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EFFECT OF CALCIUM ON THE GROWTH AND ION UPTAKE IN NaCl-STRESSED PLANTS[1]

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ABSTRACT

Peanut and kidney bean gave similar responses to salt stress showing a more severe growth reduction, as compared to spinach, leaf beet, barley cultivars and wild barley. Calcium showed the protective effect on salt injury in all plants, the effect of which was more pronounced in spinach, leaf beet and both types of barley plant than that in bean and peanut.

High concentrations of NaCl decreased the content of K, Ca and Mg in both shoot and root of all plants. Elevated Ca in the nutrient solution, however, considerably alleviated the inhibition of K uptake due to NaCl. The result suggested that the maintenance of K/Na selectivity by Ca in plant might result in the enhancement of salt tolerance.

RINGKASAN

Kacang tanah dan buncis memberikan respon yang sama terhadap stres garam, yang memperlihatkan pertumbuhan yang lebih buruk dibandingkan dengan spinasi, bit, dan barley. Pemberian kalsium dapat mengurangi tingkat kerusakan akibat stres garam; pengaruh ini lebih nyata terlihat pada spinasi, bit dan barley.

Konsentrasi NaCl yang tinggi menurunkan kandungan K, Ca dan Mg pada pucuk dan akar seluruh tanaman. Peningkatan konsentrasi Ca pada larutan hara dapat mengurangi penghambatan serapan K oleh NaCl. Selektivitas K/Na yang tinggi yang dapat dipertahankan karena adanya Ca diduga berkaitan dengan peningkatan daya toleransi tanaman.

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Table 1. The botanical names and the origin of barley used as test plants in this investigation

Plants	Botanical names	Origin
Cultivar		
OUE 002	H. *vulgare* L.	Ethiopia
Akashinriki	H. *vulgare* L.	Japan
Wild-barley		
II-5	H. *maritimum* With.	Portugal
II-6	H. *marinum* Huds.	Egypt
II-8	H. *marinum* Huds	Lebanon
II-16	H. *murinum* L.	Japan
II-22	H. *murinum* L.	Iran

After having been grown for about 3-4 weeks, the plants were then harvested and separated into shoots and roots. The tissues were dried at 100° C, and dry matter weighed. The dried tissues were then ground, and alliqout of ground tissues were ashed at 500° C, and the ashes were dissolved with diluted hydrochloric acid. The concentration of Na, K, Ca, and Mg in plants were measured by atomic absorption spectrophotometry, and P by molybdovanadophosphate method.

RESULTS

Plants Growth

Figures 1 and 2 show the effect of Ca on growth of respectively shoot and root of various plants exposed to saline conditions. High concentrations of NaCl decreased the growth of all plants, the decrease of which was principally in the same extent both in the shoots (Fig. 1) and roots (Fig. 2). Peanuts and kidney bean (Figs. 1A and 2A) were the most sensitive plants to salts stress, which showed a more severe growth reduction. As compared to peanut and bean, spinach and leaf beet showed a better growth, especially under 2.0 mM Ca addition. Moreover, barley cultivars OUE 002 (Fig. 1A) and akashinriki (Fig. 1B) and wild plants (II-5 to II-22) remained alive in 250 mM NaCl, a half concentration of sea water, in the presence of 2.0 mM Ca. Both cultivars OUE 022 and akshinriki had a similar growth response to salinity, but the response was different in wild barley plants.

Under low Ca (0.1 mM) addition, the growth of plants severely decreased as the concentration of NaCl increased. Calcium showed a protective effect on the growth from the adverse effect of NaCl, as evidenced in spinach. However, Ca had a lesser effect in alleviating salt injury in peanut and bean. In wild barley, especially in II-5, Ca became more efective in reversing the inhibitory effect of NaCl on plant growth at high concentrations of NaCl (Figs. 1A and 2A).

INTRODUCTION

A number of studies have shown that salinization in the medium causes a reduction of growth and results in an imbalanced composition of mineral nutrients, especially low potassium content (Epstein, 1961; Lynch and Lauchli, 1984; Kawasaki and Moritsugu, 1978a and 1978b). Potassium nutrition is particularly important to salt-stressed non-halophytes (Winter and Preston, 1982), since considerable evidences indicate that metabolic damages result from cytoplasmic high Na:K ratios. Calcium is an important factor in the maintenance of membrane integrity and ion-transport regulation. Epstein (1961) pointed out that Ca is essential for K/Na selectivity and membrane integrity. The elevation of Ca concentration in the nutrient solution alleviated the adverse effects of NaCl in plants by inhibition of Na uptake (Greenway and Munns, 1980; Kawasaki and Moritsugu, 1978a and 1978b; Kawasaki *et al.*, 1983). Moreover, Ca recovered the inhibition of K absorption due to high concentrations of NaCl (Kawasaki and Moritsugu, 1978a and 1978b).

The purpose of this study was to determine the effect of Ca on the growth and ion uptake in NaCl-stressed plants, and to compare the salt tolerance of various plants with special reference to mineral nutrition.

MATERIAL AND METHODS

In this experiment, peanut (*Arachis hypogea* L.), kidney bean (*Phaseolus vulgaris* L.,CV. Masterpiece), spinach (*Spinacia oleracea* L.), leaf beet (*Beta vulgaris* L. var. flavescens), barley cultivars (*Hordeum vulgare* L.) and wild barley plants (see Table 1) were used as test plants. The seeds of plants were germinated for about 7 days, then seedlings were transplanted into 3.5 l pots containing nutrient solutions. The nutrient solution consists of KNO_3 4.0 mM, $NaNO_3$ 1.0 mM, NaH_2PO_4 1.0 mM, $MgSO_4$ 1.0 mM, Fe 1.0 ppm (Fe citrate). B 0.5 ppm (H_3BO_4), Mn 0.5 ppm ($MnCl_2$), Zn 0.05 ppm ($ZnSO_4$), Mo 0.01 ppm $(NH_4)_6Mo$ 0.01 ppm $(NH_4)_6Mo_7O_{28}$ and Cu 0.02 ppm ($CuSO_4$).

Except for barley, NaCl was used at 5 levels (2, 10, 30, 50, and 100 mM), and $CaCl_2$ at 2 levels (0.1 and 2.0 mM). In barley, the concentration of NaCl was extended up to 250 mM. At a high Ca supply (2.0 mM), 6 levels of NaCl, i.e. 2, 50, 100, 150, 200, and 250 mM were added. At 0.1 mM Ca, however, the concentrations of 200 and 250 mM NaCl were omitted, since these concentrations caused severe injuries to plants, as observed in preliminary experiments.

During the first 2-3 days of the experiment, all salts except micronutrient were given at 1/4 strength of the concentration of nutrient solution. Then, plants were supplied with full strength nutrient solution. The medium pH was adjusted to 5.0 for peanut, and 5.5 for other plants, Every other day. Fe was added, and nutrient solution was aerated continuously, and renewed once a week.

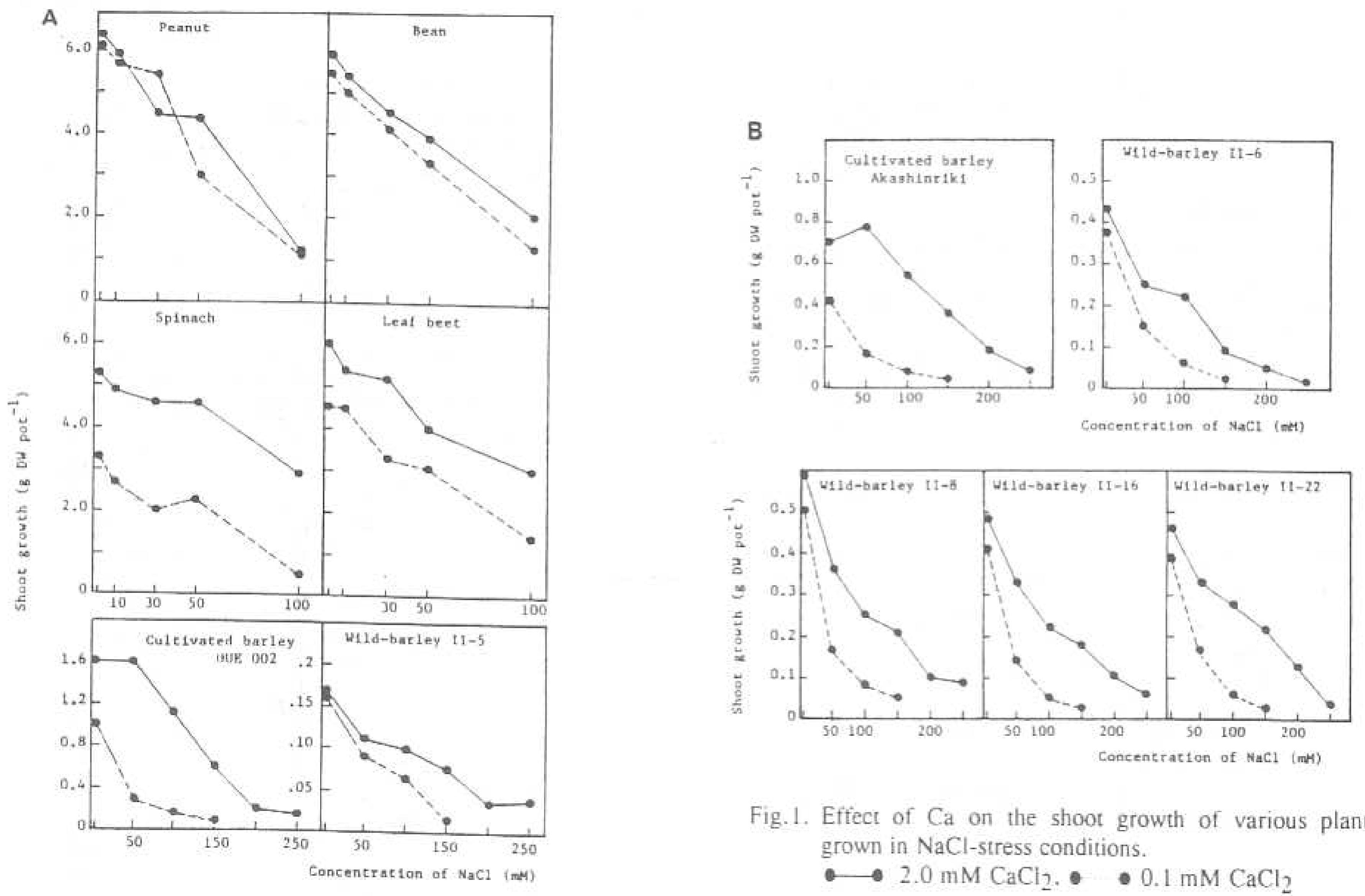

Fig.1. Effect of Ca on the shoot growth of various plants grown in NaCl-stress conditions.
●—● 2.0 mM $CaCl_2$. ● ● 0.1 mM $CaCl_2$

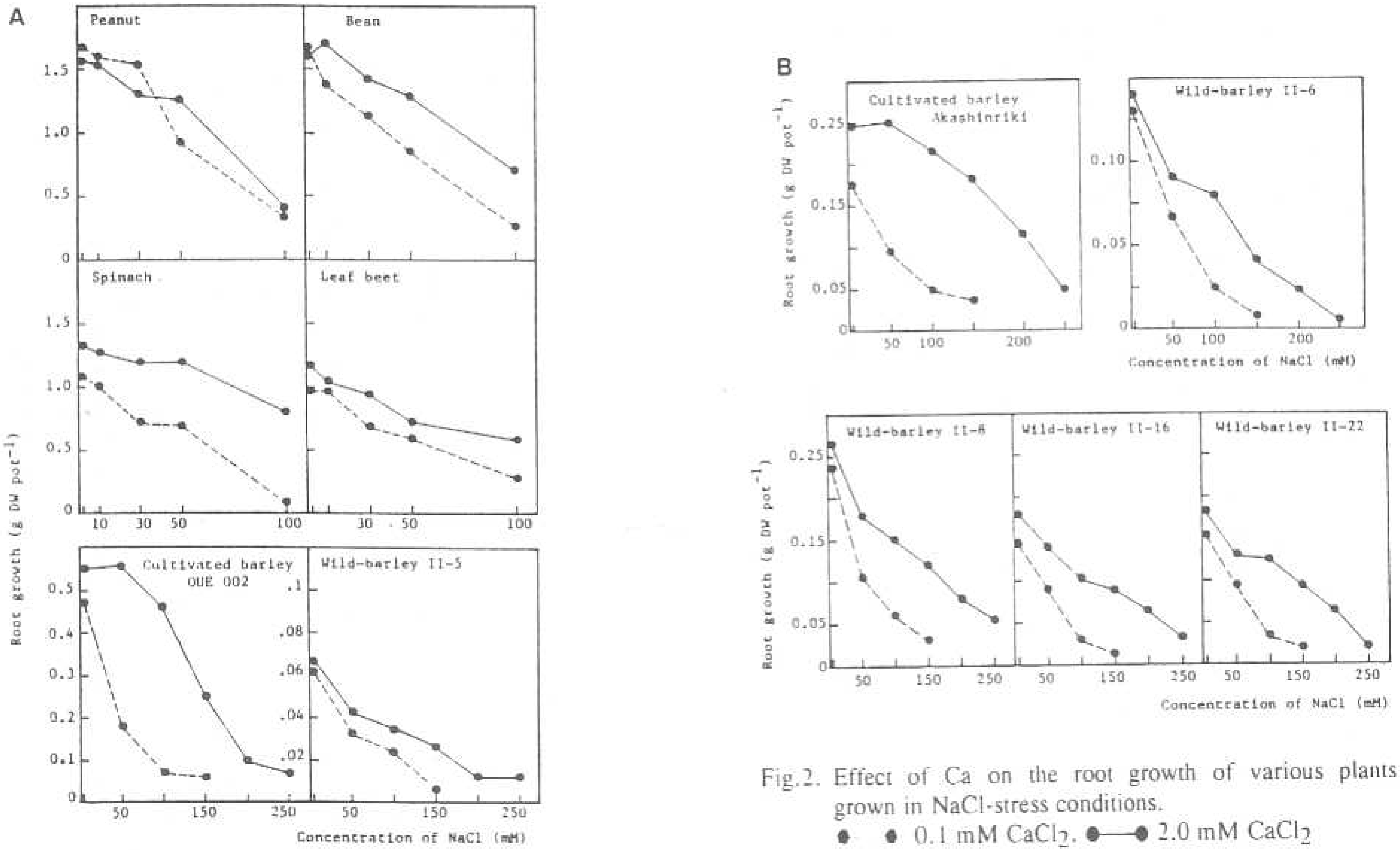

Fig.2. Effect of Ca on the root growth of various plants grown in NaCl-stress conditions.
● - - ● 0.1 mM $CaCl_2$. ●—● 2.0 mM $CaCl_2$

Na and K Absorption

Figures 3 and 4 show the effects of Ca on Na and K uptake in various plants grown in NaCl stress conditions. Peanut and bean absorbed much less Na in shoots (Fig. 3A) than in roots (Fig. 3B) when these plants were exposed to NaCl below 50 mM, thought at 100 mM NaCl, Na considerably accumulated in the shoots. In contrast to peanut and bean, other plants, i.e. spinach, leaf beet, barley cultivars and wild plants accumulated larger amounts of Na in the shoots (Figs. 3A and 4A) than in the roots (Figs. 3B and 4B).

Addition of 2.0 mM Ca decreased Na content in the shoots of all plants (Figs. 3A and 4A), though it had no clear effect in peanut. In the roots (Figs. 3B and 4B), however, Ca showed a less interference on Na uptake, as evidenced in peanut, bean and spinach. Moreover, this depressing effect of Ca on Na uptake in the roots of wild barley plants disappeared (Fig. 4B), whilst in cultivated ones (Figs. 3B and 4B), elevated Ca resulted in the increase of Na accumulation in the roots.

High concentrations of NaCl generally decreased K content in both shoot and root of various plants (Figs. 3 and 4). Elevated Ca in the nutrient solution, however, considerably alleviated the inhibition of K uptake due to NaCl, the evidence of which was observed in all plants, especially in spinach, leaf beet and barley cultivar Akashinriki.

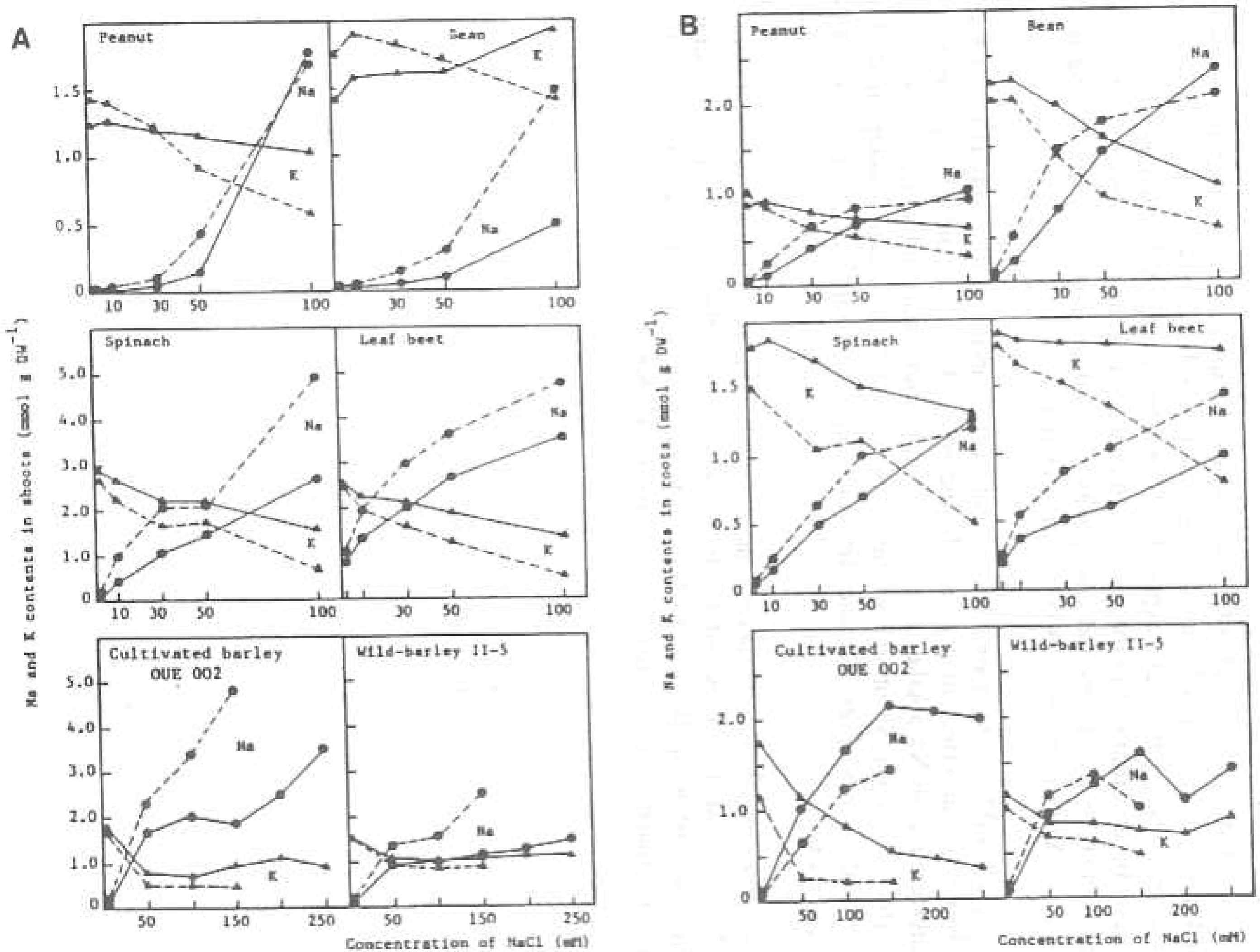

Fig.3. Effect of Ca on Na and K absorption in various plants grown in NaCl-stress conditions. (●) Na, (▲) K; ········ 0.1 mM $CaCl_2$, —— 2.0 mM $CaCl_2$.

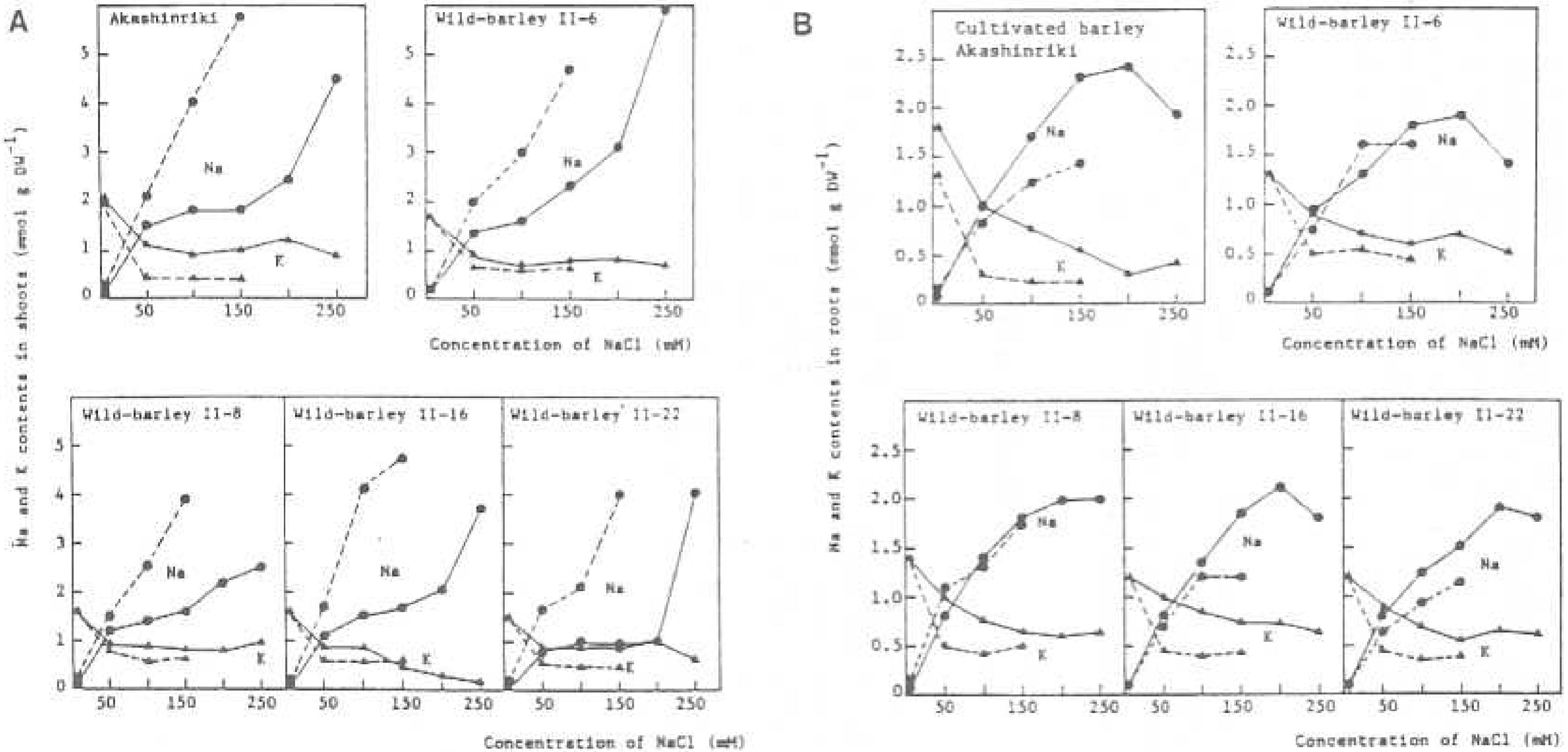

Fig.4. Effect of Ca on Na and K absorption in barley plants grown in NaCl-stress conditions. (●) Na. (▲) K; ······ 0.1 mM $CaCl_2$. —— 2.0 mM $CaCl_2$.

Ca and Mg Absorption

The effects of salinity and Ca on the absorption of Ca and Mg in plants are presented in Figs. 5 and 6. Generally, the contents of Ca and Mg in various plants declined as the concentrations of NaCl increased. Calcium showed an interference on Mg uptake in the shoots of peanut and bean (Fig. 5A), though in other plants this competitive effect of Ca appeared only at low concentrations of NaCl below 50 mM. In most of plants roots (Figs. 5B and 6B), however, Ca stimulated Mg absorption, especially in peanut, barley cultivars, and wild barley plants.

P Absorption

Except for barley akashinriki and II-8 (data not shown), there was no significant change in P absorption in the shoots of plants exposed to salinity at both levels of Ca. But this was not found for the roots of many plants (data not shown), in which P absorption increased as the concentration of NaCl was increased, especially at a low Ca supply.

DISCUSSION

The results showed the differences among plants tested, not only in growth but also in the pattern of ion uptake as their responses to salinity. For growth, peanut and bean gave similar responses to salt stress showing a more severe growth reduction, as compared to other plants. It could be observed that there were differences in growth patterns between barley cultivars and wild plants. In barley cultivars supplied 2.0 mM Ca, concentrations of NaCl below 50 mM had no effect on growth. This result was principally in agreement with that previously reported by Kawasaki and Moritsugu, (1978b).

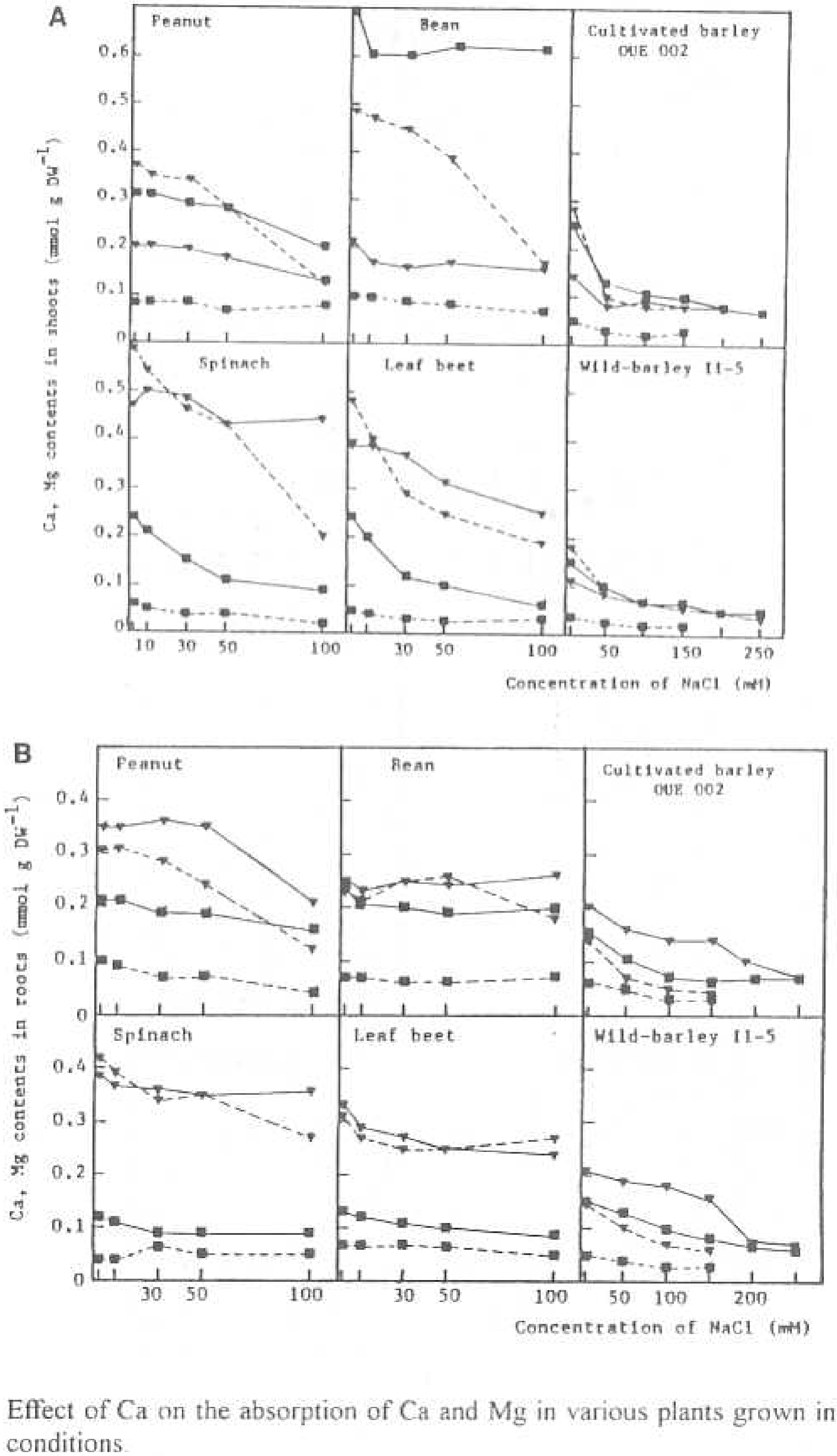

Fig.5. Effect of Ca on the absorption of Ca and Mg in various plants grown in NaCl-stress conditions.
(■) Ca, (▲) Mg; (⋯⋯) 0.1 mM $CaCl_2$, (——) 2.0 mM $CaCl_2$.

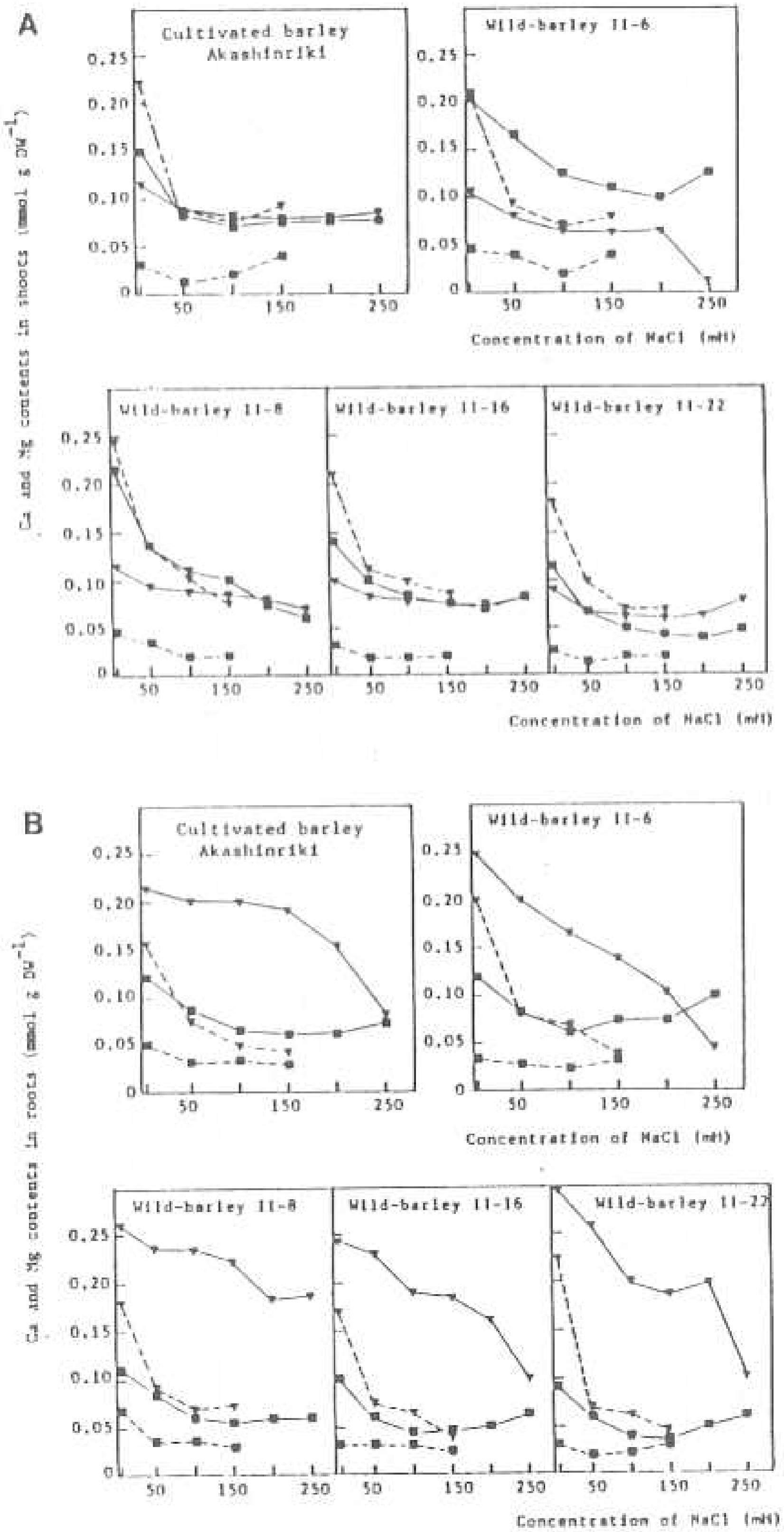

Fig.6. Effect of Ca on the absorption of Ca and Mg in barley plants grown in NaCl-stress conditions.
(■) Ca, (▲) Mg. (------) 0.1 mM $CaCl_2$, (——) 2.0 mM $CaCl_2$.

In the present study, wild barley plants have been chosen as experimental materials in order to compare their tolerance with those of cultivated plants (Table 1). Wild barley plants, however, failed to show more tolerance as compared to the cultivated ones, though barley II-5 could expressed a good tolerance at high levels of NaCl.

The protective effect of Ca on salt injury in plants was more pronounced in spinach, leaf beet and both types of barley plants, than that in bean and peanut. In bean and peanut, Ca had a less effect in diminishing the damage of growth due to high concentrations of NaCl. A similar result was also revealed by Kawasaki and Moritsugu (1978a) in bean. Among the plants used in the presents experiment, wild barley was the group of plants which showed an excellent example in expressing the alleviating effect of Ca on salt injury. This was evidenced by the fact that this protective effect of ca effectively increased as the concentration of NaCl increased.

The plants showed differences in the pattern of ion uptake, which may be expected to be associated with physiological differences in the mechanism of salt tolerance. It was evidence that spinach, leaf beet and barley responded to salt stress by accumulating higher Na in the shoots than in the roots. On the other hand, peanut and bean retained most Na in the roots. Kawasaki and Moritsugu (1978a and 1978b) revealed a similar results in bean and maize. Other workers suggested that generally in salt sensitive species, like bean (Jacoby, 1984) and maize (Yeo *et al.*, 1977), relatively less Na is found in shoots. Moreover, in *Chenophodiaceae*, like spinach and beet, osmotic adjusment in archieved mainly by accumulation of high levels of Na and Cl in the shoots, accompanied by synthesis of substantial amounts of compatible solutes such as glycinebetaine (Gorham *et al.*, 1985). Thus, the present result suggest that a high Na accumulation in the shoots of spinach, beet and barley is probably required for osmotic adjustment in the leaf cells of these plants. It is likely that the salt tolerance of spinach, beet and barley is mainly due to the efficient ion compartmentation in the leaf cells. On the other hand, peanut and bean appeared to respond to salinity mainly by Na exclusion from shoots, as also evidenced in bean (Jacoby, 1964), maize (Yeo *et al.*,1977) and rice (Yeo *et al.*, 1987). There is evidence that when adequate amounts of Ca was supplied to a saline medium, Na uptake was inhibited, whilst K uptake was restored to a certain level. The maintenance of K/Na selectivity by Ca in plants might result in the enhancement of salt tolerance, as it has been proposed by Kawasaki and Moritsugu (1978a and 1978b) and LaHaye and Epstein (1969). Moreover, Ca involvement in the maintenance of membrane integrity from adverse effect of NaCl might have led to the recovery of K uptake in plants.

The contents of Ca and Mg in plants were decreased by high concentration of NaCl. Kawasaki *et al.* (1983a) reported also the inhibiting effect of NaCl on Ca and Mg in barley plants. There was no clear effect of Ca on Mg uptake in plants grown under salinity. In one case, Ca mitigated the inhibition of Mg uptake due to NaCl as found in spinach and bean. In other case, Ca also stimulated the uptake of Mg in the roots of many plants. However, there has been no definitive information, as far known, on the interaction between Ca and Mg in plants.

There is growing evidence that high accumulation of P in the shoot of plants grown in saline enviroment often causes injury. The recent salinity-fertility studies showed a marked interactive effect salinity and nutrition inorganic P on plants (Nieman and Clark, 1976; Berstein *et al.*, 1974; Grattan and Mass, 1984). The decrease in plant growth appears to result from excessive uptake of inorganic P (Roberts *et al.*, 1984), with translocation to the leaves, leading to symptom of phosporus toxicity (Nieman and Clark, 1976). In the present investigation, however, the decrease in growth of plants exposed to saline conditions was not likely caused by excessive uptake of P, since no significant change in P content has been found in plant shoots.

CONCLUSION

1. At a low concentration of Ca (0.1 mM), plants growth was depressed with increasing concentration of NaCl. On the other hand, plant growth at 2.0 mM Ca suffered less damage. Peanut and kidney bean were the most sensitive plants to salt stress, which showed a more severe growth reduction, as compared to spinach, leaf beet and barley.

2. High concentration of NaCl decreased the content of K, Ca and Mg in both shoot and root of all plants.

3. Addition of 2.0 mM Ca depressed content in the shoot all plants and considerably alleviated the inhibition of K uptake due to NaCl.

4. Generally, there was no significant change in P absorption in the shoot of allp plants grown under saline condition at bith levels of Ca.

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