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Salt-stress effects on avocado rootstock growth. I. Establishing criteria for determination of shoot growth sensitivity to the stress

Nirit Bernstein[1], Marina Ioffe & Miriam Zilberstaine
Institute of Soils, Water and Environmental Sciences, The Volcani Center, PO Box 6, Bet-Dagan, 50-250, Israel
[1]*Corresponding author**

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Abstract

The mechanisms of shoot growth inhibition under salt-stress are not yet understood. Comparative evaluation of growth tolerance level of plant material used for mechanistic studies into stress effect on growth processes is a critical step towards establishing a plant model system for studies of stress effects on growth.

In the present study, numerous vegetative shoot-growth parameters were evaluated as criteria for determination of shoot growth sensitivity of avocado rootstocks (*Persea americana* Mill.) to salt-stress. Seven different West-Indian rootstocks were investigated for their growth response to the stress. The plants, grown in 50 L soil-filled containers, were treated with 4 *mM* Na and 6 *mM* Cl (control) or 18 *mM* Na and 20 *mM* Cl (salt) for a period of 3 years. Changes in trunk diameter, branch diameter, number of new branches, branch fresh weight (FW) and dry weight (DW), leaf length, leaf area, leaf number, leaf FW and DW and distribution of the leaf population into size classes, were quantified during the season of rapid shoot growth, and used for grading growth sensitivity.

The seven rootstocks studied were ranked according to the sensitivity of their vegetative growth to the stress. The order of sensitivity was found to be similar when the ranking was based on changes in DW and FW deposition per branch, cumulative leaf area and leaf weight per branch, or branch stem weight. Stress effects on leaf FW deposition were similar to the effect on total branch FW. Stress effects on branch stem FW were closely related to total branch DW deposition. Inhibition of cumulative leaf area per branch was less sensitive to the stress than deposition of FW or DW biomass. Biomass production per branch, or alternatively leaf biomass production per branch, is suggested as a practical parameter for evaluation of avocado rootstock shoot growth sensitivity to salt-stress.

Introduction

Sodium chloride salinity inhibits growth and reduces yield of many plant species. It is commonly hypothesized that plant growth inhibition under salt-stress is associated with altered water relations (osmotic effects), specific ion effects (excess or deficiency) or energy availability (carbohydrates) (Lazof and Bernstein, 1998; Munns, 1993). The mechanisms of growth inhibition under salt-stress are not understood. Investigations into the mechanisms underlying growth inhibition and tolerance often rely on comparative studies of lines, cultivars or transformants demonstrating differential levels of sensitivity or resistance to the sub-optimal conditions. Evaluating and grading the level of tolerance of growth processes to the stress of the different plant material used for investigation is a fundamental stage crucial for establishment of such comparative mechanistic studies.

Establishing parameters for evaluation of plant tolerance is not a trivial process. Salt-stress may have a differential effect on various growth processes in the plant. For example, reproductive and vegetative growth might be differently affected, and vegetative growth of the shoot is often more sensitive to salinity than is growth and development of the root

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* FAX No: 972-3-9604017. E-mail:nirit@volcani.agri.gov.il

(Delane et al., 1982; Weimberg et al., 1984). In the shoot, developmental process may prove to demonstrate a range of sensitivities to the stress. Ranking a collection of plant material based on their growth sensitivity to salt-stress is, therefore, not a straightforward procedure and the criteria used for determination of sensitivity level of the plant, and the specific growth process evaluated, might vary with the scientific (or the applicative) questions involved.

Avocado is very sensitive to NaCl in the root-growing medium; even low levels of salt inhibit tree growth and decrease productivity (Ben-Ya'acov and Michelson, 1995 and references therein; Bernstein, 1965). Although the species as a whole demonstrates high levels of sensitivity to the stress, a wide range of sensitivities exists between the commercial avocado races, and among the different rootstocks in each race (Bar et al., 1997; Ben-Ya'acov and Michelson, 1995 and references therein; Kadman, 1963; Oster et al., 1985, Xu et al., 2000). Overall, the West-Indian avocado race is more resistant to salinity than is the Mexican or the Guatemalan races (Cooper, 1951; Oppenheimer, 1947), with a wide range of sensitivity to salt-stress also found among the West-Indian rootstocks. Avocado is, therefore, well suited as a model system for the study of salt-stress effects on growth. The variation in sensitivity between related rootstocks allows comparative studies into the physiological mechanisms involved in growth inhibition and tolerance.

Sensitivity of avocado to salt-stress has usually been quantified as stress effect on agricultural production, or leaf-burn due to chloride or sodium accumulation. Resistant rootstocks, based on these criteria, demonstrate low levels of leaf damage under saline conditions, but the negative effect on productivity remains (Ben-Ya'acov et al., 1979). This suggests involvement of plant factors other than leaf damage in decreased productivity. In many crop plants, vegetative development is greatly inhibited by salt-stress, and in direct relation to decreased productivity. Only little information is found in the literature about the effects of salinity on specific growth parameters in avocado. Other than yield and biomass production, root/shoot ratio, tree height and trunk diameter, we know of no other tree growth factor referenced to be applied for determination of tolerance level in avocado. In the present study, we evaluate numerous shoot growth parameters for quantification of rootstock shoot growth sensitivity to salt-stress. We demonstrate a range of sensitivities to salt-stress of various shoot growth parameters of seven West-Indian rootstocks. Determination of the level of growth tolerance among these rootstocks, and their ranking according to the sensitivity level, will allow (in following studies) comparative evaluation of the relevance of alteration in physiological parameters under stress to growth inhibition or maintenance.

Materials and methods

Plant material

Seven avocado rootstocks of the west-Indian race were used in this project. The designated rootstocks were selected during the 1950s and 1960s for excellence under elevated NaCl conditions from trees grafted on seedlings rootstocks from the first commercial avocado orchards throughout Israel (Ben-Ya'acov et al., 1992). Among the seven selected rootstocks, vc 69 is the only one which is currently commercially grown in Israel. It is known as relatively resistant to salt conditions based on its performance in terms of yield production and Cl induced leaf burn. The other six selected rootstocks used in this project (vc 55, vc 131, vc 159, vc 256, vc 803, vc 804), were recently found to vary in the degree of Cl induced leaf burn (Hirschland, 1997). The extent of salinity-induced leaf burn increased from vc 256 to vc 55 in the following order $256<803<804<131<69<159<55$. The agronomic production and growth sensitivity of these rootstocks to salt-stress were not investigated.

Vegetative clones (vc) of the selected rootstocks were prepared in a commercial nursery. When the generated ungrafted plants (e.g. 'rootstocks') were 1 year old, selected uniform seedlings were transferred to 50 L growing containers and were established in a screenhouse at the Volcani Center campus. Four plants per rootstock were so selected. The growing containers were filled with a peat and tuff mixture.

Growth conditions

The avocado plants were housed in a naturally lit screenhouse, in which air temperature during the growth measurements duration varied between a maximum of 35 °C and a minimum of 17 °C. The plants were irrigated with a drip irrigation system. Nutrient solutions were given copiously to permit drainage through the pot, and avoid salt accumulation in the root zone. Na and Cl levels in the drainage solution were routinely monitored. As an added precaution, pots were leached once a month with water, followed by displacement with nutrient solution containing the appropriate salt level. The plants were irrigated every 48 h (during the summer) or every 72 h (during the fall, winter and spring) with 1/4 strength Modified Hoagland solution (Bernstein et al., 1993). Salinization began two months after the plants were transferred to the 50 L growing containers. The control solution contained 6 *mM* Cl, and 4 *mM* Na. The saline solution contained 20 *mM* Cl and 18 *mM* Na.

Growth measurements

The cumulative effect of the first 2 years of exposure to the stress on overall plant status was evaluated non-destructively prior to the beginning of the study. Plant height and stem diameter were measured, and the overall appearance of each plant was evaluated. The stressed plants were ranked on a scale of 1–5 based on reduction in canopy size and appearance of leaf burn damage (with 5 representing lack of stress effects, and 1 representing severe leaf-burn and size decrease (Table 1).

Effects of salt-stress on numerous vegetative shoot growth parameters were studied. The emergence and growth of new spring branches (spring flush) was followed during the shoot rapid growing season from May to July. The rate of branch emergence was recorded, and the total number of new branches emerging on each plant during 70 days of growth was quantified. A destructive harvest of the new spring branches 70 days following branch emergence allowed evaluation of the rates of biomass deposition into the branches (FW and DW); rates of branch stem growth; rates of cumulative leaf length, leaf area, leaf FW and leaf DW increase; and analysis of the distribution of the leaf population on each branch to size classes. Branch diameter and trunk diameter were measured as well. The seven rootstocks studied were ranked according to the sensitivity of their vegetative shoot growth to the stress.

Branch diameter was measured with a caliper, 70 days after branch emergence, at location 0.5 cm above the point of attachment to the lower order branch. The length of each leaf on the spring branches was measured as the distance from the base of the leaf lamina, to its tip. Results are presented as cumulative leaf length per branch (and represent 70 days growth). The area of individual leaves was recorded with a portable area meter (LI-COR, model Li 3000, Lambda instruments corporation). Results are presented as cumulative leaf area per branch (and represent 70 days growth). Measured values for individual leaves were used for the division of the leaf population into size classes. The fresh weight (FW) of individual leaves was recorded immediately following removal from the tree; dry weight (DW) was recorded following 48 h of desiccation in an 60 °C oven. FW and DW represent deposition into the branch during 70 days of branch growth. Trunk diameter was measured 10 cm above ground level. Marks were made on the trunk with a felt pen to ensure that the caliper was guided to the same spot on each measurement occasion. Reported diameters were measured at the beginning of the studied growing season. Diameter measurements 24 and 36 months after the initiation of the salt-treatment, when the plants were 36 and 48 month old, respectively, were used for calculations of trunk cross sectional area increase during the investigated season.

Statistical analyses

Statistical testing (of the various growth parameters) by a one-way analysis of variance (ANOVA) was performed by SAS program (SAS Institute Inc. SAS Campus Drive, Cary, NC 27513, USA). *P* values of the *F* statistics and pooled standard error (P.S.E) are presented in the figures. The letters in Table 1 represent significance by the SNK method (Student-Newman-Kleuls test) at 5% level.

Correlation coefficients (*r*) between inhibition of shoot growth and the other growth parameters and their *p*-values were calculated using SAS program.

Results and discussion

The seven examined rootstocks varied in their shoot growth response to salinity. Non-destructive measurements at the beginning of the growing season examined in this project demonstrated differential stress effects on overall tree appearance, trunk diameter and tree height (Table 1). Overall appearance of the shoot (evaluated visually according to canopy size, and leaf burn damages) was affected by salinity in all of the examined rootstocks. vc 159 was least affected by the stress, and vc 69 demonstrated the highest level of stress induced damages. A significant reduction in trunk diameter was observed in 4 of the 7 examined rootstocks (vc 69, 159, 803 and 55), and a significant reduction in tree height was observed in 5 rootstocks (vc 159, 55, 803, 804, 131).

Table 1. Status of the plants at the beginning of the experimental season, following 2 years of salinization. Plant status was evaluated on a scale of 1 (severe stress effects) to 5 (no stress effects). Data are means ± SD for 4 replicates. Means associated with a common letter within the column are not significantly different at the 0.05 level

Rootstock	Plant status	Trunk diameter (cm)			Plant height (cm)		
		Control	Salt	% of control	Control	Salt	% of control
vc 69	3.21±0.15 c	2.15±0.11	1.78±0.14	83.0 b	206.7±23.3	197.7±14.5	95.2 a
vc 256	3.65±0.20 bc	2.48±0.14	2.41±0.11	96.9 a	190.0±35.1	166.7±14.5	87.7 ab
vc 131	3.61±0.23 bc	2.25±0.32	2.23±0.18	99.2 a	210.0±30	175.1±25	83.3 b
vc 804	3.70±0.18 bc	2.20±0.01	1.91±0.14	86.9 ab	153.3±14.5	120.0±20	78.2 bc
vc 803	4.20±0.21 b	2.60±0.33	1.88 ±0.27	72.4 b	236.7±6.7	175.3±12.3	73.2 c
vc 55	4.03±0.31 b	2.39±0.14	1.99±0.17	83.4 b	263.3±18.6	196.7±24	74.7 c
vc 159	4.70±0.23 a	2.96±0.45	2.53±0.10	85.5 b	276.7±28.5	227.7±13.3	81.9 b

Destructive measurements of new spring flush branches, 70 days following their emergence on the higher order branch, allowed comparative evaluation of stress effects on specific vegetative growth parameters in the examined rootstocks. Since the plant biomass present on the branch at the time of sampling represent 70 days growth, the single destructive harvest allows evaluation of rates of biomass deposition (i.e. g/70 days). Salinity induced a decrease in the production of biomass per branch in 4 out of the 7 examined rootstocks (vc 69, vc 256, vc 131 and vc 804) (Figure 1a). The FW of the new spring flush branches of these rootstocks was reduced 32–51% (Figure 1b) and their DW was reduced 42–58% under salinization (Figure 1c). Biomass production per branch in the remaining 3 rootstocks (vc 803, vc 55, vc 159) increased under salt-stress conditions. The new spring flush branch FW was increased 37–100% (Figure 1b), and branch DW was increased 41–59% by salinization (Figure 1b).

Vegetative growth of plants is very well known to be sensitive to saline conditions (Lazof and Bernstein, 1998 and references therein). In particular, the growth of plant shoots (or a tree canopy) demonstrates high sensitivity to salt-stress. Previous studies with avocado, as well, reported inhibition of vegetative development of the shoot when the tree was irrigated with stress-induced levels of NaCl (Bar et al., 1997; Bingham et al., 1968; Downton, 1978; Oster et al., 1985; Oster and Arpaia, 1992; Steinhardt et al., 1986; Wallace et al. 1955). In light of these findings, our results for the 3 West-Indian rootstocks, (vc 803, vc 55 and vc 159), which demonstrate stimulation of vegetative shoot growth under stress conditions, might be surprising. One must note, however, that the rootstocks investigated in this study are vegetative clones of rootstocks from selected trees (grafted on seedling rootstocks), identified in commercial orchards, to excel under elevated salt conditions. Shoot parameters such as yield, extent of leaf chlorosis and leaf burn, were used as criteria for tree performance under stress. The performance of these rootstocks under salt-stress conditions, in terms of biomass production, might therefore be expected to exceed that of known commercial rootstocks.

The number of newly emerging spring vegetative flush branches, and their biomass production, are among the factors determining tree potential for next year's yield production. In three of the seven investigated rootstocks, salt-stress significantly increased the number of new spring flush branches (vc 256, 55 and 159 /Figure 2) increasing the potential for excelled yield under stress. In the remaining four rootstocks, the appearance of new branches was not significantly affected (vc 69 and 803) or was reduced by salinization (13 and 22% in vc 804 and vc 131, respectively).

The total production of biomass per plant by the new spring flush branches (a factor of the number of new emerging branches and of the biomass production per branch), was significantly increased under stress in three of the seven rootstocks (vc 803, vc 55 and vc 159) (Figure 2, insert). These 3 rootstocks demonstrated improved growth based on single branch biomass production as well (Figure 1). vc 69, which demonstrated the highest sensitivity to the stress based on biomass production per branch, demonstrated, as well, the largest reduction in total biomass production (to 50% of the non-stressed value). Interestingly, in vc 256, the stress induced increase in the number of

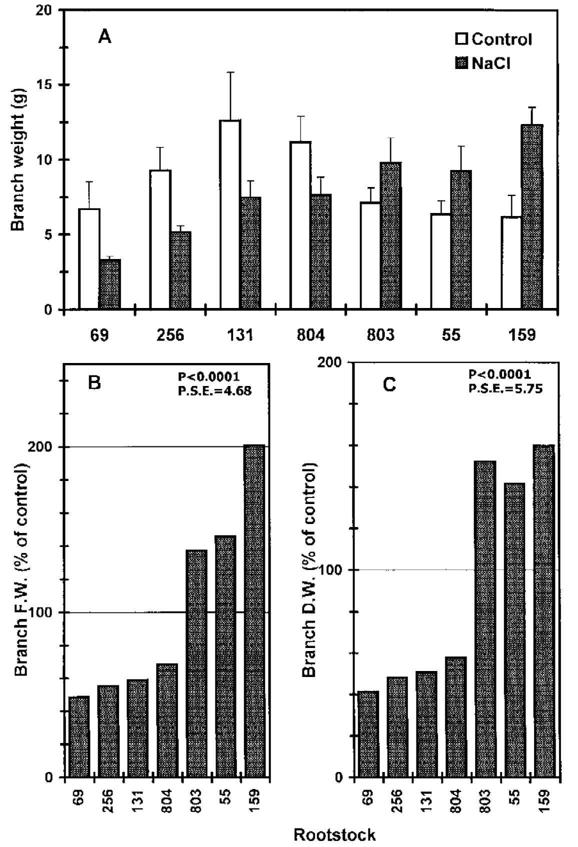

Figure 1. Effect of salinity on branch growth (new spring flush) of 7 avocado rootstocks selected to excel under elevated NaCl condition: (a) Branch fresh weight, (b) Branch F.W. (% of control), (c) Branch D.W. (% of control). The data are results of a destructive harvest of branches 70 days after branch emergence, and represent biomass deposition into a branch during 70 days of growth. Data are means of 4 replicated plants. Error bars in (a) are SD (n=4). Results from all branches (10–30) that emerged on a specific plant during the month of May were averaged to give the plant result. P-values of the F-statistic and pooled standard errors (P.S.E.) are presented in (b) and (c).

new flush branches compensated for the inhibition of biomass deposition into individual branches, so that overall biomass deposition into the plants was only slightly decreased under stress (14% /Figure 2, insert).

Except in one rootstock (vc 256), the order of sensitivity to the stress among the rootstocks was similar when the evaluation criterion was biomass production per single flush branch (Figure 1), or total shoot flush production (Figure 2, insert). Since the former is much easier quantified, we suggest it as a preferred evaluation factor for shoot growth inhibition under stress.

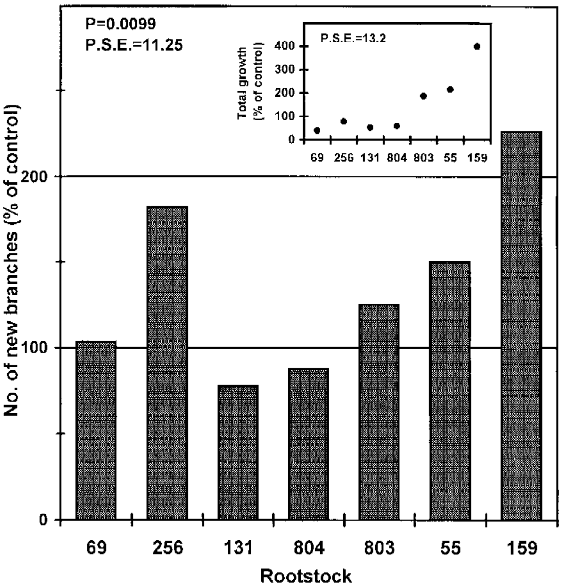

Figure 2. Effect of salinity on the number of new branches (main figure), and the cumulative biomass production of the new branches (inset). Growth of branches that emerged during the month of May was monitored during the following 70 days. Total growth represents cumulative biomass deposition into the branches that emerged over 70 days, during 70 days of branch growth. Data are means (n=4 plants), P-values of the F-statistic and pooled standard errors (P.S.E.) are presented.

Avocado is characterized by several vegetative growth flushes each year. Total shoot annual biomass production is a cumulative result of growth during a number of flushes. It is currently unknown if salt-stress affects the number of growth flushes per year, and if growth during different flushes is similarly effected.

Biomass deposition into new flush branches can be divided to deposition into photosynthetic tissue (leaves) or structural tissue (stem). Salt-stress might affect each of these components differently. Stress induced alterations of the photosynthetic leaf tissue, or the new branch stem tissue might affect tree performance differently. While the main photosynthetic tissue (leaves) determines the potential for carbohydrate production (and hence vegetative growth and yield production), characteristics of the stem tissue affect the tree structure and physical strength. The results presented in Figures 3 and 4 demonstrate the variable effect of salt-stress on leaf and branch stem parameters. These figures describe stress-induced changes in cumulative growth per branch during the 70 days following branch emergence; they can, therefore, be considered as rates (g/70 days; area/70 days; length/70 days etc.) Cumulative leaf FW, leaf length and leaf

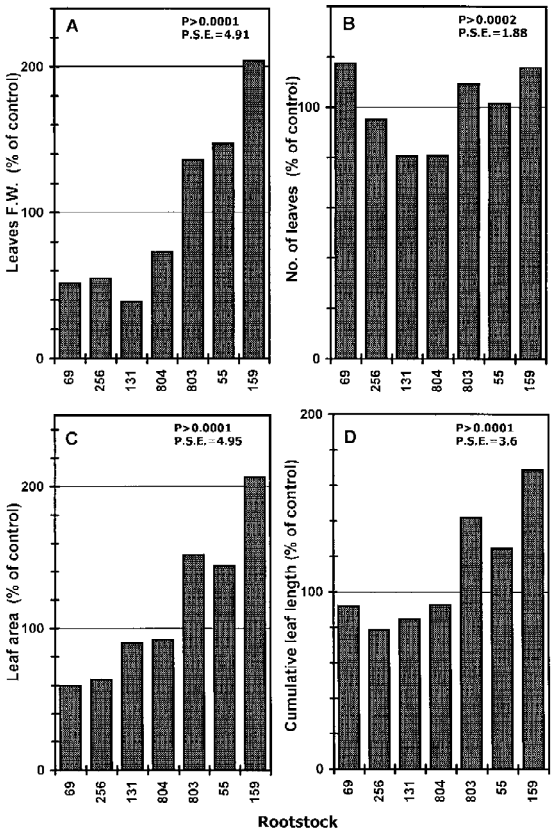

Figure 3. Effect of salinity on leaf growth, (a) Total Leaf F.W. per branch, (b) Number of leaves per branch, (c) Total leaf area per branch, and (d) cumulative leaf length per branch. The data was evaluated from biomass production during 70 days of branch growth. Data are means (n=4 plants), P-values of the F-statistic and pooled standard errors (P.S.E.) are presented.

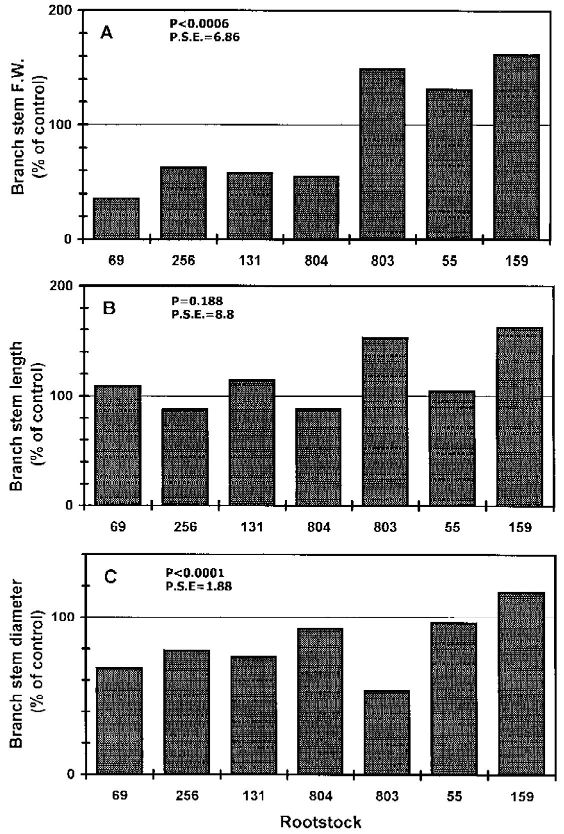

Figure 4. Effect of salinity on the branch stem growth. (a) branch stem biomass, (b) branch stem length and (c) branch stem diameter. The data was evaluated from biomass production during 70 days of branch growth. Data are means (n=4 plants), P-values of the F-statistic and pooled standard errors (P.S.E.) are presented.

area production per branch were inhibited by salt-stress in 4 of the 7 rootstocks studied (vc 69, 256, 131, and 804/ Figure 3), the same rootstocks which demonstrated inhibition of branch biomass production under stress (Figure 1). Branch stem FW of the same 4 rootstocks was inhibited as well (Figure 4a). Therefore, inhibition of biomass production per branch was a combined result of reduced biomass deposition into both leaf and stem tissues. Salinity affected leaf-tissue growth similarly to stem-tissue growth; in our 7 examined rootstocks, stress induced inhibition of leaf tissue growth was always accompanied with similar extent of stem tissue growth inhibition (as is apparent from vc 69, 256, 131 and 804/ Figure 4a). Stress induced stimulation of leaf tissue growth was similarly always accompanied by stimulation of stem tissue growth (apparent from vc 803, 55 and 159/ Figure 4a).

The cumulative leaf length increase (Figure 3d) was less sensitive to salt-stress than branch biomass production, cumulative leaf area or leaf FW increase (Figures 1 and 3c, a). It is therefore a less suitable parameter for determination of growth sensitivity. Measurements of cumulative leaf area and leaf FW increase give similar results to branch biomass production (Figures 1 and 3) regarding the order of rootstock sensitivity to salt-stress. The extents of growth reductions described by leaf area measurements however were somewhat lower than the values achieved by branch biomass production measurements (Figures 3c and 1). Measurements of total biomass production per branch, or alternatively, the somewhat more demanding measurements of leaf FW deposition per branch are, thus, suggested for determination of growth sensitivity to salt-stress.

While the stress effect on leaf FW deposition was similar to stress effects on total branch FW deposition (Figures 3a and 1b), the effect on branch stem FW deposition was closely related to total branch DW deposition (Figures 4a and 1c).

Branch stem elongation (Figure 4b) was less affected by salt-stress than overall branch growth (Figure 1b). No correlation was found between the effect of salinity on deposition of biomass into a branch, and the effect on its growth in length (Figure 4b, and Figure 7/ discussed further in later sections). In two rootstocks (vc 69 and cv 131), branch stem length was not significantly affected by the stress, while branch weight was reduced; and in vc 55 branch length was not affected by the stress while branch biomass increased 30% (Figure 4a,b).

Similar to branch stem elongation, branch stem radial expansion was also less affected by salt-stress than overall branch growth (Figures 4c and 1). Here, too, no correlation was found between the effect of salinity on deposition of biomass into a branch, and the effect on its stem diameter growth (Figure 7).

The rate of initiation of new leaves on a flush branch was affected by salt-stress (Figure 3b) differently than the rate of deposition of leaves biomass (Figure 3a) or branch biomass (Figure 1b). Salt-stress induced a significant reduction in the rate of leaf emergence per branch in 2 of the 7 examined rootstocks (vc 131 and 804) and significantly increased the rate of leaf initiation in other 3 rootstocks (vc 69, 803 and 159) (Figure 3b). Stress effects on leaf biomass deposition (Figure 3a) were not positively correlated with the effect on new leaf initiation. For example, vc 69, which suffered a large reduction in leaf biomass, demonstrated the highest increase in leaf number under stress (Figure 3a, b). And in vc 55, which showed about 45% increase in leaf biomass, leaf number was not significantly affected by stress.

Clearly, such deviation from positive correlation suggests an effect of salt-stress on the distribution of the leaf population according to size classes. This issue was further investigated in Figure 5. Additionally, such deviation rules out the application of the simple non-destructive measurement of leaf initiation rate as a criterion for estimation of salt-stress effect on shoot growth.

Salt-stress altered the distribution of leaves into size classes. The highest proportion of leaves, in both treatments, was always found in the leaf size categories between 20 and 80 cm^2 (Figure 5). In the 3 most tolerant rootstocks, which demonstrated stimulation of shoot growth under salt-stress (vc 803, 55, and 159/ Figure 1, Figure 2 insert), the proportion of leaves which was found in the size categories between 20 and 80 cm^2 was greatly reduced by stress. The proportion of leaves in the larger size categories was increased under stress and so was the maximal leaf size. Since stress induced but a relatively small increase (2–17% Figure 3b) in the number of leaves per branch in these 3 rootstocks (for vc 55 the 2% increase is not significant), we conclude that the large increase in cumulative leaf area per branch (44–106% Figure 3c) in the tolerant rootstocks was a result of the stress-induced increase in percentage of leaves in the largest size categories.

In the 4 rootstocks that demonstrated sensitivity of shoot growth to salt-stress (vc 69, 256, 131 and 804 /Figure 1) leaf size was affected differently by salt-stress. In both vc 69 and 256, the proportion of leaves in the size classes between 0 and 60 cm^2 was higher under stress, and the distribution curves condensed to the left (towards the smaller leaf categories) indicating a smaller percentage of large leaves under stress, and a stress induced reduction in maximal leaf size. While no leaves on the stressed rootstocks were larger than 120 mm^2, leaves of the size category 160–180 mm^2 were present on the non-stressed plants. The large reduction in leaf biomass per branch of these 2 rootstocks (about 50%/ Figure 3a) and in leaf area (about 40%/Figure 3c) was caused by the decrease in percentage of large leaves and maximal leaf size. The stimulation of leaf emergence rate under stress in vc 69 (which resulted in 17% increase in the number of leaves which emerged on the branch during the 70 days of observation /Figure 3b) could not compensate for the reduction in the proportion of large leaves and maximal leaf size and therefore did not prevent the reduction in leaf area.

In vc 804 and vc 131, stress effects on leaf number (Figure 3b) and leaf area per branch (Figure 3c) were similar. In vc 804, salt-stress had but little effect on the distribution of leaves to size classes. The reduction in leaf biomass (Figure 3a) and leaf area (Figure 3c), therefore resulted from reduction in leaf number (20%/Figure 3b). The larger inhibition of leaf biomass per branch in vc 131 resulted from depression in the percentage of leaves of the size classes 40–100 cm^2 (which was not compensated by the small increase in% of leaves of the size classes 120–160 cm^2 (Figure 5).

It is important to note that the data presented in Figure 5 are a description of one point in time (a destructive harvest) selected to represent growth over 70 days (following branch emergence). It was

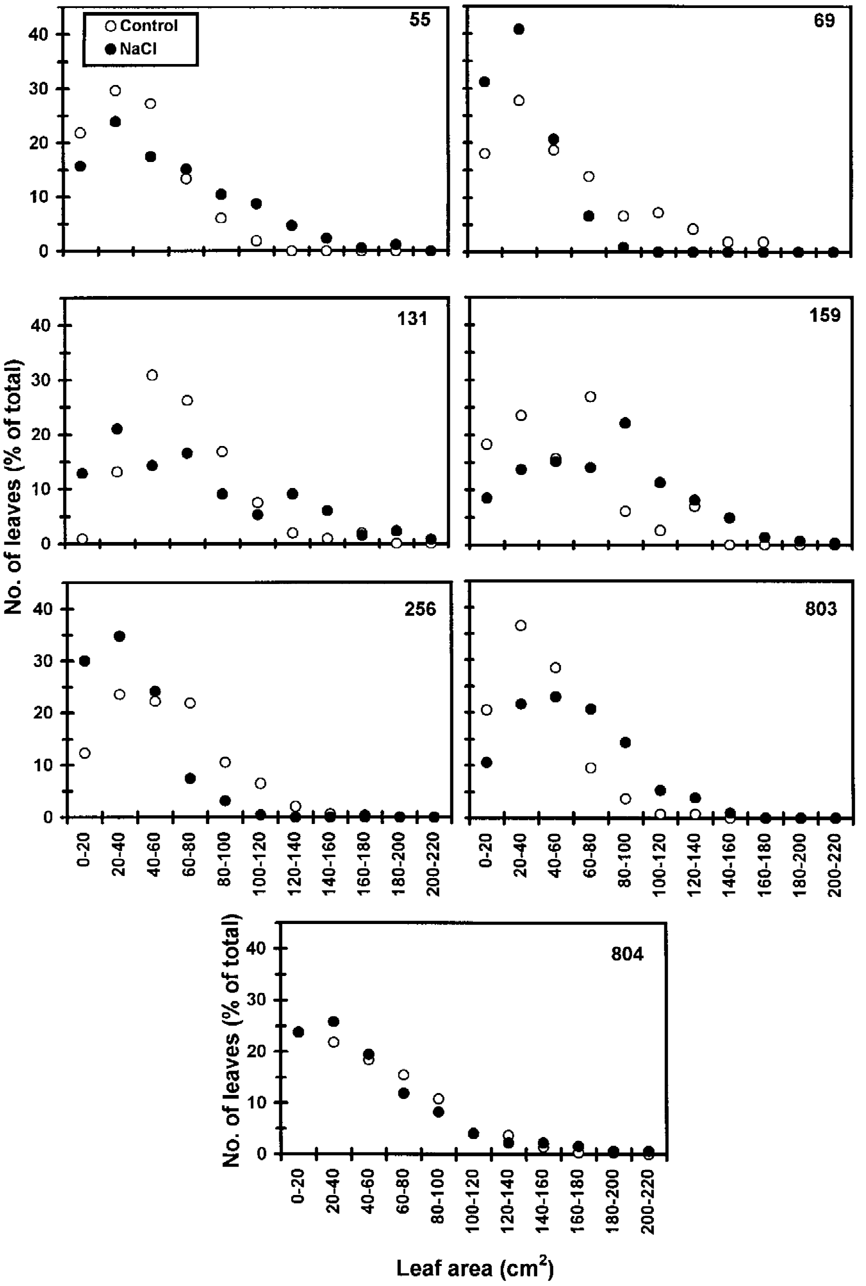

Figure 5. Effect of salinity on leaf size. The figure presents the distribution of leaves, from branches that emerged during the month of May, into size classes according to their area. Results of destructive measurements following 70 days of branch growth.

impossible to distinguish by this method whether a small sized leaf was a very young one still undergoing rapid expansion, or alternatively an older leaf whose development was arrested and expansion terminated early, as we have observed to occur occasionally under salt-stress. Non-destructive investigations of leaf expansion growth under stress throughout the leaf development duration are needed, and are currently under way in our laboratory.

Figure 6 demonstrates the effect of salt-stress on trunk cross sectional area increase, during the third year of salinization. Salinity inhibited the radial expansion of the trunk of the 7 examined rootstocks (Figure 6, inset). The extent of inhibition of trunk cross

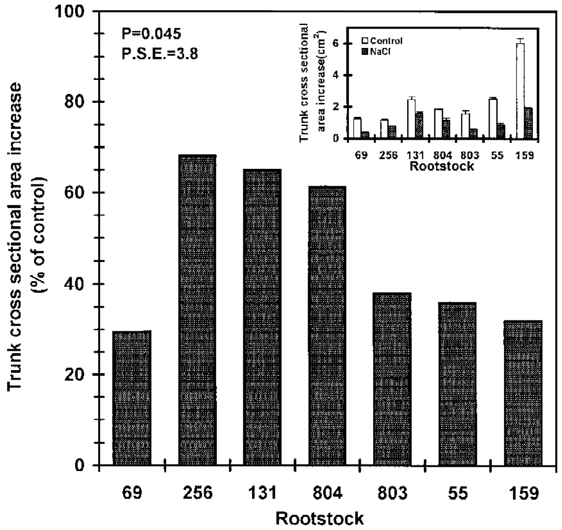

Figure 6. Effect of salinity on trunk cross sectional area increase. Presented as% of control (main figure) or cm^2 (inset). The data was evaluated from two non-destructive measurements, 24 and 36 months after the initiation of the salt-treatment, when the plants were 36 and 48 months old, respectively. Diameter was measured 10 cm above ground level. Data are means of 4 plans. SD (inset) and *P*-values of the *F*-statistic and pooled standard errors (P.S.E.) (main figure) are presented.

sectional area increase ranged from 32–70% (Figure 6). Similar effects of salinity on trunk diameter growth have been reported by several investigators (Bingham et al., 1968; Downton, 1978; Kadman, 1963; Oster et al., 1985) for other avocado rootstocks, and various rootstocks-scion combinations. Earlier measurements of trunk diameter, at the beginning of the experimental season when the trees were exposed to salinity for only 24 months /Table 1), demonstrated similar trends of stress effects on trunk radial growth, but a lower extent of growth inhibition (2–48% inhibition of trunk cross sectional area increase, calculated from the diameter data presented in Table 1 for the first to years of growth). The lower extent of growth inhibition in the earlier measurement was probably due to the smaller fraction of time that the plants were exposed to salinity in this evaluated growth duration (2 out of 3 years of growth), in comparison to the later measurement where the stress was imposed throughout the evaluated growth duration. Additionally, the extent of trunk growth inhibition might increase under longer exposure to the stress.

The stress induced reduction of rootstock trunk diameter growth, which is well known to be a good indicator of tree exposure to salt-stress, was not a suitable parameter upon which to base salt-tolerance and sensitivity. In our experimental rootstock system, no correlation was found between the extent of radial trunk expansion, and biomass production by the shoot under salt-stress. For example, the trunk expansion in vc 159, 55 and 803 was reduced under salt-stress, no less than in the other 4 investigated rootstocks, giving no indications of the high tolerance of their shoot growth to the salt-stress conditions. The salt-stress effect on trunk expansion was also not indicative of the effect on biomass deposition into branch stem tissue. The extent of trunk growth inhibition did not correlate with branch stem diameter growth (Figure 4c), branch stem FW (Figure 4a) or branch stem elongation (Figure 4b) nor with the number of new flush branches (Figure 2).

Figure 7 demonstrates the relationships between shoot-growth tolerance (calculated as inhibition of branch biomass production under stress) and changes in specific growth parameters under stress in each of the seven examined rootstocks. High positive correlation is demonstrated between shoot growth tolerance of a rootstock, and its leaf FW production per branch (r=0.993), leaf area per branch (r=0.984), cumulative leaf length per branch (r=0.973) and stem FW (r=0.953). A significantly smaller correlation coefficient was found for the number of spring flush branches per plant (r=0.846). The number of leaves per branch, stem length and stem diameter, demonstrated poor correlation with shoot growth tolerance (r= 0.448, 0.693 and 0.520, respectively).

Rootstocks are known to influence the salt-tolerance of grafted fruit trees (Maas, 1990), including avocado (Cooper, 1951; Embelton et al., 1962; Haas, 1950; Kadman, 1970; Wallace et al., 1955). Shoot (scion) biomass production in avocado decreases with increasing salinity (Oster and Arpaia, 1992) with differences among rootstocks influencing the extent of growth reduction under stress. Salt-tolerance is an arbitrary term, associated with the selected criterion upon which it was based. The rootstocks investigated in the present study where chosen for excellence under elevated NaCl-condition based on excelled fruit production, and minimized leaf burn of mature grafted trees (Ben-Ya'acov et al., 1992). Prior investigations have reported that results of such selection were rootstocks in which the salt-tolerance mechanisms include reduced transport of Cl and exclusion of Na. In the present project, we demonstrate a wide range of growth sensitivities among these selected rootstocks, resulting in growth inhibition or growth stimulation under salt-levels typical of commercial fields in Is-

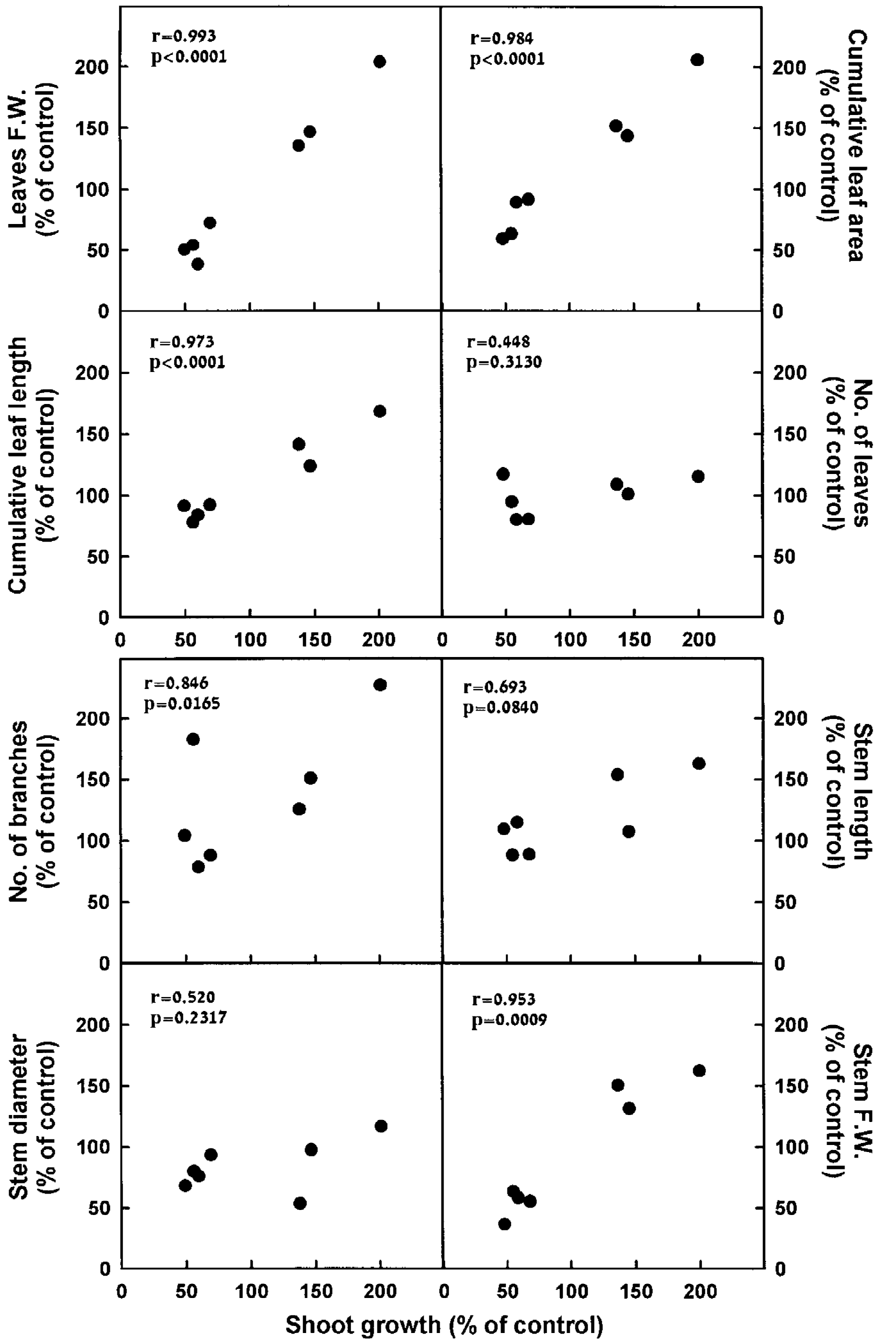

Figure 7. Correlation between inhibition of several shoot growth parameters by salinity, and extent of shoot growth reduction. Shoot growth reduction was calculated as inhibition of total branch biomass production (from data presented in Figure 1). Correlation coefficients (*r*) and *p*-values are shown in the figure.

rael. Such a variable growth response to the stress can be used for further investigation of salt-tolerance mechanisms. It might also indicate that the tolerance mechanism imposed by these rootstocks on the grafted trees, in terms of fruit production, might also be associated with the extent of growth sensitivity of the rootstock itself to salt-stress (i.e. sensitivity of root-growth to the stress). Two types of studies are suggested from this line of thinking: (a) evaluation of correlation between Na and Cl exclusion (or other ionic relations in the shoot) and level of growth tolerance and (b) examination of the correlation between root growth sensitivity, and tolerance level of the shoot. Very little information is found in the literature concerning these issues, which are presently being investigated in our laboratory, and are the topics of the following two manuscripts of this series.

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