INNSPUB

International Journal of Biosciences | IJB |
ISSN: 2220-6655 (Print) 2222-5234 (Online)
http://www.innspub.net
Vol. 11, No. 2, p. 163-169, 2017

RESEARCH PAPER OPEN ACCESS

Effect of Mono-ammonium phosphate 12-61 and salinity on growth, total chlorophyll and proline content of bean *Vicia faba L minor*, Variety Sidi Aich

Nouri Tayeb[1*], Reguieg Yssaad Houcine Abdelhakim[1], Latigui Ahmed[2], Oubachir Ahlem[3], Djebbar Naziha[3] and Bouyahia Hadj[3]

[1]*Laboratory of Biodiversity and Conservation of Water and Soils, Université ofMostaganem, Algeria*

[2] *Faculty of nature and life sciences, University of IBN Khaldoun, Tiaret, Algeria.*

[3]*University Center SALHI AHMED,Naama, Algeria.*

Key words: Salinity, MAP, Bean, Growth, Biochemical's parameters

http://dx.doi.org/10.12692/ijb/11.2.163-169 Article published on August 30, 2017

Abstract

Soil salinity is a major constraint to agricultural development in the whole world, particularly in arid and semi-arid areas. The purpose of this study is to evaluate the combined effect of four salinity levels 0, 3, 6, 9dS/m and three concentrations of Monoammonium phosphate (MAP) 12-61 on Growth, biochemical's parameters as total Chlorophyll and the content proline of bean *Viciafaba* L *minor*, variety of SIDI AICH. Sowing was carried on plastic cylinders of 40cm in height containing soil of different salinity levels (0.3.6 and 9dS/m), and treated with Mono-Ammonium Phosphate 12.61 at doses of 1 and 1.5g/ 5kg of dry soil. However, The Irrigation of plants was performed with distilled water and a Hoagland nutrient solution. Also, Saline and MAP effect were investigated on leaf and root area, stem and root length, total chlorophyll and proline content. So, the results showed that; the strong salinity of 9dS/m caused a reduction in the length of the stems and roots, the leaf and root area, total chlorophyll. Nevertheless, the treatment of plants by the MAP at the dose of 1 and 1.5g was significantly improved leaf and root area, stem length and proline content under the effect of salinity.

* **Corresponding Author:** Nouri Tayeb ✉ nouritayeb@yahoo.fr

Introduction

Soil salinity is a major environmental constraint to crop production, (Rengasamy, 2010). The arid and semi-arid lands account for one third of the world's surface, nearly 400 million hectares (Jouve and *et al.*, 2002, Baatoour and *et al.*, 2004). Worldwide, it is estimated that almost 800 million hectares of land are affected by salt, either by salinity (397 million ha) or by sodisation conditions associated with sodium levels (434 million ha). Indeed, salinity covers more than 6% of the total surface of the planet (Benbrahim, 2008) of which 3.8% is located in Africa (Manchanda, 2004). On the other hand, excessive intake of fertilizers to increase yields also causes soil salinity (Quadir and Coster, 2004). Salt stress,absolutely like many other abiotic stresses, inhibits plant growth. High concentrations of salts cause ion imbalance and hyperosmotic stress in plants (Zhu, 2001; Rontain and *et al.*, 2002). Faced with these stress conditions, plants implement adaptation and defense strategies (Neeting, 2002). The accumulation of organic compound has been demonstrated in several plant species subjected to salt stress. This accumulation varies according to species, stage of development and level of salinity. Differences in the accumulation of solutes such as proline and total soluble sugars between the control plants and the plants subjected to salt stress are very important. (El midaoui and *et al.*, 2007).Many studies confirm the inhibitory effect of salinity on biochemical processes, of which photosynthesis is the most important.

The effect on photosynthesis can be gauged from the effect on the photosynthetic pigments. The results of specific studies (Sultana and *et al.*, 2000; Tort and Turkyilmaz, 2004; Misraand *et al.*, 2006; Murillo-Amador *and et al.*, 2007; Taffouo and *et al.*, 2010) clearly indicate that salinity reduces the content of photosynthetic pigments in treated plants.

The objective of this study is to investigate the major effect of different salinity levels (0, 3, 6,9dS/m). Also for acidifying fertilizer, monoammonium phosphate (MAP) 12-61 with doses of 1 and 1.5g /5kg of dry soil on the growth and physiology of bean *Vicia faba L minor*, Variety Sidi Aich.

Materials and methods

The trial was shared between biology laboratory and environmentally controlled green-house located at Nâama University Center, Algeria (33°16'36.67"N, 0°19'8.54"O) from the period of November 2016 to May 2017.

Methodology

Preparation of the culture substrate

The culture substrate was composed of the saline soils (S1, S2 and S3) obtained by adding NaCl. In addition to the control S0 (Table. 01) The soil was analyzed for physicochemical properties before filling at 5kg cylinder^{-1}. This soil has under gone several preparations by drying and sieving to remove plant debris, for obtaining than fine element.

Table 1. Physico-chemical properties of culture substrate.

Substrate	EC (dS/m)	Textural class	OM (%)	Content of NaCl (g/kg of soil)
S0	0.24	Sandy clayloam	0.21	0.60
S1	3	Sandy clayloam	0.21	4.5
S2	6	Sandy clayloam	0.21	7.5
S3	9	Sandy clayloam	0.21	10.25

Plant material

The vegetable material used is a bean (*Vicia faba L minor*), Indeed, Sidi Aich local Variety were provided by the ITGC of Constantine, Algeria.

Seeding

The seeds are disinfected with 8% bleach for 10 minutes and rinsed several times with distilled water to remove any trace of chlorine. Sowing took place in cylinder filled with five kilograms of each substrate used. Plants grow under salinized substrates of 90 days.

Conduct of experimentation

Irrigation

Irrigation was performed with distilled water. The water holding capacity was determined by the

difference between the quantity of water supplied by irrigation and that recovered after 24h. Reserve easily water used corresponded between 30 and 60% of water retention capacity.

That is to say; 350 and 700mL cylinder^{-1}. Plants were watered three times a week, twice with deionized water and once in the nutrient Hoagland solution diluted to 1 ppt.

Salinity levels and concentration of monoamonium phosphate

Four salinity levels of 0.24dS m-1 (Control original soil), 3dSm-1, 6 dS m-1 and 9 dS m-1 were developed by using calculated amount of NaCl in each cylinder combined with three treatments of monoammonium phosphate (0g, 1g and 1. g). Table 2 shows the main chimical and physical characteristics of this acidifing fertilizer.

Table. 2. Physico-chemical properties of Mono ammonium phosphate (MAP) 12-61.

Parameters	Contents
P2O5 (%)	61
P (%)	27
N total (%)	12
N-NH_4^+ (%)	12
pH (concentration 0.1%)	4.7
EC (dS/m) (Concentration 0.1%)	0.86

Destructive analyses

After 90 days of salt and monoammonium phosphate 12-61 treatment, beans were harvested by extracting and washing roots from the soil. Then, the roots and aerial parts were separated, and leaves and roots area, the stems height, roots length, total chlorophyll and amount of proline were measured.

Biometric parameters

Leaf and root area

Leaves and roots were scanned with Epson SX218 as a model and the images are processed through *Imag J* software after the conversation across the pixels image.

Length of stems and roots

After 90 day, the plant removed from the cylinder and the aerial part of the root one is separated, In fact, the stems length of bean is measured by means of a graduated scale.

Biochemical analyses

Total chlorophyll content:

Extraction

Chlorophyll content was determined according to the method developed by Francis *et al.* Besides, it is carried out in the mixture of acetone and ethanol (75% and 25%) in volume and 80% and 20% in concentration. On the other hand, an amount of 0.01 g fresh leaf samples is added 10ml of a mixture of acetone and volumes ethanol Respectively 75 and 25% with two concentrations of 80 and 20%. However, after 10min of centrifugation at 5000 rpm at 4°C.

Determination of total chlorophyll

Spectrophotometric reading was taken at 645 and 663 nm wavelengths. So, Data were assessed in these formulates:

Chl a (µg/g MF) = 12,7x DO (663) – 2,59x DO (645) x V/(1000x W) .

Chl b (µg/g MF) = 22,9 x DO (645) – 4,68 x DO (663) x V/(1000x W).

Chl(a+b) (µg/g MF) = Chl a + Chl b

V: volume extracted solution and W the weight of fresh material of the sample

Proline amount

The proline was analyzed by the method of Bergman and Loxley (1970). The optical density was measured using a spectrophotometer at 505nm.

Experimental design

The experimental design was adopted at two main factors,First, Monoammonium phosphate with 3 doses 0, 1 and 1.5g/ 5kg of soil , the second factor is a salinity with four levels 0.24, 3, 6 and 9dS.m^{-1}.

Statistical analysis

The experiment was arranged in a completely randomized design with four replications .data collection and calculation were analyzed by STAT BOX 6.40, comparison of means were tested for significance using Student –New mean test, at 0.05 level of probability.

Results

The ANOVA results revealed very highly significant effects of Monoammonium phosphate 12-61 and

salinity factors ($p<0.001$) starting with the leaf and the root area of bean. Leaf area of plants was inversely proportional to the salt stress. Indeed, the salinity level of S3 caused a significant decrease in leaf area in all substrates compared to the control plants where the leaf area reached its minimum 14.09cm² at this salinity level in substrates treated with 1g of MAP. The addition of 1 g of mono-ammonium phosphate to substrates with salinity levels of S1 and S2 significantly improved the leaf area of plants. It is noted that the leaf area was important (67.03cm²) in the plants grown for instance: in the S1 substrates and absolutely treated with 1g of Mono-Ammonium Phosphate Nevertheless, the root surface of the bean. So, the addition of 1g of mono - ammonium phosphate in saline levels of S2 and S3 improve the root surface of the plants compared to the controls (Tables 3). The stems and roots length were significantly decreased ($p<0.001$) as the salinity level increased surely.

The lowest values of stem and root length were recorded in plants grown in substrates with salinity level S3 and 31.00cm and 9.52cm, respectively. The substrates treatment with the concentrations of MAP at 1 and 1.5g improved the stems length of the stressed plants at the salinity levels of S1, S2 and S3 (Table.4).

The ANOVA results revealed significant effects of salinity and MAP factors ($p<0.05$) on the *Bean* proline content. Proline content in the aerial part of '*Vicia faba L minor*' reached its maximum 37,40 and 33,40µmol 100mg-1 of dry matter at highly salinity level of S2 in substrates treated at 1 and 1.5g of MAP , respectively. (Table.05) . The applied salinity reported significant effects ($p<0.05$) on total chlorophyll in the bean leaves. The highest values were observed in the plants found in the substrates treated with 1.5g of MAP and stressed at 3 and 6dS/m with 19.36 and 20.94 (µg/g MF) gradually (Table.06).

Table 3. Mean of leaf and root area (cm²) of bean under the effect of salinity (dS /m) and MAP (g).

Salinitylevels	Doses of MAP (g)	Leaf area (cm²)	Root area (cm²)
So	0	44.72abc	111.81a
	1	44.89abc	49.84b
	1.5	47.11ab	49.10b
S1	0	35.18bcd	130.92a
	1	67.03a	41.08b
	1.5	46.50ab	41.51b
S2	0	29.62bcd	30.08b
	1	44.41abc	65.10b
	1.5	32.29abc	35.68b
S3	0	18.31d	14.78b
	1	14.09d	19.13b
	1.5	20.89cd	38.72b

Table 4. Mean of stem and root length (cm) of bean under the effect of salinity levels and MAP (g).

Salinitylevels	Doses of MAP(g)	Stem length (cm)	Rootlength (cm)
So	0	39.62	14.81 abcd
	1	50.50	17.53 abc
	1.5	52 .00	16.90 abc
S1	0	39.50	20.08 a
	1	49.12	11.66 cd
	1.5	49.50	15.50 abc
S2	0	29.00	18,09ab
	1	45.62	14.71 abcd
	1.5	45.00	14.14abcd
S3	0	31.00	12.76 bcd
	1	36.70	9.52 d
	1.5	44.00	14.49abcd

Table 5. Mean of proline content (µMol/100mg DM) of bean under the effect of Salinity (dS /m) and MAP (g).

Salinitylevels	Doses of MAP (g)	Proline (µMol/100mg DM)
S0	0	12,20b
	1	16,40b
	1.5	13,80b
S1	0	14,00b
	1	13,40b
	1.5	23,00b
S2	0	14,20b
	1	37,40a
	1.5	33,40a
S3	0	19,50b
	1	14,80b
	1.5	12,00b

Table 6. Mean of total chlorophyll content (µg/g MF) of bean under the effect of salinity (dS /m) and MAP (g).

Salinitylevels	doses of MAP (g)	Total chlorophyll content (µg/g MF)
S0	0	13.20
	1	11.79
	1.5	14.52
S1	0	15.21
	1	16.01
	1.5	19.36
S2	0	16.76
	1	16.53
	1.5	20.94
S3	0	14.11
	1	13.99
	1.5	16.02

Discussion

The result of this study showed that salinity was significantly reduced the leaf and root area, stem and root length at higher salinity levels (S2 and S3) compared to plants control. Benidire *et al.*, (2014) report that salinity exerts an inhibitory effect on the growth of *Vicia faba L minor*. Seedlings which results the decrease in the length of the stems as the increase function in medium salinity. Generally speaking, the growth in length decreases with the stress increase intensity according to what several authors have observed in the pea (okçu and *et al.*, 2005), the bean (Abdul *et al.*, 2011). The immediate response of salt stress reduces the expansion rate of the leaf surface. On the other hand; this expansion stops if the salt concentration increases (Wang and *et al.*, 2000). Also, the total chlorophyll content was decreased under stongly salinity levels Afroz and *et al.* (2005) noted a decrease in chlorophyll concentrations in mustard greens under salt stress conditions. Seeman and Critchley (1985) noted that; the decrease in the concentration of foliar chlorophylls and lower activity of ribulose1.5 bisphosphate carboxylase/oxygenase (rubisco) in bean irrigated with a solution enriched with NaCl.

According to the results, the low level salinity S1 (3dS/m) did not trigger any salt stress. Because according to Kasmi and al (2012), Latigui and Dellal (2009, this threshold is not harmful for vegetable crops. The doses of 1 and 1.5g of MAP were significantly improved the chlorophyll and plants proline content to salinity level of S2. Hence, this explained the high concentration of proline in response to salt stress. In fact, the results were consistent with those of Reguieg and *et al.* (2012). The confirmed of the osmotic adjustment were major physiological traits of tolerance to environmental stresses. Generally, It was achieved through an accumulation of osmoregulatory compounds leading to a reduction of osmotic potential, allowing the maintenance of turgor potential (Sen, 2010; Ingweye *et al.*, 2010). Nouri, (2012) working on the bean under salt stress conditions, explained the resistance of the bean to salt stress by an accumulation of proline in the various organs of the plant. The addition of Monoammonium Phosphate to substrates affected by different levels of salinity (S2 and S3) significantly improve the growth of plants by the increase of leaf area, root area, stem length of the bean *Vicia faba* L

minor under strongly stress. Heavy or frequent P fertilizer applications may be necessary to maintain adequate crop growth and yield (Pierzynski, 1991). MAP is a good bottom fertilizer where the phosphorus is completely soluble in water has a desirable acidifying effect for the basic and saline soils whose pH becomes strongly acid (Lhoucaine, 2000).

Conclusion

From the results, it is concluded that the high salinity levels was significantly reduced the growth of Bean *Vicia faba* L minor. But it was tolerated salinity to S1 (3dS/m) and application of monoammonium phosphate in substrate at the doses of 1 and 1.5g improved the growth and biochemical parameters of bean under salinity stress.

Acknowledgement

The authors are thankful to the laboratory of Biodiversity and Conservation of Water and Soils, University of Mostaganem.

References

Abdul Qados AMS. 2011. Effect of salt stress on plant growth and metabolism of bean plant *Vicia faba* L. Journal of the Saudi Society of Agricultural Sciences **10**, 7-15.
https://doi.org/10.1016/j.jssas.2010.06.002.

Afroz S, Firoz M, Hayat S, Siddiqui MH. 2005. Exogenous application of gibberellic acid counteracts the ill effect of sodium chloride in mustard. Turkish journal of biology **29,** 233-236.

Baatour O, M'rah S, Ben Brahim N, Boulesnem F, Lachaal M. 2004. Physiological response of the gestation (*Lythyrus sativus*) to the salinity of the environment. Review of the arid regions **1,** 346-358.

Benbrahim KF, Ismaili M, Benbrahim SF, Tribak A. 2004. Problèmes de dégradation de l'environnement par la désertification et la déforestation: impact du phénomène au Maroc. Science et changements planétaires/Sécheresse **15(4),** 307-320.

Benidire L, Daoui K, Fatemi ZA, Achouak W, Bouarab L, Oufdou K. 2015. Effect of salt stress on germination and seedling of *Vicia faba* L. Journal of Materials and Environmental Science **6(3),** 840-851.

Bergman I, Loxley R. 1970. New spectrometric method for the determination of proline in tissues hydrolysates. *Analytical Chemistry* **42**, 702-706.

El Houcine. 2000. National program for the transfer of technology in agriculture, Bultin.Institue of agricutural and veterinary. University of hassan II.

El midaoui M, Benbella M, Aït houssa A, Ibriz M, Talouizte A. 2007. Contribution to the study of some mechanisms of adaptation to salinity in cultivated sunflower (*Helianthus annuus* L.). Review Hommes Terre et Eaux **136,** 29-34.

Ingweye JN, Kalio GA, Ubua JA, Umoren EP. 2010. Nutritional Evaluation of Wild Sicklepod (*Senna obtusifolia*) Seeds from Obanliku, South-Eastern Nigeria. American Journal of Food Technology **5,** 1-12 http://scialert.net/abstract/?doi=ajft.2010.1.12.

Jouve P, Corbier-Bartuaux C, Cornet A. 2002. Combating Desertification in Development Projects. A Scientific Perspective on the SFM Experience in Sub-Saharan Africa and the Maghreb Mimosa (Eds). French Development Agency (AFD) and French Scientific Committee on Desertification CSFD 162 p.

Kasmi A, Latigui A, Mettai K, Sahli B, Dilem A. 2012. Use of Sewage sludge and Fiber palm Co-compost as Components of Substrates *Lycopersicum esculentum* and *Cucumismelo* cultivated in Soil less crop. American journal of plant Physiology **7(2),** 97-103.
https://doi.org/10.3923/ajpp.2012.97.103.

Manchanda G, Garg N. 2008. Salinity and its effects on the functional biology of legumes. Acta Physiologica Plant **30,** 595-618.
https://doi.org/10.1007/s11738-008-0173-3.

Misra N, Ansari MS, Gupta AK. 2006. Differential response of scavenging of reactive oxygen species in green gram genotype grown under salinity stress. American journal of plant physiology **1,** 41-53 https://doi.org/10.1016/j.plantsci.2005.02.013.

Murillo-Amador B, Yamada S, Yamaguch T, Puente ER, Serrano NA, Hernandez LG, Aguilar RL, Dieguez ET, Garibay AN. 2007. Salinity toxicity influence of calcium silicate on growth physiological parameters and mineral nutrition in two legume species under salt stress, Journal of Agronomy and Crop Science **193(6),** 413-421.
https://doi.org/ 10.1111/j.1439-037X.2007.00273.x.

Netting A. 2002. pH, abscissic acide and the integration of metabolism in plants under stressed and non –stressed conditions –II-Modifiaction in mode of metabolism induced by variation in the tension of the water column and stress. Journal of Experimental Botany **53(356),** 151-173.

Nouri T. 2012. Response of the bean *vicia faba* L to saline stress, case of asandysoilmodified by bentonite. Magister thesis, university of Mostaganem Algeria.

Okçu G, Kaya MD, Atak M. 2005. Effect of salt and drought stresses on germination and seedling growth of pea (*Pisum sativum* L.). turkish journal of agriculture and forestry 29, 237- 242.

Pierzynski GM. 1991. The chemistry and mineralogy of phosphorus in excessively fertilized soils. Critical reviews in *environmental science and technology* **21,** 265-295.

Quadir M, Coster JD. 2004. Crop and irrigation management strategies for saline-sodic soils and waters aimes at environmentally sustainable agricultures. Science of the Total Environment **323,** 1-19.
https://doi.org/10.1016/j.scitotenv.2003.10.012.

Reguieg Yssaad HA, **Latigui** A, **Nouri** T**, Bessafi L.** 2012. Effect of salt stress and bentonite on the germination and proline content of *Vicia faba* L. Plant var. 'Semilla violeta' and 'Reine mora'.American journal of plant physiology **7,** 212-219.
http://scialert. net/abstract/?doi=ajpp.2012.212.219.

Rengasamy P. 2010. Soil processes affecting crop production in salt affected soils. Functional Plant Biology **37(7),** 613-620.
https://doi.org/10.1071 /FP09249

Rontain D, Basset G, Hanson AD. 2002. Metabolic engineering of osmoprotectant accumulation in plants. Metabolic engineering **4,** 49-56**.**
https://doi.org/10. 1006/mben.2001.0208

Seeman JR, Criteheey C. 1985. Effect of salt stress on the growth, ion content, Phaseo/us vulgaris L. Planta **164,** 151-162.
https://doi.org/10.1007/BF00396077

Sen S. 2010. S-nitrosylation process acts as a regulatory switch for seed germination in wheat. American Journal of Plant Physiology **5,** 122-132. https://doi.org/10.3923/ajpp.2010.122.132.

Sultana N, Ikeda T, Itoh R. 1999. Effect of NaCl salinity on photosynthesis and dry matter accumulation in developing rice grains. *Environmental* and Expérimental Botany journal **42,** 211-220.

Taffouo VD, Wamba OF, Yombi E, Nono GV, Akoe A. 2010. Growth, yield, water status and ionic distribution response of three bambara groundnut (*Vigna subterranean* (L.) verdc.) landraces grown under saline conditions, International Journal of Botany **6(1),** 53-58.
http://dx.doi.org/10.3923/ijb.2010.53.58.

Tort NB, Turkyilmaz A. 2004. Physiological investigation on the mechanisms of salinity tolerance in some barley culture forms. Journal Faculty Science Ege University **27,** 1-16.

Wang Y, Nil N. 2000. Changes in chlorophyll, ribulose biphosphate carboxylase oxygenase, glycine betaine content, photosynthesis and transpiration in Amaranthus tricolor leaves during salt stress. Journal of Horticultural Science *and* Biotechnology **75,** 623-627.

Zhu JK. 2001. Plants salt tolerant. Trends Plant Science **6,** 66-71