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Effect of Salinity on Tomato Fruit Ripening[1]

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ABSTRACT

Tomato (*Lycopersicon esculentum* Mill) plants from various cultivars growing on half-strength Hoagland solution were exposed at anthesis to 3 or 6 grams per liter NaCl. Salinity shortened the time of fruit development by 4 to 15%. Fruits of salt-treated plants were smaller and tasted better than did fruits of control plants. This result was obtained both for ripe fruits tested on the day of picking and for those picked at 100% development and allowed to ripen at room temperature for 9 days. Percentage of dry weight, total soluble solids, and titratable acidity; content of reducing sugars, Cl^-, Na^+, and various pericarp pigments; and electrical conductivity of the juice were higher in fruits of saline-treated plants than they were in those of control plants, while the pH was lower. Ethylene and CO_2 evolution rates during ripening; as well as the activities of pectin methyl esterase, polymethylgalacturonase, and polygalacturonase; were also higher in fruits of the saline-treated plants. The treatment with 6 grams per liter NaCl shortened the fruit shelf life considerably.

Although much work has been done on the effect of salinity on various aspects of plant growth and development (13), very little attention has been paid to the effect of salinity on fruits (19). In the tomato, the effect of salinity on fruit ripening is not known, even though its influence on other aspects of the plant has been investigated (12, 16).

Inasmuch as the tomato is an economically important crop throughout the world (15) and inasmuch as the water available for irrigation in our part of the country—the Negev desert—is brackish, we investigated the effect of salinity on tomato fruit ripening. The degree of salinity in our experiment was the same as that in the natural wells of the region.

MATERIALS AND METHODS

Plant Material. The different experiments were carried out on some or all of the following cultivar of the tomato (*Lycopersicon esculentum* Mill): Hosen Eilon (228); 202.206; 364.365; Arava (S5) Rutgers; All-round; Exhibition; and Moneymaker. Two-week-old seedlings which had been germinated on vermiculite were transferred to half-strength Hoagland solution aerated with compressed air. The plants were trained to one stem, and one or two fruits were allowed to develop at each cluster, as described previously (7, 8). Flowers were hand pollinated at full anthesis and tagged. The stage of fruit development is expressed as percentage of time from anthesis, where 100% of development is taken as the time elapsing from anthesis to the day when the first red color appears in fruits of a given population (7).

Table I. *Effect of NaCl in the Nutrient Solution on Time from Full Anthesis to the First Appearance of Color in Tomato Fruits*

Cultivar	Time from Full Anthesis to Appearance of First Color		NaCl
	Control	NaCl (3 g/l)	
	d		*% of control*
228	46 ± 0.36[a]	44 ± 0.57	96
202.206	49 ± 0.41	45 ± 0.44	92
364.365	59 ± 0.80	50 ± 0.75	85

[a] Value ± SE.

Table II. *Effect of NaCl in the Nutrient Solution on the Relative Taste of Tomato Fruits*

The variety used in this test was S5. The two tests (A and B) were run separately, and should not be compared.

Test	Taste Score[a]			Preference[b]		
	Control	NaCl (3 g/l)	NaCl (6 g/l)	−	0	+
		mean				
A. Vine-ripened	1.69[c]	2.57[d]	2.23[d]	3	4	19
B. Room-ripened (9 d)	1.37[c]	2.41[d]	2.41[d]	0	3	22

[a] Taste scores were defined as follows: 3, best; 2, medium; 1, worst. A score of 1 means that the tomato is of inferior taste to the others, but it can still be a tasty tomato.

[b] The values in the preference test table represent the number of tasters who preferred the control (−), who found no difference between the fruits (0), or who preferred the fruits of the saline-treated plants (+).

[c,d] Values having the same letter (c or d) are not significantly different.

Salinity Treatment. Either 3 or 6 g/l of salt were added to the nutrient solution upon the appearance of the first flower. The electrical conductivities of the control Hoagland solution and the salt-supplemented nutrient medium were 1.5 and 6.6 mmho[2], respectively.

Fruit Size, Gas Exchange, and Pigments. Since tomato fruits reach their maximal size at the beginning of the ripening stage, fully ripened fruits may be considered to be full sized; thus, the fruits were weighed when completely ripe. Evolution rates of CO_2 and ethylene were measured as described (11). Pigments were analyzed according to the following procedure: pericarp discs (11 mm in diameter) were extracted with acetone:hexane (4:5, v/v), and the pigment profile was obtained by a Unicam Sp 800 spectophotometer (1).

Activity of Pectinases. The enzymes were extracted from the

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[2] Abbreviations: PME, pectin methylesterase; PG, polygalacturonase; PMG, polymethylgalacturonase; TSS, total soluble solids; mmho, millimho.

Table III. *Effect of NaCl in Nutrient Solution on Chemical Composition of Tomato Fruits at Various Stages of Ripening*

The fruits were harvested and analyzed at the indicated d after 100% of development.

Cultivar	Time after 100% of Development	Total Acidity		Reducing Sugars		TSS		pH		Electrical Conductivity		Cl^-	
		Control	NaCl (3 g/l)	Control	NaCl (3 g/l)	Control	NaCl (3 g/l)	Control	NaCl (3 g/l)	Control	NaCl (3 g/l)	Control	NaCl (3 g/l)
	d	*meq/g fresh wt*		*mg glucose per g fresh wt*		*%*				*mmho/cm*		*μmol/g fresh wt*	
228	7	0.053	0.131	23.3	31.7	4.78	6.70	4.27	4.13	4.62	10.10	1.2	27.3
	14	0.044	0.094	17.7	26.5	3.80	5.36	4.43	4.18	4.52	8.00	1.4	23.6
	30	0.035	0.080	19.7	30.4	3.53	6.09	4.51	4.32	4.96	10.77	1.8	27.2
202.206	7	0.069	0.120	27.7	32.9	3.90	5.94	4.10	4.01	4.99	6.23	9.3	24.9
	14	0.058	0.130	28.0	32.8	4.75	6.20	4.26	4.08	4.93	6.64	8.3	28.4
	30	0.049	0.065	28.9	32.0	3.68	6.00	4.41	4.30	4.86	9.42	3.4	31.8
364.365	7	0.062	0.121	24.6	27.3	3.85	5.73	4.33	4.07	6.10	10.49	2.0	13.5
	14	0.061	0.125	23.3	26.2	3.78	5.50	4.28	4.06	6.40	7.93	3.5	16.4

Table IV. *Effect of NaCl in the Nutrient Solution on Fruit Fresh and Dry Weight*

	Cultivar	Control		NaCl, 3 g/l		NaCl, 6 g/l	
		Fruit Weight	Control	Fruit Weight	Control	Fruit Weight	Control
		g ± SE	*%*	*g ± SE*	*%*	*g ± SE*	*%*
Fresh weight							
	228	154.5 ± 15.0	100	125.0 ± 10.4	81.0	99.2 ± 10.4	64.2
	Rutgers	164.8 ± 14.0	100	153.8 ± 13.0	93.3	120.2 ± 7.6	72.9
	202.206	95.0 ± 5.2	100	63.4 ± 5.0	66.7	41.8 ± 2.8	44.0
Percentage dry weight							
	228	5.55 ± 0.78	100	6.67 ± 0.30	120		
	364.365	6.60 ± 0.10	100	7.76 ± 0.30	118		

pericarp of frozen (−20°C) fruits which had been harvested at various stages of development. Forty grams of frozen tissue were diced and homogenized for 5 min in 120 ml 1.0 N NaCl. The homogenate was squeezed through eight layers of cheesecloth and centrifuged for 10 min at 600*g* (Sorval, SS-34 rotor). Part of the supernatant served as the crude extract of PME (E.C. 3.2.1.11), and the remainder was dialyzed against water at 1°C for 24 h. The dialyzed extract contained the crude PG (E.C. 3.2.1.15) and the PMG enzyme fractions (14). PME activity was measured as described by Zauberman and Schifman-Nadel (20). PG and PMG activities were measured as described by Mizrahi *et al.* (10), with polygalacturonic acid (Sigma, catalog no. P-1879) and pectin (Sigma, catalog no. P-2135) as substrates, respectively.

Fruit Quality. Organoleptic evaluation was performed as described previously (9). The preference test was performed as follows. Each taster was requested to taste two samples, one consisting of fruit slices from control plants and the other consisting of slices from salt-treated plants (3 g NaCl per L). The taster was then asked to indicate which of the samples he preferred. Preference for fruits of salt-treated plants was designated by a plus sign, for control fruits by a minus sign, and for neither by a zero. Titratable acidity, TSS, and reducing sugars were also measured as already described (10).

Firmness Measurements. Measurements of firmness were performed with a Hamson firmness meter, as described previously (2, 6).

Measurements of Dry Matter. Frozen fruits (−20°C) were cut into small pieces (1 cm^3), lyophilized, and weighed.

Chemical Analyses. For chemical analyses, 10 g of tissue were homogenized with 5 ml of water in a VirTis homogenizer. The homogenate was centrifuged in a Sorval centrifuge (10,000 rpm for 10 min in an SS-34 rotor), and the analyses (pH, electrical conductivity, total acidity, TSS, reducing sugars, Na^+, and Cl^-) were performed on the supernatant solution. Cl^- ions were measured in a Büchler-Cotlove automatic titrator and chloridometer, and Na^+ ions were measured with a flame photometer.

Covariance Analysis. Covariance analysis was performed by regression analyses of the various chemical constituents of the fruit, using weight and time as quantitative explanatory variables and cultivars and treatment as qualitative (dummy) explanatory variables.

RESULTS

The different cultivars yielded similar results for the various parameters measured. Representative results are given in the Tables and Figures.

Duration of Fruit Development and Taste. NaCl treatment applied to the whole plants shortened the time between full anthesis and the initiation of the ripening process by 4 to 15% (Table I). Fruits which developed on the saline-treated plants tasted better than did fruits of the control plants. This was true for vine-ripened fruits as well as for fruits harvested at 100% of development and allowed to ripen for 9 d at 20°C (Table II). A preference test showed this difference in taste to be highly significant.

Chemical Composition and Fresh and Dry Weight. Salt-treated plants bore fruits characterized by increased values of TSS, reducing sugars, and total acidity, as well as of the electrical conductivity and Cl^- concentration (Table III). The pH was, on the other hand, lowered (Table III). The concentration of Na^+ was measured in later experiments, and it proved to be equivalent to that of the chloride ion. The fresh weight of the fruits of saline-treated plants was lower than that of the fruits of control plants

Table V. *Regression Coefficient of the Various Chemical Components of Tomato Fruits According to Cultivar, Salinity Treatment, Fresh Weight, and Time after the Initiation of Ripening*

t Values over 2.021 and 2.7 are significantly different at 0.05 and 0.01 levels respectively. **, Effects which are significant at the 1% level. NS, Effects which are not significant at the 5% level. Intercept (a) represents the value of the parameters for the All-round cultivar exposed to NaCl treatment. The cultivar effect and the control-*versus*-treatment effect represent deviations from the intercept. b, Regression coefficient; R^2, coefficient of determination, is the square of the multiple correlation coefficient, and it is the proportion of the total variance accounted for in the relevant regression model.

Explanatory Variables	Dependent Variable					
	Total acidity		Reducing sugars		TSS	
	b	*t*	b	*t*	b	*t*
Cultivar 228	0.023	2.77**	−16.75	−14.5**	1.306	3.36**
Cultivar 202.206	0.037	5.76**	−13.18	−12.5**	1.32	4.42**
Control *versus* treatment	−0.052	−9.13**	− 3.97	− 5.1**	1.48	−5.55**
Fruit fresh weight (g)	−0.052	-0.56^{NS}	− 0.53	-0.03^{NS}	−4.23	-0.96^{NS}
Time after 100% development	−0.00027	-0.44^{NS}	0.081	0.8^{NS}	−0.004	0.14^{NS}
Intercept[a]	0.089	12.03**	29.38	28.1**	4.84	13.90**
R^2	80		86		66	

Table VI. *Effect of NaCl in the Nutrient Solution on the Pigment Concentration in the Tomato Pericarp*

Values in the Table are o.d. units ± SE at 505 nm of an extract from 1 g pericarp dissolved in 15 ml acetone:hexane (5:4 v/v) mixture. Fruits were sampled 5 d after 100% of development.

Cultivar	Pigment Content		
	Control	NaCl, 3 g/l	NaCl, 6 g/l
	o.d. units ± SE		
228	0.80 ± 0.07	1.41 ± 0.10	1.88 ± 0.11
Rutgers	0.68 ± 0.09	1.34 ± 0.09	1.84 ± 0.06

Table VII. *Effect of NaCl in the Nutrient Solution on Shelf Life of Tomato Fruit*

Fruits were harvested at the first appearance of color and were kept at 20°C and 85 to 95% RH until they reached 3 scale unit on a Hamson firmness meter.

Cultivar	Shelf Life		
	Control	NaCl, 3 g/l	NaCl, 6 g/l
	d ± SE		
228	17 ± 0.5	16 ± 0.8	12 ± 1.2
Rutgers	10 ± 1.1	9 ± 0.2	7 ± 0.7

Table VIII. *Effect of NaCl in the Nutrient Solution on PME Activity in Fruits of Tomato Cultivar 228 at Various Stages of Ripening*

Time after 100% Development	PME Activity		Increase
	Control	NaCl, 3 g/l	
d	*meq CH_3 released per g fresh wt/h*		%
7	0.509 ± 0.068	0.835 ± 0.076	64
14	0.640 ± 0.080	0.770 ± 0.040	20
30	1.020 ± 0.052	1.960 ± 0.140	92

(Table IV, A), but the percentage of dry matter was higher (Table IV, B).

To investigate the extent to which the saline effect on these parameters stems from its effect on fruit size *per se*, covariance analysis was performed on parallel data of fresh weight, total acidity, reducing sugars, and TSS recorded in individual fruits harvested at different times after 100% of development from plants from of three cultivars exposed to 3 g/l NaCl as compared with fruits nontreated plants (Table V). The actual results were similar to those obtained in Tables III and IV. Covariance analysis was performed by regression analyses of the various chemical constituents of the fruit, using weight, time after initiation of ripening, cultivars, and salinity treatment as explanatory variables. Table V clearly demonstrates that the most significant explanatory variable was the salinity treatment, while the effect of fruit weight was not significant.

Table IX. *Effect of NaCl in the Nutrient Solution on PG and PMG Activity in Tomato Cultivar 228*

Time after 100% Development	Substrate	Activity		Increase
		Control	NaCl, 3 g/l	
d		*μmol reducing sugars released per g fresh wt per h*		%
7	Na-polypectate	9.13 ± 1.8	11.41 ± 1.60	24.9
14	Pectin	13.53 ± 3.5	19.72 ± 0.94	45.8
30	Na-polypectate	14.74 ± 2.42	31.30 ± 4.78	112.0

Pigments. Fruits of the salt-exposed plants were redder than were those of the control plants, as confirmed by pigment concentration measurements (Table VI). This effect of salinity was intensified when the NaCl content of the root medium was increased from 3 to 6 g/l.

Shelf Life. Shelf life is a parameter of commerical importance. For the estimation of shelf life, fruits were harvested at 100% of development and kept at 20°C and 85 to 95% RH until they reached three scale units on a Hamson firmness meter (these fruits were too soft to be sold). It can be seen from Table VII that 6 g/l NaCl shortened the shelf life considerably.

Activity of Pectolytic Enzymes. Inasmuch as it has been proposed that shelf life is a function of the activities of the enzymes that degrade the cell walls, especially pectin (3, 4), we measured PME, PG, and PMG activity at various stages of ripening (Tables VIII and IX). Salinity applied to the plants was found to increase PME activity in fruits at all stages of ripening. The activity of PG and PMG, which were later proved to be the same enzyme (unpublished data), was determined by two techniques—reduction of substrate viscosity (Fig. 1) and increase of the reducing sugar groups released from the polymer (Table IX). Activities were

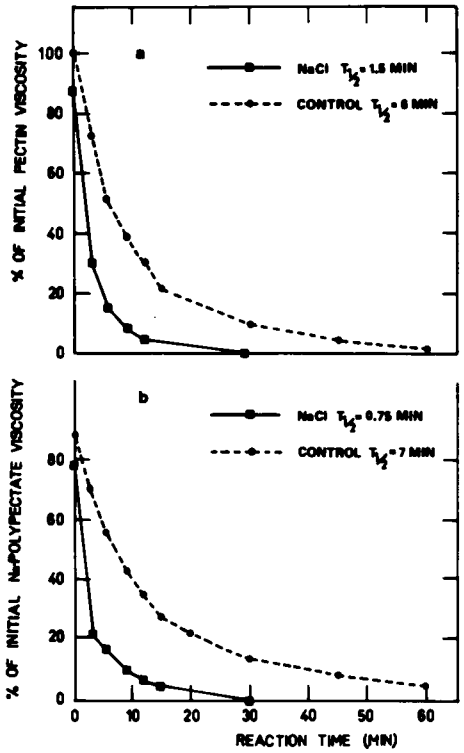

FIG. 1. Effect of NaCl (3 g/l) in the nutrient solution of PMG (top) and PG (bottom) in fruits of tomato cultivar 228, as measured by a viscosity test. $t_{1/2}$ Values are defined as the time required to reach 50% of the initial substrate viscosity.

found by both methods to be higher in fruits of salt-treated plants, as compared with activities in fruits of the control plants.

Ethylene and CO_2 Evolution. Ethylene and CO_2 evolution rates were measured in fruits harvested at 90% of development (Fig. 2). It can be seen that more ethylene and more CO_2 evolved from fruits harvested from the salt-treated plants than evolved from the control fruits.

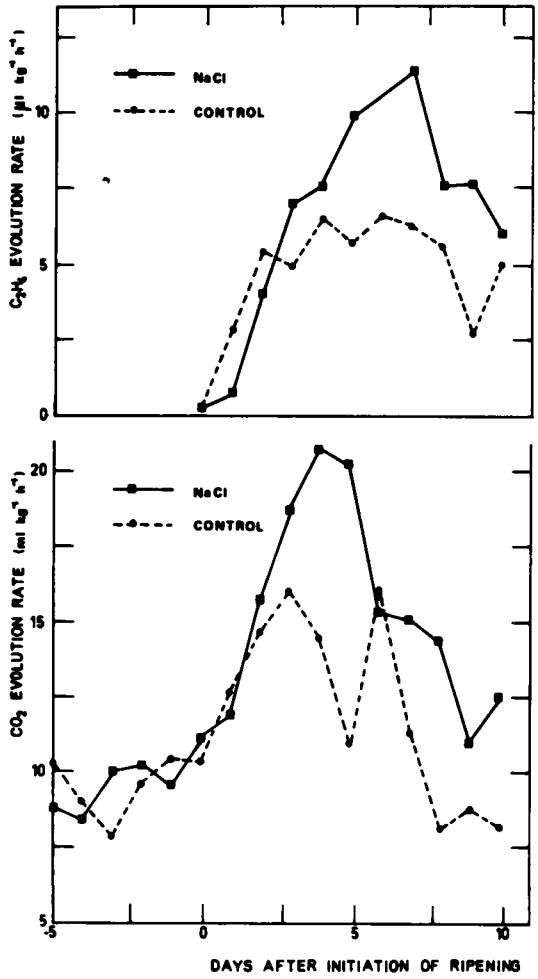

FIG. 2. Effect of NaCl (3 g/l) in the nutrient solution on ethylene and CO_2 evolution rates in fruits of tomato, cultivar All-round, harvested at 90% of development. Day 0 is the first d during which a significant increase in ethylene occurred, which indicates the initiation of the ripening process. All of the measurements were done at constant temperature of 20 ± 1°C.

DISCUSSION

The life span of the tomato fruit may be divided into two stages: (a) the developmental stage, during which the fruits increase in size; and (b) the ripening stage, which starts at the end of stage 1 (7) and terminates in tissue breakdown. Salt treatment accelerated both of these stages (Tables I and VII), and, hence, shortened the fruit life span. This effect can be considered a mechanism of adaptation to stress conditions, which enables the plant to disperse its seeds faster when exposed to stress. These results are similar to those obtained with guava fruit (19).

All the parameters of the fruit ripening process were found to be increased in the fruits of salinity-exposed plants (Tables II–IX; Figs. 1 and 2). The fruit pH was, however, slightly decreased in accordance with the increase in total acidity.

Some of the ripening parameters fluctuated during the ripening stage. Thus, quantitative comparison of fruits of salt-treated plants with control plants on the basis of their chronological age can lead to misinterpretation of the results. To solve this problem, we compared the results at various time intervals after the initiation of ripening and showed that salinity always enhanced the ripening parameters (Tables III, VI, VIII, and IX; Fig. 2). This enhancement means that, under conditions of salinity, the ripening process is more intensive.

It may be assumed that the enhancement of the ripening process stems from the effect of salinity on the reduction of the fruit size, especially in cases in which the percentage of dry weight was increased. However, Table V demonstrates that all these parameters measured were not significantly dependent on fruit fresh weight, while being significantly dependent on the salinity treatment. In support of this conclusion is the fact that the fruit protein content in both treatments was similar (data not shown) (11).

From an applied point of view, the results show that the quality—in terms of chemical constituents (mainly sugars and acids), pigments, and especially taste—of fruits from saline-treated plants is superior to that of those from control plants. The improved taste under salinity may stem from salinity-increased acids and sugars and/or from an increase of other flavoring compounds, a question which should be studied further. Thus, there is compensation, in terms of quality, for the reduction in yield (Table IV). Unfortunately, the improvement of quality is accompanied by a reduction of the shelf life (Table VII), probably as a result of the increase in activity of pectolytic enzymes (Fig. 1; Tables VII and IX), which have been suggested to be key enzymes regulating tomato fruit firmness (3, 4).

It is possible that salinity affects all of the ripening parameters independently. However, it is possible that, because salinity shortens the fruit life span and influences all of the ripening parameters, it affects the regulatory site of fruit ripening. The existence of a ripening regulatory site is also supported by genetic evidence. For example, in the *rin* and *nor* tomato mutations, in

which ripening is inhibited, all of the ripening parameters are also affected (17). Almost complete inhibition of fruit ripening occurs when these genes are present in homozygous condition, while partial inhibition is manifested in the heterozygotes (5, 18). When plants in *rin* and *nor* tomato mutants were exposed to salinity, the *nor* fruits demonstrated activation of all the ripening parameters, while the *rin* was not affected (11). The existence of a regulatory site of fruit ripening, which is affected by salinity in the *nor* mutant, is thus demonstrated.

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