Root Growth and Yield of Differing Alfalfa Rooting Populations under Increasing Salinity and Zero Leaching

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ABSTRACT

Accumulation of salinity in the root zone can be detrimental to sustained crop production. Irrigation, even with moderately saline water, pushes accumulated salts deeper into the root zone, allowing roots to proliferate in regions of relatively low salinity. Two alfalfa (*Medicago sativa* L.) subpopulations with low- and high-fibrous rooting characteristics, MnPL-9-LF and MnPL-9-HF, were used to test the effectiveness of increased rooting on yield when plants were irrigated with saline water but without leaching. Treatments were three levels of heterogeneous root zone salinity predicted by the SOWACH model to represent 10, 20, and 30 yr of irrigation with saline water. Plants were grown for five successive harvests in 10-cm-diam., 130-cm-deep cylinders. The treatments were constructed with NaCl and gypsum. As soil became depleted to 50% extractable water, irrigation water with an electrical conductivity (EC) of 2.8 dS m^{-1} was applied. By the fifth harvest, soil solution EC from the top to the bottom of the profile ranged from 3 to 12 dS m^{-1} for the control and from 3 to 23 dS m^{-1} for the highest salinity treatment. Root production of the high-fibrous root type was stimulated more at low and medium salinity than that of the low-fibrous root type. Across salinity treatments, final root length density (cm root length per cm^3 soil volume) was 24% higher for the high-fibrous root type, and herbage yield of the high-fibrous root type was 14% higher than that of the low-fibrous root type. Differential rooting was greatest in the upper half of the root zone. High fibrous rooting in alfalfa is a trait with potential usefulness as a salinity stress avoidance mechanism.

A COMMON PROBLEM in irrigated agriculture is the gradual buildup of salts in the root zone. Periodic leaching with low-saline water can greatly reduce the concentration of soluble salts, but may have undesirable consequences for users of downstream drainage water.

One strategy for reducing downstream impacts of low quality water is to forego leaching and store salt in the lower portion of the root zone. Irrigating in small amounts with increased frequency keeps the water content high and salinity low in the upper root zone. A root zone of heterogeneous salinity develops under irrigation without leaching, and yield reductions occur as salt accumulates first in the lower then the upper root zone, which is particularly salt sensitive (van Schilfgaarde et al., 1974; Jame et al., 1984; Smith, 1993).

For long-term productivity, perennial crops such as alfalfa must be able to adapt to increasing heterogeneous root zone salinity. The root zone of an established stand of alfalfa under irrigation extended to 2.5 m (Dudley et al., 1994). It may be advantageous for deep-rooted crops such as alfalfa to exploit the lower average salinity of the upper root zone preferentially as salinity increases (Minhas and Gupta, 1993).

Selection in the field for root traits in alfalfa under varying environmental conditions has had a positive effect on yield. Typically, nondormant varieties of alfalfa are strongly tap rooted (Smith, 1993), while increased dormancy is associated with greater branching of the tap root and greater fibrous root mass (Barnes et al., 1988). To improve winter survival, Saindon et al. (1991) selected for root yield within two alfalfa cultivars. This resulted in increased root branching and was correlated with higher forage yield. After evaluating the root growth of several genotypes differing in yield, a positive correlation was shown between shoot growth and highly branched root architecture. A larger root system or one that has an architecture better suited for soil resource acquisition was also proposed for improving the yield of beans (Lynch and van Beem, 1993).

To improve nodulation and thus nitrogen fixation, which occurs primarily on fibrous roots, Viands et al. (1981) produced two subpopulations of alfalfa that differed significantly in their rooting characteristics, a low-fibrous (MnPL-9-LF) and high-fibrous (MnPL-9-HF) subpopulation. It was our hypothesis that as salts accumulated in the root zone, the alfalfa subpopulation with greater fibrous rooting would have higher forage yield under increasing heterogeneous salinity, because these plants would generate more root mass in the less-saline regions of the upper root zone. The objective of this study was to compare the forage yield and root growth of subpopulations with different rooting characteristics under a range of heterogeneous salinity levels.

MATERIALS AND METHODS

Construction of Rooting Cylinders

Randomly selected plants from two near-isogenic subpopulations of alfalfa selected for low- (MnPL-9-LF) and high-fibrous (MnPL-9-HF) rooting characteristics (Viands et al., 1981) were grown in cylinders constructed from polyvinylchloride (PVC) pipe. The design of the cylinders was adapted from LeNoble et al. (1996). Each cylinder was 10 cm diam. and 130 cm deep, with a wall thickness of 3 mm. One side was replaced with an 8-cm-wide, flat, 3.2-mm-thick, clear PVC window the length of the cylinder, bonded in place with Weld-On epoxy (Industrial Polychemical, Gardena, CA). Caps made of PVC with an inside diameter of 10 cm were bonded to the

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Abbreviations: EC, electrical conductivity; EC_e, electrical conductivity of saturation extract of soil paste; ESW, extractable soil water; PVC, polyvinylchloride; RLD, root length density; TRI, traced root intensity.

Table 1. Amounts of salts added to soil and initial EC_e of saturated ion extract of soil paste of root zone increments.

Salt species	Depth	Salinity level: Control	Low	Medium	High
	cm	g salt kg^{-1} soil			
NaCl	0–15	0.000	0.000	0.099	0.263
	15–30	0.000	0.000	0.099	0.263
	30–60	0.000	0.272	0.591	0.931
	60–90	0.000	0.611	1.165	1.760
	90–120	0.000	0.871	1.750	2.616
$CaSO_4 \cdot 2H_2O$	0–15	0.000	0.000	0.069	0.168
	15–30	0.000	0.000	0.069	0.168
	30–60	0.000	0.147	0.170	0.195
	60–90	0.000	0.142	0.149	0.144
	90–120	0.000	0.108	0.077	0.045
		dS m^{-1}			
EC_e	0–15	1.25	1.25	2.15	3.30
	15–30	1.25	1.25	2.15	3.30
	30–60	1.25	3.60	5.15	7.55
	60–90	1.25	5.55	8.75	12.05
	90–120	1.25	6.95	10.95	19.00

bottom of cylinders. Before capping the bottom of cylinders and adding soil, holes were drilled in the caps and covered with wire mesh to promote aeration.

Cylinders were packed with a 2.5-cm-deep layer of gravel, then soil of varying salinities (see below) was added in 10-cm-deep increments to a bulk density of approximately 1.25 g cm^{-3}. The soil used was a 2-mm-sieved Kidman fine sandy loam (coarse-loamy, mixed, superactive, mesic Calcic Haploxerolls) from the Ap horizon. The EC was 1.25 dS m^{-1}, determined from saturation extract of the soil paste (Rhoades, 1996). The soil was deficient in phosphorus, so 30.8 mg kg^{-1} P was added to the top 15 cm of soil, and P was applied after each harvest as 100 mL of a solution containing 0.16 m*M* KH_2PO_4 and 0.84 m*M* K_2HPO_4 (pH 7.2). Cylinders were wrapped in aluminum foil to exclude light from the clear window, and placed at 25° from vertical to promote root growth along the soil–window interface (Glinski et al., 1993; LeNoble et al, 1996).

A control and three root zone treatments of increasing salinity were constructed by mixing soil with predetermined amounts of NaCl and gypsum (Table 1) in a cement mixer. The sulfate salt species and watering regimen were chosen to represent irrigation conditions prevalent in the Great Basin and Intermountain West regions. Electrical conductivities for the low, medium, and high salinity treatments were selected on the basis of predictions of crop-water balance and salt accumulation over a 10-, 20-, or 30-yr period of saline irrigation and crop transpiration by the soil water chemistry (SOWACH) model (Dudley and Hanks, 1991).

Experimental Design and Plant Growth Conditions

The factorial experiment was designed as a randomized complete block with three replications, with two alfalfa root type populations and four salinity treatments as the whole plot factors. Repeated harvests or soil depths were analyzed as subplot factors. Minitab (Minitab Inc., 1992) was used for analysis of variance, and significance was determined at $P = 0.05$ unless noted otherwise. Least significance differences (LSDs) between means were calculated where appropriate.

The study was conducted in a greenhouse maintained at 20 ± 5°C (day) and 15 ± 5°C (night) with a 16-h photoperiod. Supplemental lighting was provided by high pressure sodium lamps to an average photosynthetic photon flux of 500 μmol m^{-2} s^{-1}. Cylinder location within blocks in the greenhouse was re-randomized once every 4 wk to minimize the effect of environmental gradients.

Table 2. Final soil solution electrical conductivities (EC_e) of root zone sections determined from saturation extracts of soil pastes.

Root type (population)	Depth	Salinity level: Control	Low	Medium	High
	cm	dS m^{-1}			
Low fibrous (MnPL-9-LF)	0–15	3.00†	3.10	3.00	3.00
	15–30	3.35	3.45	3.72	3.60
	30–60	3.80	4.15	4.10	4.00
	60–90	5.45	7.00	6.75	7.70
	90–120	12.15	16.00	21.50	23.15
High fibrous (MnPL-9-HF)	0–15	2.60	3.30	3.70	3.25
	15–30	3.30	3.80	3.90	4.00
	30–60	3.80	4.30	4.45	4.45
	60–90	4.65	6.85	9.00	8.30
	90–120	11.85	16.50	18.05	20.50

† The LSD 0.10 to compare root types at a given depth and salinity is 1.73.

Alfalfa seeds were treated with the fungicide Apron ([*N*-(2,6-dimethylphenyl)-N-(methoxyacetyl)-alanine methyl ester] Ciba-Geigy Corp., Greensboro, NC) at 2.5 g per kg seed, and inoculated with a commercial alfalfa inoculant (Nitragin, Milwaukee, WI) plus a mixture of four salt-tolerant strains (USDA 1027, 1029, 1030, 1031) of *Sinorhizobium meliloti* (USDA Soybean and Alfalfa Research Laboratory, Beltsville, MD).

Application of Irrigation Water and Forage Harvest

Cylinders were watered to 0.01 MPa (container capacity) at a soil water content of 23.4% with saline irrigation water on 20 Feb. 1995. Irrigation water had an EC of 2.8 dS m^{-1}, and the concentration of salts in the irrigation water was 9.33 m*M* $CaSO_4$, 5.36 m*M* $MgSO_4$, 1.00 m*M* Na_2SO_4, and 5.41 m*M* NaCl. The final electrical conductivity of saturation extract of soil paste (EC_e) of the original medium-salinity treatment was higher in the lowest quarter of the rooting container than the original high-salinity treatment due to inadvertent leaching when cylinders were brought to container capacity. Therefore, with the exception of Table 1, the medium- and high-salinity treatments refer to the actual final salinity of treatments as reported in Table 2.

On 13 March 1995, sufficient tap water (EC 0.38 dS m^{-1}) was added to flush the top 20-mm soil layer of each cylinder and leach residual salinity. Twenty alfalfa seeds were sown directly in the cylinders and germinated without supplemental lighting. The soil was misted until germination, and seedlings were watered with an additional 50 mL tap water 3 wk after planting. Plants were thinned to one per cylinder 3 wk after emergence, and the first saline irrigation was applied 5 wk after emergence. Plant shoots were cut to a height of 10 cm when they reached the late flowering growth stage (Fick and Mueller, 1989), which occurred at 3- to 5-wk intervals, for a total of five harvest periods ending 13 Oct. 1995.

Cylinders were irrigated with the 2.8 dS m^{-1} solution described above when plants depleted 50% extractable soil water (ESW) (Carter and Sheaffer, 1983). Total ESW was calculated from the difference between cylinder mass at container capacity and the mass of the same cylinder containing air-dried soil at the time of packing. Water deficit was restored with a drip from 4-L carboys while rooting cylinders were held vertically. The soil surface was approximately 4 cm below the rim of containers, so no runoff occurred.

Root Measurements at the Soil–Window Interface

Before each harvest, lengths of roots at the soil–PVC window interface within each salinity zone were traced onto acetate sheets using permanent felt-tip pens, with a different color for each harvest (Snapp and Shennan, 1992). The uppermost root zone was further divided into an upper and lower half because fibrous roots are most prolific high in the root zone. The lengths of all traced roots within five sections of the rooting container (0–15, 15–30, 30–60, 60–90, and 90–120 cm deep) were determined by means of digitizing software (Jandel Corp., San Rafael, CA).

Root Measurements in the Bulk Soil

Following the fifth harvest, cylinders were sawn open lengthwise opposite the clear PVC window, and the soil and roots were separated into the four original zones of heterogeneous salinity; the uppermost zone was divided into upper and lower 15-cm increments, as for root tracings. A full cross-sectional subsample 5 cm in height was removed from the center of each section of the root zone for soil analysis. The remaining soil and root mass were separated using a hydropneumatic root washing machine (Gillison's Variety Fabrication Inc., Benzonia, MI) where roots were washed against a 0.5 mm sieve. Recovered roots were stored in 10% (v/v) aqueous isopropanol until they were hand sorted to remove debris. This sorting process left less than 5% debris by length and weight in root samples. Root length from the bulk soil was determined with a root length scanner (Comair, Melbourne, Australia), and root mass was determined after drying roots at 70°C for 48 h. Total root length per volume of soil (cm cm^{-3}) in each section of the rooting container, including roots at the soil–window interface, is reported as root length density (RLD).

Tap root diameter was measured 1 cm below the crown during destructive sampling following the fifth harvest. Nodules visible at the clear PVC window of the cylinders and judged to be active by appearance were counted just before destructive sampling. The final EC_e of soil taken from the center of each of the five root zones (at 7.5, 22.5, 45, 75, and 105 cm) was determined using saturated extracts of soil paste, and mineral composition of the extracts was determined by inductively-coupled plasma spectroscopy by USU Analytical Laboratories.

RESULTS AND DISCUSSION

Change in Soil Solution Salinity with Irrigation

The final EC_e of the soil solution (Table 2) increased with depth and was greatest at 90 to 120 cm. The difference between final and initial EC_e (Table 3) decreased with increasing salinity, as high salinity limited water uptake by roots. In fact, in the medium and high salinity treatments, irrigation with 2.8 dS m^{-1} water decreased salinity below initial levels in sections above 90 cm. There was also a significant interaction of root type with depth for final EC_e. In the medium and high-salinity treatments, more salinity accumulated at 90-120 cm in the low-fibrous root type (Table 2). However, these differences at 90-120 cm appear to be due more to greater leaching from higher root zones of the low-fibrous root type than to differences in overall residual salinity (Table 2). This is supported by data reported later in this section for higher shoot mass, higher root length density, and higher water use of the high-fibrous root type (Table 4).

Table 3. Difference between final and initial soil solution electrical conductivities (EC_e) of individual root zone sections determined from saturation extracts of soil pastes.

Root type (population)	Depth	Salinity level: Control	Low	Medium	High
	cm	dS m^{-1}			
Low fibrous (MnPL-9-LF)	0–15	1.75	1.85	0.85	−0.30
	15–30	2.10	2.20	1.57	0.30
	30–60	2.55	0.55	−1.05	−3.55
	60–90	4.20	1.45	−2.00	−4.35
	90–120	10.90	9.05	10.55	4.15
High fibrous (MnPL-9-HF)	0–15	1.35	2.05	1.55	−0.05
	15–30	2.05	2.55	1.75	0.70
	30–60	2.55	0.70	−0.70	−3.10
	60–90	3.40	1.30	0.25	−3.75
	90–120	10.60	9.55	7.10	1.50

Table 4. Mean number of irrigations prior to each harvest.

Salinity level	Root type (population)	Harvest number: 1	2	3	4	5	Total
		No. Irrigations					
Control	Low fibrous	1.3	3.0	3.8	3.3	2.8	14.2
	High fibrous	1.0	3.0	3.7	3.8	3.3	14.8
Low	Low fibrous	1.3	3.5	3.8	3.3	3.3	15.2
	High fibrous	1.7	3.5	3.8	4.3	4.2	17.5
Medium	Low fibrous	1.7	3.0	3.7	2.8	3.7	14.9
	High fibrous	1.8	3.3	4.0	3.8	4.0	16.9
High	Low fibrous	1.2	3.0	3.3	3.0	2.8	13.3
	High fibrous	1.2	4.2	3.3	3.5	3.2	15.3
	LSD†	NS‡	NS	NS	1.5*	1.3§	

* Indicates significance at $P \leq 0.05$.
† Comparison of root type at a fixed salinity level.
‡ NS, not statistically significant at $P \leq 0.10$.
§ Indicates significance at $P \leq 0.10$.

The composition of salts in the soil solution after five harvests also changed with depth and treatment salinity (Table 5). Sodium chloride values were similar in the upper half of rooting containers, ranging from 3.8 to 6.5 m*M*, but were very high in the lowest section of rooting containers for all salinity levels, ranging from 61 to 177 m*M*. On a molar basis, sodium chloride made up only one-quarter of irrigation water salt, whereas sulfate-based salts accounted for the remainder of the salinity. As in a field study using a similar irrigation water and zero leaching (Dudley et al., 1994), the ratio of Na to Ca increased with depth until sodium salts predominated in the lower half of the rooting zone. In that field study, gypsum crystals were observed in root channels and the increase in Na relative to Ca was attributed to precipitation of gypsum.

The decrease in the difference between the initial and final EC_e and the increase in the Na to Ca ratio with depth is consistent with precipitation of gypsum ($CaSO_4$ $2H_2O$). As an indication of the potential for gypsum precipitation, the salts in the saturation extracts (Table 5) were speciated with the computer program MINTEQA2 (Allison et al., 1991). The saturation indices for gypsum (the ratio of the ion activity product to the solubility constant; see e.g., Stumm and Morgan, 1981, p. 236) were slightly greater than one in the 60-90 and 90-120 depth segments (data not shown), indicating that gypsum precipitated in the columns. The values of the saturation index are expected to be slightly greater than 1.0 if gypsum is present because complexes of Ca with

Table 5. Mineral compositions calculated from saturation extracts of soil pastes after five alfalfa harvests.

Salinity level	Root type (population)	Depth	Salt species				Na:Ca
			$NaCl$	Na_2SO_4	$MgSO_4$	$CaSO_4$	
		cm	mM				
Control	Low fibrous (MnPL-9-LF)	0–15	4.9	1.1	5.8	7.8	0.69
		15–30	6.3	1.3	6.1	8.0	0.82
		30–60	5.0	2.8	8.0	10.8	0.79
		60–90	9.1	4.9	11.3	9.7	1.50
		90–120	73.8	2.3	18.6	14.2	5.52
	High fibrous (MnPL-9-HF)	0–15	4.0	1.5	4.6	5.7	0.87
		15–30	6.3	1.3	6.1	8.0	0.82
		30–60	5.0	2.8	8.0	10.9	0.79
		60–90	10.3	6.9	12.3	10.3	2.05
		90–120	60.8	2.8	26.5	13.1	5.09
Low salt	Low fibrous (MnPL-9-LF)	0–15	4.9	1.1	5.8	7.8	0.69
		15–30	4.0	1.9	6.5	10.0	0.63
		30–60	5.0	2.8	8.0	10.9	0.79
		60–90	23.4	1.8	16.5	12.6	2.13
		90–120	110.9	1.0	21.4	18.6	5.42
	High fibrous (MnPL-9-HF)	0–15	4.0	1.9	6.5	12.2	0.63
		15–30	4.2	2.6	7.3	14.7	0.63
		30–60	5.3	4.1	9.7	14.8	0.92
		60–90	23.4	1.8	16.5	12.6	2.13
		90–120	95.4	1.0	17.9	20.8	4.68
Medium salt	Low fibrous (MnPL-9-LF)	0–15	4.9	1.1	5.8	10.2	0.69
		15–30	4.2	2.6	7.3	14.7	0.63
		30–60	5.0	2.8	8.0	13.3	0.79
		60–90	23.4	1.8	16.5	12.6	2.13
		90–120	177.4	1.0	18.6	22.7	7.89
	High fibrous (MnPL-9-HF)	0–15	3.8	2.3	7.3	10.9	0.58
		15–30	5.0	2.8	8.0	10.9	0.79
		30–60	6.5	3.4	8.9	9.7	0.97
		60–90	32.8	1.0	23.6	12.2	2.86
		90–120	101.5	11.0	11.7	8.8	7.07
High salt	Low fibrous (MnPL-9-LF)	0–15	4.9	1.1	5.8	7.8	0.69
		15–30	3.8	2.3	7.3	10.9	0.58
		30–60	5.0	2.8	8.0	10.9	0.79
		60–90	26.3	1.0	17.3	13.1	2.16
		90–120	150.8	0.0	15.6	17.3	7.94
	High fibrous (MnPL-9-HF)	0–15	6.3	1.3	6.1	7.8	0.82
		15–30	5.0	2.8	8.0	10.9	0.79
		30–60	6.5	3.4	8.9	9.7	0.97
		60–90	30.5	3.5	20.2	11.5	3.26
		90–120	115.0	0.0	14.0	13.0	6.40

carbonates and soluble organic matter were not included in the speciation computations.

Calcium is essential to plant cell ion regulation, and when present in saline irrigation water, helps to maintain both soil structure and plant metabolism (Marschner, 1995). Calcium helps plants exclude salts by lowering cell permeability to sodium and by enhancing the activity of the sodium pump in the cell membrane (Rengasamy, 1987). However, when sodium concentrations become sufficiently high, roots cannot persist.

Forage Shoot DM Production

Across salinity levels and summed for harvests, shoot DM (Table 6) of the high-fibrous root population was 14% greater than that of the low-fibrous root type. There was a significant root type-by-harvest interaction for shoot DM, with the high-fibrous root type having higher yield for the three salinity treatments for Harvests 4 and 5 (Table 6). At Harvest 5, mean shoot DM of the high-fibrous root population was 29% higher than that of the low-fibrous population.

Forage shoot DM production in alfalfa has been related to the mean EC_e of the root zone in containers, but mean salinities were higher in those studies (Shalhevet and Bernstein, 1968; Ingvalson et al., 1976; Maas and Hoffman, 1977). Shalhevet and Bernstein (1968) established alfalfa in 50-cm-deep containers and increasingly salinized the upper and lower portions of the root zone independently. Yield was negatively correlated with the average salinity of the two zones, which ranged from 1 to 18 dS m^{-1}. Others have demonstrated that the upper portion of the root zone is the most salt-sensitive (Bingham and Garber, 1970; Francois, 1981; Jame et al., 1984). Francois (1981) reported an 80% yield reduction at an EC of 8.4 dS m^{-1} in the upper 30 cm when the predominant salts were NaCl and $CaCl_2$.

Although yield and average EC_e of the upper 30 cm of the rooting container were positively correlated in the present study ($r = 0.57$, $P < 0.05$), it is likely that the EC_e, which was maintained between 3 and 4 dS m^{-1} in this region (Table 2), was not high enough to negatively impact yields. According to Maas (1986), alfalfa is not negatively affected by sodium and calcium salts at an EC of 4 dS m^{-1}. Similarly, Mehanni and Rengasamy (1990) found that yield of alfalfa grown in saline soils with NaCl and gypsum with an average EC

Table 6. Shoot dry mass at each harvest for the low and high fibrous root types at four salinity levels. Stems were harvested at 10 cm above the soil surface.

Root type (population)	Harvest	Salinity level				
		Control	Low	Medium	High	Mean
		g plant^{-1}				
Low fibrous	1	6.1	5.7	7.0	6.2	6.2†
(MnPl-9-LF)	2	9.5	9.2	7.0	8.1	8.5
	3	9.5	9.3	8.4	9.4	9.2
	4	9.2	8.2	6.7	7.8	8.0
	5	7.8	7.9	8.4	7.1	7.8
	Cumulative	42.1	40.3	37.5	38.6	
High fibrous	1	5.3	6.3	5.9	5.9	5.9
(MnPl-9-HF)	2	8.3	9.9	9.2	8.0	8.9
	3	9.3	10.3	10.3	8.9	9.7
	4	9.9	11.2	11.4	9.9	10.6
	5	8.5	11.4	10.7	9.8	10.1
	Cumulative	41.4	49.1	47.5	42.5	

† The LSD 0.05 to compare the root types at a given harvest is 2.1.

of 4 to 5 dS m^{-1} in the top 15 cm of soil was comparable to yields under nonsaline conditions.

Both the predominance of sulfate salts in this study and the watering regimen may have contributed to salt tolerance and maintenance of yields as soil salinity increased. MacAdam et al. (1997) found that leaf area of 12-wk-old alfalfa plants watered with a 646 mg L^{-1} sulfate solution was higher than that of alfalfa watered with either 175 mg L^{-1} or 862 mg L^{-1} sulfate solutions. Similarly, Anand et al. (2000) found that two alfalfa genotypes irrigated with 4.0 dS m^{-1} water that contained both Cl^- and $SO^{2-}{}_4$ salts had higher net photosynthesis than plants irrigated with tap water.

Carter and Sheaffer (1983) determined that alfalfa growing on coarse-textured soils produced shoot DM equal to that of well-watered plants if it was watered at 50% depletion of ESW, but yield suffered at lower ESW levels. Therefore, plants were watered upon depletion of ESW to 50% to avoid confounding salinity effects with drought stress. This may explain why our results differ from those of studies in which yield differences were attributed to salinity stress at relatively low EC.

Greenhouse conditions can also ameliorate the effect of salinity upon yield, and higher salt tolerance has been achieved in greenhouse studies than in field studies (Chang, 1960; Bernstein and Francois, 1973). This may be due to the higher relative humidity or better temperature control in a greenhouse in the summer that reduces water stress and evapotranspiration needs.

Root Growth

Root Length at the Soil–Window Interface

Root growth that occurred at the soil–growth cylinder window interface from harvest to harvest is reported as cumulative traced root intensity (TRI) (Fig. 1). In earlier experiments utilizing slant tube methodology, TRI has proven useful for in situ observations of root morphology (Rutherford and Curran, 1981; McMichael et al., 1992), but TRI has not been as reliable for relative quantification of root growth as more direct methods. Cumulative TRI increased with depth to 90 cm then decreased in the lowest section of the rooting container, where the accumulation of salts, particularly sodium, was greatest (Table 5).

Root Length in Bulk Soil

There was a significant difference between the RLD (cm root length per cm^3 soil; Fig. 2) of the low- and high-fibrous root types across salinity treatments. Compared to the control, salinity stimulated root extension, particularly in the upper 30 cm of the high-fibrous root population. The low-fibrous root type responded to salinity with increased root extension only at the highest salinity level.

Root Length Densities: Bulk Soil vs. Soil–Window Interface

We investigated the difference in RLD (Fig. 2) and cumulative TRI (Fig. 1) with depth and salinity treatment (Fig. 3). The proportion of roots at the PVC window was calculated by dividing the traced root density (cumulative TRI divided by the 0.3 cm viewing depth; Glinski et al., 1993) by the RLD determined from the bulk soil only.

Only 40 to 75% of the actual RLD in the upper 30 cm of the soil was represented by TRI, whereas in the lower 90 to 120 cm, roots in the medium and high salinity treatments were over-represented by 35 to 45% (Fig. 3). This salinity-by-depth interaction was significant, but there was no significant effect of root type on the percentage of roots at the window, so TRI did not discriminate between root morphologies when used to quantify root growth.

Others have also demonstrated from data obtained from similar observation windows that bulk soil rooting was underestimated. Utilizing slant tubes made of clear polyethylene to study the roots of ‘Penncross’ creeping bentgrass [*Agrostis stolonifera* L. var. *palustris* (Huds.) Farw.], Glinski et al. (1993) found, as we did, that TRI tended to increase with depth relative to actual RLD, and concluded this occurred because the plant growth angle of 20 to 25° degrees from vertical forced roots to grow into a smaller volume with depth.

The more saline the soil environment, the more roots

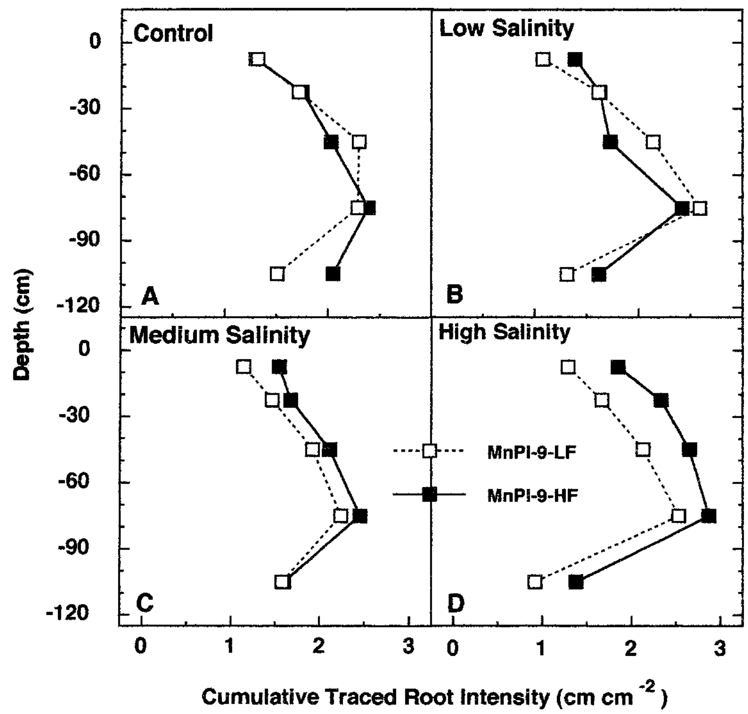

Fig. 1. Cumulative traced root intensity (TRI) at the soil–window interface determined at each of five harvests. New root growth at the soil–window interface was traced at the time of each harvest, and data were summed for this figure. Data were significant at $P < 0.10$, and the LSD to compare root types is 0.19.

tended to grow along the PVC root observation window compared with the bulk soil. This could indicate that some advantage was conferred by proximity to the window. Even though irrigation was applied with cylinders held vertically, water likely permeated the soil toward the window when cylinders were placed back at 25° from vertical. This would raise the soil water potential at the soil–window interface, thereby easing water extraction. There may also have been an attraction of roots to the clear PVC of the window, such as the attraction

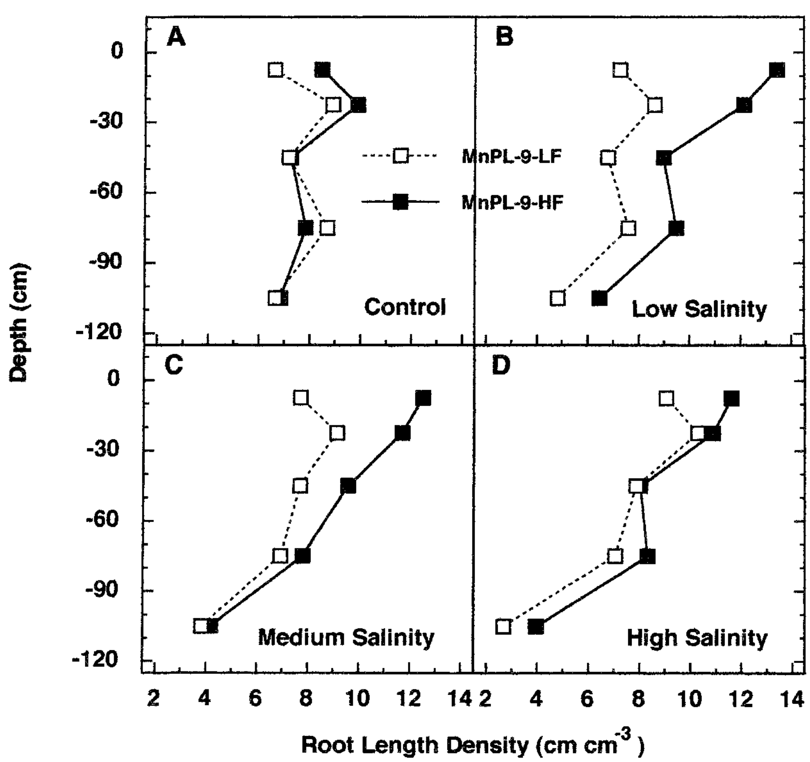

Fig. 2. Root length density (RLD) in bulk soil plus roots at the soil–window interface determined at destructive harvesting following five growth periods and harvests. The LSD0.05 to compare root types is 1.46.

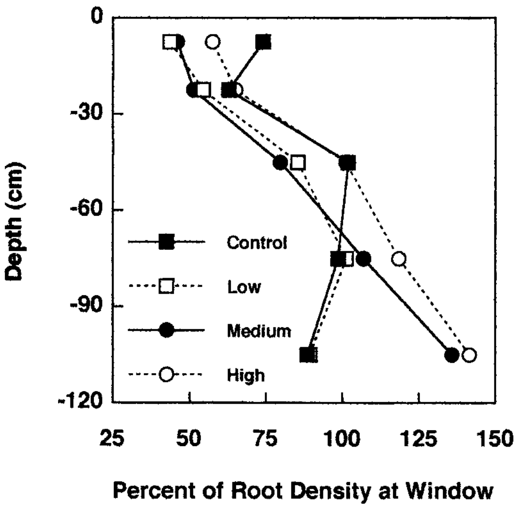

Fig. 3. Percentage of roots concentrated at the soil–window interface following five harvests, the quotient of cumulative traced root density (TRI divided by the 0.3-cm viewing depth) and root length density in the bulk soil. The LSD0.05 to compare salinity treatments at a given root depth is 22.

Voorhees (1976) hypothesized for roots to the Plexiglas window in his study.

Specific Root Dry Mass

Although RLD of the two root types was significantly different, specific root DM (g root per cm^3 soil volume) recovered from the soil did not differ significantly for the two root types (Fig. 4). Similarly, Snapp and Shennan (1992) found that root DM did not differ in response to salinity. Since fibrous roots increased significantly with salinity (Fig. 2) and tap root diameter did not decrease (see below) our data suggest the high-fibrous population produced longer, thinner fibrous roots in response to increasing salinity.

Tap Root Diameter

Tap root diameter of the high-fibrous root type (1.20 cm) was significantly greater than for the low-fibrous root type (1.07 cm) across salinity treatments (LSD 0.05 = 0.09). In the field, Barnes et al. (1988) associated a larger diameter tap or primary root with a high-fibrous root system, while a smaller root is indicative of a low-fibrous or tap root-dominated root system. Our findings in the greenhouse are in agreement with those from the earlier field study. This trait was not influenced by salinity treatment, and demonstrates that the expected characteristics of the two root types were displayed in this study under greenhouse conditions.

Nodulation

The original objective in selecting alfalfa for high-fibrous rooting was to enhance nitrogen fixation ability (Viands et al., 1981). The number of nodules in the field in that study was positively correlated (r^2 = 0.61) with fibrous root score. In our study, however, the number of nodules visible and active at the PVC window did not differ for the two root types, but was stimulated by salinity from a low of 22 for the control to a high of 52 for the high salinity treatment (LSD0.05 = 19). Since root length data taken at the PVC window tended to underestimate roots in the upper half of the root zone

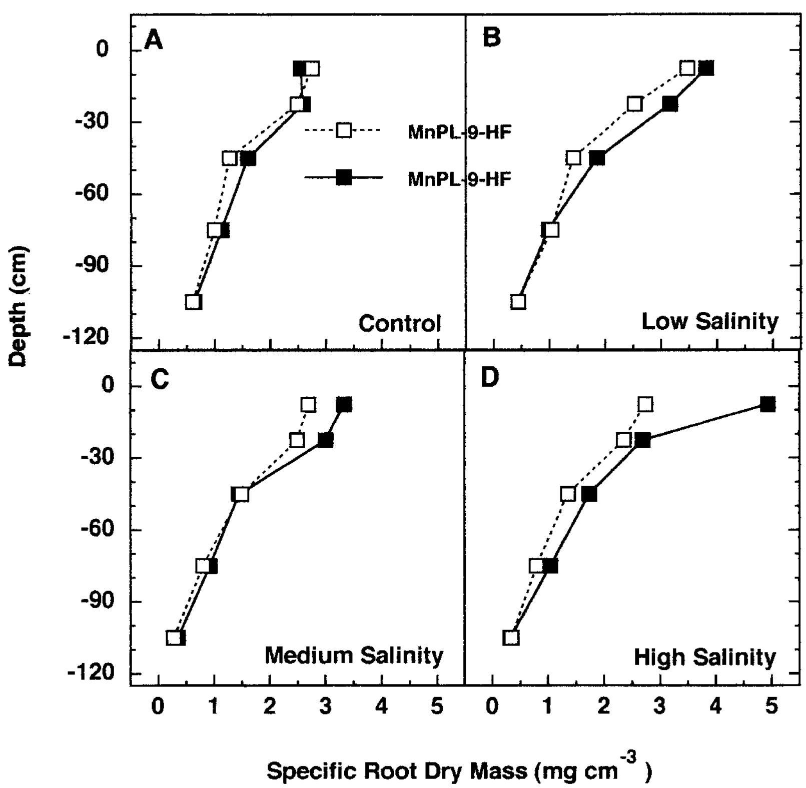

Fig. 4. Specific root dry mass following five harvests. Root types were not significantly different.

(Fig. 3) where root length density was also concentrated (Fig. 2), observations at the window may also have underestimated nodule number. The inoculation of all alfalfa seed with a mixture of four salt-tolerant strains of *Sinorhizobium meliloti* likely helped plants adapt to the saline environment, as nitrogen is often limiting in saline conditions (Khan et al., 1994). The ability to retain or especially to increase nodulation with increasing salinity would be critical to the maintenance of yield under long-term saline irrigation.

CONCLUSIONS

This study was designed to determine the response of alfalfa populations differing in fibrous root growth to the salinity predicted to accumulate over 10, 20, or 30 yr of irrigation with saline water and without leaching. Root production in both populations (Fig. 2) was stimulated by salinity, but the high-fibrous population was significantly more responsive at low and medium salinity than the low-fibrous population. Shoot DM production of the high-fibrous root type was also stimulated by salinity (Table 6), suggesting that the yield of alfalfa irrigated with water enriched in sulfate salts could be sustained for many years in varieties with the high-fibrous rooting characteristic, even in the absence of leaching of the root zone, because of its ability to increase root production in the less saline regions of the upper root zone. Field experiments should be conducted to test this promising finding.

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REFERENCES

Allison, J.D., D.S. Brown, and K.J. Novo-Gradac. 1991. MINTEQA2/PRODEFA2, a geochemical assessment model for environmental systems: Version 3 user's manual. EPA/6003-91/021, USEPA, Athens, GA.

Anand, A., M.J. Baig, and P.K. Mandal. 2000. Response of alfalfa genotypes to saline water irrigation. Biol. Plant 43:455–457.

Barnes, D.K., N.R. Degenhart, D.M. Smith, and R.N. Sahi. 1988. Root morphology of alfalfa plant introductions and North American cultivars. p. 6 *In* Report of the 31st North American Alfalfa Improvement Conf., Beltsville, MD.

Bernstein, L., and L.E. Francois. 1973. Leaching requirement studies: Sensitivity of alfalfa to salinity of irrigation and drainage water. Soil Sci. Soc. Am. Proc. 37:931–943.

Bingham, F.T., and M.J. Garber. 1970. Zonal salinization of the root system with NaCl and boron in relation to growth and water uptake of corn plants. Soil Sci. Soc. Am. Proc. 34:123–126.

Carter, P.R., and C.C. Sheaffer. 1983. Alfalfa response to soil water deficits. I. Growth, forage quality, yield, water use, and water use efficiency. Crop Sci. 23:669–675.

Chang, C.W. 1961. Effects of saline irrigation water and exchangeable sodium on soil properties and growth of alfalfa. Soil Sci. 91:29–37.

Dudley, L.M., and R.J. Hanks. 1991. Model SOWACH, soil-plant-atmosphere-salinity management model user's manual. 1991 Report. Utah Agric. Exp. Stn. Res. Rep. 140.

Dudley, L.M., R.J. Hanks, J.W. MacAdam, R.W. Mace, and A.P. Low. 1994. Use of saline waste water from electrical power plants for irrigation. 1993 Report. Part 1. Soil, water and crop yields. Utah Agric. Exp. Stn. Res. Rep. 150.

Fick, G.W., and S.C. Mueller. 1989. Alfalfa: Quality, maturity and mean stage of development. Info. Bull. 217 Cornell Univ., Ithaca, NY.

Francois, L.E. 1981. Alfalfa management under saline conditions with zero leaching. Agron. J. 73:1042–1046.

Glinski, D.S., K.J. Karnok, and R.N. Carrow. 1993. Comparison of reporting methods for root growth data from transparent interface measurements. Crop Sci. 33:310–314.

Ingvalson, R.D., J.D. Rhoades, and A.L. Page. 1976. Correlation of alfalfa yield with various index of salinity. Soil Sci. 122:145–153.

Jame, Y.W., V.O. Biederbeck, W. Nicholaichuk, and H.C. Korven. 1984. Salinity and alfalfa yield under effluent irrigation in southwestern Saskatchewan. Can. J. Plant Sci. 64:323–332.

Khan, M., M. Silberbush, and S.H. Lips. 1994. Physiological studies on salinity and nitrogen interaction in alfalfa. I. Biomass production and root development. J. Plant Nutr. 17:657–668.

LeNoble, M.E., D.G. Blevins, and R.J. Miles. 1996. Prevention of aluminium toxicity with supplemental boron: II. Stimulation of root growth in an acidic, high-aluminium subsoil. Plant Cell Environ. 19:1143–1148.

Lynch, J., and J.J. van Beem. 1993. Growth and architecture of seedling roots of common bean genotypes. Crop Sci. 33:1253–1257.

Maas, E.V. 1986. Salt tolerance of plants. Appl. Agric. Res. 1:12–26.

Maas, E.V., and G.H. Hoffman. 1977. Crop salt tolerance – current assessment. J. Irrig. Drain. Div. Am. Soc. Civ. Eng. 103:115–134.

MacAdam, J.W., D.T. Drost, L.M. Dudley, and N. Soltani. 1997. Shoot growth, plant tissue elemental composition, and soil salinity following irrigation of alfalfa and tall fescue with high-sulfate waters. J. Plant Nutr. 20:1137–1153.

Marschner, H. 1995. Mineral Nutrition of Higher Plants. 2nd ed. Academic Press, London.

McMichael, B.L., D.R. Upchurch, and H.M. Taylor. 1992. Transparent wall techniques for studying root growth and function in soil. J. Plant Nutr. 15:753–762.

Mehanni, A.H., and P. Rengasamy. 1990. Establishment and production of lucerne on a salinised clay soil irrigated with moderately saline groundwater. Aust. J. Exp. Agric. 30:203-207.

Minhas, P.S., and R.K. Gupta. 1993. Conjunctive use of saline and non-saline waters. I. Response of wheat to initial salinity profiles and salinisation patterns. Agric. Water Manage. 23:125–137.

Minitab, Inc. 1992. Minitab reference manual, Release 9. Sowers Printing Co., Lebanon PA.

Rengasamy, P. 1987. Importance of calcium in irrigation with saline-sodic water – a viewpoint. Agric. Water Manage. 12:207–219.

Rhoades, J.D. 1996. Salinity: Electrical conductivity and total dissolved solids, p. 417–435 *In* D.L. Sparks et al. (ed.) Methods of soil analysis. Part 3. SSSA Book Ser. 5. SSSA. Madison, WI.

Rutherford, M.C. and B. Curran. 1981. A root observation chamber for replicated use in a natural plant community. Plant Soil. 63:123–129.

Saindon, G., R. Michaud, and C.A. St-Pierre. 1991. Breeding for root yield in alfalfa. Can. J. Plant Sci. 71:727–735.

Shalhevet, J., and L. Bernstein. 1968. Effects of vertically heterogeneous soil salinity on plant growth and water uptake. Soil Sci. 106: 85–93.

Smith, S.E. 1993. Salinity and the production of alfalfa (*Medicago sativa* L.). p. 431–448. *In* M. Pessarakli (ed.) Handbook of crop stress. Vol. 1. Marcel Dekker, New York.

Snapp, S.S., and C.S. Shennan. 1992. Effects of salinity on root growth and death dynamics of tomato, *Lycopersicon esculentum* Mill. New Phytol. 121:71–79.

Stumm, W., and J.J. Morgan. 1981. Aquatic chemistry. 2nd ed. Wiley, New York.

van Schilfgaarde, J., L. Bernstein, J.D. Rhoades, and S.L. Rawlins. 1974. Irrigation management for salt control. J. Irrig. Drain. Div. Am. Soc. Civ. Eng. 100:321–328.

Viands, D.R., D.K. Barnes, and G.H. Heichel. 1981. Nitrogen fixation in alfalfa: Responses to bidirectional selection for associated characteristics. USDA-ARS Tech. Bull. 1643.

Voorhees, W.B. 1976. Root elongation along a soil-plastic container interface. Agron. J. 68:143.