Ion accumulation in different organs of green bean genotypes grown under salt stress

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ABSTRACT

Salt-tolerant Gevas Sirik 57 (GS57) genotypes and salt-sensitive 4F-89 French variety, previously determined in our preliminary study, were used in the study. The genotype and the variety exposed to 100mM NaCl application developed different mechanisms to be protected against toxic effects of Na^+ ion. Salt-sensitive 4F-89 French variety let Na^+ accumulate in all organs. On the contrary, salt-tolerant GS57 did not avoid salt and acted selectively among ions; the majority of toxic ion Na^+ accumulated in old leaves and shoots and the plants did not transport them into young leaves. K^+ accumulation was high in organs in which Na^+ concentrations were low, and vice versa; Na^+ content was low in young and high in old leaves of GS57, but K^+ content was opposite. Ca^{2+} content in young leaves of GS57 and 4F-89 decreased; still its content was the highest of all examined ions found in young leaves. This indicated that beans can develop different mechanisms to accept and adapt high levels of salt. Storing toxic ion (Na^+) in old leaves and having a limited transmission of salt into young leaves serves as a protection from detrimental effects of salt.

Keywords: *Phaseolus vulgaris*; green bean genotypes; salt stress; plant parts; ion accumulation

Plants try to grow and develop by adapting themselves to unfavorable conditions in their environment and by developing some defense mechanisms against all kinds of biotic or abiotic stress factors. Levitt (1980), who provided important information about salt stress in plants and responses of plants against it, reported that handling the salt stress salt could be overcome by avoiding it and/or by developing a tolerance mechanism for it. Tal (1983) and Lauchli (1986) agreed with this concept. They proposed that the low permeability of plant root cell for salt kept salt away from plant shoot. Root cells of plants that can tolerate high levels of salt should have as high permeability as possible. Furthermore, plants can move salt from shoot by pumping Na^+ ion out of cell as a salt avoiding mechanism. Efflux of Na^+ ion from cytoplasm via Na^+-pump helps to maintain plant tolerable limits of Na^+ level (Yang et al. 1990). Another method is to decrease salt levels that are accumulated by fast growth, in other words to dilute salt in growth. Keeping salt ions away accumulated in plasma cell by isolating them in vacuoles is another mechanism that protects plants from harmful effects of salt (Tattini et al. 1994, Zhang and Blumward 2001).

The tolerance to salt and adaptation to it was also observed; plants can develop it by reducing the transmission of Na^+ ion from plant roots to shoot and leaves (Poljakoff-Mayber et al. 1987).

Na^+ and Cl^- ions are well distributed among organs and tissues of green parts in plants well tolerating salt. Storing Na and Cl^- ions mainly in old leaves and having a limited transmission of salt into young leaves are well-known characteristics of salt-tolerant plants. These plants contain more K^+ in young leaves than in old leaves; this balance is achieved by transporting the K^+ from old leaves through phloem into young leaves (Wolf et al. 1991).

In our previous study (Yasar 2006), 13 different green bean genotypes were used. 7 genotypes were selected from the Basin of Van Lake, the other 6 genotypes are local varieties grown in different regions of Anatolia. They were grown under 100mM NaCl and were classified according to fresh weight of green part of plants, degree of

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oxidative damage (MDA) in lipids and some ions (Na^+, K^+, Ca^{2+}) contents and ratios (K/Na, Ca/Na). At the end of the study, one salt tolerant (GS57) and one salt-sensitive variety (4F-89) were selected according to these growth parameters.

The objective was to determine the exact mechanism for salt-tolerance in GS57 and salt-sensitive 4F-89 by individual examination of the levels of ions in root, shoot, and different leaf levels of bean. Determining of the mechanism involved in ion balance regulation in bean could point out physiological targets for breeding program aimed at increasing the salt tolerance of this crop.

MATERIAL AND METHODS

Plant material. As the plant material, one salt-tolerant (Gevas Sirik 57) and one salt-sensitive (4F-89 French) local Turkish green bean varieties, whose salt tolerance characteristics were determined in our previous study (Yasar 2006), were used.

Plant growth and treatments. The green bean seeds were germinated in plastic pots (40 × 25 × 5 cm) filled with perlite in growth chamber with 16/8 h light/dark photoperiod, at 25 ± 2°C and 70% humidity. Seedlings were irrigated with Hoagland nutrient solution. Plants were grown in these pots until the emergency of the first two leaves. Afterwards, seedlings were transplanted to hydroponic culture. For hydroponic culture, plastic developing dishes (40 × 25 × 5 cm) filled with Hoagland nutrient solution were used. The nutrient solution was renewed once a week. Seedlings were grown under control conditions until the emergency of the fourth leaf; at the same time salt stress treatment was initiated. In the salt treatment, the first increment of salt containing 50mM NaCl was added and other increments of given concentrations were added daily until the salt concentration reached the final treatment level of 100mM NaCl. Treatments were replicated four times, each repetition having 15 plants, and arranged in a completely randomized design. 10 days after the salt treatment, six plants were harvested from each genotype at random. Whole plant weights were measured and these plants were separated into root, shoot and leaves for ion analysis.

Determination of ion content. For ion determination, fresh samples of root, shoot, first two old leaves and first two young leaves of one shoot were extracted in concentrated 0.1N nitric acid. Na^+, K^+ and Ca^{2+} contents were determined by flame photometry in the samples from green bean plants (Taleisnik and Grunberg 1994). Relative ion accumulation (Na^+, K^+ and Ca^{2+}) in whole plant (wp) was calculated as described by Taleisnik and Grunberg (1994).

RESULTS AND DISCUSSION

0mM and 100mM NaCl were applied to GS57 (salt-tolerant genotype) and 4F-89 (salt-sensitive genotype). Then, we tried to reveal ion distribution mechanisms of salt-sensitive and salt-tolerant beans by determination of fresh plant weight, accumulation of Na^+, K^+, Ca^{2+} ions in root, shoot, old and young leaves and a relative accumulation of these ions in the whole plant (Tables 1 and 2).

100mM NaCl applied salt-tolerant and salt-sensitive bean genotypes demonstrated very different development patterns. While 4F-89 had a high decrease in fresh plant weight, GS57 fresh weight

Table 1. Fresh whole plant weights (FW_{wp}) and relative ion accumulation in whole plants (wp) of salt-sensitive and salt-tolerant green bean varieties with 0 and 100mM NaCl treatment

Varieties	Salt treatment	FW_{wp} (g)	Na^+_{wp}	K^+_{wp}	Ca^{2+}_{wp}
			(µg/100 mg wp)		
4F-89	0	5.971 bA*	241.35 bB	410.60 aA	215.55 aA
	100	2.004 bB	553.00 aA	273.25 bB	211.32 aA
GS57	0	9.566 aA	281.60 aB	373.35 bA	178.73 bA
	100	6.868 aB	539.25 bA	308.50 aB	197.25 bA

*Different letter case represents different means of genotypes in the same dose level (comparison of genotypes)
*Different upper case letter represent different means of fresh weight and ions in the same genotypes and dose level (comparison of parameters)

Table 2. In roots, shoots and old and young leaves of salt-sensitive and salt-tolerant green bean genotypes Na^+, K^+ and Ca^{2+} ions accumulations (µg/mg fresh weight)

Ion	Salt treatment (mM)	Varieties	Fresh leaf	Old leaf	Shoot	Root
Na	0	4F-89	2.686 bBII*	3.692 bAII	1.298 aDII	1.978 bCII
		GS57	3.113 aBI	4.028 aAII	1.430 aDII	2.693 aCII
	100	4F-89	4.757 aBI	4.672 bBI	6.278 aAI	6.412 aAI
		GS57	3.170 bCI	6.645 aAI	6.320 aAI	5.434 bBI
K	0	4F-89	6.219 aAI	5.416 aBI	2.110 aDI	2.687 aCI
		GS57	5.066 bAI	4.868 bAI	2.442 aBII	2.558 aBI
	100	4F-89	3.745 bAII	3.472 aBII	1.934 bCI	1.778 bCII
		GS57	4.157 aAII	3.341 aBII	2.986 aCI	1.856 aDII
Ca	0	4F-89	4.303 aAI	3.703 aBI	0.235 aCII	0.381 aCI
		GS57	3.589 bAI	2.762 bBI	0.238 aDII	0.560 aCI
	100	4F-89	3.590 aAII	2.699 bBII	1.733 bCI	0.431 aDI
		GS57	2.540 bBII	2.922 aAI	1.939 aCI	0.449 aDI

*Different letter case represents different means of genotypes in the same dose level (comparison of genotypes)
*Different upper case letters represent different means of organ in the same genotypes and dose level (comparison of organs)
*Different numbers represent different means of dose level in the same genotypes and organs (comparison of doses)

decreased less (Table 1). Total K^+ and Na^+ ions accumulation and distribution among the organs indicate the level of tolerance of beans to salt. If a plant is not able to be selective in transporting ions taken from its root to growing ends or did not transport them at a certain rate, the rate of decrease in growth may be high. Similar patterns were reported by Wolf et al. (1991) with barley, and Perez et al. (1993) and Taleisnik and Grunberg (1994) with tomato.

Both salt-sensitive and salt-tolerant genotypes had a higher Na^+ accumulation in whole plant and organs than control when Na^+ accumulation was investigated in genotypes under salt stress. However, there was no increase in Na^+ deposition in the young leaves of GS57; the highest increase was observed in the roots and shoot of plants. But whereas Na^+ accumulation was low in young leaves, it was very high in old leaves and the body of salt-tolerant GS57. The highest Na^+ accumulation, both overall and in organs, was observed in 4F-89 among plants grown under stress (Table 2, Figure 1). Salim and Pitman (1983) with mung bean plants (*Vigna radiata* L.), Wolf et al. (1991) with barley, Soliman and Doss (1992), Perez-Alfocea et al. (1993) and Cuatero and Fernandez-Munoz (1999) with tomato, and Santa-Maria and Epstein (2001) with wheat, reported similar results and indicated that the distribution of Na^+ ions varies among organs and tissues of green parts of plants that tolerate salt well. They also noted that transportations of Na^+ ions into young leaves were limited because the plants keep them in old leaves, which is known as the most desirable characteristics of salt-tolerant plant. Moreover, it was reported that K^+ ion levels were higher in young leaves than in old leaves in these plants; this balance was achieved by transporting K^+ in old leaves to young leaves through phloem.

Similar to total plant K^+ concentrations, K^+ concentrations in all organs of all varieties, except plant shoot, decreased more compared with control. K^+ accumulation was high in organs in which Na^+ concentrations was low, and vice versa. While Na^+ content was low in young and high in old leaves of GS57, K^+ content was contrary. In 4F-89 Na^+ content was similar in old and young leaves and at the same time it was lower, compared to other organs; in roots and shoots it was relatively high. K^+ content in this variety was lower in all organs compared to GS57 (Table 2, Figure 1). These results indicate that there was a competition between Na^+ and K^+ regarding their uptake. The salt-tolerant genotype has a greater

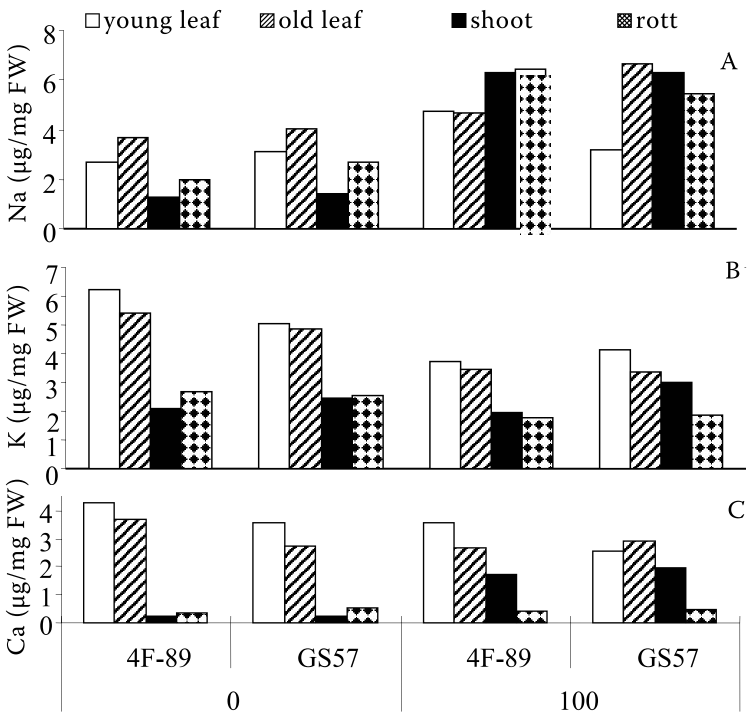

Figure 1. Comparison of different organs (young leaf, old leaf, shoot and root) × variety (4F-89 and GS57) × doses (0–100mM NaCl) of green bean for sodium (Na; Panel A), potassium (K; Panel B) and calcium (Ca; Panel C) accumulation; means followed by the same letter are not significantly different at 0.05 probability level, based on DMR

K^+ accumulation capacity. Similar results were reported with different tomato varieties (Caro et al. 1991, Cuartero et al. 1992, Perez-Alfocea et al. 1993, Taleisnik and Grunberg 1994, Zhang and Blumward 2001, Daşgan et al. 2003).

Ca^+ content in young leaves of GS57 and 4F-89 decreased. Ca^+ accumulation in young leaves of 4F-89 French was greater compared with its other organs and with young leaves of GS57. Ca^+ content was the highest in young leaves, followed by old leaves, shoot, and roots. Ca^+ content decreased only in old leaves of 4F-89, but it did not change the tolerance of the genotype. Ca^+ accumulation in the shoot of both genotypes did not vary (Table 2, Figure 1). Similar results were reported with different plants by Cramer et al. (1986), Lauchli (1990), Cuartero et al. (1992), Rengel (1992), Huang and Redmann (1995), Zhang and Blumward (2001), Daşgan et al. (2002).

It was observed that beans can develop different mechanism to accept and adapt to high levels of salt. GS57 kept Na ions mainly in old leaves by taking them from roots, and limited their transport into young leaves. A higher concentration of K^+ was observed in young leaves than in old leaves in this genotype.

REFERENCES

Caro M., Cruz V., Cuartero J., Estan M.T., Bolarin M.C. (1991): Salinity tolerance of normal-fruited and cherry tomato cultivars. Plant Soil, *136*: 249–255.

Cramer G.R., Lauchli A., Epstein E. (1986): Effects of NaCl and $CaCl_2$ on ion activities in complex nutrient solutions and root growth of cotton. Plant Physiol., *81*: 792–797.

Cuartero J., Fernandez-Munoz R. (1999): Tomato and salinity. Sci. Hort., *78*: 83–125.

Cuartero J., Yeo A.R., Flowers T.J. (1992): Selection of donors for salt-tolerance in tomato using physiological traits. New Phytol., *121*: 63–69.

Daşgan H.Y., Aktaş H., Abak K., Cakmak I. (2002): Determination of screening techniques to salinity in tomatoes and investigation of genotype responses. Plant Sci., *163*: 695–703.

Hung J., Redman R.E. (1995): Solute adjustment to salinity and calcium supply in cultivated and wild barley. J. Plant Nutr., *18*: 1371–1389.

Lauchli A. (1986): Responses and adaptation of crops to salinity. Acta Hort., *190*: 243–246.

Lauchli A. (1990): Calcium, salinity and plasma membrane. In: Leonard R.J., Hepler P.K. (eds.): Calcium in Plant Growth and Development. In: Proc. 10th An.

Revers. Symp. Plant Physiology, Am. Soc. Plant Physiol., Rockville: 26–35.
Levitt J. (1980): Responses of Plants to Environmental Stresses. Vol. II. 2nd ed. Academic Press, New York.
Perez-Alfocea F., Estan M.T., Caro M., Bolarin M.C. (1993): Response of tomato cultivars to salinity. Plant Soil, *150*: 203–211.
Poljakoff-Mayber A., Symon D.E., Jones G.P., Naidu B.P., Paleg N.G. (1987): Nitrogenous compatible solutes in native sought Australina plants. Aust. J. Plant Physiol., *14*: 341–350.
Rengel Z. (1992): The role of calcium in salt toxicity. Plant Soil, *149*: 229–233.
Salim M., Pitman M.G. (1983): Effects of salinity on ion uptake and growth of mung bean plants (*Vigna radiata* L.). Aust. J. Plant Physiol, *10*: 395–407.
Santa-Maria G.E., Epstein E. (2001): Potassium/sodium selectivity in wheat and the amphiploid cross wheat × *Lophopyrum elongatum*. Plant Sci., *160*: 532–534.
Soliman M.S., Dos M. (1992): Salinity and mineral nutrition effects of growth and accumulation of organic ions in two cultivated tomato varieties. J. Plant Nutr., *15*: 2789–2799.
Tal M. (1983): Selection for stress tolerance. In: Evans D.E., Sharp W.R., Ammirato P.V., Yamada Y. (eds.): Handbook of Plant Cell Culture. Vol. 1. Collier Macmillan Publ., London: 461–487.
Taleisnik E., Grunberg K. (1994): Ion balance in tomato cultivars differing in salt tolerance. I. Sodium and potassium accumulation and fluxes under moderate salinity. Physiol. Plant., *92*: 528–534.
Tattini M., Coradeschi M.A., Ponzio C., Traversi L. (1994): Responses of olive plants to salt stress. In: Abstr. XXIVth Int. Hort. Congr. Kyoto-Japan ISHS.
Wolf O., Munns R., Tonnet M., Jeschke W.D. (1991): The role of the stem in the partitioning of Na^+ and K^+ in salt-treated barley. J. Exp. Bot., *42*: 278–282.
Yang Y.W., Newton R.J., Miller F.R. (1990): Salinity tolerance in sorghum. I. Whole plant response to sodium chloride in *S. bicolor* and *S. halepense*. Crop Sci., *30*: 775–781.
Yasar F. (2006): Effects of salt stress on ion and lipid peroxidasion content in green beans genotypes. Asian J. Chem. (Submitted)
Zhang H.X., Blumward E. (2001): Transgenic salt-tolerant tomato plants accumulate salt in foliage but not in fruit. Nat. Biotechnol., *9*: 765–768.

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