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Effects of Certain Salts and Their Combinations on Germination and Seedling Development of *Securigera securidaca* Linn

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ABSTRACT. Effects of NaCl, Na_2SO_4 and $MgSO_4$ and their combinations on germination and seedling development of bitter lentil were studied. Both of these processes were inhibited by the three salts used. Seedling development, however, was more sensitive to salt treatment than germination. The effect of $MgSO_4$ on germination was essentially osmotic, while that of NaCl and Na_2SO_4 was presumably due to toxic or ionic actions. Both Na and Cl were more toxic than Mg and SO_4. Magnesium sulfate combined with NaCl and with Na_2SO_4 did not only overcome the inhibitory effects of NaCl and Na_2SO_4 on germination and seedling growth, but there occurred also a remarkable synergism in both processes, as a result of such combinations.

Salinity is a major problem in crop production affecting many kinds of plants, especially those which are sensitive to the presence of high concentrations of salts in the soil. Saline habitats are usually dominated by NaCl, but many other salts such as $MgSO_4$, Na_2SO_4 and KCl may also be present. Nonhalophytes differ in their response to different concentrations of the various electrolytes in their environment (Bernstein and Hayward 1958, Flowers *et al.* 1977, Greenway and Munns 1980).

The ability of seeds to germinate in saline environments is essential for good establishment of the plants in saline soils. *Securigera securidaca* is a herb in the family Leguminosae, and is found in the western parts of Asia, and in some parts of the Middle East, North Africa and southern Europe (Ghosh and Dutta 1950). Seeds of bitter lentil are used in folk medicine as a remedy for diabetes (Ghosh and Dutta 1950). In an attempt to propagate this plant in Iraqi soils where salinity is high, the study of its tolerance to various salts and their combinations became necessary.

Material and Methods

Seeds of bitter lentil, *Securigera securidaca* Linn., were obtained from the Botany Department of the Ministry of Agriculture and Agrarian Reform, Abu-Ghraib, Iraq. All germination tests were carried out in 9 cm glass Petri dishes lined with two layers of Whatman No. 1 filter paper, and moistened with 5 ml of the test solution. Four replications of 25 seeds per dish were used in all experiments. The effects on germination and seedling development of NaCl, Na_2SO_4 and $MgSO_4$ separately and in some combination, were studied. The concentration of the salt solutions was expressed in terms of osmotic pressure (OP), and the OP was calculated using the Van't Hoff formula (described by Pauling and Pauling 1975). A stock solution with an OP of 16 atmospheres was prepared for each of the three salts, and successive dilution, with OP varying by one atmosphere, were made in each case. For preparing the mixtures of the salts, equal volumes from the two given salt solutions of the same osmotic pressure were combined. Distilled water was used as a control. All Petri-dishes were kept in an incubator (Germinator Model No. 1200) with a front glass panel exposed to room light conditions at a constant temperature of 20°C. Germination was defined as emergence of the radicle from the seed coat. Germination counts were made daily and the percentage germination represents the final count on the 7*th* day when the seedling lengths were also measured.

Results and Discussion

1. Germination

Germination of the seeds of bitter lentil was 100 percent in water, but this percentage was reduced to different extents in solutions of NaCl, Na_2SO_4 and $MgSO_4$, depending on the nature of the salt and its osmotic pressure (Fig. 1 and Table 1). First there was a slight reduction in the germination percentage in each of these salt solutions, at the lowest concentration investigated, *i.e.*, 1 atm. The germination percentages were 92 in NaCl, 89 in $MgSO_4$ and 85 in Na_2SO_4. Although reduction in germination at this concentration appeared to be primarily due to an osmotic effect (diffusion pressure deficit gradient), it is suggested that NaCl has perhaps some beneficial effect (Clarkson and Hanson 1980, Greenway and Munns 1980, Carlson *et al.* 1983).

The second important observation in the present work is that as the concentration of the three salts increased to 2 atm., the germination percentage in NaCl dropped sharply to 61, compared to 85 percent in Na_2SO_4 and 88 percent in $MgSO_4$. This might be explained on the basis that accumulation of Na and Cl was continued at a faster rate, reaching the toxic or the so called "ion excess" level (Eaton 1942, Ryan *et al.* 1975, Miller and Chapman 1978, Greenway and Munns 1980, Clarkson and Hanson 1980).

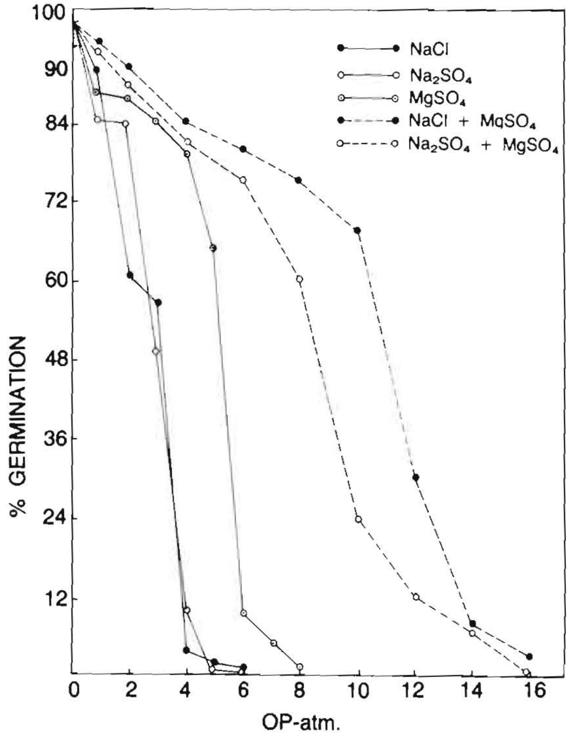

Fig. 1. Seed germination of bitter lentil as affected by osmotic concentrations of different salts and their

Table 1. Seed germination of bitter lentil as affected by osmotic pressure of various salts and their combinations.

OP (atm)	Germination percentage together with standard error					
	Treatments					
	H_2O	NaCl	Na_2SO_4	$MgSO_4$	Na_2SO_4 + $MgSO_4$	NaCl + $MgSO_4$
0.0	100					
1		92.0 ± 1.0	85.3 ± 2.8	89.2 ± 0.7	96.0 ± 1.0	96.0 ± 1.0
2		61.3 ± 3.1	85.3 ± 1.2	88.0 ± 1.7	90.6 ± 0.6	93.3 ± 0.7
3		57.3 ± 3.1	49.3 ± 2.9	85.3 ± 1.0		
4		4.0 ± 0.0	10.6 ± 1.5	80.0 ± 1.0	85.3 ± 1.2	84.0 ± 1.0
5		2.7 ± 0.6	2.7 ± 0.6	65.7 ± 2.2		
6		1.3 ± 0.6	0	10.6 ± 1.6	74.6 ± 1.6	81.3 ± 1.2
7		0		5.3 ± 1.5		
8				1.3 ± 0.6	60.0 ± 6.1	76.0 ± 1.0
10				0	24.0 ± 4.6	68.0 ± 3.6
12					12.0 ± 1.0	30.7 ± 5.5
14					6.7 ± 1.5	8.0 ± 1.0
16					0	2.7 ± 1.2

It is known that NaCl has high permeating characteristics and that less tolerant species might accumulate more Na and Cl than the tolerant ones (Bernstein and Hayward 1958, Greenway and Munns 1980, Hafeez and Marshall 1981). Of course we do not know how the toxic effect of the ions is exerted; but the possibility includes effects on membrane permeability and on enzyme activities (Greenway and Munns 1980).

At the next level of concentration, *i.e.*, 3 atm. a sharp reduction in germination percentage occurred in Na_2SO_4 (49 percent germination), while germination in $MgSO_4$ remained high (85 percent). As in the case of NaCl, one is inclined to speculate that in Na_2SO_4 the uptake of Na was increased with an increase in its concentration, reaching also the toxic level. The effect of SO_4 in this case cannot be assessed but it is presumed that the sulfate ion is easily assimilated into organic compounds within the cells (Clarkson and Hanson 1980). The effect of $MgSO_4$, on the other hand, remained essentially osmotic, as its inhibitory action on germination increased slowly and gradually up to 5 atm. It is not easy to distinguish between osmotic and specific ion effects, but the progressive and gradual decrease in germination with increasing osmotic pressure can be explained in terms of classical osmotic theory (Bernstein and Hayward 1958).

It appears, therefore, that the germinating seeds of bitter lentil are very sensitive to salinity, especially with respect to NaCl and Na_2SO_4 in which very little germination takes place at OP levels above 3 atm.

Synergism between $MgSO_4$ and the other salts

Results related to the combined effect on germination of $MgSO_4$ with NaCl and with Na_2SO_4 are shown in Figure 1 and Table 1. In each of these combinations, $MgSO_4$ did not only mitigate the inhibitory effect of NaCl and Na_2SO_4, but there occurred, in addition, a considerable synergistic effect between $MgSO_4$ and the other two salts. The pattern of this synergism was essentially similar in both cases, and its magnitude increased as the concentration of the mixtures increased. It is noticed, however, that the magnitude of such synergism was higher in the case of $MgSO_4$ with NaCl.

The fact that the inhibitory effect of one salt could be overcome by another salt has been known for a long time (Harris and Pitman 1918, 1919). Yet it is not easy to explain this unique synergism observed in the present work. It is possible that Mg reduces somehow the uptake of Na or/and causes its leakage out of the cell. It is also possible that Mg causes more compartmentation of the different ions within the cells which in turn leads to more favourable water balance and turgor pressure necessary to growth. This is probably a characteristic of many non halophytes in their adaptation to salinity (Flowers *et al.* 1977, Greenway and Munns 1980). However, this explanation, though tempting, remains highly speculative.

2. Seedling Growth

Although the effects of the salts and their combinations on early seedling growth can not be easily separated from their effects on germination, and although such effects in both cases followed in general the same pattern (Fig. 1 and Fig. 2), yet there are several significant differences between the two processes in their response to salt treatments. First, the inhibitory effect of the three individual salts on seedling growth was remarkably higher than that on germination, even in the lowest concentration, *i.e.*, 1 atm. At this low concentration, for example, the average reduction of germination in the three salts was about 11 percent, compared with an average reduction of about 85 percent in seedling growth (Table 1 and Table 2). Another point of interest here is that the inhibitory effect of $MgSO_4$ on seedling development was similar to the effects of NaCl and Na_2SO_4.

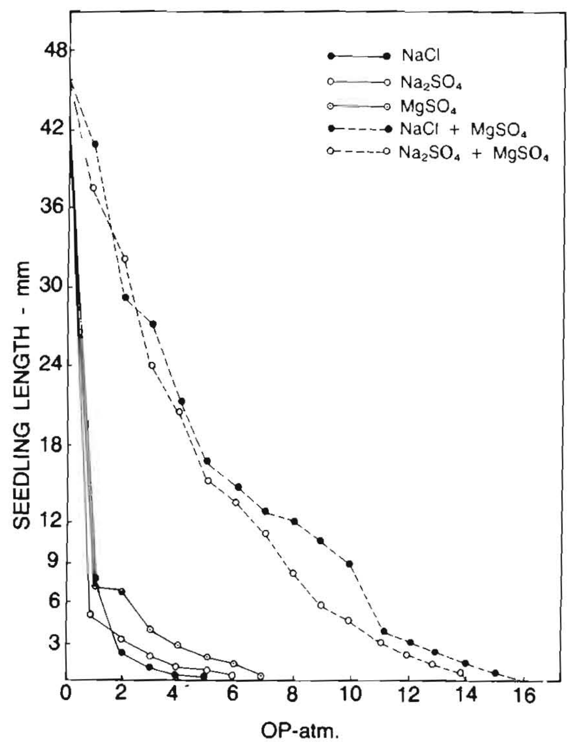

Fig. 2. Seedling development of bitter lentil as affected by osmotic concentration of different salts and their combinations.

The process of germination of course differs from seedling growth in that it usually involves an inhibitional component. Germination also involves active cell division as the main component of differentiation, whereas early seedling development is characterized by actively expanding cells with large vacuoles. There is also evidence that ion concentrations are related to cellular differentiation (Bernstein and Hayward 1958), and it is, therefore, expected that different patterns of ion distribution and concentration occur in the two main types of differentiating cells. But whether the high sensitivity of the expanding cells to salt treatments resulted from unfavourable water balance, or from the toxic effects of the ions, remains an unresolved question.

Table 2. Seedling development of bitter lentil as affected by osmotic pressure of various salts and their combinations.

OP (atm)	Seedling length (mm) together with standard error					
	Treatments					
	H_2O	NaCl	Na_2SO_4	$MgSO_4$	Na_2SO_4 + $MgSO_4$	NaCl + $MgSO_4$
0.0	42.6 ± 2.5					
1		8.3 ± 1.6	5.3 ± 0.6	8.0 ± 1.7	38.3 ± 2.7	41.3 ± 3.1
2		1.7 ± 0.3	4.0 ± 1.7	7.0 ± 1.0	33.0 ± 3.0	29.3 ± 7.9
3		1.3 ± 0.3	2.3 ± 0.2	4.0 ± 1.0		
4		0.5 ± 0.0	0.8 ± 0.3	3.0 ± 1.0	21.0 ± 1.7	21.7 ± 2.1
5		0.2 ± 0.3	0.2 ± 0.3	2.0 ± 1.0		
6		0.2 ± 0.3	0	1.0 ± 0.5	13.6 ± 2.5	15.6 ± 1.5
7		0		0.5 ± 0.3		
8				0.5 ± 0.3	8.0 ± 2.6	12.3 ± 1.5
10					4.6 ± 1.1	9.0 ± 1.7
12					2.3 ± 0.8	4.0 ± 1.0
14					0.6 ± 0.8	2.7 ± 0.6
16					0.3 ± 0.8	1.0 ± 1.0

Synergism

The synergism between $MgSO_4$ and the other two salts on seedling development is also most conspicuous (Fig. 2 and Table 2), and the pattern of this synergistic interaction in both cases is similar. Unlike that observed in germination, however, the magnitude of synergism here is strikingly higher at the lower concentrations. Whether Mg was responsible for overcoming the inhibition of the other two salts remains again speculative. The explanation offered earlier concerning synergism between $MgSO_4$ and the other two salts on germination, though conjectural, might be applied to seedling development as well.

References

Bernstein, L. and **Hayward, H.E.** (1958) Physiology of salt tolerance, *Ann. Rev. Plant Physiol.* **9**: 25-46.

Carlson, J.R., Ditterline, R.E., Martin, J.M., Sands, O.C. and **Lund, R.E.** (1983) Alfalfa seed germination in antibiotic agar containing NaCl, *Crop Science* **23**: 882-884.

Clarkson, D.T. and **Hanson, J.B.** (1980) The mineral nutrition of higher plants, *Ann. Rev. Plant Physiol.* **31**: 239-298.

Eaton, F.M. (1942) Toxicity and accumulation of chloride and sulfate salts, *J. Agric. Res.* **64**: 357-399.

Flowers, T.J., Troke, P.E. and **Yeo, A.R.** (1977) The mechanisms of salt tolerance in halophytes, *Ann. Rev. Plant Physiol.* **28**: 89-121.

Ghosh, B.R. and **Dutta, A.** (1950) The constituents of *Securigera securidaca* Linn., *Indian Journal of Pharmacology.* **12**: 233-235.

Greenway, H. and **Munns, R.** (1980) Mechanisms of salt tolerance in nonhalophytes, *Ann. Rev. Plant Physiol.* **31**: 149-190.

Hafeez, K.A. and **Marshall, C.** (1981) Salt tolerance within populations of chewing fescue (*Festuca rubra* L.). *Commun. in Soil Sci. Plant Anal.* **12**: 1271-1281.

Harris, F.S. and **Pitman, D.N.** (1918) Soil factors affecting the toxicity of alkali, *J. Agric. Res.* **15**: 287-319.

Harris, F.S. and **Pitman, D.N.** (1919) Relative resistance of various crops to alkali, *U.S. Utah Agric. Exper. Sta. Bull.* No. 168.

Miller, T.R. and **Chapman, S.R.** (1978) Germination responses of three forage grasses to different concentrations of six salts, *J. Range Manage.* **31**: 123-124.

Pauling, L. and **Pauling, P.** (1975) *Chemistry: The Osmotic Pressure of Solutions.* Freeman and Company, San Francisco. U.S.A. 767 P.

Ryan, J., Myamato, S. and **Stroklein, J.L.** (1975) Salt and specific effects on germination of four grasses. *J. Range Manage.* **28**: 61-64.

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تـأثير بعض الأمـلاح منفرداً ومـزدوجاً عـلى إنبات ونمو بادرات نبات العدس المر

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درست تـأثيرات كلوريـد الصوديـوم، كـبريتـات الصـوديـوم وكـبريتات المغنيسيـوم بصورة منفصلة أو مـزدوجة عـلى إنبات بذور نبات العدس المر ونمـو بادراتـه. ولقد وُجـد أن الأملاح الثلاثة عرقلت إنبات البذور ونمو البادرات بدرجات متفاوتة، ولكن عملية نمو البادرات كانت أكـثر حساسيـة للأمـلاح من عمليـة الإنبـات ويبـدو أن تـأثـير كـبريتـات المغنيسيـوم عـلى العمليتـين كان بسبب ارتفـاع الضغط الازمـوزي للمحلول، بينما كان تأثير كل من كلوريد الصوديوم وكبريتات المغنيسيـوم ناتجاً عن السمية الأيونية لهذين الملحـين. وظهر أيضـاً أن كلا من أيـون الصوديـوم والكلوريـد كـان أكـثر سمّيـة من أيـون المغنيسيوم والكبريتات. وعند دمج كبريتات المغنيسيوم مع كل من كلوريد الصوديوم وكبريتات الصوديوم زال التأثـير السّمي للملحـين الأخيرين كليـاً، في كـل من عمليتي الإنبـات ونمـو البادرات.

والأهم من هذا هو أن تأثير كبريتات المغنيسيوم مع كل من الملحين الأخرين وعلى كلتا العمليتين كان متدائباً.