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Changes in micronutrients, dry weight and plant growth of soybean (*Glycine max* L. Merrill) cultivars under salt stress

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This study was carried out to determine the effects of salt stress on the growth, dry weights and micronutrient contents of soybean cultivars grown in green house conditions. Twelve soybean cultivars (Omaha, A-3127, Mancon, Stresland, LN-89-3264, NE-3297, Ap-2292, Althow, Irigious, S-4520, Amsoy-71 and Cisne) were exposed to salinity treatments (150 mM NaCl and Control). Shoot, leaf and root dry weights of all cultivars at 45-day-old plants were determined. Micronutrient contents (Fe, Mn, Cu and Zn) of leaves, stems and roots were also analyzed. Salinity stress negatively affected soybean cultivars and the extent of effects varied depending on the salt tolerance of the cultivars. Generally, salinity reduced the plant growth and dry weights. Fe, Mn, Cu and Zn concentrations were higher in roots compared with those in leaves and shoots in salt applied samples. It was determined that, micronutrient contents showed some variation in different organs of soybean cultivars as a result of salt application to growing environment. Iron (Fe), manganese (Mn) and copper (Cu) content increased in the samples with salt applications except in some cultivars. On the other hand, when mean data of cultivars were considered, zinc (Zn) content was not significantly affected by salt stress.

Key words: Soybean, *Glycine max* (L), salt stress, dry weight, micronutrient accumulation.

INTRODUCTION

Soybean is a major food and oil crop in most countries where salinity problems exist or might develop. Large areas of formerly arable land are being removed from crop production every year due to increasing soil salinity. Use of saline irrigation water and application of fertilizer are the main factors responsible for increasing soil salinity (Epstein et al., 1980). Reducing the spread of salinization and increasing the salt tolerance of high yielding crops are important global issues. Soybean is moderately salt tolerant, and may be cultivated in a light moderate saline soil (Grieve et al., 2003).

Salinity reduces leaf number, leaf area, shoot dry weight and number of crowns, leading to low yields (Hamdy et al., 1993; Essa, 2002; Li et al., 2006; Sharifi et al., 2007). Moreover, salinity causes physiological and biochemical changes in plants. In the plants, these chan-

ges appear depending on the effects of ions and solutes in the root zone on water activity in the cell and physiological and biochemical functions of the cell (Greenway and Munns, 1980) reducing turgor, limiting photosynthesis (Schwarz and Gale, 1981; Walker et al., 1981) and increasing ion deficiency due to inadequate transport mechanisms (Hasegawa et al., 1986).

Most of the salt stress in nature is due to sodium chloride salts (Levitt, 1980). Salinity can damage the plant through its osmotic effect, which is equivalent to a decrease in water activity through specific toxic effects of ions and by disturbing the uptake of essential nutrients (Laüchli and Epstein, 1990; Marschner, 1995; Dorais et al., 2001). However, there are different ideas related to how salinity affects the micronutrient composition of plants. It was declared that the micronutrients are generally less affected by salt stress compared with macronutrients (El Fouly and Salama, 1999; Hu and Schmidhalter, 2001; Turhan and Eris, 2005). In addition, some researchers suggest that salinity increases the Zn and Mn concentrations in plant (Cornillon and Palloix,

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1997; Alpaslan et al., 1998; Villora et al. 2000). On the contrary, Sanchez-Raya and Delgado (1996) declared that salinity generally decreases Fe and Mn transport from seed to seedling in sunflower and transport of Fe to aerial part also markedly reduces.

Some nutritional disturbances are expected under saline condition, resulting in high ratios of Na/Ca and Na/K. In presence of excess NaCl in medium, Na and Cl are accumulated in plant organs, and these saline ions can affect other mineral elements uptake through competitive interactions or by affecting the ion selectivity of membranes, which causes nutrient deficiencies in plants (Bohra and Döffling, 1993). Under saline conditions, a non-uniform distribution of ions in the successive leaves within the shoots and between the leaf blade and sheath has been observed frequently (Boursier and Läuchli, 1989; Jeschke and Wolf, 1988; Munns et al., 1988; Yasar et al. 2006).

The objectives of the present article are to investigate the effect of salinity on the shoot and root length; dry root, dry shoot and dry leaf weight in soybean cultivars and to determine the micronutrient distribution in the successive roots, shoot and leaves under salinity stress.

MATERIAL AND METHODS

An experiment was conducted in pots filled with soil in the greenhouse of the Horticulture Department of Agriculture, Faculty of Yuzuncu Yil University, Van, Turkey during April-June, 2007. The experiment was carried out using a complete randomized design with control and sodium chloride (NaCl) (0 and 150 Mmol) treatments replicated three times. Twelve soybean (*Glycine max* L. Merrill) cultivars (Omaha, A-3127, Mancon, Stresland, LN-89-3264, NE-3297, Ap-2292, Althow, Irigious, S-4520, Amsoy-71 and Cisne) were used as experimental material.

Ten seeds of each cultivar were sown directly in plastic pots containing 4 kg of loam soil. Surface soil was collected from an agricultural field and passed through a 2-mm mesh screen. The texture of the soil based on sand clay silt, total organic matter 1.96%, total salt 0.035%, pH 7.30, total nitrogen 0.9%, available phosphorus 28 mg kg⁻¹ dry soil, exchangeable potassium 180 mg kg⁻¹ dry soil. All pots were fertilized with urea as a nitrogen fertilizer equivalent to 150 kg ha⁻¹ and triple-superphosphate (80 kg P₂O₅ ha⁻¹) were incorporated into the soil before seeding. The daily air temperature ranged from 30°C (maximum at day) to 10°C (minimum at night), with the daily average temperature being about 25°C. Relative humidity fluctuated between 30 and 85%; the average value was about 60%.

Salinity treatments: Non-salt-treated plants were kept as controls and salt-stressed plants were subjected to 150 mM NaCl 30 d after sowing. The salinity treatments were maintained until final harvest. The pots were randomly arranged in a green house and rearranged several times during the growth period. After sowing, soils were watered immediately and watering was carried out regularly on daily interval during experiment (45 days). Plants were irrigated until saturated, with the excess solution allowed to drain into collection pans.

Thinning was carried out 15 days after planting, leaving four plants from each pot; 30 days later, salt-treatment started. After 45 days, plants were lifted and samples were washed in deionized water to remove salts from the tissue surfaces. Plant and root height (cm) was measured. After this, leaves, roots and shoots were separated. Their dry weights were determined after drying for

48 h at 75 - 80°C in forced air oven.

For micronutrients determination, dry samples of roots, shoots and leaves were extracted in concentrated HNO₃ and HClO₄. Fe, Mn, Cu and Zn contents were determined by atomic absorption spectrometry (AAS) according to Kacar (1984).

Data were analyzed by an analysis of variance using SAS (1985) software to test the significance of the main effects. Means were compared using LSD multiple range tests. Terms were considered as significant at the level of $P < 0.05$.

RESULTS AND DISCUSSION

The effects of salinity treatment on the plant heights, root length, dry root weight, dry shoot weight and dry leaf weight of twelve soybean cultivars are shown in Table 1. While root dry weight and shoot dry weight were affected by salinity, the difference between cultivars was not significant ($P > 0.05$). However, the difference of plant height, root length and leaf dry weight in both salt treatment and cultivars was significant ($P < 0.01$). The results showed that salt stress caused significant reductions in all growth variables including dry weights for all soybean cultivars except Mancon.

Plant heights recorded 45 days after planting were significantly affected by salt treatment (Table 1). Plants of the Ne-3297 cultivar were taller than that of the other cultivars in control group. Salinity decreased the plant height of soybean cultivars, but it did not affect that of Mancon and Irigious. Plant height of Althow was more affected (25% reduction compared to controls) than of the other cultivars. A comparison of the responses of the different cultivars indicated that root length was reduced significantly by salt stress except Mancon. Althow was more affected (19% reduction compared to controls) than the other cultivars in point of root length. Root dry weight was reduced by salinity in all cultivars except for Mancon. Shoot dry weight was reduced by salinity in all cultivars, but observed decrease was less in Mancon and Irigious cultivars. This shows that root was more affected than shoot by the salt treatment. For example, in the present experiment, root dry weight was more affected than shoot dry weight by salt stress. Leaf dry weight decreased significantly by salt treatment in all cultivars except Mancon. It seems that salinity affected (25% reduction compared to controls) the leaf dry weight of Althow more significantly than that of other cultivars.

Salt treatment affects differently early growth stages of plants. Salinity has both osmotic and specific ion effects on plant growth (Dionisio-Sese and Tobita, 2000). In present study, salt stress caused a significant decrease in plant height, dry weights of root, shoot and leaf of cultivars (Table 1). Reduction in plant growth as a result of salt stress has also been reported in several other plant species (Ashraf and McNeilly, 1990; Mishra et al., 1991; Ashraf and O'leary, 1997; Turkmen et al., 2008). The uptake of some mineral nutrients dissolved in water is also restricted in plants. Thus, growth and development of plants are inhibited due to occurring defect in meta-

Table 1. Effects of NaCl treatment on leaf, shoot and root dry weights, plant height and root length in soybean cultivars.

Variett	Plant height ^a		Root length ^a		Root dry ^b weight		Shoot dry weight ^b		Leaf dry weight ^a	
	0	150	0	150	0	150	0	150	0	150
Omaha	22.0 de	18.7 f	19.7 bc	19.0 ac	0.29	0.26	0.29	0.27	0.62 ef	0.52 e
A-3127	22.5 de	18.8 f	17.8 c	16.3 c	0.27	0.25	0.29	0.27	0.62 f	0.57 ce
Mancon	28.3 a	29.0 a	20.0 bc	20.3 ab	0.27	0.28	0.30	0.29	0.70 bd	0.75 a
Stresland	23.3 ce	21.7 df	19.0 bc	17.7 bc	0.29	0.26	0.30	0.27	0.65 cf	0.61 cd
LN-89 3264	21.7 de	18.3 f	18.8 bc	16.7 bc	0.30	0.24	0.28	0.24	0.58 f	0.52 e
Ne-3297	30.3 a	26.5 ac	18.0 c	17.8 bc	0.29	0.26	0.30	0.26	0.70 bd	0.55 ce
Ap-2292	29.7 a	27.7 ab	19.7 bc	17.0 bc	0.30	0.24	0.29	0.27	0.63 df	0.60 cd
Althow	28.2 a	21.3 ef	21.3 ac	17.3 b	0.30	0.23	0.30	0.24	0.71 bc	0.54 de
Irigious	23.8 bd	24.3 be	23.7 a	22.3 a	0.30	0.24	0.28	0.27	0.73 b	0.68 b
S-4520	27.8 ab	25.7 ad	22.0 ab	21.7 a	0.29	0.26	0.30	0.28	0.69 be	0.62 bc
Amsoy 71	27.0 ac	23.3 ac	20.3 ac	20.0 ac	0.29	0.26	0.31	0.29	0.91 a	0.75 a
Cisne	19.3 e	17.7 e	18.0 c	16.2 c	0.26	0.24	0.28	0.26	0.61 f	0.50 e
Mean	25.3	22.8	19.8	18.5	0.29	0.25	0.29	0.27	0.68	0.60
LSD	2.69		2.33		Ns		Ns		0.038	

^aMean values indicated by the same letter are not significant different ($p < 0.05$).

^bNot significant.

bolism. Some investigators thought that because of ion accumulation by changing membrane permeability, metabolism was negatively influenced (Cramer et al., 1985; Grieve and Fujiyama, 1987). Most crop plants suffer after exposure to saline conditions and showed decline in growth. The deleterious effect of salinity was suggested as a result of water stress, ion toxicities, ion imbalance, or combination of all these factors (Kurt et al., 1986).

The findings related to micronutrient contents of roots, shoots and leaves of plants are shown in Table 2. As shown from the table, differences in amounts of Fe, Mn, Cu and Zn are significant among all cultivars. Significant differences were determined among varieties in terms of Fe, Mn, Cu and Zn contents (Table 2). Fe, Mn, Cu and Zn concentrations were higher in roots compared with shoots and leaves in the salinized samples.

When compared to control plants, salt treatment caused significant increases in Fe content in roots (except for Stresland and Ne-3297), shoots (except for LN-89-3264, Althow, Irigious and S-4520) and leaves (except for Omaha, LN-89-3264 and Ne-3297) of all varieties. Similar result was reported for tomato cultivars, where Fe concentration decreased in some cultivars and increased in the others under salt stress (Martinez, 1987). However, S-Raya and Delgado (1996) suggested that Fe transport decreased from seed to seedling under salt stress in sunflower. On the other hand, Lazof and Bernstein (1999) have determined that salinity had no effect on Fe content of lettuce leaf.

In the root part of plants, Mn content decreased for Omaha, Ne-3297 and Althow in salt stress treatment; however, its concentration increased in the other cultivars. Mn content decreased with salt stress in shoot of plants in the Omaha, Mancon, Ne-3297 and Irigious.

However, it increased in the others cultivars shoots. Mn content of leaves decreased in the LN-89-3264, Ne-3297 and Irigious, but increased in the other cultivars. Wang and Han (2007) reported that salinity significantly increases the uptake and concentration of Mn in the shoots and leaves of alfalfa plants. On the other hand, it was found that salinity had no effect on Mn content of root and aerial part of strawberry (Turhan and Eris, 2005).

Copper contents of plant organs were different under salt stress. While it decreased in roots of A-3127, LN-89-3264, Ne-3297, Althow and Amsoy-71, it increased in the roots of the other cultivar. Salt application decreased the Cu concentration in shoots of Omaha, Althow, Irigious and S-4520, while there was increase in the other cultivars as a result of the treatment. Cu decreased in leaves of A-3127, Mancon, Stresland, LN-89-3264, AP-2292 and Amsoy-71, but increased in the other cultivars. Wang and Han (2007) determined that salinity reduced the uptake and concentration of Cu in alfalfa plants but significantly increased Zn content in the roots, shoots and leaves. On the other hand, Alpaslan et al. (1998) and Martinez et al. (1997) observed that salinity increased Mn content in rice, wheat and tomato plants.

Zinc contents of plant organs were different under salt stress; while it decreased in roots of A-3127, LN-89-3264 and Althow, it increased in the other cultivars. It also decreased in shoots of Omaha, Mancon, Ne-3297 and Irigious, but increased in the other cultivars. Zn decreased in leaves of Omaha, A-3127, Stresland, LN-89-3264, Ne-3297, Ap-2292 and Irigious, but increased in the other cultivars. Previously, conflicting results have been obtained in other plants in terms of Zn content under salt stress conditions. In most cases, salinity in-

Table 2. Micronutrient accumulations (ppm) under salt treatment and non salt treatment In roots, shoots and leaves of soybean varieties.

M.N	P. O	T	Omaha	A-3127	Mancon	Stresland	LN-89-3264	Ne-3297	Ap-2292	Althow	Irigious	S-4520	Amsoy 71	Cisne	T.M	P.O.M
Fe	Root	0	81.0cd	74.8 de	59.5 fg	115.3 a	85.8 c	87.3 c	79.6 cd	74.3 de	97.3 b	54.5 g	57.3 g	67.1ef	77.8	
		150	108.7 a	99.3 bc	82.9 ef	92.7 cd	92.4 cd	78.8 f	88.9 de	98.3 bc	103.2 ab	61.8 h	70.7 g	94.3 cd	83.6	83.6 A
	Shoot	0	48.1 e	48.9 e	63.2 cd	69.5 c	88.0 a	78.6 b	57.3 d	64.4 cd	58.6 d	44.3 e	63.5 cd	59.0 d	61.9	
		150	48.2 hi	68.3 ef	77.3 bd	86.3 a	74.4 ce	84.9 ab	60.8 fg	41.6 i	54.2 gh	40.5 i	80.1 ac	71.6 de	65.7	63.8 C
	Leaf	0	77.6 ab	70.7 bc	80.5 a	67.4 cd	79.4 a	61.4 df	64.9 ce	70.9 bc	54.7 f	53.7 f	61.4 df	57.2 ef	66.7	
		150	59.9 e	86.1 ac	80.8 bd	87.4 ab	78.9 cd	51.4 f	76.4 d	92.5 a	65.1 e	60.3 e	75.4 d	62.5 e	73.1	69.8 B
Mn	Root	0	243.3 a	165.9 ef	157.0 f	199.5 b	182.6 cd	230.9 a	140.1 g	173.0 de	195.3 bc	196.4 bc	196.6 bc	163.4 ef	187.1	
		150	216.3 ce	237.8 ab	209.7 df	237.3 ab	224.0 bd	197.0 f	231.5 b	137.6 g	247.3 a	234.5 ab	229.4 bc	208.9 ef	217.6	202.3 A
	Shoot	0	60.6 cd	39.5 e	83.0 ab	48.7de	67.9 c	89.8 a	65.8 c	33.6 e	47.5 de	34.3 e	74.6 bc	40.9 e	57.2	
		150	35.9 c	77.3 b	44.6 c	48.7 c	88.3 ab	84.4 ab	77.1 b	34.4 c	45.2 c	39.5 c	94.2 a	44.0 c	59.5	58.3 C
	Leaf	0	106.3 ab	102.5 b	97.2 bd	85.4 d	86.9 cd	82.1 d	101.3 bc	104.7 b	104.4 b	112.4 ab	85.4 d	120.1 a	99.1	
		150	117.7 bc	127.2 ab	110.3 c	92.5 de	84.2 e	78.9 e	102.8 cd	133.0 a	86.9 e	135.8 a	116.6 bc	138.1 a	110.3	104.7 B
Cu	Root	0	7.33 c	8.49 a	3.61 f	7.64 bc	5.34 e	6.84 cd	8.32 ab	6.96 cd	4.78 e	8.59 a	7.12 cd	6.39 d	6.78	
		150	7.53 cd	7.05 de	4.91 g	9.54 b	4.98 g	7.99 c	11.2 a	5.87 f	6.39 ef	10.65 a	7.83 cd	7.56 cd	7.62	7.20 A
	Shoot	0	5.98 b	5.05 cd	7.10 a	5.28 bc	3.99 e	6.75 a	4.41 de	4.27 de	5.07 cd	4.38 de	7.30 a	3.16 f	5.19	
		150	4.21 d	6.29 b	6.71 b	6.28 b	4.59 d	6.61 b	7.04 b	5.50 c	4.48 d	4.15 d	8.93 a	3.83 d	5.75	5.47 B
	Leaf	0	3.60 g	4.40 f	5.17 df	5.50 be	4.68 ef	4.78 ef	6.29 ab	5.81 ad	5.36 ce	6.16 ac	5.44 ce	6.54 a	5.31	
		150	5.27 bc	4.14 d	4.12 d	4.17 d	4.64 cd	5.89 b	4.86 cd	7.95 a	5.72 b	7.74 a	5.16 bc	7.30 a	5.58	5.44 B
Zn	Root	0	227.3 a	238.7 a	107.0 f	202.2 bc	205.3 b	225.4 a	132.7 e	189.0 c	142.4 e	127.7 e	161.2 d	167.1 d	177.2	
		150	268.7 a	227.3 b	133.0 ef	228.9 b	177.3 c	184.9 c	184.9 c	142.2 de	155.3 d	135.1 ef	121.1 f	218.1 b	181.4	179.3 A
	Shoot	0	100.7 a	41.5 f	48.59 ef	80.8 bc	96.8 a	91.5 ab	78.6 bc	48.7 ef	75.3 cd	60.9 de	61.7 de	79.8 bc	72.1	
		150	53.4 ce	45.7 ef	61.54 cd	108.1 a	112.5 a	101.1 a	82.5 b	46.9 de	66.8 c	51.0 de	85.6 b	84.8 b	74.9	73.5 C
	Leaf	0	123.0 a	90.1 d	119.6 ab	112.6 ac	94.1 d	88.6 d	111.5 ac	89.7 d	90.5 d	104.7 bd	93.1 d	97.9 cd	101.3	
		150	107.2 bc	78.4 ef	139.1 a	95.3 cd	84.6 df	72.5 f	92.3 de	109.4 bc	85.3 df	112.2 b	118.2 b	122.4 b	101.4	101.3 B

Mean values indicated by the same letter are not significant different ($p < 0.05$).

Abbreviations: M.N, micro nutrient; P.O, plant organs; T, treatment; T.M, treatment mean; P.M.O, plant organs mean.

creases the content of Zn in plant tissue; for example in pepper (Cornillon and Palloix, 1997), wheat and rice (Alpaslan et al., 1998), Zucchini (Villora et al., 2000), strawberry (Turhan and Eris, 2005) and alfalfa (Wang and Han, 2007).

Significant differences were determined among

the soybean cultivars for plant growth and micronutrient contents of plant tissues under the salt stress. It can be concluded that salt stress negatively affected all yield components studied in soybean cultivars except for Mancon. Some soybean varieties maintained higher growth under

saline conditions. The effect of salinity on the micronutrient composition of plant tissues was differently affected depending on the cultivar. Salt stress caused ion imbalance in the soybean cultivars. The results indicate changes in micronutrient contents and distribution in the plant tissues

as a result of salt stress.

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