# EFFECT OF NaCI ON THE GERMINATION, SEEDLING AND SOME METABOLIC CHANGES IN SWEET BASIL (OCIMUM BASILICUM)

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## (Received February 8, 1993; revised December 13, 1994)

The effect of different salinity levels (up to 120 mM NaCl) on germination, Seedling growth, dry matter and some related metabolic parameters of sweet basil (*Ocimum basilicum*) was studied. During germination it tolerated salinity up to 60 mM NaCl. However, seedling growth and dry matter production were decreased by NaCl greater than 40 mM NaCl. Soluble carbohydrate and proline concentration of seedling increased with increase of salinity. However, soluble protein and amino acids were decreased by salinity. In long term (36 days) experiment, growth decreased with increasing salinity levels, except at 5 and 10 mM NaCl where growth was increased. Water content in the whole test plant increased at 5 mM NaCl and decreased as salinity increased to 50 mM NaCl. These results may lead to the conclusion that, sweet basil is a medium tolerant crop and is considered as glycophyte.

Key words: Ocimum basilicum, Salinity, Germination, Growth.

#### Introduction

It has been generally recorded that salinity adversely affects seed germination and seedling growth as well as some relevant metabolic processes of glycophytic plants [1-5]. However, the directions and magnitude of these changes varied according to the level and duration of salinization treatment as well as the plant species used. This variation in species response and the need to select some of our economic plants for cultivation in saline soils, necessitated a series of investigations to test their salinity tolerance and to adapt in their physiological adaptation to salinity.

Plants apparently rely on several mechanisms by which they adapt to salinity stress. These include accumulation of organic molecules like soluble carbohydrates, soluble proteins,proline and possibly other compounds which may act as non-toxic cytoplasmic osmotica in various salt-tolerant plants [2,6,7].

Reehan (sweet basil) Ocimum basilicum is regarded as one of the most important medicinal plant, highly valued for its aromatic oils [8]. In the kingdom of Saudi Arabia, reehan is a legendary plant admired for the exquisite scent of leaves and flowers. Traditionally it is grown in almost every home garden. Thus keeping in view its traditional value economic and medicinal importance, the aim of the present study is to provide information on seed germination, some metabolic changes in seedlings of sweet basil grown under different salinization levels.

## **Materials and Methods**

The seeds of sweet basil (*Ocimum basilicum*) were obtained from the Botanical Garden, King Abdulaziz University, Jeddah. The germination experiments were performed as described by Maftoun and Sepaskhah [9]. The following salinity levels were used: 0.0 (control), 5, 10, 20, 40, 60, 80, 100, 120 mM NaCl in 1/10 Hoagland solution [10]. Twenty seeds were placed on absorbent pads in Petri dishes to which 30 ml of the experimental solution was added. Seeds were considered to be germinated after the radicle had emerged from the testa. After 10 days of germination the length of the plumule and radical of germinated seeds were measured. The fresh plumule and radical were then dried in an aerated oven at 70°C during which successive weighing was carried out until a constant dry weight was reached. Free proline [11], total free amino acids [12], carbohydrate [13] and soluble protein [14] in the seedlings (10-days old) were determined.

Plant growth. The seeds were germinated on two layers of filter paper in flat trays 3 cm deep, and irrigated with distilled water until they were large enough to handle (after 14 days). The seedlings were then transported to boxes (2 liters) which had been painted black, containing Hoagland solution [15]. Seedling were suspended above the solution by supporting them with non-absorbent cotton inside holes in small depressions in the box lid. The plants were grown in growth cabinets with a 16 hr photoperiod of approximately 280  $\mu$ mol m<sup>-2</sup>S<sup>-1</sup>, the temperature, ranging between 27°C during the dark periods. Salinity treatments were imposed (when the seedlings were 21 days old) by adding salt; 0,5,10,20 and 50 mM NaCl in 1/2 Hoagland solution. After 15 days from salinization, the fresh shoots and roots were dried in an aerated oven at 80°C until a constant dry weight was reached. The relative water content was determined according to Bars and Weatherley [16], Zidan [17] for details. Shoot/root ratio was

calculated. Three replicates were used and the data were statistically analysed to calculate the least significant difference (L.S.D.).

## **Results and Discussion**

The final germination percentage of sweet basil seeds remained unchanged up to the level of 60 mM NaCl (Table 1). Above this level, germination decreased with the increased NaCl level. The highest level used (120 mM NaCl) completely inhibited the germination of sweet basil seeds.

Low salinity (5-40 mM NaCl) stimulates the plumule and radicle length (Table 2) as well as production of dry matter in sweet basil seedlings. Above these levels, the values of dry matter were reduced with the increase of the NaCl concentration. The water content of seedlings of the test plant decreased whatever the salinization level used, however, the higher concentrations (60-100 mM NaCl) slightly increased these values (Table 2). Salinity induced a significant increase in the content of soluble carbohydrates of seedlings at all salinity levels above 20 mM NaCl (Table 3). The soluble protein

TABLE 1. EFFECT OF VARIOUS CONCENTRATIONS OF NaCl

SALINITY ON THE GERMINATION OF SWEET BASIL SEEDS.

NaCl mM	24 hr	48 hr	72 hr
0.0	$80 \pm 3.0$	100	100
100A 5	$81 \pm 5.0$	100	100
10	$81 \pm 3.1$	100	100
20	$71 \pm 5.0$	$91 \pm 5.2$	100
40	$69 \pm 5.2$	$78 \pm 2.5$	100
60	$61 \pm 4.6$	$72 \pm 1.5$	100
80	$34 \pm 4.7$	$62 \pm 2.2$	$71 \pm 5.1$
100	r [6] arstong oldi	$31 \pm 3.8$	$47 \pm 3.5$
120	- digni	ord) were determ	a gab (91) agun

(Data expressed as % of control). All values are the means of three replicates  $\pm$  SE of means.

TABLE 2. EFFECT OF VARIOUS CONCENTRATIONS OF NaCl ON	
Plumule and Radicle Length (cm/seedling), Water	

CONTENT	AND DRY	MATTER (	g 100 SEED	LINGS).

NaCl	Plumule	Radical	Dry	Water
(mm)	length	length	matter	content
0.0	3.37	4.3	5.89	94.1
5	3.57	4.36	6.59	93.4
10	4.76	5.3	6.81	93.2
20	5.07	5.77	7.84	92.2
40	5.46	6.16	7.97	92.0
60	3.25	4.20	5.67	94.3
80	3.01	4.1	5.13	94.9
100	2.40	3.6	5.77	94.2
LSD at 5%	0.40	0.73	0.83	0.73
	and the second			

content remained statistically unchanged up to 20 mM NaCl and then showed a significant decrease. There is a significant increase in the concentration of proline in seedling at all salinity levels above 10 mM NaCl. The pattern of changes in the total free amino acids of the seedlings was opposite to that obtained for changes in proline (Table 3).

All salinity levels decreased the dry matter production of shoots and roots of test plant grown for 36 days (Table 4). It was also observed that the values of shoot/root ratio decreased whatever salinity used. The water content of the whole plant increased at all salinity levels when compared to the control (Table 4).

The observed tolerance in the rate of germination and the final germination percentage and consequently seedling growth of the test plant subjected to the lower and moderate salinization were found to be associated with more or less constant values of water content. However, other authors [15, 18] pointed out that the decrease in final germination percentage was always associated with a decrease in water absorption. This behaviour of the water content of seedlings was found to be linked with a pronounced accumulation of organic solutes (soluble carbohydrates and proline) which might play an

TABLE 3. EFFECT OF VARIOUS CONCENTRATIONS OF NaCl Salinity on Carbohydrate and Protein (mg per g dry wt.), Proline and Other Amino Acids (unot es der g dry wt.)

NaCl (mM)	Soluble carbohydrate	Soluble protein	Proline	Amino acids
0.0	89.4	86.3	3.9	30.8
5	91.4	89.7	4.8	27.8
10	91.3	88.2	5.5	25.4
20	92.9	83.7	6.2	21.6
40	98.0	78.9	7.4	21.5
60	106.8	77.8	8.1	19.3
80	109.0	69.5	9.8	17.7
100	110.2	60.6	10.8	15.8
LSD at 5%	7.3	6.8	1.7	3.2

TABLE 4. EFFECT OF VARIOUS CONCENTRATIONS OF NaCl Salinity on Dry Matter (mg/3 plants), Water Content (G/G d.wt.)

(0/0 D. W1.)					
NaCl	Dry matter		Shoot/Root	Water	
(mM)	Shoot	Root		content	
0.0	18.86	4.95	3.81	8.7	
5	15.2	4.14	3.67	12.4	
10	15.0	4.28	3.50	11.8	
20	13.2	4.65	2.84	11.6	
50	11.3	4.28	2.64	10.9	
LSD at 5%	1.7	0.43	0.38	0.73	

important role in increasing the internal osmotic pressure (compatible solutes) [1,6]. The losses in protein were accompanied by increases in soluble carbohydrates. This leads to the conclusion that salt tolerance seems to be linked with an equilibrium and interconversion between carbohydrates and nitrogen metabolism, whereas saline injury leads to metabolic disturbances in both components. Similar results were obtained by other authors [19,20]. In the present study, the pattern of changes in proline was opposite that of other amino acids, indicating that the increase in proline is at the expense of other amino acids through an effect of salinity in promoting their conversion [21, 22]. The significance of proline accumulation in response to salt has been contentious. Chandler and Thorpe [23] have suggested that proline may either enhance salt tolerance or be a symptom of cell damage during salt/water stress. Also Shah et al. [24] suggested that proline accumulation is important for cell growth only when a certain level of salt stress is attained, and this level will depend on the presence or absence of other protectant mechanisms in the tissue.

Dry matter production of seedling remained more or less unaffected up to 40 mM NaCl. Moreover, 10 and 20 mM NaCl stimulate a pronounced increase in seedling growth (root and shoot lengths) and dry matter production of test plant seedlings as compared with control plants. Different workers [2, 25] recorded a promotion in the dry weight of some salinized seedlings. The dry matter production of the whole plant decreased at salinity levels were found to be generally lowered when compared to the control. This inhibitory effect of high salinity level may be due to an inadequate respiratory system to provide the energy for active transport, or there may be an insufficient number of carriers required for the fast rate of ion uptake for cell elongation under saline conditions [26,27]. The recorded increase in water content of salinized plants was in agreement with the results obtained by other workers [28, 29] working with glycophytic plants. They stated that salinity increased the concentration of the hormone abscissic acid, which in turn induces stomatal closure, and as a result the rate of transpiration decreases and consequently an increase in the water content of plant tissues take place.

Finally, it can be said that the tolerance of this experimental plant during seedling growth might be linked to the accumulation of soluble carbohydrates and proline, which in turn increased their ability for water absorption under salinity. However, during its growth it is a medium tolerant crop and is considered as glycophyte.

### References

- 1. J.B. Drossopoulos, A.J. Karamanos and C.A. Niavis, Ann. Bot., **59**, 173 (1987).
- 2. M.A. Shaddad and M.A. Zidan, Beitr, Trop. Landwirtsch,

Vet. Med., 27, 187 (1989).

- C.R. Hampson and G.M. Simpson, Can. J. Bot., 68, 524 (1990).
- 4. S. Hardegree and W.E. Emmerich, Ann. of Bot., **66**, 587 (1990).
- 5. U. Schmidhalter and J.J. Oertli, Plant and Soil, **132**, 243 (1991).
- T.J. Flowers, P.P. Troke and A.R. Yeo, Ann. Rev. Plant Physiol., 28, 89 (1979).
- R. Munns, C.J. Brady and E.W.R. Barlow, Aust. J. Plant Physiol., 6, 379 (1979).
- 8. J.B. Chogo and G.J. Grank, Nat. Prod., 44, 308 (1981).
- M. Maftoun and A.R. Sepaskhah, Can. J. Plant Sci., 58, 295 (1978).
- D.R. Hoagland and I.D. Arnon, Calif. Agric. Exp. Sta. Cir., 347 (1950).
- L.S. Bates, R.P. Waldran and I.D. Teare, Plant and Soil, 39, 205 (1973).
- 12. S. Moore and W. Stein, J. Biol. Chem., 17, 367 (1948).
- 13. F.W. Fales, J. Biol. Chem., 193, 113 (1951).
- O.H Lowery, N.J. Roserbrough, A. Al-Farr and R.J. Randall, J. Biol, Chem., **193**, 265 (1951).
- M.M. Heikal, A.M. Ahmed and M.A. Zidan, Bull. Fac. Sci. Assiut Univ., 9, 15 (1981).
- H.D. Barrs, P.E. Weatherby, Aust. J. Biol. Sci., 15, 413 (1962).
- 17. M.A. Zidan, Arab Gulf J. Sci. Res., 11, (2), 201 (1993).
- 18. A. Dell' Aquila, Ann. Bot., 69, 167 (1992).
- 19. P.S. Thakur and V.R. Rai, Biologia Plantarum, 24, 96 (1982).
- 20. G. Singh and V. R.Rai, Biologia Plantarum, 24, 7 (1982).
- 21. S.F. Boggess, C.R. Stewart, Q'spinall, and L.E. Paleg, Plant Physiol., **58**, 398 (1976).
- 22. A.D. Hanson, C.E. Nelsen, A.R. Pedersen and E.H. Everson, Crop Science, **17**, 720 (1977).
- 23. S.F. Chandler and Ta. A. Thorpe, Plant Cell Reports, 6, 176 (1987).
- 24. S.H. Shah, S.J. Wainwright and M.J. Merrett, New Phytol., 166, 37 (1990).
- 25. M.M. Heikal, M.A. Shaddad and A.M. Ahmed, Biol. Plant., 24, 124 (1982).
- H. Greenway and R. Munns, Annu. Rev. Plant Physiol., 32, 149 (1980).
- 27. A.R. Yeo, Physiol. Plant, 58, 214 (1983).
- M.A. Zidan, Photosynthetic Activity, Mineral Composition and Growth of some Legumes as Affected by Salinity, M.Sc., Thesis, Assiut University, Egypt, (1979), pp. 150.
- 29. A. Poljkoff-Mayber and J. Gale, *Ecological Studies 15: Plants in Saline Environments* (Springer-Verlag Berlin Heidelberg New York, 1975), pp. 197-205.