50% for most major crop plants (Bartels and Sunkar, 2005). The abiotic stresses that include a component of cellular water deficit, usually noted are salinity and low temperature stresses can also severely limit crop production (Ansari and Lin, 2010). Abiotic stresses, such as drought, salinity, extreme temperatures, chemical toxicity and oxidative stress are serious threats to agriculture and result in the deterioration of the environment. Abiotic stress is the primary cause of crop loss worldwide. (Boyer, 1982; Bray et al., 2000). This problem is intensified in coastal areas due to sea water intrusion. This results from reduced ground water levels as the water demand exceeds the annual groundwater recharge (Ben Asher et al., 2002). As reported above, some of the emerging regions in risk of increasing levels of salinization of their soils are located in the Mediterranean Basin (Nedjimi et al., 2006; Beltrao et al., 2014), Australia (FAO, 2008), Central Asia (Hamidov et al., 2013), the Middle East (Ben Asher et al., 2002), and Northern Africa (Yensen, 2008). Salinity is one of the rising problems causing tremendous yield losses in many regions of the world, especially in arid and semiarid regions. The use of salt-tolerant crops does not remove the salt and hence halophytes that have the capacity to accumulate and exclude the salt can be an effective way (Hasanuzzaman, 2014).

Intensive irrigation of agricultural crops with high level of water mineralization causes salts to accumulate in the root zones, which adversely affects the crop productivity. In order to reduce such negative impact, a regulated

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deficit irrigation (RDI) technique was adopted to combat salinization in the arid and semi-arid environments by reducing the water application during certain growth stages of the crops (Cameron et al., 2006).

When RDI is not feasible, halophyte crops might be a solution for the salinization of agricultural land. These crops can be irrigated by, for example, seawater, salt contaminated phreatic sheets, brackish water, wastewater or drainage water from other plantations (Grieve and Suarez, 1997; Yensen, 2008; Ben Asher et al., 2012b).

Hence, our aims were to choose a salt removing crop, tolerant to salinity and at the same time with interest as a food crop, and to test its drought tolerance through its response to several water regimes. Tetragonia (*T. tetragonioides*) was the selected crop. In the previous experiment was demonstrated its capability as a high biomass horticultural leaf crop, producing a plant dry weight of 40,000-50,000 DM kg ha⁻¹, if the plant population density is around 75,000 plants ha⁻¹ (Neves et al., 2008).

MATERIALS AND METHODS

Experimental procedure. The experimental work was conducted in the greenhouse of Horto at the University of Algarve, Faro, Portugal (37°02'37.1N 7°58'30.8W), from February to April 2010. The salt removing species *T. tetragonioides* were the selected species. Four leaf plants were transplanted to 7 L (litre) capacity and 0,25 m depth randomized pots on February, 10. The number of plants per pot was eight, with three replications. The species were irrigated with tap water every three days until the beginning of the treatments (February, 1 – March, 8). A nitrogen fertigation treatment was started on March, 8 (daily applied with concentrations of 2 mM NO₃⁻ and 2 mM NH₄⁺, being the cumulative amount of NO₃NH₄ (g plant⁻¹) to the end of the experimental studies (April, 22). The electrical conductivity (EC_w) of irrigation water was 0,6 dS m⁻¹ and pH 7.

The treatments consisted of three irrigation regimes in a randomized complete block design with three replicated treatments based on replenishing the 0,25 m deep (depth of the pots) to field capacity when the soil water level dropped to 70 % (T1, wet treatment), 50% (T2, medium treatment), and 30% (T3, dry treatment) of the available water capacity (aw). This concept has been developed by Veiheimeyer and Hendrickson (1931), where 'aw' is the range of available water that can be stored in soil and is available for growing crops. It was assumed by the same authors that the soil available water content readily to plants (θ_{aw}) is the difference between the volume of water content at field capacity (θ_{fc}) and at the permanent wilting point (θ_{wp}), as follows:

$$\theta_{aw} = \theta_{fc} - \theta_{wp} \quad (1)$$

The watering volume was estimated to replenish of the soil profile to field capacity at a depth of 0,25 m. The volumetric soil water content - m³ water / m³ soil (m³ m⁻³) – was determined just before the water application (see Table 2).

To control soil water along the soil profile, the irrigation frequency and water amounts, the pots were weighted every day. The soil water content was monitored periodically, gravimetrically measured for a 0,00–0,25 m depth.

The plants were harvested after the treatments (April, 26), washed with distil water for a few minutes and wiped with paper. Then the fresh weight (FW) was measured. The fresh samples were dried in a forced drought oven at 70° C for 48 hours and the dry weight (DW) was measured. Plant materials were collected for chemical analyses. The soil electrical conductivity (ECs) and pH were measured before and after the experiment.

Climate conditions in the greenhouse. The average climatic data during the experimental period in the greenhouse were: maximal relative humidity – 88,4 %, minimal relative humidity – 11,3 %, maximal temperature – 45,8 °C and minimal temperature – 11,4 °C.

During the experimental period, the relative humidity of the greenhouse was increased, and the maximal temperature decreased.

Plant growth, chemical and biochemical analyses. During the vegetation period we measured the stem length, number of nodes and number of leaves of *T. tetragonioides* every seven days.

The plants' leaves were analysed on total growth and mineral compositions (Na, Cl, N, K, P, Ca and Mg). Dried leaves and stems were finally grounded and analyzed by using the dry-ash method. The levels of Na and K were determined by a flame photometer and the remaining cations (Na, K, Ca and Mg) were assessed by atomic absorption spectrometry. Chloride ions were determined in the aqueous extract by titration with silver nitrate according to the method of Radojevic and Bashkin (1999). Plant nitrogen (N) content was determined by the Kjeldhal method. Phosphorus was determined by the colorimetric method according to the vanadate – molybdate method. All mineral analyses were only performed on the leaves.

The analysis of pigments was done on four discs with a size of 0,66 cm and a total area of 1,37 cm². For sugars there were ten discs, with the size of a disc of 0,66 cm and a total area of 3,42 cm². The amount of photosynthetic pigments (chlorophyll a, b total and carotenoids) was determined according to the method of Lichtenthaler (1987). Shoot samples (0,25 g) were homogenized in acetone (80%). The extract was centrifuged at 3.000 g and

absorbance was recorded at wavelengths of 646,8 and 663,2 nm for chlorophyll assay and 470 nm for carotenoids assay by the Varian Cary 50 UV-Vis spectrophotometer. Chla, Chlb, ChIT and carotenoids were calculated. Soluble sugars (glucose) in leaves were extracted as described by Dubois et al. (1956). The change in absorbance was continuously followed at 340 nm using an Anthos hat II microtiter-plate reader (Anthos Labtec Instrument, Hanau).

Statistical analyses. Data (n = 4) were examined by a one – way ANOVA test. Multiple comparisons of the means of data between different irrigation water regimes within the plants were performed using the Duncan’s test at a P<0.05 significance level (all tests were performed with SPSS Version 17.0 for Windows program).

Soil. Table 1 shows the soil texture and soil parameters before the experiment. According to FAO, based on the USDA particle-size classification, the soil texture was sandy clay loam. The soil parameters show that the range in the soil’s pH value is slightly alkaline and that the electrical conductivity (ECs) is 1,1 dS m⁻¹ (non - saline soil) at 25 °C.

Table 1 - Soil parameters before the experiment

Soil texture		Soil parameters		
Sand (%)	58,9	Field capacity θ_{fc} (m ³ m ⁻³)	0,238	pH (H ₂ O) 7,7
Silt (%)	18	Wilting point θ_{wp} (m ³ m ⁻³)	0,119	EC _e * (dS m ⁻¹) 1,1
Clay (%)	24,1	Available soil water θ_{aw} (m ³ m ⁻³)	0,119	
Classification	Sandy Clay Loam	Bulk density (g cm ⁻³)	1,41	

EC_e* - Electrical conductivity of the extract of a saturated soil paste (dS m⁻¹)

Table 2 shows the volumetric soil water content - m³ water / m³ soil (m³ m⁻³) - just before the water application. The volumetric soil water content in soil ranges between 0,202 – 0,155 m³ m⁻³.

Table 2 - Volumetric soil water content - m³ water / m³ soil (m³ m⁻³) - just before the water application

Treatment	Determination	Θ (m ³ m ⁻³)
T1	$\theta_{wp} + 0,70 \cdot \theta_{aw}$	$\Theta_1 = 0,202$
T2	$\theta_{wp} + 0,50 \cdot \theta_{aw}$	$\Theta_2 = 0,178$
T3	$\theta_{wp} + 0,30 \cdot \theta_{aw}$	$\Theta_3 = 0,155$

RESULTS

Effect of irrigation water regimes on plant growth. Table 3 shows the irrigation water regimes’ effects on the *T. tetragonioides*’ growth (stem length, number of nodes and number of leaves). A significant effect on the stem length can be seen. In the beginning of the experiment, the stem length of the crop showed very low variations between T1 and T2 treatments. During the last three weeks of the experimental period, the stem length increased showing equal differences between each two treatments – T1 and T2, and T2 and T3 (Δ stem length ~.5 cm). The number of nodes and number of leaves was also higher in treatment T1.

Table 3 - Effect of irrigation water regimes on stem length, nodes and number of leaves of *T. tetragonioides*

Treatment	<i>Tetragonia tetragonioides</i>		
	Stem length (cm)	Number of nodes	Number of leaves
T1	38,8 ± 1,9 ^a	22,5 ± 0,6 ^a	9,9 ± 0,58 ^a
T2	34,2 ± 0,5 ^b	18,1 ± 0,6 ^b	8,1 ± 0,37 ^b
T3	29,3 ± 1,5 ^c	19,2 ± 0,8 ^b	9,5 ± 0,22 ^a

Fresh (FW) and dry (DW) weights. The fresh weight (FW) of *T. tetragonioides* species shows low variations among treatments. There was a low increase of the fresh weight of stem, leaves and seeds in treatment T1. Surprisingly the obtained results in treatment T3 were slightly higher than in treatment T2.

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The obtained results of dry matter show that the stem, leaves and seeds of treatment T1 were slightly higher than in the other treatments. There were very low variations of dry matter between T2 and T3 treatments (Table 4).

Table 4. Fresh and dry weight of species *T. tetragonioides*

Treatment	Fresh weight, g				Dry weight, g			
	Stem	Leaves	Seeds	Total mass	Stem	Leaves	Seeds	Total mass
T1	539,9	450,9	50,2	1041,0	45,6	29,4	7,7	82,7
T2	478,6	401,0	34,1	913,7	36,2	23,4	6,5	66,1
T3	497,9	433,9	35,1	966,9	38,4	22,5	6,3	67,2

Effect of irrigation water regimes on mineral composition in leaves. Table 5 shows the effects of water application treatments on the mineral composition of *T. tetragonioides*' leaves, as follows: the total nitrogen leaf content of the species showed low variations among treatments. There was an enhancement of chloride and sodium concentration with the decrease of water content. There was a general decrease of phosphorus, calcium, potassium, iron and magnesium in the leaf content under drought conditions.

Table 5 – Mineral composition of leaves of *T. tetragonioides*

Treatment	Leaf mineral composition (%)			
	Na	Cl	Mg	Ca
T1	3,4 ± 0,19 ^c	1,3 ± 0,07 ^c	0,43 ± 0,03 ^a	0,0006 ± 0,04 ^a
T2	4,3 ± 0,25 ^b	3,0 ± 0,22 ^b	0,36 ± 0,02 ^b	0,0004 ± 0,02 ^b
T3	4,4 ± 0,42 ^a	3,5 ± 0,09 ^a	0,35 ± 0,01 ^b	0,0004 ± 0,03 ^b
	N	K	P	Fe
T1	0,34 ± 0,02 ^b	4,2 ± 0,23 ^a	3,1 ± 0,02 ^a	0,0001 ± 0,02 ^b
T2	0,34 ± 0,01 ^b	3,7 ± 0,17 ^b	2,7 ± 0,04 ^b	0,0002 ± 0,03 ^a
T3	0,37 ± 0,01 ^a	4,1 ± 0,14 ^{ab}	2,7 ± 0,02 ^b	0,0001 ± 0,01 ^b

Effect of irrigation water regimes on chlorophyll content in leaves. The response of chlorophyll content in leaves of *T. tetragonioides* to the different water regimes is shown in Table 6. The results show that the chlorophyll content was higher in treatment T2 and lower in treatments T1 and T3. These results are in agreement with the findings obtained by Pirzad et al. (2011) where the minimum amount of chlorophyll a, chlorophyll b and total chlorophyll were obtained from the wettest and driest treatment in *Matricaria chamomilla* L. potted plants. Similar results were obtained by Bradford and Hsiao (1982) and Chartzoulakis et al. (1993).

Table 6 – Chlorophyll in leaf content of *T. tetragonioides*

Treatment	Chlorophyll content					
	C _a , mg m ⁻²	C _b , mg m ⁻²	C _{a+b} , mg m ⁻²	C _a , mg g ⁻¹ (DW)	C _b , mg g ⁻¹ (DW)	C _{a+b} , mg g ⁻¹ (DW)
T1	232,1 ± 12,1	80,9 ± 5,8	312,9 ± 17,6	26,5 ± 1,4	9,2 ± 0,7	35,8 ± 1,9
T2	289,9 ± 7,3	110,1 ± 4,5	400,1 ± 11,4	32,3 ± 1,3	12,2 ± 0,5	44,5 ± 1,7
T3	253,9 ± 13,2	91,6 ± 5,6	345,6 ± 18,7	29,8 ± 1,6	10,7 ± 0,6	40,5 ± 2,1

Effect of irrigation water regimes on carotenoid content in leaves. Carotenoids in all higher plants are synthesized and located in the chloroplast along with the chlorophyll. Table 7 shows the carotenoid content of the leaves of *T. tetragonioides* under different irrigation water regimes. The maximum leaf carotenoid content was 8,44 mg g⁻¹ DW in treatment T2. On the other hand, for wetter and drier treatments (T1 and T3) the carotenoid content was lower, respectively, 7,2 and 7,9 mg g⁻¹ DW. Lower carotenoid content was also obtained for stress water regimes of some fenugreek varieties (Hussein and Zaki, 2013). Moreover, a decrease of leaf carotenoid content in green beans was attributed to water stress and the Vegetation Index (NDVI) showed the highest correlations with the chlorophyll (a, b and total) and carotene content of leaves (Koksal et al., 2010).

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Table 7 - Carotenoid in leaf content of *T. tetragonioides*

Treatment	Leaf carotenoid content	
	Car, mg m ⁻²	Car, mg g ⁻¹ (DW)
T1	62,9 ± 3,2	7,2 ± 0,3
T2	75,8 ± 2,2	8,4 ± 0,4
T3	67,6 ± 4,1	7,9 ± 0,5

Effect of irrigation water regimes on soluble carbohydrates content in leaves. The irrigation water regimes had a slight effect on the soluble carbohydrates' content in leaves of the species *T. tetragonioides*. The glucose and soluble carbohydrates content in leaves increased in the wet (T1) and dry (T3) treatments, respectively glucose – 0,58 and 0,57 mg ml⁻¹ and soluble carbohydrates – 1,71 and 1,67 mg. These results are confirmed by Redillas et al. (2012). There was decreased glucose (0,54 mg ml⁻¹) and soluble carbohydrates (1,59 g) content in leaves in the medium (T2) treatment (Table 8).

Table 8 - Soluble carbohydrates in leaf content of *T. tetragonioides*

Treatment	Soluble carbohydrates				
	Glucose, mg ml ⁻¹	Area, cm ²	Solub. carb, mg	DW, cm ²	Solub. carb, g
T1	0,57 ± 0,05	3,42 ± 0,0	1,67 ± 0,14	0,001 ± 0,0	1,9 ± 0,15
T2	0,54 ± 0,03	3,42 ± 0,0	1,59 ± 0,08	0,001 ± 0,0	1,8 ± 0,09
T3	0,58 ± 0,03	3,42 ± 0,0	1,71 ± 0,09	0,001 ± 0,0	2,0 ± 0,12

Crop yield. *T. tetragonioides* produced significant amounts of dry matter, which ranged from 82,7 to 66,1 g plant⁻¹. The partition of the plant dry matter to plant organs was changed by the effect of the irrigation water regimes (Table 9). The fact that the species was irrigated during the vegetation period T1 (70 % - wet treatment), significantly increased the dry biomass of the species at the harvest time averaging 6616 kg ha⁻¹. The dry matter of the species decreased when the soil water decreased in treatments T2 (50% - medium treatments) and T3 (30% - dry treatment). There was no significant difference between treatments. The obtained results confirmed that the species *T. tetragonioides* is tolerant to drought condition. The yield of the crop shows that the drought had less effect than the salinity (6616 – 5288 kg DM ha⁻¹). These results are confirmed by the previous study of Bekmirzaev et al. (2011).

Table 9 – Crop yield of *T. tetragonioides*

Treatment	<i>Tetragonia tetragonioides</i>				
	FW	DW	Yield, %	FW kg ha ⁻¹	DW kg ha ⁻¹
T0	1041,0	82,7	7,94	208200	16540
T1	913,7	66,1	7,23	182740	13220
T2	966,9	67,2	6,95	193380	13440

DISCUSSION

The experimental results showed several effects of the water irrigation regimes on the growth, mineral composition and photosynthetic pigments of *T. tetragonioides*, as follows:

- Plant growth (stem, leaves and seeds) increased slightly with enhancement of the water level (near the field capacity), the growth difference between the drier water regimes is very low. This increase is probably due to the increase of stomatal conductance, and consequently, transpiration and CO₂ fixation are higher. Hence, it is not surprising that experimental results in which the only variable was water application agree quite well on this supposed theory.

- Leaf mineral composition of chloride and sodium, respectively, the main responsible ions for the soil salinization and alkalization in arid and semi-arid regions, enhanced with the decrease of the soil water content. These contents were very high in relation to other plants, showing their high capacity as salt removing species.

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- There was a general low decrease of phosphorus, calcium, potassium, iron and magnesium in leaf content under drought conditions, probably due to the chloride and potassium competition.
- The total nitrogen leaf content of species showed a very low variation probably due to the same fertigation for all irrigation treatments.
- The minimum carotenoids' amount of chlorophyll a, chlorophyll b and total chlorophyll were obtained from the wettest and driest treatment in *T. tetragonioides* plants, probably due to higher plant senescence provoked by these regimes.
- The glucose and soluble carbohydrates content in leaves increased in the driest treatments and enhanced tolerance to drought condition.
- The yield of the species increased in the wettest and driest treatments.

CONCLUSIONS

As concluding remarks, it can be suggested that the *T. tetragonioides* are species tolerant to drought conditions. Its capacity as a halophyte and salt removing species when the soil water content decreased has been shown, suggesting its use in arid and semi-arid regions. Moreover, growth and yield differences in the several irrigation regimes were very low, which suggests the other important advantage of these species - its cultivation under dry conditions, when used as a leafy vegetable for human consumption or for animal feeding. Nevertheless, more research is needed in order to test the plant development under drier conditions, in arid and semi-arid climates.

ACKNOWLEDGEMENTS

The authors are thankful to the financial support of ERASMUS MUNDUS ECW Lot 8, 2010-2011 and to CIEO (Research Centre for Spatial and Organizational Dynamics), University of Algarve, Portugal.

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