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EFFECT OF CULTIVARS ON UPTAKE AND TRANSLOCATION OF SODIUM AND CHLORIDE IN OLIVE (*Olea europaea* L.) PLANT*

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ABSTRACT

In this study the relative effects of cultivars on uptake and translocation of Na^+ and Cl^- in olive (*Olea europaea*) were studied. For this purpose, a control and 3 levels of NaCl (2560, 5120 and 7680 mg L^{-1} NaCl) were applied to 8 olive cultivars grown in sand culture supplied with half strength Hoagland's solution. Experiment was carried out 95 days then plants were harvested and separated into organs. Thin root, thick root, shoot and leaf were analyzed for Na^+ and Cl^- content. Dry matter (DM) of root, shoot and leaf were determined. Shoot elongation (SE) was also measured. Important differences were found in the average SE and DM of cultivars. These parameters affected inversely each other and salinity effect was related to SE rather than DM accumulation. Most of cultivars were similar in uptake and translocation of Na^+ and Cl^-. In general, concentration of both elements in thin root was greatest. However, there were quantitative differences in initial Na^+ and Cl^- uptake levels of cultivars. The difference was greater for Na^+ and, Frontoio & Picholine cultivars accumulated at least 4-5 times higher Na^+ in thin roots than other cultivars. However, ion translocation process was not related to initial uptake level in thin roots. The findings suggested that olive cultivars differed in uptake and translocation of Na^+ and Cl^-. Probably, salt exclusion mechanism is operative within the root system especially in thin roots. This parameter might be used as a clue in order to understand salinity tolerance mechanism of *Olea europaea* cultivars.

Key Words: *Olea europaea* L., salinity, cultivar, shoot elongation, dry matter.

Zeytin (*Olea europaea)* Bitkisinde Sodyum ve Klorun Alınması ve Taşınması Üzerine Çeşitlerin Etkisi

ÖZET

Bu çalışmada zeytin (*Olea europaea)* çeşitlerinin Na^+ ve Cl^-'ün alınması ve taşınması üzerine olan etkileri araştırılmıştır. Bu amaçla, kontrol ve 3 farklı NaCl dozu (2560, 5120 ve 7680 mg L^{-1}) kum kültüründe yarı yarıya seyreltilmiş Hoagland çözeltisi verilerek yetiştirilen 8 farklı zeytin çeşidine uygulanmıştır. Deneme 95 gün boyunca sürdürülmüş ve daha sonra bitkiler hasat edilerek organlarına ayrılmıştır. Bitki dokularında Na^+ ve Cl^- analizleri yapılarak kök, gövde ve yapraklarda kuru madde miktarı belirlenmiştir. Ayrıca, sürgün boyu ölçülmüştür. Çeşitler kuru madde miktarı ve bitki boyu yönünden farklılık göstermişlerdir. Bu parametreler birbirini olumsuz yönde etkilemiş ve tuz etkisi kuru madde miktarından çok sürgün boyu ile ilişkili bulunmuştur. Çeşitlerin büyük bir çoğunluğu Na^+ ve Cl^- elementlerinin ilk alımı ve taşınması yönünden benzer bulunmuştur. Na^+ ve Cl^-, uygulama dozlarıyla orantılı olarak kök ortamından alınmış ve toprak üstü organlara taşınmıştır. Genel olarak, her iki elementin miktarı ince köklerde en fazla bulunmuştur. Aynı zamanda, çeşitlerin Na^+ ve Cl^- elementlerini ilk alım düzeyleri arasında da farklılıklar belirlenmiştir. Bu farklılık Na^+ elementinde daha fazla olmuş ve Frontoio & Picholine çeşitleri ince köklerinde diğer çeşitlere göre en az 4-5 kat daha fazla Na^+ depolamıştır. Buna karşın elementlerin taşınan miktarları ince köklerdeki ilk alım dereceleri ile ilgili bulunmamıştır. Sonuçlar, çeşitlerin Na^+ ve Cl^- elementlerinin alınması ve taşınmasına olan tepkilerinin farklı olduğunu ortaya koymuştur. Olasılıkla, tuzun alınmasını denetleyen mekanizma kök içerisinde, özellikle de ince köklerde etkin olarak çalışmaktadır. Bu parametrenin zeytin çeşitlerinin tuza dayanıklılık mekanizmalarının anlaşılmasında bir ipucu olarak kullanılabileceği düşünülmektedir.

Anahtar Sözcükler: *Olea europaea*, tuzluluk, çeşit, bitki boyu, kuru madde

INTRODUCTION

NaCl salinity is a widespread problem in arid and semiarid regions, seriously reducing plant productivity. Genotypes may differ appreciably in their protoplasmic salt resistance and ability to absorb certain ions such as Na^+ and Cl^-. The second characteristic may be used advantageously to limit the absorption of harmful ions by sensitive plants.

Olive is estimated to be moderately tolerant to salt (Hartman et al., 1966; Maas, 1986) and is generally cultivated in areas where water is the main limiting factor for agricultural production (Tattini et al., 1994). Cultivar specify, however, is extremely variable (El Gassar et al., 1979; Tattini et al., 1992, Tattini et al. 1997) and genotypic responses of olive to NaCl stress has not been extensively investigated (Tattini et al., 1994). For some plants, especially woody perennials (such as citrus and grapevines), Na^+ is retained in the woody roots and stems, and it is the Cl^- that accumulates in the shoot and is most damaging to the plant (often by inhibiting photosynthesis) (Flowers, 1988). However, for many plants (such as graminaceaus crops), Na^+ is the primary cause of ion-specific damage (Tester and Davenport, 2003). Olive productivity is reduced only by 10% when the

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electrical conductivity (EC) of soil solution is 4-6 dS m^{-1} and this value can be as high as 6-8 dS m^{-1} in soils of high calcium status. Olive can, however, tolerate even higher EC values when NaCl represents a small part of the soluble salts (Therios and Misopolinos, 1988). In recent years, increasing interest has been focused on the potential use of saline water for crop production and its efficient use as irrigation water.

The objectives of this study were to evaluate the differential response of 8 olive cultivars to NaCl-induced salinity, especially with respect to uptake and translocation of Na$^+$ and Cl$^-$ and growth parameters. The practical implication is to provide data enabling the expansion of olive cultivation in areas with saline water.

MATERIAL AND METHODS

Plant Material

Eight olive cultivars, namely Arbequina, Barnea, Nabali, Leccino, Souri, Phicoline, Maalot and Frontoio were used as test plants in the experiment. At the initiation of the experiment, homogeneous seedlings which were taken from commercial nursery cut at about 15 cm lenght and planted in 3 liter containers with coarse sand of 0.6-0.8 mm particle size.

Salt Treatments

The experiment was set up according to a completely randomized block design with 6 replicates and 1 plant per pot making a total of 6 plants per replicates. Seedlings were grown for 2 months by using half-strength Hoagland's solution (Hoagland and Arnon, 1950) until they reached about 30 cm. After 2 months, control and 3 different NaCl treatments (2560, 5120 and 7680 mg L^{-1} NaCl which are equal to 4 dS m-^{1}, 8 dS m^{-1} and 12 dS m-1) and half concentrated Hoagland's solution were applied together to the buckets twice a day. The conductivity of irrigation water was nearly 1 dS m^{-1} (640 mg L^{-1} NaCl).

Chemical and Physiological Analyses

At the beginning of the experiment homogenous seedlings were chosen. In addition, SE was expressed as percent, in relation to initial length, in order to eliminate differences in the initial size and vigor of the different cultivars. Plant heights were measured weekly. Fifty five days after salt was applied, significant leaf drop was seen in cv. Arbequina applied 5120 and 7680 mg L^{-1} NaCl and this cultivar was removed for mineral analyses. Salinization continued without symptoms of damage to other cultivars for a total of 95 days then plants were harvested and separated into organs (thin root, thick root, shoot and leaf). For analyses, plant samples were placed in paper bags without wash, dried in a forced-air oven at 70^{o}C for 72 hours. The samples were then ground in a stainless steel Wiley mill. The ground samples were wet digested in a mixture of nitric acid:perchloric acid (HNO_3:$HClO_4$) (4:1) and then Na$^+$ content in the digest was determined by using flame photometry (Jenway PFP7). Cl$^-$ contents of the samples were determined by chloride meter (Jenway PCLM 3). For this purpose, 0.1 g ground sample were put into glass tube, 10 ml distilled water were added and agitated for two hours. Extracts were kept into fridge for 12 hours then 0.5 ml extract were put into beaker containing buffer solution and stirred. The results were determined as digitally (Kacar, 1972).

Statistical Analyses

Analysis of variance procedures were performed for obtained data according to Littel and Hills (1978). Mean separation was performed with least significant difference (LSD) at P<0.05.

RESULTS AND DISCUSSION

Shoot elongation (SE) and dry matter (DM) accumulation are two important expressions of growth. Salinity affected on average SE of the cultivars in different extent (Figure 1).

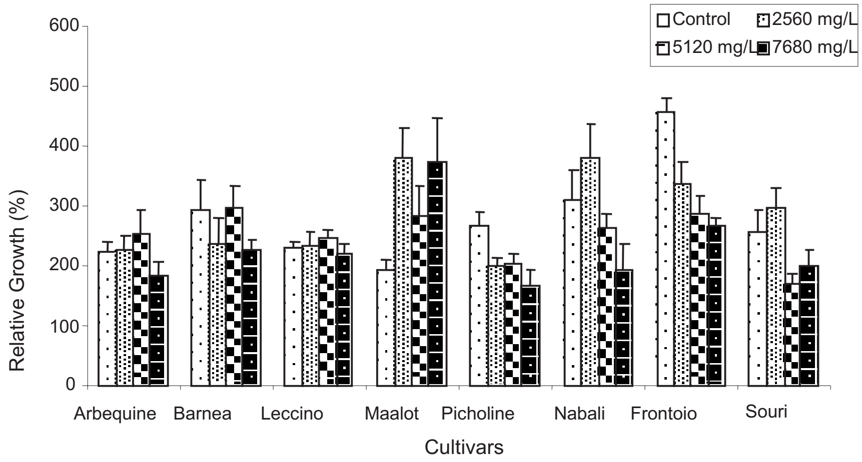

Figure 1. Effect of cultivar on shoot elongation (%) of olive plants under different NaCl supplies.

This effect was statistically significant in Phicoline, Frontoio and Souri (Table 1). In point of plant growth, Frontoio, Nabali and Maalot were significantly vigorous than Barnea, Souri, Picholine, Arbequina and Leccino. Frontoio was the most vigorous cultivar and SE of vigorous cultivars like Frontoio and Nabali were affected by the salinity in greater extent. However, salinity had no effect on SE of the least vigorous cultivar Leccino. Different researchers stated that different olive cultivars have been found to vary in the degree of their response to high salinity (Therios and Misopolinos, 1988; Benlloch et al., 1991). According to Tattini (1994) growth reduction by salt treatment was significantly higher in Leccino than Frontoio under saline conditions. Lewitt (1980) stated that growth reduction following salt treatments in olive is generally attributed to excessive salt accumulation in growing tissues. As reported by Tattini et al. (1992) growth reduction of olive plants is related to leaf Na^+ and Cl^- accumulation. Tester and Davenport (2003) reported that all salts can affect plant growth, but not all inhibit growth. In addition, salts do not act alone in the soil, but interact in their effects on plants; some of these interactions are simple (e.g. interaction between Na^+ and Ca^{+2}), whereas some are complex (e.g. carbonates, and their effects via increased soil pH). The most common effect of salinity is growth inhibition by Na^+ and Cl^-.

Another important approach is to assess the effects of salinity on plant growth to measure accumulation of dry matter in plant. In general, treatments decreased total DM of plants. This effect was statistically significant in Phicoline, Maalot, Souri and Frontoio (Table 2). Most probably, DM and SE are inversely related each other. For instance, Leccino accumulated more DM (ca. 140 g/plant) than other cultivars in control (Figure 2). However, SE of this cultivar was less than other cultivars (Figure 1).

Table 1. Relative shoot elongation (%) of some olive cultivars affected by increasing NaCl supplies

NaCl Treatment	Shoot Elongation		
(mg L^{-1})	Phicoline	Frontoio	Souri
Control	267.9	454.7	255.2
2560	210.6	333.2	302.2
5120	215.8	263.3	174.9
7680	200.8	273.3	214.4
LSD	38.68*	67.76	62.20

* Values are means of six replications. Means separations by Least Significant Difference (LSD) at P 0.05

The first NaCl treatment, 2560 mg L^{-1}, affected total DM accumulation of Arbequine, Leccino, Phicoline and Frontoio. However, Barnea, Maalot, Nabali and Souri were affected only at the highest treatment, 7680 mg L^{-1} NaCl (Figure 2).

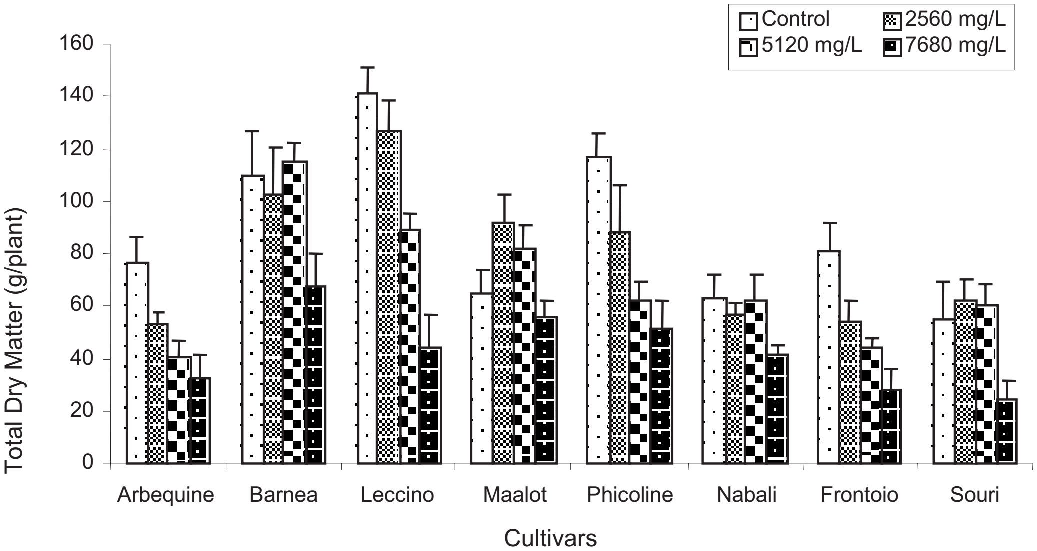

Figure 2. Effect of cultivar on total dry matter (g $plant^{-1}$) of olive plants under different NaCl supplies.

Table 2. Dry matter content (%) of some olive cultivars affected by increasing NaCl supplies

NaCl Treatment	Phicoline		
(mgL^{-1})	Root	Shoot	Leaf
Control	22.62	26.86	50.49
2560	18.43	27.83	53.71
5120	22.64	27.97	49.37
7680	29.31	28.54	42.12
LSD	3.58*	ns	4.01
		Maalot	
Control	18.31	32.42	49.23
2560	19.52	30.71	49.75
5120	23.69	28.83	47.46
7680	31.01	22.32	46.64
LSD	4.16	2.44	ns
		Souri	
Control	25.04	27.15	47.78
2560	20.04	28.38	51.55
5120	21.05	27.94	50.99
7680	28.86	19.66	51.45
LSD	ns	2.77	ns
		Frontoio	
Control	23.53	31.09	45.35
2560	18.41	32.36	49.19
5120	22.04	29.13	48.80
7680	25.26	25.16	49.55
LSD	ns	2.13	ns

* Values are means of six replications. Means separations by Least Significant Difference (LSD) at P<0.05

Presumably, salinity effect was related to SE rather than DM accumulation. This was clearly demonstrated from the response of cultivars Maalot&Nabali and Barnea&Picholine couples (Figure 2). These couples had similar total DM in the control (ca. 64 g plant^{-1} for Maalot&Nabali and 110 g plant^{-1} for Barnea&Picholine), but their DM accumulation were inhibited by salinity in different extent. Maalot accumulated only ca. 86% DM at the highest salinity, compared to 65% in Nabali, and Barnea accumulated 62% DM at the highest treatment, compared to 45% in Picholine. The effects of salinity on leaf, shoot and root DM were similar (Figure 3, 4 and 5).

Tattini et al. (1994) stated that salt resistance is inversely related to genotype's vigour. According to Staples and Toenniessen (1984), there is no correlation between extent of Cl$^-$ retranslocation and growth depression caused by salinity in several species. With regard to Na$^+$, however, there were significant correlations between decrease in DM production and Na$^+$ retranslocation from leaves, and, in particular, efflux of Na$^+$ from the roots. As reported by Therios and Misopolinos (1988) several hypotheses have been proposed to explain adverse effects of salinity on plant growth, such as salt exclusion mechanism, reduced

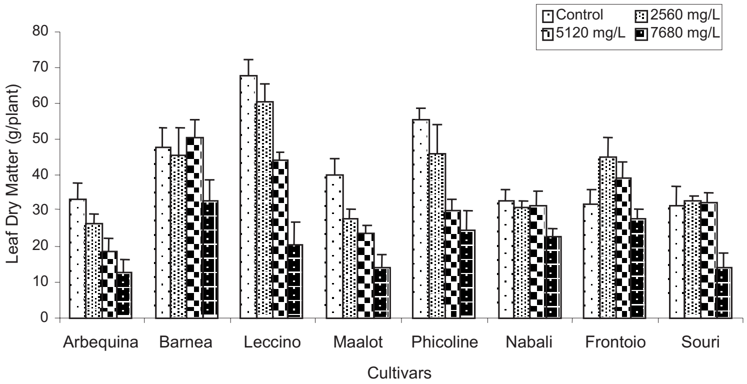

Figure 3. Effect of cultivar on leaf dry matter (g plant^{-1}) of olive plants under different NaCl supplies.

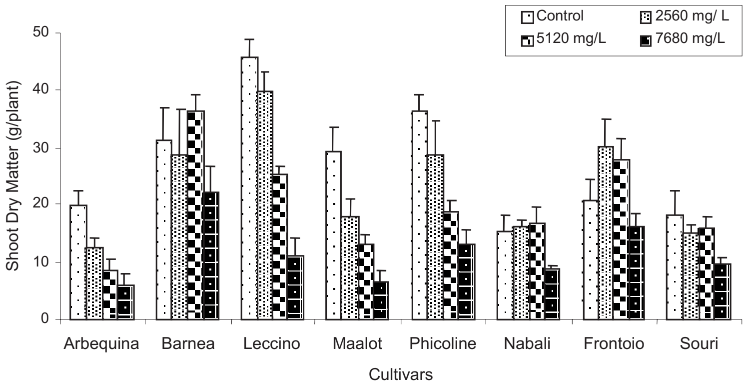

Figure 4. Effect of cultivar on shoot dry matter (g plant^{-1}) of olive plants under different NaCl supplies.

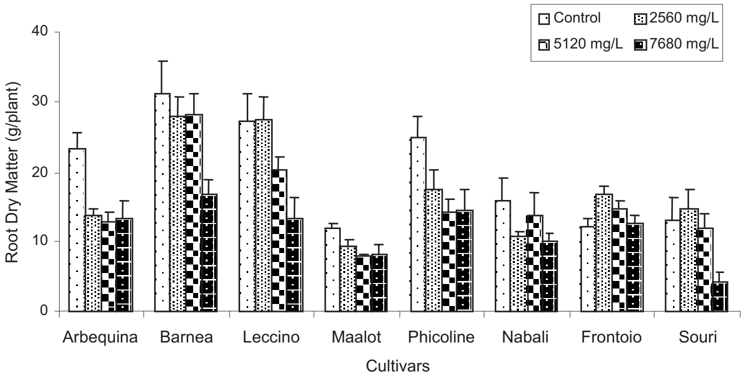

Figure 5. Effect of cultivar on root dry matter (g plant^{-1}) of olive plants under different NaCl supplies.

root permeability and water availability, enhancement of stomatal resistance, reduced translocation of assimilates to roots, amount of cytokonins reaching the tops, lower protein synthesis and decreased activity of enzymes, such as PEP and RuBP carboxylas. Salinity also affects the organelle ultrastructure, and mitochondria and distortion of tonoplast.

What the exact mechanism is of the differential response to salinity of the 8 cultivars tested is not known and this might be a subject for further research.

Most of the cultivars were similar in uptake and accumulation of Na$^+$ and Cl$^-$ (Figure 6 and 7). In general, concentrations of both elements in thin root was greatest, and in thick root and shoot less, than in leaves. Higher thin root concentration in olive cultivars might be resulted from low translocation potential (i.e. low mobile elements) or by a feedback control, from demand by vegetative growth, which regulated the uptake and translocation from root to canopy (Hale and Orcutt ,1987).

It is generally accepted that halophytes accumulate large quantities of ions (Na$^+$ and Cl$^-$) in their tissues in order to adapt to a saline environment (Flowers et al., 1977), whereas in contrast, mesophytes are generally known to limit the uptake of these ions (Greenway and Munns, 1980; Wyn Jones, 1981). Preferential accumulation of either Na$^+$, Cl$^-$, or both is known to account for salt tolerance in crop species and specific injury due to the accumulation of these ions rather than osmotic stress, which was suggested to be the major factor for salt sensitivity (Gratten and Grieve, 1999; Jacoby, 1999).

There were quantitative differences in Na$^+$ and Cl$^-$ uptakes of cultivars. The difference was greater in Na$^+$ and, Frontoio&Picholine couple accumulated at least 4-5 times higher Na$^+$ than other cultivars. Most probably, Na$^+$ accumulation is not related to SE and/or DM accumulation. For instance, Frontoio was vigorous but a low DM accumulator, Picholine accumulated fairly high DM but moderately vigorous (Figure 1 and 2). However, these cultivars had slightly less Cl$^-$ concentration in their tissues than the others (Figure 7). Although Frontoio and Picholine have similar Na$^+$ content in thin roots, they translocated Na$^+$ to upper plant parts in different extent (Figure 6). Relative translocation of Na$^+$ was lowest in Frontoio. Na$^+$ was retained mostly in thin roots in this cultivar. Therefore, Na$^+$ translocation process is not related to initial uptake level in thin roots. Except Phicoline, thin root Na$^+$ content of other cultivars was lower. According to Shibli and Al-Juboory (2002), there is no other olive cultivar that is better adapted to water and salinity stress in the Mediterranean Region than Nabali. Tattini et al. (1997) stated that Frontoio is salt tolerant and Leccino is salt sensitive olive cultivars.

Most probably salt tolerance of a plant depends on the regulation of ion transport (Ashraf, 2002) and different olive cultivars differed in uptake and translocation of NaCl (Tattini, 1994). For instance, Frantoio significantly depressed in Na$^+$ translocation from root to shoot with respect to the sensitive Leccino. As reported by Tattini et al. (1992) mechanisms of salt resistance in *O. europaea* are likely due to a control of net salt import to the shoot. The mechanism located within the root system and prevented the salt translocation, rather than the salt absorption.

Most plant species take up Cl$^-$ very rapidly and in considerable amounts. Uptake rate depends primarily on concentration in the nutrient or soil solution. There is considerable evidence that uptake is metabolically controlled (Mengel and Kirkby, 1982). According to Staples and Toenniessen (1984), higher plants might be classified as salt excluders and salt includers. Salt excluders possess mechanisms that ensure that salt reaches to shoot only in very small amounts. In contrast, salt includers absorb salt and store it at high amounts in stem and leaves. It is evident that, whatever the strategy by which a plant is able to adapt to salinity, transport phenomena plays a significant role.

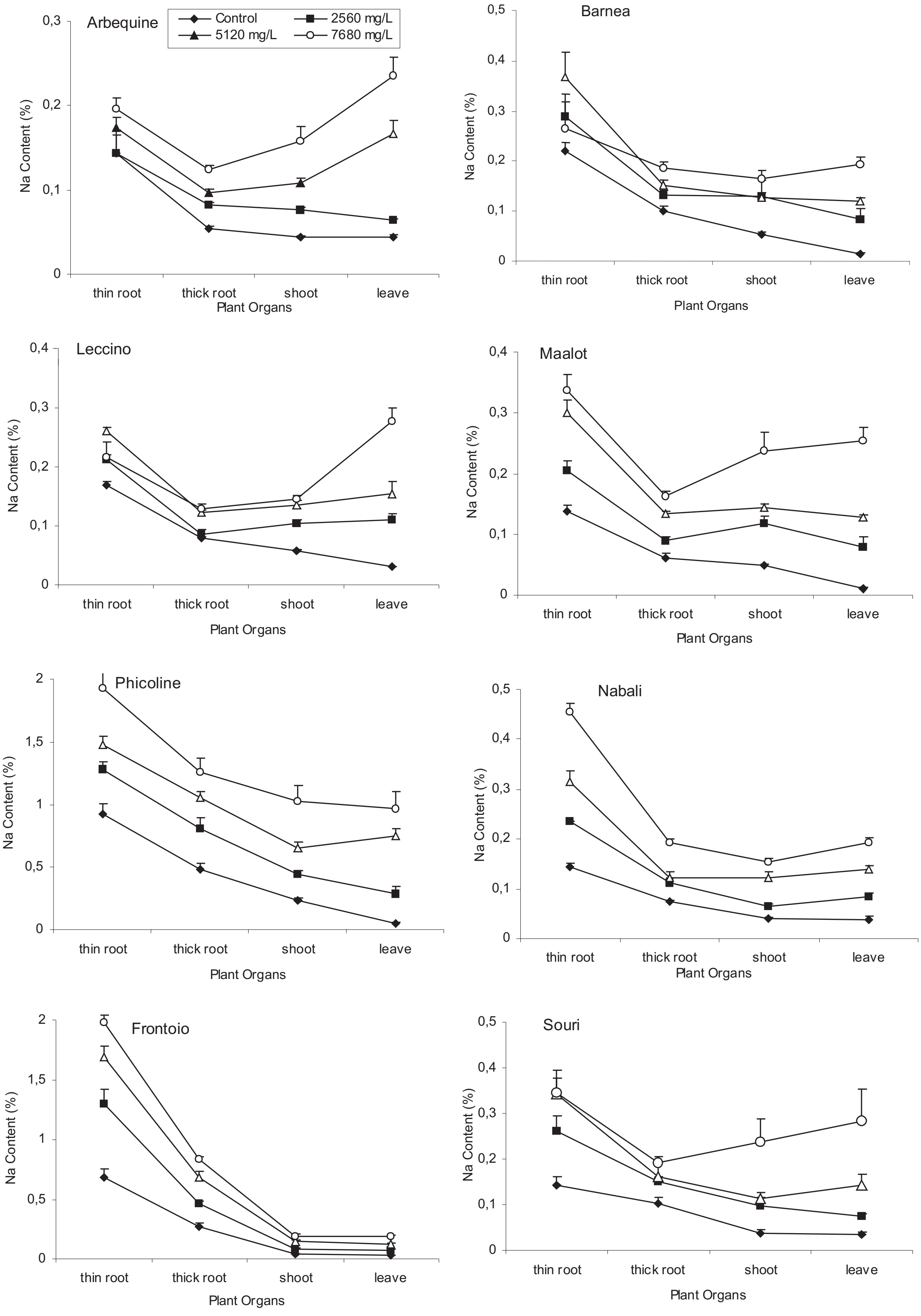

Figure 6. Uptake and translocation of Na$^+$ in different olive cultivars under different NaCl supplies.

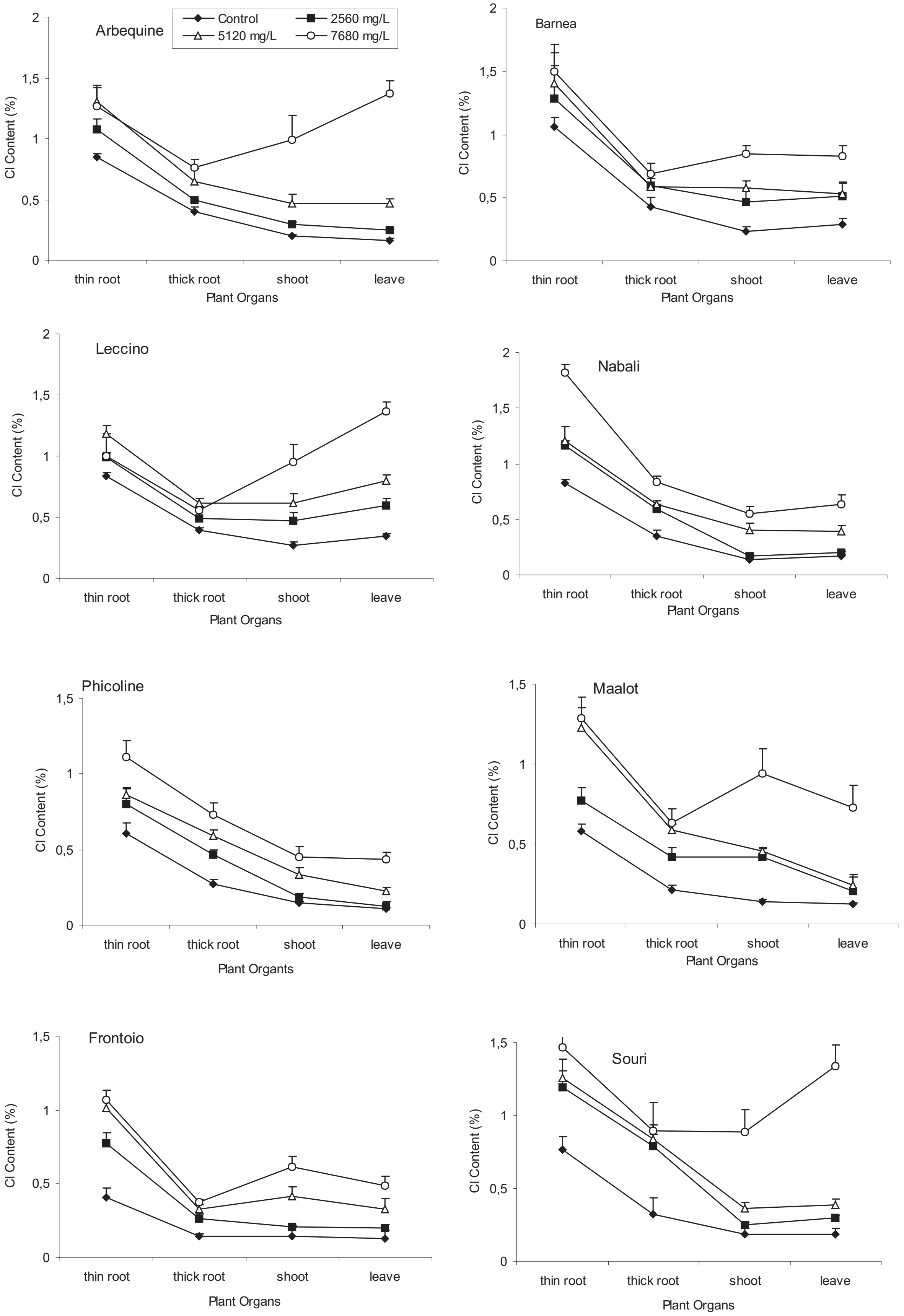

Figure 7. Uptake and translocation of Cl- in different olive cultivars under different NaCl supplies.

CONCLUSIONS

The results suggested that olive cultivars tested in the research differed in uptake and translocation of Na$^+$ and Cl$^-$. Difference was greater for Na$^+$. Frontoio and Picholine cultivars accumulated at least 4-5 times higher Na$^+$ in thin roots than others. Concentrations of both elements in thin roots were greater, and in thick root and shoot less, then in leaves. Higher thin root concentration was evaluated as a result of low translocation potential or feedback control. It is clear that salt exclusion mechanism is operative within the root system especially in thin roots. More vigorous cultivars were inhibited more significantly by NaCl treatments than intermediate and low vigorous cultivars. DM accumulation of some cultivars was evaluated as an account of SE by the increasing NaCl supplies. Salinity effect was related to SE rather than DM. SE determined as a good characteristic which might be help for identifying salt tolerance level of olive cultivars.

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