5-1996

The Effect of Salinity Level upon the Yield, Root Growth, and Water Extraction of Contrasting Rooting Subpopulations of Alfalfa (*Medicago sativa*) Under Conditions of Zero Leaching

Laura A. Vincent
Utah State University

THE EFFECT OF SALINITY LEVEL UPON THE YIELD, ROOT GROWTH AND WATER EXTRACTION OF CONTRASTING ROOTING SUBPOPULATIONS OF ALFALFA (*Medicago sativa*) UNDER CONDITIONS OF ZERO LEACHING

by

Laura A. Vincent

A thesis submitted in partial fulfillment
of the requirements for the degree

of

MASTER OF SCIENCE

in

Plant Science
(Plant Physiology)

Approved:

Dr. Jennifer W. MacAdam Major Professor	Dr. Lynn M. Dudley Committee Member
Dr. Martyn M. Caldwell Committe Member	Dr. James P. Shaver Dean of Graduate Studies

UTAH STATE UNIVERSITY
Logan, Utah
1996

ABSTRACT

The Effect of Salinity Level upon the Yield, Root Growth, and Water Extraction of Contrasting Rooting Subpopulations of Alfalfa (*Medicago sativa*) Under Conditions of Zero Leaching

by

Laura A. Vincent, Master of Science

Utah State University, 1996

Major Professor: Dr. Jennifer W. MacAdam
Department: Plants, Soils, and Biometeorology

A major problem in irrigated agriculture in the Western U.S. is the gradual accumulation of salinity in the plant root zone. These nonuniformly saline soils contain increasing amounts of salinity with depth, and salt accumulation is accelerated in situations where leaching is minimized. Root growth and thus plant yield is limited in these soils due to decreased water uptake. We studied the root growth of two subpopulations of alfalfa differing in their ability to produce fibrous roots to determine if altering root morphology would increase plant yield and water extraction, in an irrigated saline soil.

Soil profiles for a control and three treatments with increasing salinity were packed in to PVC cylinders fitted with a flat window down one side for root measurements. A single alfalfa plant was grown from seed in each cylinder,

and irrigated with water enriched primarily in sulfate salts. Alfalfa plants were grown for five successive harvests in a greenhouse, and water extraction was measured in the control and high Salinity treatment by time-domain reflectometry. Final electrical conductivities of the soil ranged from 3.0 to 23 dS m^{-1}. The yield of the high fibrous root subpopulation was not reduced by the soil salinity by the fifth harvest, while that of the low fibrous subpopulation was reduced 22%. Root growth of the high fibrous subpopulation was significantly increased by as much as 54% in the upper 30 cm of the root zone, compared to that of the low fibrous subpopulation. Water extraction was higher in the upper, least saline portion of the root zone for the high fibrous root subpopulation. The results of this study support the use of alfalfa with increased fibrous root production under saline irrigation with minimal leaching.

(119 pages)

ACKNOWLEDGMENTS

I would like to express my gratitude to Utah Power and Light for the support of this research project. I would also like to thank my committee members, Drs. Lynn Dudley and Martyn Caldwell, for support and accessibility throughout the program, and especially my advisor and mentor Dr. Jennifer MacAdam. Other faculty and staff of the Department of Plants, Soils, and Biometeorology deserve recognition for providing equipment and expertise in aid of this project.

My friends and family have been so supportive while attaining this degree and deserve special thanks. My parents, Linda and Cameron Adams, and good friend, Brannan Vaughan have been wonderful, and without their patience and understanding this would not have been possible.

Laura Vincent

CONTENTS

LIST OF TABLES

LIST OF TABLES (CONTINUED)

LIST OF TABLES (CONTINUED)

LIST OF FIGURES

LIST OF FIGURES (CONTINUED)

CHAPTER I

INTRODUCTION AND LITERATURE REVIEW

Accumulation of salts in the plant root zone under irrigation is a widespread problem in semi-arid and arid regions; throughout the world, almost one-third of all irrigated land is salt-affected (Johnson et al., 1992). Alfalfa (*Medicago sativa*) production occurs on a total of 33 million where it is commonly grown under irrigation hectares (Smith, 1993). Because of the high amount of irrigated alfalfa produced and the declining quantity of arable land (Smith, 1993), there is considerable interest in optimizing growth of alfalfa under saline conditions. Also, as competition increases for high quality water, it is essential to explore alternative sources of irrigation water for agricultural crops. One source of potential importance in the Western U.S. is lower quality saline water.

It was determined by Jury et al. (1977) that future cooling water requirements for electrical generation plants may seriously conflict with agricultural water requirements. Electrical power plants utilize water in a cooling process where water evaporates as it is recycled, concentrating salts naturally present in the water (Dudley et al., 1993). Land application of this water is a practical means of disposal (Zhartman and Gichuru, 1984) and provides an alternative to evaporation ponds. Cooling tower water contains

sufficient salt to be classified as saline irrigation water, but the combination of salts and other elements present determines the usefulness of the water.

Past research has shown that long term irrigation of crops with saline water is feasible (Rhoades et al., 1976; Rhoades et al., 1989; Thellier et al., 1990), with the consideration that irrigation management strongly influences crop response to nonuniform saline soils (Ingvalson et al., 1976). The management practice most commonly utilized is to leach salts out of the root zone (Bower et al., 1969; Bernstein and Francois, 1973; Bernstein et al., 1975; Lonkerd et al., 1979, Francois, 1981; Smith and Hancock, 1986). However, leaching is not always an option in areas where the groundwater is already saline (Mehanni and Rengasamy, 1990) or where future water supplies are protected by regulations prohibiting the addition of saline water to aquifers.

In order to survive in nonuniform saline environments, plants must adapt to a system that is continually changing (Maas, 1986). Short-term exposure to soil solutions above a salinity threshold decreases plant water potential, which negatively affects water uptake by roots and translocation to shoots, and causes plant water stress. Long-term exposure results in premature leaf senescence, (caused by excessive ion accumulation in leaves), a decrease in photosynthate production, a reduction in growth, and often plant death (Munns, 1993). Depending upon the predominant salts the maximum ("threshold') concentration of salts alfalfa can tolerate in a uniform saline soil, without a decrease in growth, is equal to an electrical conductivity (EC) of 4 dS m^{-1} (Maas,

1986). Plant death usually occurs at EC's of 32-35 dS m^{-1} (Bernstein and Francois, 1973).

Roots are the first organ of the plant to be affected in saline environments (Waisel and Breckle, 1987). Previous work has shown that roots can control ion accumulation and leaf growth, while some believe they may even contain the mechanism of salt tolerance (Munns, 1993). Roots are important to the exclusion of salts, as well as in determining how they accumulate within the soil and the plant. The effect of salinity on rooting, particularly on root growth, has been studied previously (Snapp and Shennan, 1992; Waisel and Breckle, 1987). However, utilizing populations with contrasting root growth to study the ability of crop plants to adapt to salinity is a novel approach to this problem.

Root yield has been used as a criterion for selecting plants with tolerance to environmentally stressful conditions (Eissa et al., 1983; Noble et al., 1984; Saindon et al., 1991; Lynch and vanBeem, 1993). Saindon et al. (1991) proposed that breeding for root yield in alfalfa would increase winter survival rates. Noble et al. (1984) identified several populations of alfalfa with differing salt tolerances, and determined that increasing root dry mass and shoot dry mass were closely associated with increased salt tolerance. Thus increasing root growth is a promising area for breeding and physiological studies.

Plant roots can be sensitive to the amount and form of nitrogen available in the soil environment. In a study utilizing three soil nitrogen levels, Trimble et al. (1987) found that nitrogen regimen influenced herbage, root yield and root

morphology of alfalfa. A high application of nitrogen reduced root yields and increased branching or fibrousness, while low nitrogen treatments contained nodulated plants with a more typical tap root system. Under normal field conditions, where alfalfa is inoculated with *Rhizobium*, root morphology assumes a response similar to that of low nitrogen application, provided that nodulation is uninhibited. Typically, non-winterhardy varieties of alfalfa are tap-rooted, and breeding for winterhardiness often alters root morphology, by increasing branching of the tap root and the number of fibrous roots (Barnes et al., 1988).

Selection for alfalfa root traits to encourage nitrogen fixation was conducted by Viands et al. (1981). They utilized a broad based gene pool, MnPl, and conducted two cycles of bi-directional, recurrent phenotypic selection for nodule mass and root characteristics such as fibrous root mass. The breeding program produced two subpopulations, MnPl-9-LF and MnPL-9-HF, with significantly higher (MnPl-9-HF) or lower (MnPl-9-LF) root mass per plant. Their objective was to improve the physiology of alfalfa through selection for morphological traits supporting nitrogen fixation. The studies presented in this thesis provide evidence that the morphological trait of high fibrousness could also confer a production advantage in nonuniform saline soils.

Previous researchers have shown that water uptake of alfalfa roots in the upper 30 cm comprises approximately 40% of the total water extracted throughout the root zone (Jame et al., 1984). Studies suggest that this upper

portion of the root zone is the most sensitive to salinity (Lunin and Gallatin, 1965; Bingham and Garber, 1970; Francois, 1981). Francois (1981) grew tap-rooted alfalfa in lysimeters of different depths and irrigated with saline water at zero leaching. Accumulation of salinity was greatest at the base of the root zone, but not drastically reduced until salt accumulated in the upper portion of the root zone. The period of time before alfalfa yields was reduced was dependent upon the lysimeter depth, because soil salt storage potential increased with lysimeter depth. In a saline root zone where salt concentrations increase with depth, alfalfa that is capable of producing more fibrous roots in the upper, least saline portion of the root zone could potentially extract more water. Thus growth of high fibrous rooted alfalfa could be less affected by salinity.

We propose that alfalfa rooting subpopulations with low and high fibrous rooting characteristics will differ in root distribution and water uptake patterns in nonuniform saline soil conditions. Our objectives were to study the effect of contrasting root structures upon the yield, root growth, and root water extraction of two rooting subpopulations differing in their capacity for fibrous root production.

CHAPTER II

ABSTRACT

Salt accumulation in the root zone can be detrimental to alfalfa (*Medicago sativa* L.) growth in semi-arid and arid regions. Even without leaching, the upper portion of the root zone is less saline, and if alfalfa roots could proliferate in low salinity regions of the root zone, high rates of production could be sustained for many years. Two alfalfa subpopulatons, MnPI-9-LF and MnPI-9-HF, with low and high fibrous rooting characteristics, respectively, were used to determine the effects of saline irrigation upon yield and root growth without leaching. Alfalfa plants were grown for five successive harvests in 1.3 m long cylinders with a clear, flat window along one side for root measurements. Soil packed in the cylinders was premixed with NaCl and gypsum salts to reconstruct a control and three heterogeneous profiles of increasing salinity. Irrigation water with an EC of 2.8 dS m^{-1} was applied at 50% extractable soil water depletion to replace water removed by evapotranspiration. By the fifth harvest, salts had accumulated throughout the root zone with electrical conductivities ranging from 3 to 12 dS m^{-1} for the control, to 3 to 23 dS m^{-1} for the highest salt treatment. After five harvests, yield of the low fibrous root type in the highest salt treatment was reduced 22%, while those of the high fibrous root treatment were not reduced. The high fibrous root type had significantly greater root length density in the upper 30 cm of the root zone. Traced root intensity

(TRI) measurements taken at the clear PVC window revealed that the high fibrous root type had higher TRI values than the low fibrous root type. From these results we may conclude that the high fibrous root type is better suited to nonuniform saline conditions without leaching.

INTRODUCTION

A common problem in irrigated agriculture is the gradual build up of salts in the root zone. Without leaching, long-term yield reductions occur as salt accumulation extends to the upper portion of the root zone, which is particularly salt sensitive (Schilfgaarde et al., 1974; Jame et al., 1984; Smith, 1993). Plants could utilize the higher soil water potentials in the upper portion of the root zone (Minhas and Gupta, 1993) if root growth were concentrated in this area.

Selection in the field for root traits in alfalfa (*Medicago sativa*) has been shown to have a positive effect upon yield in varying environmental conditions. To improve winter survival of alfalfa, Saindon et al. (1991) selected for root yield in two cultivars. They found that increased root branching was correlated with higher yield, and suggested that further improvements in winter survival were possible through breeding. 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). After evaluating several genotypes with differing yields for variation of growth within the root system, they concluded there was a correlation between shoot growth and root architecture.

Typically, non-dormant varieties of alfalfa are tap-rooted (Smith, 1993), and winterhardiness is associated with greater branching of the tap root and greater fibrous root mass (Barnes et al., 1988). To improve nitrogen fixation,

Viands et al. (1981) selected two subpopulations of alfalfa that significantly differed in their rooting characteristics, a low fibrous (MnPI-9-LF) and high fibrous (MnPI-9-HF) subpopulation. We propose that the alfalfa subpopulation with greater fibrousness will yield more in a nonuniform saline root zone, because these plants will be able to generate more root mass in the least saline regions of the root zone. The objective of this study was to compare the yields and root growth of two contrasting rooting subpopulations under conditions of increasing salinity without leaching.

MATERIALS AND METHODS

Two alfalfa (*Medicago sativa* L.) near isogenic subpopulations selected for low fibrous (MnPl-9-LF) and high fibrous (MnPl-9-HF) rooting characteristics (Viands et al., 1981) were planted in cylinders constructed from PVC pipe. Each cylinder was 1.3 m long with a 10-cm diameter and a wall thickness of 30 mm. One side was replaced by an 8-cm-wide, flat, 32 mm thick, clear, PVC window bonded in place using weld-on epoxy (Industrial Polychemical, Gardena, CA). Caps made of PVC with an inside diameter of 10 cm were bonded to the bottom of cylinders. Prior to packing soil, holes were drilled in the caps and covered with wire mesh. The cylinders were packed with a 2.5 cm layer of gravel to allow soil drainage and promote aeration. Soil was then packed in 10 cm increments to a bulk density of approximately 1.25 g cm^{-3}. The soil used was a 2-mm-sieved Kidman fine sandy loam from the Ap Horizon (coarse-loamy, mixed, mesic Calcic Haploxeroll) obtained in Smithfield, Utah. Cylinders were wrapped in aluminum foil to exclude light from the clear face, and placed on an A-frame at 25° from vertical to promote root growth along the soil-window interface (Fig. 1).

Four root zones with heterogeneous salinity were constructed to include a control and three increasingly saline treatments (low, medium and high salt), by mixing predetermined amounts of NaCl and gypsum with the soil in a cement mixer. The amounts of salts added to the soil for each desired EC_e are provided

in Table 1. Electrical conductivities were determined based upon predictions of crop water balance and salt accumulation at a10, 20 and 30 year period and associated yield decrements by a soil water chemistry (SOWACH) model (Dudley and Hanks, 1991). The electrical conductivity of soil saturated paste extracts (EC_e) of the control, low salt, medium salt, and high salt treatments at experiment initiation are reported in table 2.

The study was conducted in a greenhouse that was maintained at 20± 5°C day and 15± 5°C night temperatures with a 16 hour photoperiod. Supplemental lighting was provided by high pressure sodium lights at an average photosynthetic photon flux density of 500 μmol m^{-2} s^{-1}. Cylinder location in the greenhouse was re-randomized once every four weeks to minimize the effect of environmental gradients.

Nitrogen at two levels, zero and 20 mg kg^{-1}, was added to the top 15 cm of soil to determine if root morphology would be affected by nitrogen concentration (D.K. Barnes, pers. comm.). Added nitrogen had no effect on nodulation or any other root parameter.

Soils were determined to be deficient in P, thus P equivalent to 70 mg kg^{-1} P_2O_5 was mixed with the top 15 cm of soil, and phosphate was applied after each harvest as 100 ml 0.16 mM KH_2PO_4 and 0.84 mM K_2HPO_4 (pH 7.2).

Alfalfa seed was treated with the fungicide Apron (Ciba-Geigy, Ltd. Switzerland) at 2.5 g per kg of seed, and inoculated with a commercial inoculant (Nitragin, Milwaukee, WI) as well as a mix of four salt-tolerant strains

(USDA 1027, 1029, 1030, 1031) of *Rhizobium meliloti* (USDA Soybean and Alfalfa Research Laboratory, Beltsville, MD).

Cylinders were watered to a container capacity (0.1 bar) of 23.4% with saline irrigation water on 20 February 1995. Irrigation water had an EC of 2.8 dS m^{-1}. The concentration of salts was 9.33 mM $CaSO_4$, 5.36 mM $MgSO_4$, 1.00 mM Na_2SO_4, and 5.41 mM NaCl, which is the composition of water following its use in an evaporative system at an electrical power plant in Huntington, Utah.

Salinity was flushed from the top 2-cm soil layer of each cylinder with approximately 40 ml tap water, and seeds were sown in PVC rooting cylinders on 13 March 1995 and germinated under natural lighting. Seedlings were thinned to one plant per cylinder 21 days after emergence. The first saline irrigation was applied after plant establishment, or five weeks after emergence. Plants were cut to a height of 10 cm when they reached the late flowering growth stage (Fick and Mueller, 1989), which occurred at 3-5 week intervals for a total of five harvest periods ending 13 October 1995.

Plants were watered when cylinders reached 50% extractable soil water depletion (ESW) (Carter and Sheaffer, 1983). Total ESW was calculated from the difference between cylinder mass at container capacity and the mass of cylinders containing air-dried soil at the time of packing. Water was applied in a drip from 4 L carboys while cylinders were held upright to prevent soil channeling. Volumetric soil water content was also measured in six replicates of the control and the high saline treatments by time-domain reflectometry (TDR)

(Environmental Sensors Inc., San Diego, CA). These data are discussed in the following chapter.

Root measurements at the soil-window interface

At the end of each of five harvest periods and just prior to cutting alfalfa plants, the lengths of roots at the soil-PVC window interface was traced onto acetate sheets using permanent felt-tip pens. Root distributions displayed at the soil-window interface were traced with a different color at each harvest (Snapp and Shennan, 1992). The length of traced roots within each section of the root zone (0-15, 15-30, 30-60, 60-90, 90-120 cm deep) was determined using a root digitizer (Jandel Corp., SigmaScan).

Destructive Analysis

Following the fifth harvest, cylinders were sawed open longitudinally opposite the clear PVC face, and the soil and roots were divided into five sections. A 5-cm-long subsample was removed from the center of each section of the root zone for soil analysis. The remaining soil and root mass was separated by a pneumatic 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 samples. Actual root length was determined by a root

length scanner (Comair, Melbourne, Australia) and root mass was determined after drying at 70°C for 48 hours.

Tap root diameter was measured 1 cm below the crown after the fifth harvest. Nodules that were active and visible at the clear PVC face of the cylinders were counted just prior to destructive sampling. The final EC_e of the soil was determined by saturated paste extracts (Table 3), and mineral composition was determined by inductively-coupled plasma spectrometry (ICP) by the USU Soil Testing Lab (Table 4).

The final EC_e of the original medium salinity treatment were higher in the bottom segment of the root zone than the original high salinity treatment, probably due to inadvertent leaching when cylinders were brought to container capacity. Therefore, with the exceptions of Tables 1 and 2, the medium and high salt treatments refer to the actual relative final salinity of treatments.

The experiment was designed as a split-plot arranged in a randomized complete block with six replications. Data from all procedures were compared within and between treatments using analysis of variance (ANOVA), (Minitab Inc., 1992). Response and interactions of nitrogen level, salt treatment, root type and, when applicable, depth, were tested using the pooled residual as the error term. Significance was determined by p-values, and least significance differences (LSD) between means were calculated.

RESULTS AND DISCUSSION

Yield

Salinity levels reduced average cumulative shoot dry mass of the low fibrous (MnPl-9-LF) root type by 22% (Fig. 2). Whereas, the shoot dry mass of the high fibrous (MnPl-9-HF) root type was higher for saline than control treatments ($p \leq 0.26$). Differences in shoot dry mass of alfalfa subpopulations MnPl-9-LF and MnPl-9-HF (Table 5) were greater for harvest four and five.

Researchers in the past have related yield to the mean EC_e of the root zone, but these studies achieved higher salt levels (Shalhevet and Bernstein, 1968; Ingvalson et al., 1976; Mass and Hoffman, 1977). Shalhevet and Bernstein (1968) established alfalfa in 50-cm long containers and increasingly salinized the upper and lower portions of the root zone. They found a positive correlation of yield with the average salinity of the two zones, which ranged from 1 to 18 dS m^{-1}. Still others have shown that the upper portion of the root zone is the most salt sensitive (Bingham and Garber, 1970; Francois, 1981; Jame et al., 1983). Although, yield and average EC_e of the upper 0 to 30 cm of the root zone were not well correlated ($r^2 = 0.32$, $p \geq 0.05$), it is likely that the EC_e of 3 to 4 dS m^{-1} in this study were not high enough in this region to 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 alfalfa could be grown in saline soils with NaCl and gypsum with an average EC of 4-5

dS m^{-1} in the top 0-15 cm of soil, while Francois (1981) reported 80% yield decrements at EC of 8.4 dS m^{-1} in the upper 30 cm when the predominant salts were NaCl and $CaCl_2$. In the study reported here, EC_e in the lowest sections of the root zone were as high as 23.15 dS m^{-1} with no negative effect on yield for the high fibrous root type.

In addition to the predominance of Na and Ca salts in this study, other factors such as the watering regimen may have contributed to increased salt tolerance and yields. Plants were watered upon depletion of extractable soil water (ESW) to 50% to avoid confounding salinity effects with drought stress. Carter and Sheaffer (1983) determined that moderate application of water on alfalfa growing on coarse-textured soils at 50% depletion of ESW was a threshold for maintenance of favorable plant water status. It may be possible that this watering regimen masked some symptoms of salt stress at the EC_e achieved in this study.

It has also been found in other studies that greenhouse conditions can ameliorate salt effects upon yield. Salt tolerance in greenhouse studies conducted by several researchers (Chang, 1960; Bernstein and Francois, 1973) have been higher than field studies. The higher relative humidity in the summer that the greenhouse provides reduces water stress and evapotranspiration needs.

Soil Solution Salinity

Composition of soluble salts deposited in the soil by the irrigation water is described in Table 4. Sodium chloride gradually increased with depth and treatment, but was consistently very high at 90 to 120 cm, ranging from 60.84 to 177.38 mM. On a molar basis, sodium chloride salt made up only a small portion of the salinity in the irrigation water, whereas sulfate-based salts accounted for the remainder of the salinity. Yet, as in other studies (Dudley et al., 1994) the ratio of Na/Ca shows that sodium predominated at lower depths. This ratio is also useful because the addition of calcium has been shown to alleviate some of the symptoms of salt stress (Cramer et al., 1986; Rengasamy, 1987; Evlagon et al., 1992). Calcium is essential to plant cell ion regulation, and when present in saline irrigation water, helps both soil structure and plant metabolism. Calcium cations help 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 can not persist.

Root Growth

Measurement at the Soil-Window Interface

Root lengths ascertained at the soil-window interface are reported as traced root intensity (TRI), which is traced root length per area of root viewing window (cm cm^{-2}). Earlier experiments utilizing slant tube methodology to

determine TRI have proven it very useful for in situ qualitative observations of root morphology and ecology (Rutherford and Curran, 1981; McMichael et al., 1992), provided the data regard quantification of root growth using TRI compared with the other acceptable approaches.

The interaction of salinity treatment and rooting depth on TRI was highly significant ($p = 0.003$) reflecting the effect of both salinity and normal alfalfa root development on root distribution at the soil-window interface. Cumulative TRI for the five harvests is presented in Figure 3. Traced root intensity declined sharply for all treatments at the bottom of the root zone, where salt, and particularly sodium, accumulation was greatest (Table 4). There were significant differences ($p \leq 0.08$) between the two root types, particularly in the upper 60 cm at the highest salinity treatment.

Measurement of Roots in Bulk Soil

Measured root length in each section of the root zone is reported as actual root length density (RLD), which is root length per volume of soil (cm cm^{-3}). There was a significant difference between the RLD of the low fibrous and high fibrous root types ($p \leq 0.03$). The interaction of root type and depth ($p \leq 0.01$) and salt and depth ($p = 0.0$) were highly significant. Analysis within salinity treatments indicates a significant difference in the interaction of root and depth for the low salt ($p \leq 0.05$) and the medium salt ($p \leq 0.001$) treatments (Figs. 4b and 4c).

The effect of salinity treatment on the distribution of RLD of the low fibrous and high fibrous rooting subpopulations are shown separately to better demonstrate the interaction of root type and depth (Fig. 5). Root length density of the low fibrous root type in the lower 90-120 cm decreased from 6.7 cm cm^{-3} for the control to 2.7 cm cm^{-3} for the high salt treatment (Table 7), representing a 40% decrease, while RLD values in the upper 0-15 cm increased 37%, from the low to the high salt treatment.

The RLD for the high fibrous root type show a different distribution pattern (Fig. 5b) from the low fibrous root type relative to salt level. The values for the control closely resemble those of the low fibrous root type (Table 7). However, in the upper 0-15 cm, the low salt treatment shows the largest value for RLD and the values gradually decrease with salt concentration. Also, in contrast to the low fibrous root type, in all salt treatments, the RLD values in 0-15 cm were all greater than the 15-30 cm. Similarly, the RLD of the high fibrous root type for the high salt treatment was 58% lower than the control in the bottom 90-120 cm of the root zone, while at its greatest difference from the control, the low salt was 59% higher in the 0-15 cm depth.

Thus both subpopulations showed a root distribution change in response to salinity. As expected, the high fibrous root type was able to concentrate more roots in the less saline upper portion of the root zone increased 59% compared with the control than the low root type with an increase of only 37% in this

portion of the root zone (Fig. 6). Therefore, performance of the high fibrous root type was superior at low-to-moderately saline conditions used in this study.

Furthermore, unlike the saline treatments, root growth of the control plants was not concentrated in the upper, least saline portion of the root zone (Figure 4a). Regardless of root type, the control distribution pattern was quite even throughout the profile; the three salt treatments had quite different root distributions (Figure 4b-d). It has been noted in the past that there is a disadvantage when salt is applied after plant establishment because of root development and proliferation (Shalhevet and Bernstein, 1968). Thus, it would have been interesting to compare the growth of another treatment established on a heterogeneous saline root zone, but irrigated with non-saline water.

Comparison of Soil-Window Interface and Bulk Soil Root Determinations

Traced root length data and actual root length data were compared to determine the effect of growth at a 25° angle on the percentage of roots concentrated at the observation window. Percent roots at the window was calculated by dividing the traced root length density (traced root intensity x 3 mm viewing depth; cm cm^{-3}) (Glinski et al., 1993) by the actual root length density.

Statistical analysis revealed there was no significant effect of root type on the percent roots at the window ($p \leq 0.45$). However, salt ($p \leq 0.03$) and the interaction of salt and depth ($p \leq 0.001$) were highly significant (Table 8). The

graph of percent roots at the window as a function of depth (Fig. 7) demonstrates that both root types responded to the growth angle by concentrating fewer roots at the 0 to 30 cm depths, and more in 90 to 120 cm depth. Thus, tracing roots at the window quantified 60% of the roots in 0-15 cm of bulk soil of the low fibrous subpopulation and 50% of the roots of the high fibrous subpopulation. However, in the bottom 90-120 cm, root tracings over-represented the two root types by 12% and 15% for the low and high fibrous subpopulations, respectively. Thus, from the graph, TRI in the region from 30 to 90 cm appeared to best describe the actual root distributions for both subpopulations. Because in this experiment both root types responded similarly to the growth angle, this suggests that TRI does not discriminate between root morphologies when used as a tool to quantify root growth.

Others have shown the values obtained from the clear PVC window underestimated bulk soil rooting. Utilizing slant tubes made of clear polyethylene to study the roots of Penncross creeping bentgrass, Glinski et al. (1993) found that TRI tended to increase with depth relative to actual RLD; our results are in agreement with this. The rationale they provided was that the plant growth angle of 20 - 25° degrees from vertical forced roots to grow into a smaller area with depth.

The percentage of roots at the window were graphed by salinity treatment as a function of depth (Fig. 8) to best describe the effect of salinity upon estimates of traced root intensity. Again, the traced root distributions of all

treatments were underestimated in the top 0 to 15 and 15 to 30 cm depths. The percentage of roots at the window increased with salt level in the bottom half of the root zone at 60 to 90 and 90 to 120 cm. Thus the more saline the soil environment, the more roots tended to grow along the PVC root observation window compared with the bulk soil. The fact that at the greatest depth, the high salt treatment had 53% more roots concentrated at the window than the control indicates some advantage was conferred by proximity to the window.

A possible explanation is that even though irrigation was applied at 90° (Fig. 1) the water permeated the soil towards the face when cylinders were placed back at 25° from vertical. This would raise the soil water potential, thereby easing water extraction. Another, though less likely explanation, may be an attraction to the PVC window. Voorhees (1976) hypothesized an attraction for the roots to the window, which for his study consisted of plexiglas. However, because we utilized PVC, which is the same material as the cylinder, a similar interaction is unlikely and the response of all treatments would have been similar.

Dry Root Mass

Statistical analysis of the mass of dried roots recovered from the volume of the soil revealed that the difference between root types was nonsignificant ($p \leq 0.30$) (Table 9). Similarly, Snapp and Shennan (1992) found that root weight does not provide information on root morphological responses to salinity.

However, the interaction of salt and depth ($p = 0.03$) was significant. The root mass distributions by depth for all treatments (Fig. 9) show that salinity increased root mass in the upper 0-15 and 15-30 cm for both root types, and decreased it in the bottom 90-120 cm. In the high salt treatment (Fig. 9) the low fibrous root type had 55% less density of root mass than the control in the bottom 90 to 120, while the high fibrous had 48% less.

There was a stronger correlation between dry root mass and cumulative yield ($r^2 = 0.97$) ($p \leq 0.006$) than between RLD and yield ($r^2 = 0.64$) ($p \geq 0.05$). Thus dry root mass played an important part in shoot dry mass by harvest five.

Specific root length

The length of roots per root mass (cm g^{-1}) is important in characterizing the morphology of roots. The higher the specific root length value, the more root length per mass, suggesting a more fibrous root system. Conversely, a lower value of specific root length suggests a more tap-rooted system.

There were no significant differences in specific root length between root types and salt treatments (Table 10), only depth ($p = 0.0$) was significant. This indicates that both the RLD and root biomass of the high fibrous root type increased proportionately relative to the low fibrous.

As a function of root type (Fig. 10a) or salt treatment (Fig. 10b), specific root length gradually shifted from tap to fibrous roots with increasing soil depth.

Tap Root Diameter

Statistical analysis of tap root diameter showed a significant difference between the low and high fibrous root type ($p \leq 0.02$), with the high fibrous root type having a larger tap root diameter regardless of salinity treatment ($p = 0.45$).

In the field, Barnes et al. (1988) have associated a larger diameter tap or primary root with a high fibrous root system, while a relatively 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 field. This trait was not influenced by salt level, and proves, along with supporting root data, that the expected characteristics of the two root types were displayed in this study.

Nodulation

The objectives in selecting for high fibrous rooting was to enhance nitrogen fixation ability (Viands et al., 1981). The number of nodules in the field in that study were well correlated ($r^2 = 0.61$) with fibrous root score. However, we found that the number of nodules visible and active at the PVC window was unaffected by root type.

Nodule number was not affected by initial application of nitrogen nor were any of the root parameters, (i.e., RLD, TRI and dry root mass).

There was a significant ($p \leq 0.05$) positive effect of salinity level on nodule number (Fig. 11). The inoculation of alfalfa seed with a mixture of four salt-tolerant strains of *Rhizobia meliloti* may have helped plants adapt to the

saline environment, as nitrogen is often limiting in saline conditions (Khan et al., 1994). The control had robust nodules, but significantly fewer than the salt treatments, prompting the question of whether the increase in nodulation with increasing salinity contributed to the stimulation of yield with salinity in the high fibrous subpopulation (Fig. 2).

Root to Shoot Ratio

There was a strong positive correlation ($r^2 = 0.70$) ($p \leq 0.01$) between root dry mass and cumulative shoot dry mass. Statistical analysis of the root:shoot ratio showed no significant differences by root type ($p \leq 0.59$) or salt treatment ($p \leq 0.55$). Although, nitrogen was significant ($p = 0.03$), indicating that it conferred some advantage in the ratio between roots and shoots. However, utilizing yield from Harvest 5 only, the root:shoot ratio had slightly different results. Again, neither root type ($p \leq 0.12$) or salt ($p \leq 0.30$) level were significant, but the interaction of the two factors was highly significant ($p = 0.004$), suggesting the amount of dry root mass and salt influenced the proportion to yield.

CONCLUSIONS

We may conclude from this study that irrigation with electrical plant cooling water enriched in sulfate salt for the production of alfalfa can be sustained without yield decrement for many years, even without leaching. Alfalfa yields were, in fact, stimulated by heterogeneous root zone salinity. The root characteristics of the MnPI-9-LF and MnPI-9-HF subpopulations displayed in the greenhouse were consistent with those seen in the field. However, specific root length indicated that the high fibrous root subpopulation had more roots rather than a proliferation of significantly finer roots. Both responded to salinity by altering their root distribution, but the high fibrous root type had the ability to concentrate more roots in the upper, least saline portion of the root zone. Future research efforts should be focused on understanding the physiological basis for the changes observed in root morphologies.

REFERENCES

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 Conference, 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. 1960. Effects of saline irrigation water and exchangeable sodium on soil properties and growth of alfalfa. Soil Sci. Soc. Am. ????

Cramer, G.R., A. Lauchli, and E. Epstein. 1986. Effects of NaCl and $CaCl_2$ on ion activities in complex nutrient solutions and root growth in cotton. Plant Physio. 81:792-797.

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

Dudley, L.M., Hanks, R.J., MacAdam, J.W., Mace, R.W., 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. Sta. Res. Rpt. 150.

Evlagon, D., I. Ravina, and P.M. Neumann. 1992. Effects of salinity stress and calcium on hydraulic conductivity and growth in maize seedling roots. Isr. J. Bot. 39:239-247.

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

Francois, L.E. 1981. Alfalfa management under saline conditions with zero leaching. Ag. 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.

Lynch, J., 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. ASCE 103:115-134.

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

Mehanni, A.H. and P. Rengasamy. 1990. Establishment and production of lucerne on a salinised clay soil irrigated with moderatelly 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 patters. 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.

Rutherford, M.C. and B. Curran. 1981. A root observation chamber for replicated use in a natural plant community. Plant and 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.

Schilfgaarde, J.V., L. Bernstein, J.D. Rhoades, and S. L. Rawlins. 1974. irrigation management for salt control. J. Irrig. Drain. Div. ASCE 100:321-328.

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.

Snapp, S.S., and C.S. Shennan. 1992. Effects of Salinity on Root Growth and Death Dynamics of Tomato, *Lycopersicon esculentum* Mill. New Phyto. 121:71-79.

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. Ag. J. 68:143.

Table 1. Amounts of salts added for desired EC_e.

Salt	Depth	Low Salt	Medium Salt	High Salt
	cm	---------- g salt kg^{-1} of soil ----------		
NaCl	0-15	0.000	0.099	0.263
	15-30	0.000	0.099	0.263
	30-60	0.272	0.591	0.931
	60-90	0.611	1.165	1.760
	90-120	0.871	1.750	2.616
$CaSO_4$ $2H_2O$	0-15	0.000	0.069	0.168
	15-30	0.000	0.069	0.168
	30-60	0.147	0.170	0.195
	60-90	0.142	0.149	0.144
	90-120	0.108	0.077	0.045

Table 2. Initial soil solution electrical conductivities of individual root zone segments determined from saturation soil paste extracts.

Depth	Treatment			
	Control	Low Salt	Medium Salt	High Salt
cm	---------- dS m^{-1} ----------			
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

Table 3. Final soil solution electrical conductivities of individual root zone segments determined from saturation soil paste extracts.

Root Type	Depth	Treatment			
		Control	Low Salt	Medium Salt	High Salt
	cm	---------- dS m^{-1} ----------			
MnPI-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
MnPI-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

Table 4. Mineral compositions calculated from saturated paste extracts following Harvest 5.

Treatment	Root Type	Depth	NaCl	Na2SO4	MgSO4	CaSO4	Na/Ca
		cm	---------- mM ----------				
Control	MnPI-9-LF	0-15	4.88	1.08	5.76	7.76	0.69
		15-30	6.29	1.25	6.13	7.95	0.82
		30-60	4.96	2.75	8.04	10.79	0.79
		60-90	9.11	4.89	11.30	9.68	1.50
		90-120	73.82	2.29	18.62	14.21	5.52
	MnPI-9-HF	0-15	4.01	1.49	4.58	5.68	0.87
		15-30	6.29	1.25	6.13	7.96	0.82
		30-60	4.96	2.75	8.04	10.94	0.79
		60-90	10.32	6.88	12.30	10.31	2.05
		90-120	60.84	2.80	26.53	13.05	5.09
Low Salt	MnPI-9-LF	0-15	4.88	1.08	5.76	7.76	0.69
		15-30	3.95	1.85	6.46	10.04	0.63
		30-60	4.96	2.75	8.04	10.94	0.79
		60-90	23.35	1.75	16.54	12.62	2.13
		90-120	110.94	1.00	21.38	18.57	5.42
	MnPI-9-HF	0-15	3.95	1.85	6.46	12.15	0.63
		15-30	4.20	2.57	7.34	14.74	0.63
		30-60	5.33	4.14	9.66	14.80	0.92
		60-90	23.35	1.75	16.54	12.62	2.13
		90-120	95.36	1.00	17.88	20.81	4.68
Medium Salt	MnPI-9-LF	0-15	4.88	1.08	5.76	10.24	0.69
		15-30	4.20	2.57	7.34	14.74	0.63
		30-60	4.96	2.75	8.04	13.33	0.79
		60-90	23.35	1.75	16.54	12.62	2.13
		90-120	177.38	1.00	18.64	22.74	7.89
	MnPI-9-HF	0-15	3.75	2.31	7.26	10.91	0.58
		15-30	4.96	2.75	8.04	10.94	0.79
		30-60	6.46	3.42	8.86	9.69	0.97
		60-90	32.75	1.04	23.56	12.19	2.86
		90-120	101.54	11.00	11.71	8.78	7.07
High Salt	MnPI-9-LF	0-15	4.88	1.08	5.76	7.76	0.69
		15-30	3.75	2.31	7.26	10.91	0.58
		30-60	4.96	2.75	8.04	10.94	0.79
		60-90	26.31	1.00	17.32	13.13	2.16
		90-120	121.43	1.00	22.50	10.09	9.18
	MnPI-9-HF	0-15	6.29	1.25	6.13	7.76	0.82
		15-30	4.96	2.75	8.04	10.94	0.79
		30-60	6.46	3.42	8.86	9.69	0.97
		60-90	30.49	3.46	20.23	11.48	3.26
		90-120	149.72	1.36	21.06	13.75	9.25

Table 5. Shoot dry mass at each harvest for the low and high fibrous root types under four salt treatments.

Treatment	Root type	Harvest Number					
		1	2	3	4	5	Cumulative
		---------- g ----------					
Control	MnPI-9-LF	6.11	9.52	9.55	9.17	7.81	42.15
	MnPI-9-HF	5.33	8.32	9.35	9.89	8.47	41.36
Low Salt	MnPI-9-LF	5.68	9.21	9.28	8.24	7.89	40.30
	MnPI-9-HF	6.27	9.93	10.32	11.23	11.39	49.15
Medium Salt	MnPI-9-LF	6.96	7.03	8.39	6.72	8.41	37.51
	MnPI-9-HF	5.93	9.24	10.28	11.41	10.69	47.54
High Salt	MnPI-9-LF	6.18	8.07	9.40	7.84	6.16	33.11
	MnPI-9-HF	5.89	7.96	8.93	9.94	9.75	42.47

Table 6. Cumulative mean traced root intensity for two alfalfa rooting subpopulations and four salt treatments.

Treatment	Depth	Root Subpopulation	
		MnPL-9-LF	MnPL-9-HF
	cm	------- cm cm^{-2} -------	
Control	0-15	1.32	1.30
	15-30	1.72	1.75
	30-60	2.30	2.03
	60-90	2.29	2.39
	90-120	1.51	2.05
Low Salt	0-15	1.08	1.39
	15-30	1.62	1.63
	30-60	2.14	1.73
	60-90	2.59	2.42
	90-120	1.31	1.62
Medium Salt	0-15	1.15	1.54
	15-30	1.46	1.67
	30-60	1.92	2.11
	60-90	2.24	2.45
	90-120	1.58	1.60
High Salt	0-15	1.29	1.85
	15-30	1.67	2.33
	30-60	2.13	2.65
	60-90	2.53	2.87
	90-120	0.92	1.38
	$LSD_{0.10}$ [a]	0.51	

[a] least significant difference at the 0.10 level between a fixed depth, comparing salt treatments and root type.

Table 7. Mean root length density for two alfalfa rooting subpopulations and four salt treatments.

Treatment	Depth	Root Subpopulation	
		MnPL-9-LF	MnPL-9-HF
	cm	--------- cm cm^{-3} ---------	
Control	0-15	6.59	8.46
	15-30	8.89	9.88
	30-60	7.18	7.25
	60-90	8.69	7.85
	90-120	6.66	6.85
Low Salt	0-15	7.26	13.43
	15-30	8.64	12.13
	30-60	6.78	8.98
	60-90	7.60	9.60
	90-120	4.83	6.54
	$LSD_{0.05}$ [a]	2.04	
Medium Salt	0-15	7.70	12.49
	15-30	9.12	11.68
	30-60	7.70	9.57
	60-90	6.93	7.81
	90-120	3.83	4.17
	$LSD_{0.05}$ [b]	2.06	
High Salt	0-15	9.05	11.61
	15-30	10.29	10.88
	30-60	7.88	8.03
	60-90	7.05	8.33
	90-120	2.67	3.97
	$LSD_{0.05}$ [c]	3.05	

[a] least significant difference at the 0.05 level for the low salt treatment, for a fixed depth, comparing salt treatment and root type.
[b] least significant difference at the 0.05 level for the medium salt for a fixed depth, comparing salt treatment and root type.
[c] least significant difference at the 0.05 level for four salttreatments, for a fixed depth, comparing salt treatment and root type.

Table 8. Percent roots concentrated at the PVC window for two alfalfa rooting subpopulations and four salt treatments.

Treatment	Depth	Root Subpopulation	
		MnPL-9-LF	MnPL-9-HF
	cm	%	
Control	0-15	66.92	51.06
	15-30	64.68	59.17
	30-60	106.69	93.38
	60-90	87.69	101.53
	90-120	75.38	100.00
Low Salt	0-15	49.59	34.40
	15-30	62.57	44.85
	30-60	105.16	64.25
	60-90	113.82	85.23
	90-120	90.68	83.62
Medium Salt	0-15	49.48	41.07
	15-30	53.29	47.77
	30-60	83.25	73.46
	60-90	108.09	104.87
	90-120	137.34	128.06
High Salt	0-15	47.51	53.23
	15-30	53.94	71.42
	30-60	90.10	110.09
	60-90	119.72	114.90
	90-120	114.98	115.91
	$LSD_{0.05}$ [a]	32.17	

[a] least significant difference at the 0.05 level for four salt treatments, for a fixed depth, comparing salt treatment and root type.

Table 9. Mean root dry mass per volume of soil for two alfalfa rooting subpopulations and four salt treatments.

Treatment	Depth	Root Subpopulation	
		MnPL-9-LF	MnPL-9-HF
	cm	--------- g cm^{-3} ---------	
Control	0-15	0.275	0.255
	15-30	0.248	0.258
	30-60	0.125	0.162
	60-90	0.102	0.113
	90-120	0.062	0.063
Low Salt	0-15	0.348	0.380
	15-30	0.253	0.317
	30-60	0.143	0.187
	60-90	0.103	0.118
	90-120	0.043	0.056
Medium Salt	0-15	0.255	0.493
	15-30	0.258	0.270
	30-60	0.162	0.173
	60-90	0.113	0.105
	90-120	0.063	0.032
High Salt	0-15	0.275	0.333
	15-30	0.235	0.300
	30-60	0.135	0.143
	60-90	0.080	0.090
	90-120	0.033	0.033
	$LSD_{0.05}$ [a]	0.120	

[a] least significant difference at the 0.05 level for four salt treatments, for a fixed depth, comparing salt treatment and root type.

Table 10. Mean specific root length for two alfalfa rooting subpopulations and four salt treatments.

Treatment	Depth	Root Subpopulation	
		MnPL-9-LF	MnPL-9-HF
	cm	--------- cm g^{-1} ---------	
Control	0-15	23.12	36.62
	15-30	37.15	40.20
	30-60	61.93	50.08
	60-90	95.47	92.75
	90-120	120.98	113.20
Low Salt	0-15	21.47	36.13
	15-30	34.90	43.42
	30-60	47.15	50.03
	60-90	73.00	88.18
	90-120	110.75	134.95
Medium Salt	0-15	31.22	25.92
	15-30	40.13	46.12
	30-60	59.28	60.18
	60-90	91.35	80.35
	90-120	118.32	137.55
High Salt	0-15	36.20	37.00
	15-30	44.00	38.50
	30-60	57.32	67.00
	60-90	99.12	103.03
	90-120	111.92	131.97

Not statistically significant.

Table 11. Mean tap root diameter, number of nodules, and root to shoot ratio for two alfalfa rooting subpopulations and four salt treatments.

Treatment	Root Subpopulation	Tap Root Diam.	Nodules	Root:Shoot
		cm	#	
Control	MnPL-9-LF	1.05	20.17	1.31
	MnPL-9-HF	1.21	24.83	1.21
Low Salt	MnPL-9-LF	1.13	32.17	1.27
	MnPL-9-HF	1.2	31.67	1.00
Medium Salt	MnPL-9-LF	1.10	42.50	1.04
	MnPL-9-HF	1.19	36.17	1.05
High Salt	MnPL-9-LF	0.99	37.67	1.18
	MnPL-9-HF	1.21	66.83	1.01
	LSD0.05 [a]	0.22	20.17	NS

[a] least significant difference at the 0.05 level comparing salt treatment and root type.
NS is not statistically different.

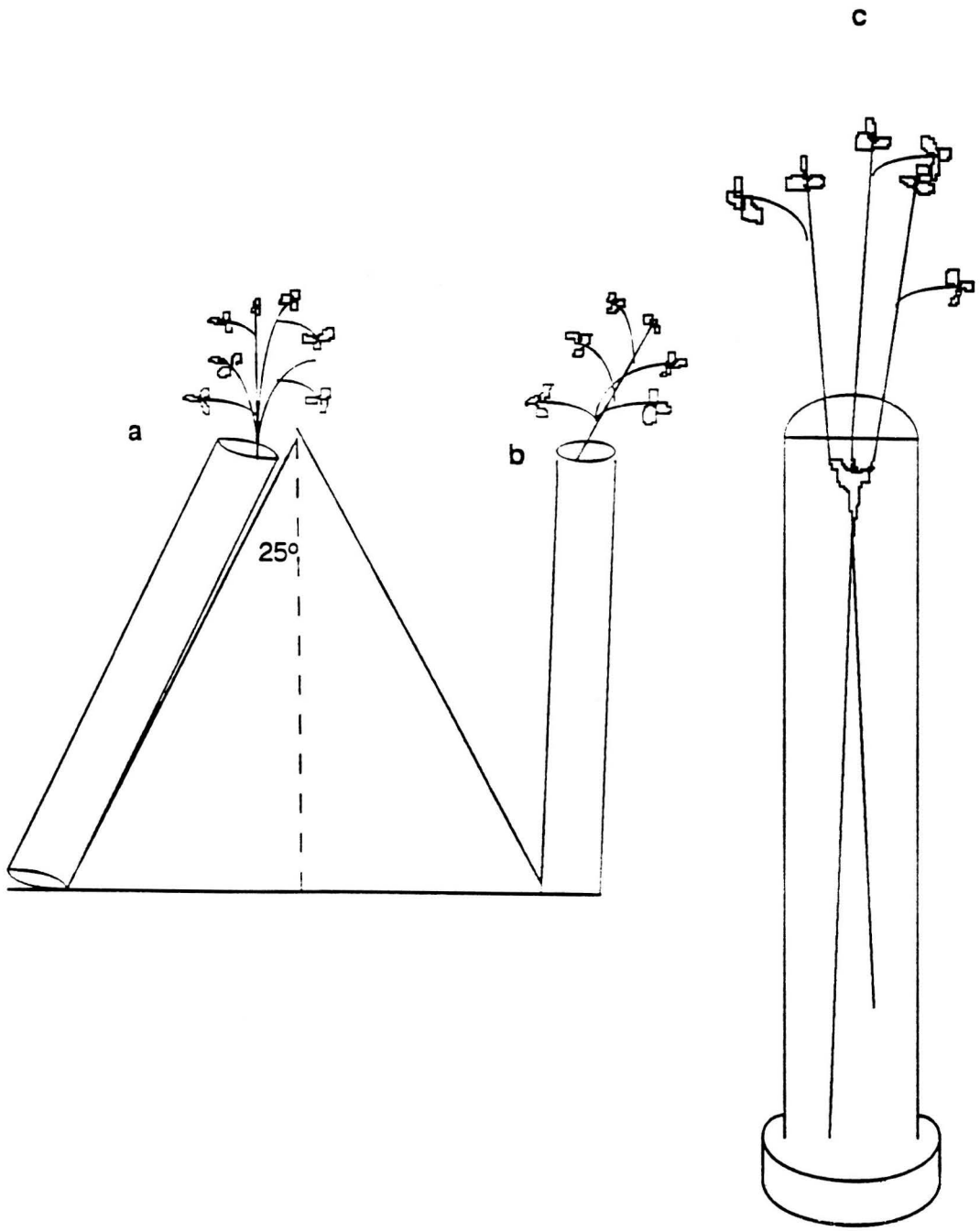


Figure 1. Schematic of, a; PVC cylinders with plants growing at an angle of 25° from vertical, b; cylinders standing upright for watering, and c; alfalfa roots visible through the PVC window.

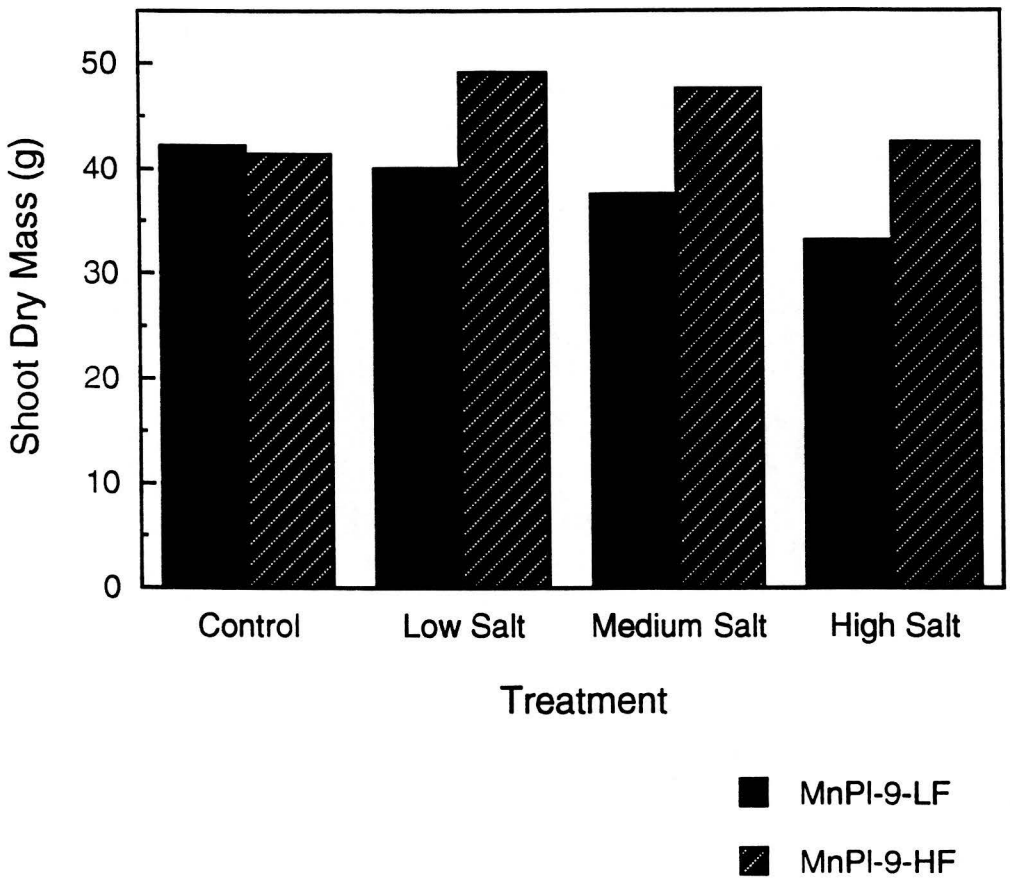


Figure 2. Cumulative shoot dry mass from five harvests.

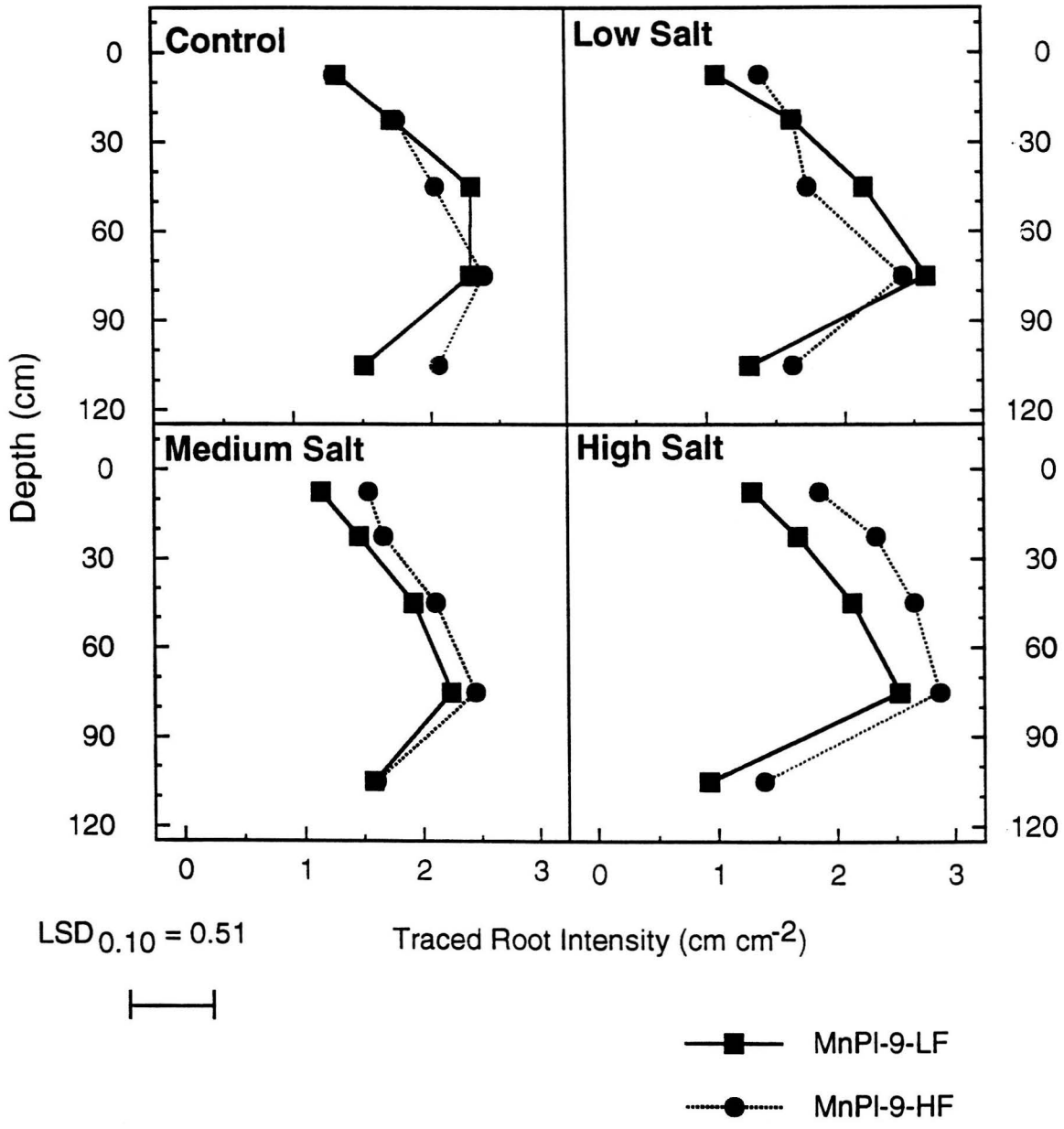


Figure 3. Cumulative mean traced root intensity after five harvests.

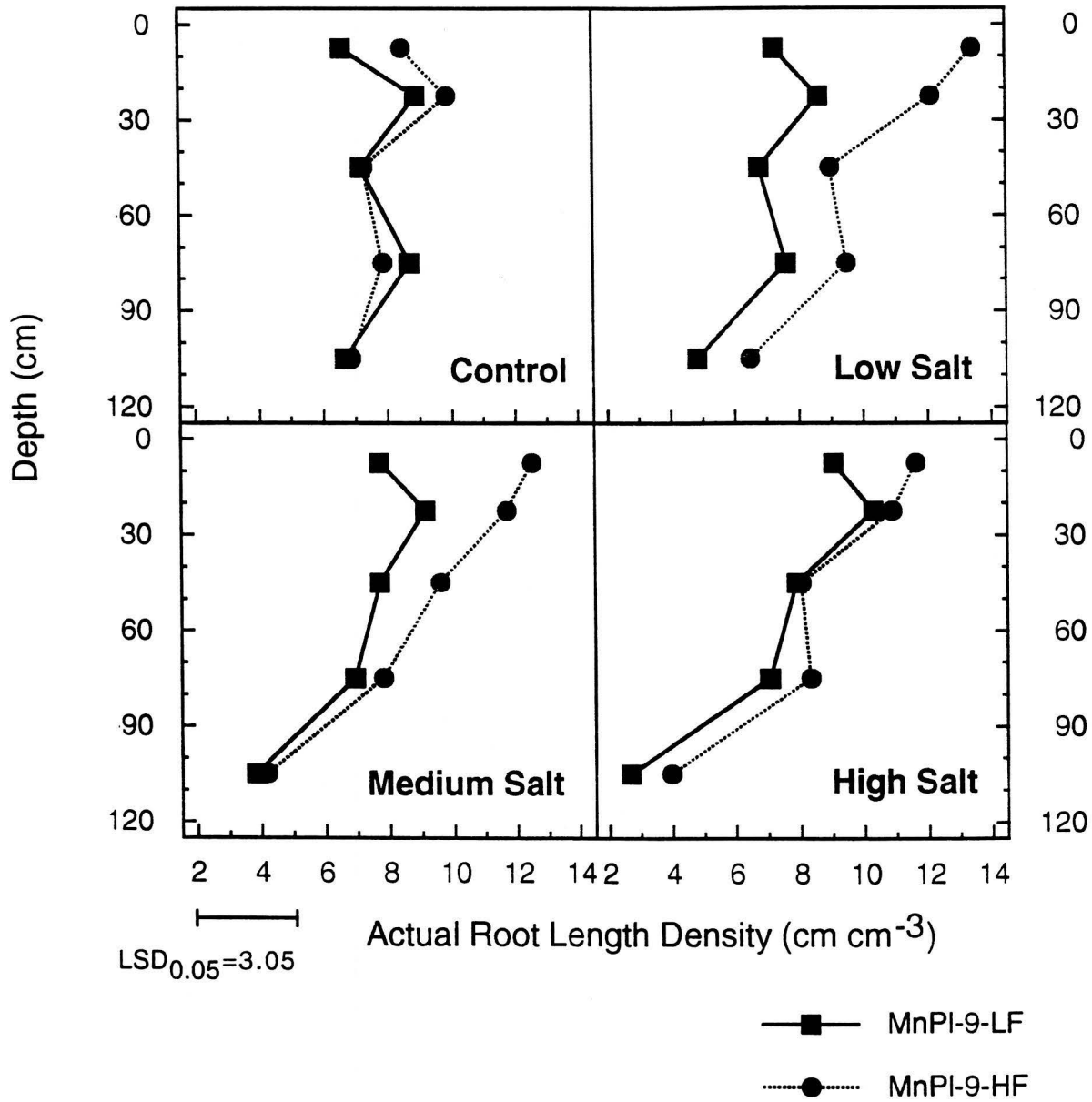


Figure 4. Mean actual root length density after five harvests.

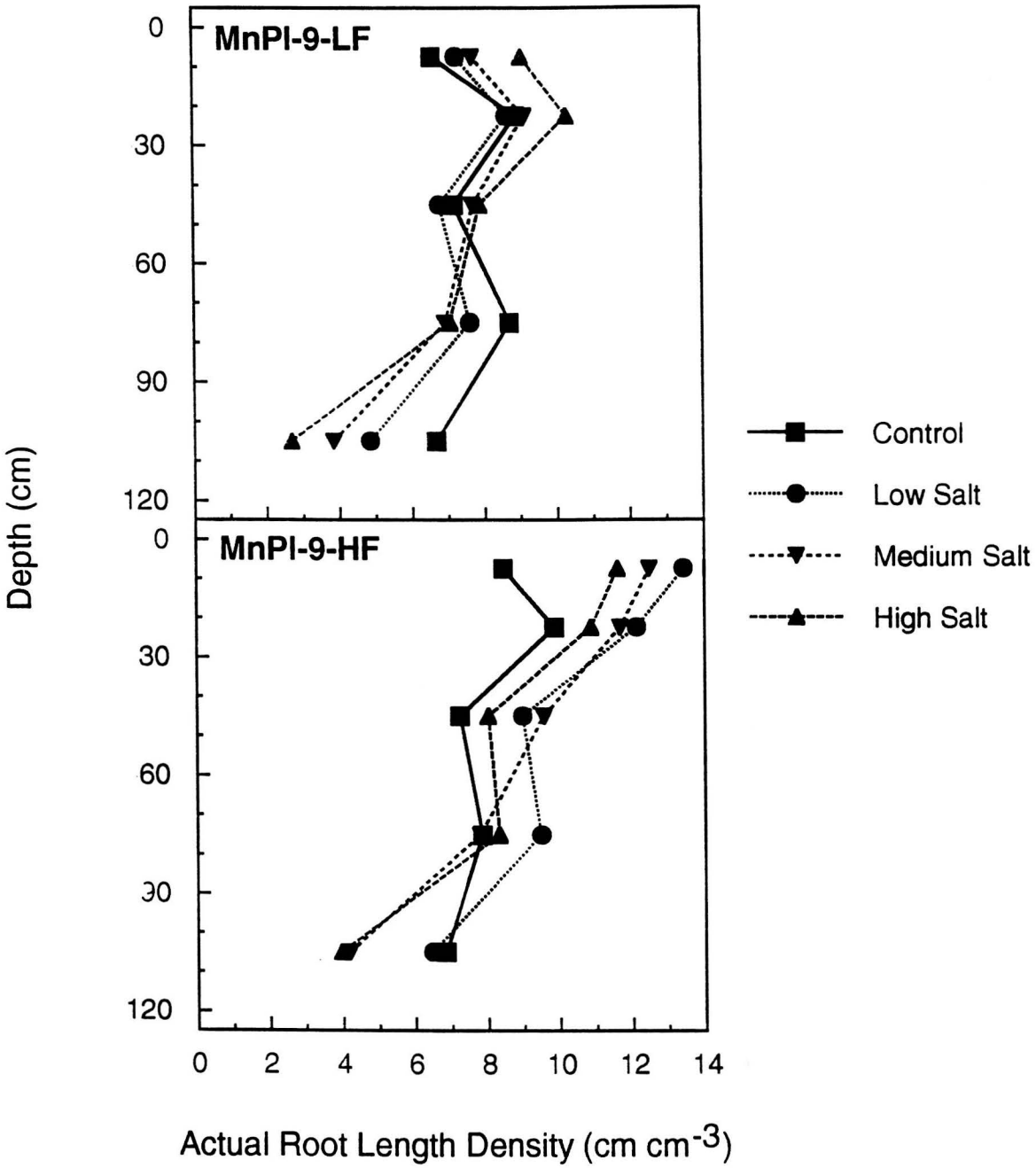


Figure 5. Mean actual root length density after five harvests.

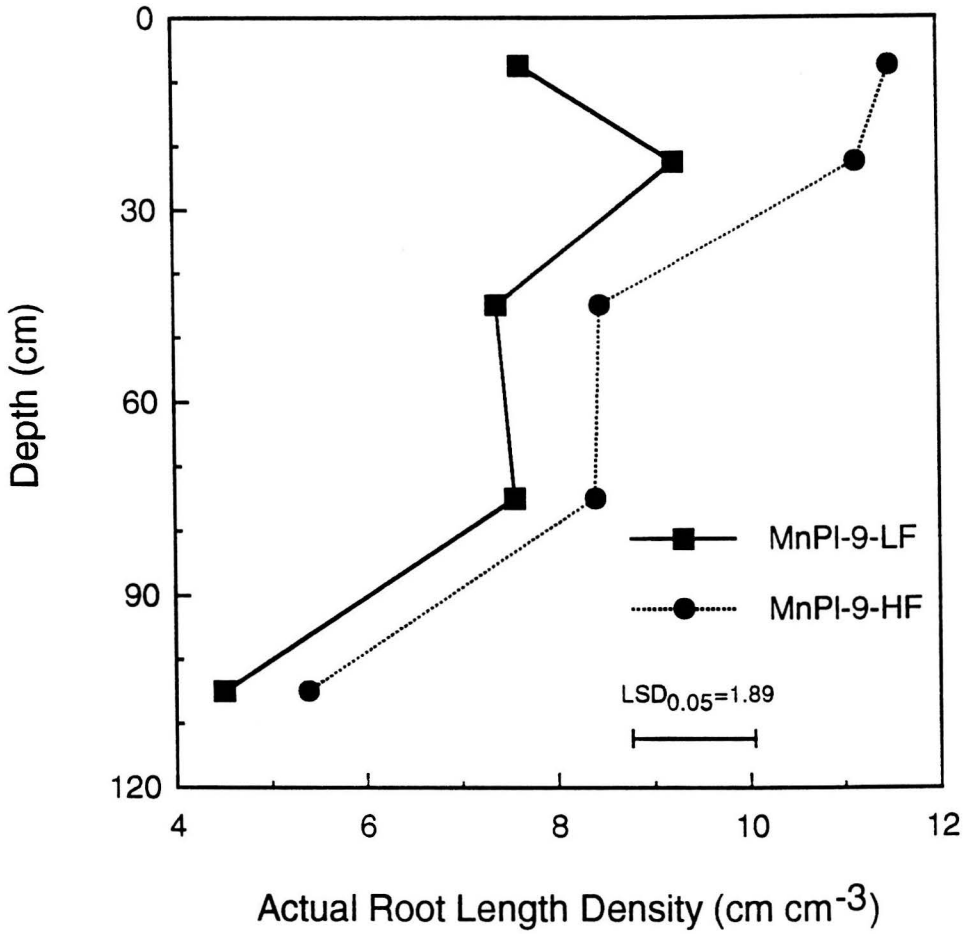
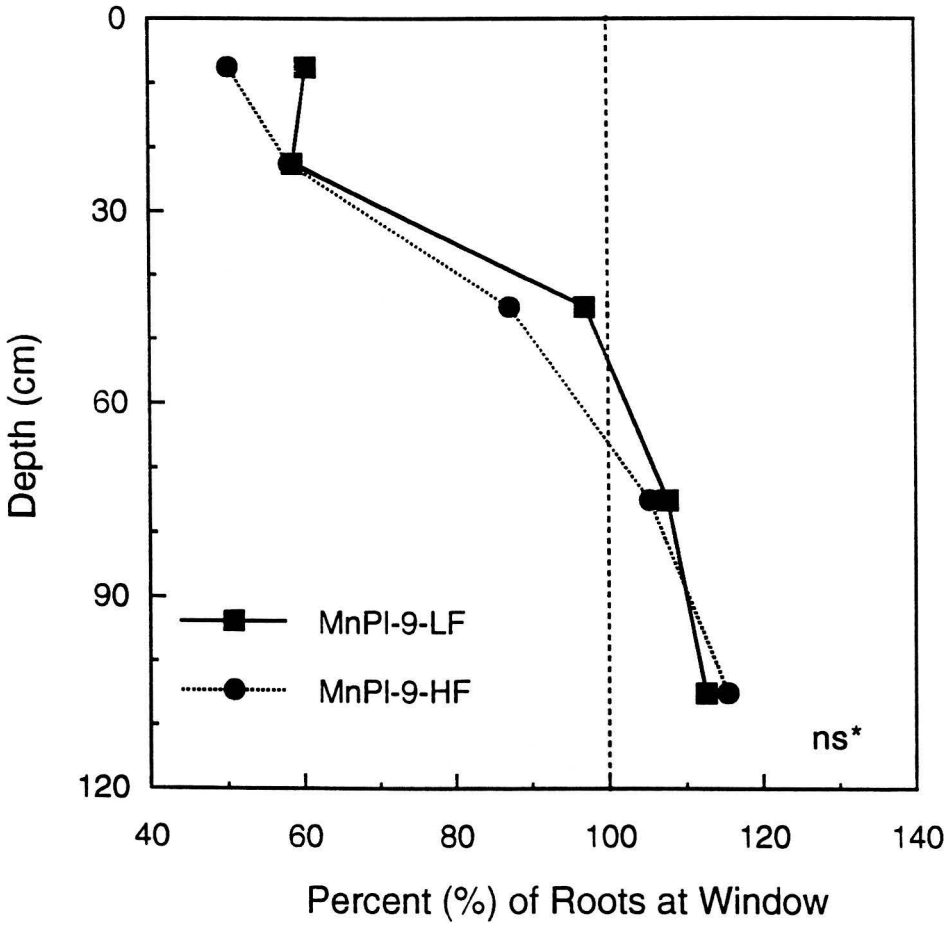


Figure 6. Comparison of mean actual root length densities between root types MnPl-9-LF and MnPl-9-HF across depth over salt.



* not statistically significant.

Figure 7. Proportion of roots concentrated at the root viewing window across depth and over salt treatments.

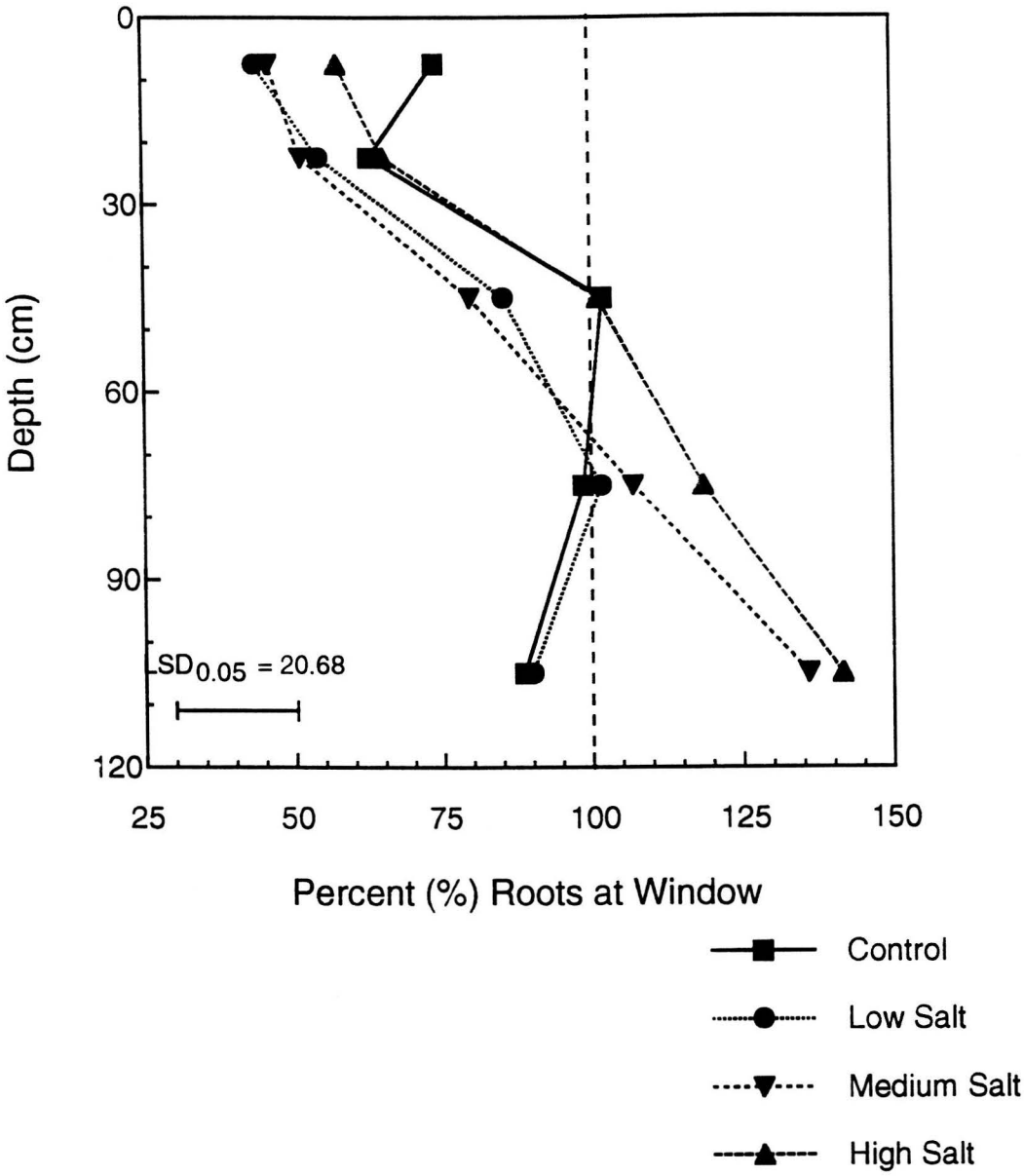


Figure 8. Mean proportion of roots concentrated at the root viewing window across root type by depth and salt treatment.

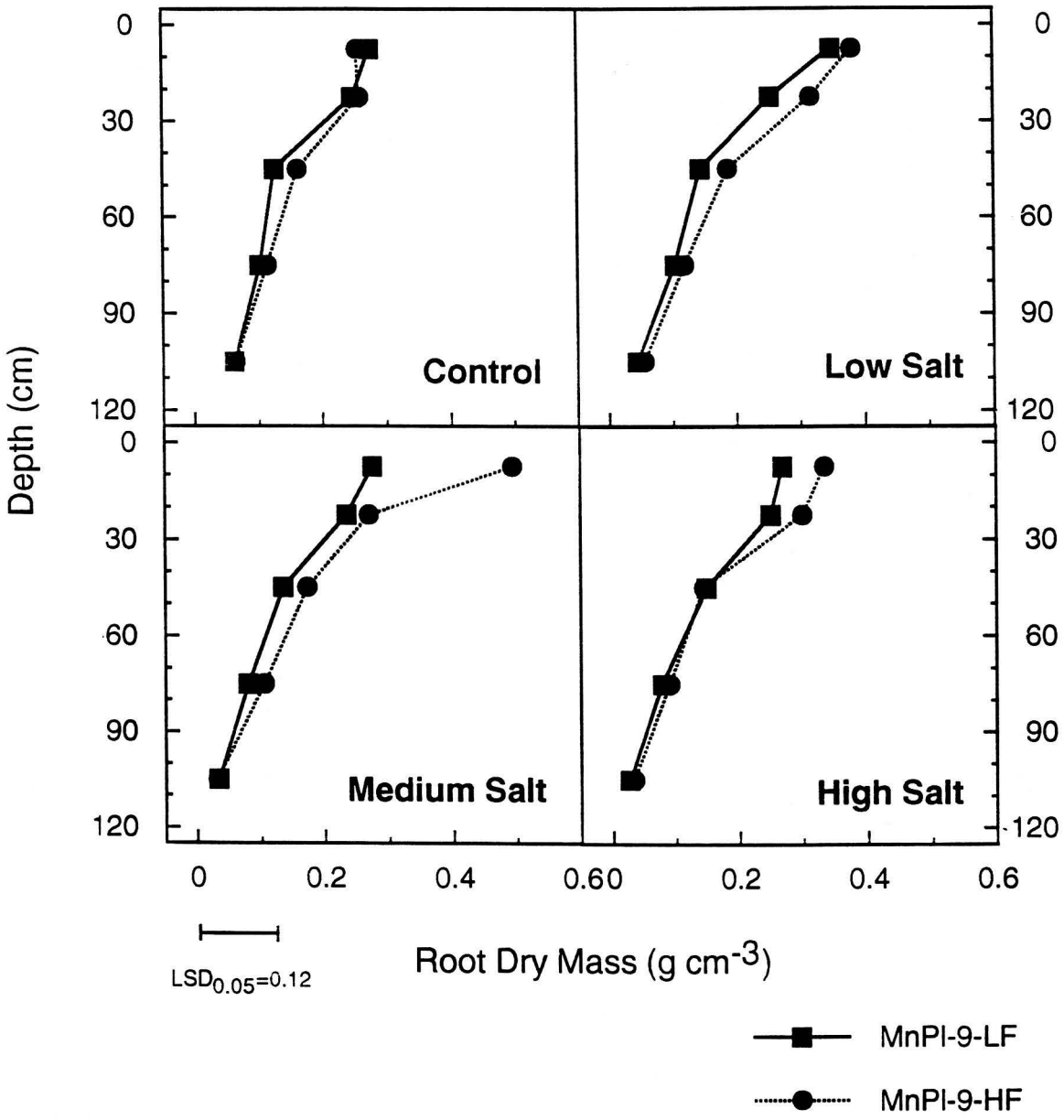


Figure 9. Mean root dry mass per volume of soil comparisons for all salt treatments.

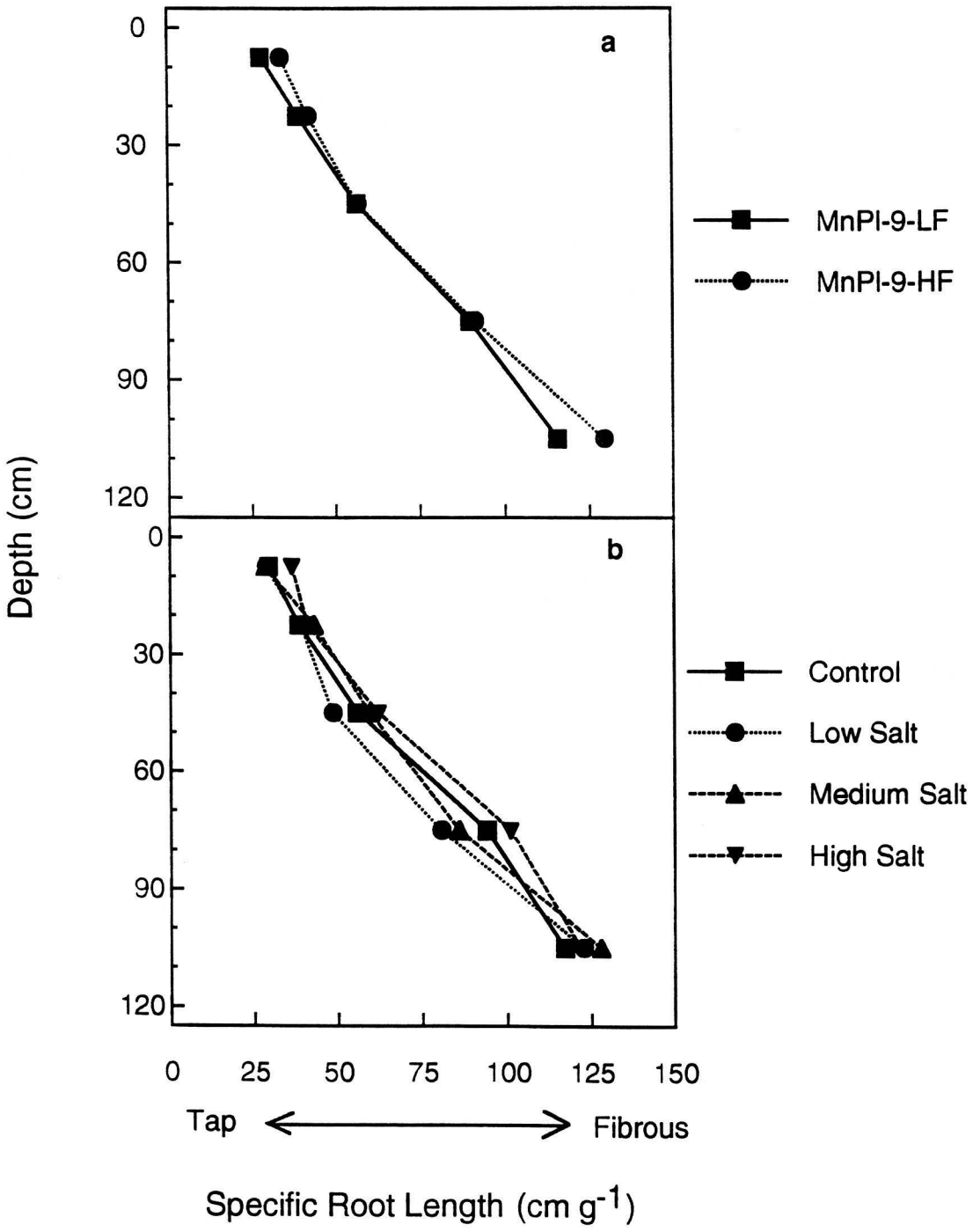


Figure 10. Specific root length, in cm root length per gram root weight, a; comparison between two root types across depth, b; across salt level.

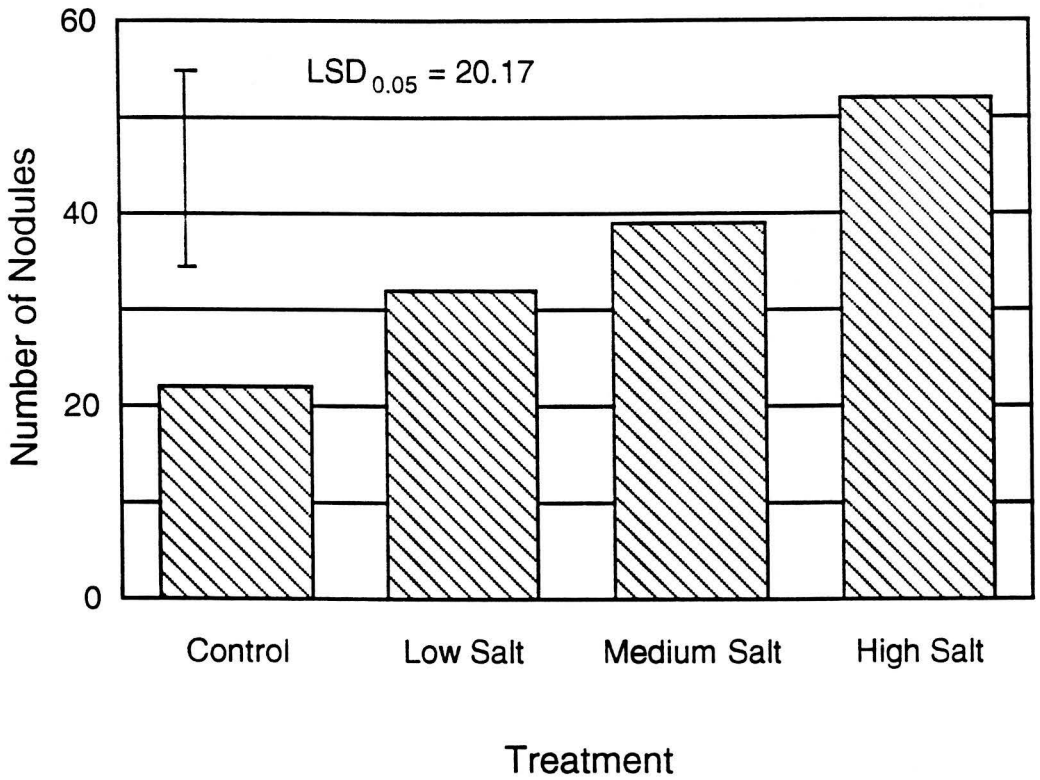


Figure 11. Mean number of nodules on alfalfa roots for four salt treatments.

CHAPTER III

ABSTRACT

Water use is an important aspect of alfalfa production in nonuniform saline soils. Evapotranspiration, water use efficiency and root water extraction were the useful parameters chosen to quantify water use by two alfalfa rooting subpopulations differing in fibrous root production. Seed of near isogenic lines of alfalfa, MnPI-9-LF and MnPI-9-HF, were planted in a control and three increasingly saline treatments. A single alfalfa plant was grown in each PVC cylinder in the greenhouse for a total of five harvests. Cylinders were weighed regularly to monitor water use, and saline irrigation water was applied at amounts equal to that lost by evapotranspiration. Time-domain reflectometry (TDR) probes were packed into three replications of each rooting subpopulation of the control and high salt treatments, and changes in soil water content of five depths were monitored throughout the study. In the highest salinity treatment, cumulative evapotranspiration was reduced by 5% for the low fibrous subpopulation relative to the control, and by less than 1% for the high fibrous subpopulation. Irrigation frequency was increased for the high fibrous root type ($p \leq 0.05$). TDR measurements were positively correlated ($r^2 = 0.90$; $p \leq 0.05$) with gravimetric samples for the upper four depths, but the high salinity of the lowest root zone segment (90 to 120 cm) resulted in an overestimation of soil water content in high salinity treatment. The low fibrous and high fibrous root

types differed in their ability to extract water in the most saline treatment. Patterns of water use suggest that the high fibrous root type was more efficient and ultimately better suited to production under saline irrigation without leaching.

INTRODUCTION

The ability of plant roots to extract water from saline soil environments will determine plant adaption to salinity. Water use and water use efficiency are useful parameters in comparing the performance of different plants in the production of shoot dry mass. Salinity usually decreases water use or evapotranspiration (ET) (Hanks et al. 1977; Francois 1981) and increases water use efficiency (WUE) (Bernstein and Francois, 1973; McCree and Richardson, 1987). However, we need a more specific understanding of the interaction of root morphology with water uptake by plant roots (Wraith and Baker, 1991), particularly in saline situations where sulfate salts predominate.

In situ monitoring of soil-water uptake by plant roots has been a promising and expanding area of research since the initial application of time-domain reflectometry (TDR) to measure soil volumetric water content by Topp et al. (1980). Measurements from TDR have compared well to those determined by gravimetric sampling (Topp and Davis, 1985). It is an advantageous method because of the minimal disruption of plants and surrounding soil, and the high spatial resolution that can be achieved within the root zone. Wraith and Baker (1991) have successfully utilized TDR to monitor root water uptake from sorghum plants, and were able to determine water uptake from various depths throughout the root zone.

Past research has shown that root growth and configuration influences the exploitation of available nutrients and water by plants (Gerard, 1978). Previous studies have also shown that species differ in their ability to extract water, however, detecting differences between cultivars is more difficult. Thomas et al. (1985) conducted a field study utilizing two barley cultivars to explain crop yields in terms of root growth and water extraction. They found that the barley cultivars, which were early and late maturing, differed in their efficiency of utilizing extracted water for biomass production.

Our objectives were to determine the water use and extraction of two alfalfa subpopulations with low and high fibrous rooting characteristics by using change of cylinder weight and TDR.

MATERIALS AND METHODS

Seed of alfalfa subpopulations MnPI-9-LF and MnPI-9-LF with low and high root systems were sown on 13 March 1995 in PVC cylinders with constructed nonuniform soil profiles of increasing salinity as described in Chapter II of this thesis. Seedlings were thinned to one plant per cylinder 21 days after emergence (DAE). The first saline irrigation was applied after plant establishment on 39 DAE. Plants were grown for five successive harvests and the experiment was ended on 13 October 1995, 211 DAE.

Cylinders were weighed every 2 to 3 days to monitor water use throughout the experiment. The composition of saline irrigation water was described in Chapter II, and irrigation was applied in amounts equal to that transpired by the plant when 50% of the extractable soil water (ESW) was depleted. Change in weight of the cylinder from its weight at container capacity was used as a basis for water application, and no compensation was made for added root or shoot weight during the study. However, the mean dry weight for combined shoot and root material in a given cylinder at Harvest 5 was approximately 11 g. The mean change in weight of a cylinder from container capacity to 50% ESW was 1.50 kg.

Time-domain reflectometry (TDR) probes were calibrated for five depths (0-15, 15-30, 30-60, 60-90 and 90-120 cm) before packing into rooting cylinders with the probe provided by the manufacturer (Environmental Sensors, Inc., San

Diego, CA). In addition, one TDR probe was calibrated to the Kidman fine sandy loam soil used in this study for a full wetting and drying cycle, and data compared satisfactorily to results of gravimetric samples and TDR probes from a cable tester (Tektronix, Beaverton, OR).

Time-domain reflectometry probes were packed into three cylinders each of the low fibrous and high fibrous root types for both the control and highest saline treatments. Probes were rod-shaped and measured 1.5 m, with an active length of 1.2 m. They were made of stainless steel, epoxy and high density plastic with a rubber tip at the bottom end for insertion into the soil, but the tip was removed for this study. Soil was packed tightly around probes to prevent air from interfering with measurements.

Comparisons Between Gravimetric and TDR Measurements

Volumetric water content measurements of each of the five depths were taken every 2 to 3 days by TDR throughout the experiment, as well as prior to water application and harvesting of plant shoots. Final TDR measurements were taken just prior to destructive analysis of the bulk soil in the cylinders following the fifth harvest. Soil samples from cylinders were dried for 24 hours at 105° for gravimetric soil water content determinations, and compared to final readings from TDR probes.

Linear regressions were calculated individually for the five depths for the TDR data and gravimetrically determined SWC. These correlations were highly

significant for all but the lowest segment of the soil profile which had an r^2 of 0.31 ($p \leq 0.06$) (Table 14).

In the 90 to 120 cm depth, measurements of SWC determined by TDR were often higher than the SWC at container capacity, especially for the high salinity treatment. Readings were also (0.2 m m^{-3}) above the theoretical levels possible for a Kidman fine sandy loam soil. The discrepancy between gravimetric SWC and TDR measurements can be attributed to the effect of salinity on TDR, which was highest in the 90 to 120 cm depth (Table 3).

To accurately use TDR to measure volumetric water content, it is necessary to calibrate probes independently for electrically-conducting soils (White et al., 1994), because the presence of electrolytes in the soil water affects the slope and intercept of commonly used calibration equations. However, in this experiment, salts not only were mixed with the soil prior to packing the TDR probes, but they were also applied in the irrigation water throughout the experiment. It was therefore not possible to satisfactorily calibrate the probes to the very high electrical conductivities that developed in the lowest section of the root profile.

Evapotranspiration and Water Use Efficiency

Water use (ET) was recorded at each irrigation application. Cumulative water use and shoot dry mass for each harvest period was used to calculate

water use efficiency (WUE). Both ET and WUE were analyzed for each harvest and for cumulative water use and cumulative yield.

Water Extraction

The amount of water extracted from each depth in the soil for a representative irrigation period just prior to each harvest was calculated from TDR data for SWC and ET of each cylinder. Evapotranspiration was measured as the difference between the weight of cylinders and their known weight at container capacity. The volume of water in each cylinder at container capacity was apportioned to the five depths based on the amount of air-dried soil packed into each depth. The amount of water extracted from each depth was based on TDR determinations of volumetric water content, except for the 90 to 120 cm depth, which was calculated by difference. Data were converted to percentage of water extracted per depth to facilitate comparisons.

Statistical Analysis

The experiment was designed as a split-plot arranged in a randomized complete block with six replications. Data from all procedures were compared within and between treatments using analysis of variance (ANOVA), and student's t-test (Minitab Inc. 1992). Response and interactions of nitrogen level, salt treatment, root type and, when applicable, depth, were tested using the

pooled residual as the error term. Significance was determined by p-values and the least significance differences (LSD) between means were calculated.

RESULTS AND DISCUSSION

Evapotranspiration and Water Use Efficiency

In conditions of zero leaching, water is removed from the soil only by evapotranspiration (ET). Statistical analysis revealed that ET did not differ between the low fibrous and high fibrous root subpopulations for the cumulative amount of water applied throughout the study ($p \leq 0.28$) or for the period between Harvests 4 and 5 ($p \leq 0.39$). Cumulative ET correlated well with cumulative yield ($r^2=0.98$) ($p \leq 0.05$). Also, although nonsignificant, ET of the high fibrous subpopulation was highest for the low salinity treatment, then decreased with higher salinity treatments, remaining at least as high as the control. This is the same trend that was seen for the high fibrous subpopulation for both yield and RLD.

Previous container and field studies have shown that ET decreases with an increasingly saline root zone (Lunin and Gallatin, 1965; Bingham and Garber, 1970; Hanks et al., 1977; Francois, 1981). Francois (1981) also found that evapotranspiration decreased with yield, but only when significant differences occurred between salinity treatments. This study shows that the growth of alfalfa with a capacity for fibrous root production is stimulated by moderate sulfate-based salinity.

Statistical analysis showed that both ET and WUE differed significantly for alfalfa harvests ($p \leq 0.0$, $p \leq 0.001$) which is to be expected as root systems

developed. There was a significant interaction between root type and harvest number ($p \leq 0.05$) for WUE, indicating that the effect of root type was not the same for each harvest. However, WUE of the high fibrous root subpopulation was significantly higher ($p \leq 0.07$) than the low fibrous subpopulation for all salinity treatments by the fifth harvest.

There were no significant differences between salinity treatments for cumulative ET ($p \leq 0.20$) or cumulative WUE ($p \leq 0.60$). However, while the WUE of most treatments declined for successive harvests, the decline in WUE of the high fibrous subpopulation stopped and values began to increase for saline treatments by the fourth or fifth harvest (Table 15).

Irrigation Frequency

The number of saline irrigation applications were statistically tested for each harvest (Table 15). For the fourth harvest, the two rooting subpopulations significantly differed ($p \leq 0.05$), with the high fibrous root type receiving more frequent irrigations than the low fibrous root type. The high fibrous root type depleted container capacity to 50% ESW faster than the low fibrous root type, and thus required irrigation more frequently.

The high fibrous root type had more root growth concentrated in the upper, lower salinity, portion of the root zone and so was able to extract available water from this region more efficiently. Thus irrigations were essentially recharging the upper portion of the root zone. Although this ability to

uptake water is advantageous, it also accelerates the accumulation of salinity through an increase in water use. Although not significant, comparison of final values of soil salinity for the low and high fibrous root types for the medium and high salinity treatments show that more salinity accumulated in the four upper portions of the root zone of the high fibrous subpopulation (Table 3).

At the fifth harvest, the low and medium salinity treatments had the most frequent irrigations, while the control and highest salinity treatments received the least ($p \leq 0.06$). Francois (1981) also reported a decrease in the frequency of irrigations with increasing salinity and attributed this decline to differences in yields. As plant growth is adversely affected by salinity, less water is required for evapotranspiration needs.

Water Extraction

Soil Water Content

Soil water content fluctuations based on TDR for each depth are shown from emergence through termination of the experiment for representative samples of the low fibrous and high fibrous subpopulations in the control (Fig. 12) and high salinity treatments (Fig. 13). Similar data for all 12 cylinders that had TDR probes are included in Appendix B. In Figure 12, the 0 to 15 cm depth shows the greatest fluctuation in SWC. From 0 DAE to approximately 25 DAE there is little change in SWC in the 15 to 30, 30 to 60, 60 to 90, or 90 to 120 cm

depths of both root types, probably because plant roots were still proliferating into these regions during this time.

Figure 13 compares the SWC of the low and high fibrous root types of the high salinity treatment. The series of graphs show a very similar pattern to that of the control in terms of activity in the 0 to 15 cm depth, and in the timing of root proliferation into the lower depths; however, there are important differences. The SWC of the high salinity treatment is reduced more frequently, but in the 90 to 120 cm depth, SWC of the high salinity treatment, indicates little or no water extraction. Thus it appears that less water extraction was impaired in the bottom 90 to 120 cm in the high than the low fibrous treatment.

Water Extraction

Water extraction as determined by TDR differed significantly ($p \leq 0.04$) for the control and high salinity treatments for the second and third harvest ($p \leq 0.04$). However, the amount of water extracted by the two root subpopulations was not significantly different ($p \leq 0.15$). The fact that more water was extracted by the high salt treatment suggests that the higher water potential of the soil solution stimulated root development; in Chapter II, it was seen that root length density increased with salinity (Fig. 4).

In the high salinity treatment the high fibrous root type was able to extract a larger percentage of water from the upper 90 cm of soil than the low fibrous

root type from Harvest 3 (Table 17). Water uptake is considered to be a function of root length (Vetterlein, 1993).

It is clear from Figure 4b and c of Chapter II that the proliferation of roots by the high fibrous subpopulation was greater in the upper root zone, and from Table 15, that ET was higher ($p \leq 0.25$) for the high fibrous root subpopulation. This is the difference in root water uptake capability of the two isogenic root types that we expected to be expressed under salinity stress. In the most saline portion of the root zone, 90 to 120 cm, there was no measurable water extraction from this depth by Harvest 5. This indicates that the solute potential of the soil solution in this region had been sufficiently high to inhibit water uptake.

In the control treatment rooting differences also expressed: the high fibrous subpopulation extracted water more readily from higher in the root profile, as would be expected from RLD (Fig. 6, Ch. II). The added root competition in the upper regions of the root zone for the high fibrous root type increased the rate of water depletion and this means that in a production system, as in our greenhouse study, irrigations must be shorter but more frequent as roots become concentrated in the upper, least saline portion of the root profile.

CONCLUSIONS

The salinity levels utilized in this study caused evapotranspiration and water use efficiency for the two rooting subpopulations to peak and begin to decline by the third or fourth harvest. The frequency of irrigation applications was increased for the high fibrous root type, indicating that it extracted water more efficiently from the root zone. The soil water contents also appeared to be affected by salinity, and the amount of water extracted in the bottom of the root zone was lower for the higher fibrous root type than for the low fibrous root type. Water extraction for the high fibrous root type increased with moderate salinity because it was able to extract a greater percentage of water from the upper portion of the root zone than the low fibrous root type. This supports the further investigation and use of alfalfa with higher fibrous roots in moderately saline field conditions with zero leaching.

REFERENCES

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.

Brown, S.C., P.J. Gregory, and A. Wahbi. 1987. Root characteristics and water use in mediterranean environments. *In* Drought tolerance in winter cereals. Ed. J.P. Srivastava, E. Porceddu, E. Acevedo and S. Varma. pp. 275-283. Wiley, Chichester, New York, Toronto.

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

Gerard, C.J. 1978. Root growth along plexiglas surfaces by sugarcane under different soil salinity conditions. Ag. J. 70: 639-643.

Hanks, R.J., T.E. Sullivan, and V.E. Hunsaker. 1977. Corn and alfalfa production as influenced by irrigation and salinity. Soil Sci. Soc. Am. J. 41:606-610.

Lunin, J., and M.H. Gallatin. 1965. Zonal salinization of the root system in relation to plant growth. Soil Sci. Soc. Am. Proc. 29:608-612.

McCree, K.J. and S.G. Richardson. 1987. Salt increases the water use efficiency in water stressed plants. Crop Sci. 27:543-47.

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

Thomas, S. Fukai, and G.L. Hammer. 1995. Growth and yield response of barley and chickpea to water stress under three environments in south-east Queensland. II. Root growth and soil water extraction pattern. Aust. J. Agric. Res. 46: 34-48.

Topp, G.C., and J.L. Davis. 1985. Time-domain reflectometry (TDR) and its application to irrigation scheduling. Adv. Irrig. 3:107-127.

Topp, G.C., J.L. Davis, and A.P. Annan. 1980. Electromagnetic determination of soil water content: Measurements in coaxial transmission lines. Water Resour. Res. 16:574-582.

Vetterlein, D., H. Marschner, and R. Horn. 1993. Microtensiometer technique for in situ measurement of soil matric potential and root water extraction for a sandy soil. Plant and Soil. 149:263-273.

White I., J.H. Knight, S.J. Zegelin, and G.C.Topp. 1994. Comments on 'Considerations on the use of time-domain reflectometry (TDR) for measuring soil water content' by W.R. Whally. Euro. J. Soil Sci. 45:503-508.

Wraith, J.M. and J. M. Baker. 1991. High-resolution measurement of root water uptake using automated time-domain reflectometry. Soil Sci. Sci. Am. J. 55:928-932.

Table 12. Comparison of gravimetric soil content and volumetric water content from TDR determined at the time of destructive sampling.

Treatment	Root Type	Meausurement Method	Gravimetric Soil Water Content					
			0-15	15-30	30-60	60-90	90-120	Container Capacity
					%			%
Control	MnPI-9-LF	gravimetric	7.4	8.6	10.0	10.0	13.0	21.1
		TDR	5.8	10.1	10.2	10.6	25.0	
	MnPI-9-HF	gravimetric	9.4	11.1	14.1	15.0	16.7	21.3
		TDR	6.6	13.2	14.0	16.2	32.2	
High Salt	MnPI-9-LF	gravimetric	9.5	13.0	13.2	13.5	18.8	21.8
		TDR	6.7	13.8	12.7	15.4	51.2	
	MnPI-9-HF	gravimetric	8.3	9.9	10.4	12.5	18.1	20.7
		TDR	5.9	13.6	12.6	15.6	52.8	
		r^2	0.862	0.721	0.79	0.742	0.313	
		p-value	0.0001	0.0005	0.0001	0.0003	0.0587	

Table 13. Mean evapotranspiration prior to each harvest and water use efficiency of two alfalfa rooting subpopulations.

Treatment	Root type	Harvest Number 1	2	3	4	5	Cumulative
		Evapotranspiration (L)					
Control	low fibrous	2.29	3.93	4.42	4.05	3.30	17.98
	high fibrous	1.88	3.48	4.22	4.58	3.99	18.14
Low Salt	low fibrous	2.34	3.83	4.54	3.78	3.66	18.16
	high fibrous	2.73	4.23	4.73	5.29	4.53	21.46
Medium Salt	low fibrous	2.65	3.13	4.10	3.28	4.19	17.35
	high fibrous	2.82	4.33	4.22	4.77	4.12	20.24
High Salt	low fibrous	2.27	3.82	3.83	3.94	3.24	17.09
	high fibrous	2.37	3.90	4.23	3.99	3.61	18.10
		Water Use Efficiency (g L^{-1})					
Control	low fibrous	2.82	2.76	2.10	2.22	2.32	2.31
	high fibrous	2.99	2.49	2.23	2.11	2.13	2.30
Low Salt	low fibrous	2.60	2.46	2.02	2.24	2.17	2.21
	high fibrous	2.36	2.33	2.15	2.17	2.57	2.28
Medium Salt	low fibrous	2.69	2.38	2.05	2.03	1.98	2.16
	high fibrous	2.12	2.13	2.44	2.44	2.61	2.35
High Salt	low fibrous	3.09	2.10	2.38	2.00	2.19	2.23
	high fibrous	2.55	1.97	2.06	2.47	2.71	2.29

Table 14. Mean number of irrigations during each harvest period of two alfalfa rooting subpopulations.

Treatment	Root type	Harvest Number 1	2	3	4	5
Control	low fibrous	1.3	3.0	3.8	3.3	2.8
	high fibrous	1.0	3.0	3.7	3.8	3.3
Low Salt	low fibrous	1.3	3.5	3.8	3.3	3.3
	high fibrous	1.7	3.5	3.8	4.3	4.2
Medium Salt	low fibrous	1.7	3.0	3.7	2.8	3.7
	high fibrous	1.8	3.3	4.0	3.8	4.0
High Salt	low fibrous	1.2	3.0	3.3	3.0	2.8
	high fibrous	1.2	4.2	3.3	3.5	3.2
	LSD	NS	NS	NS	1.5 [a]	1.3 [b]

NS is not statistically significant.

[a] least significant difference comparing root type at a fixed salt level, at the 0.05 level.

[b] least significant difference comparing salt level at a fixed root type, at the 0.10 level.

Table 15. The percent of water extracted from each depth for all five harvests for a representative low and high fibrous root type of the control and high salt treatment.

Treatment	Root type	Depth	Amount of Water Extracted				
			Harvest Number				
			1	2	3	4	5
		cm			%		
Control	MnPI-9-LF	0-30	60.8	49.6	42.2	43.7	48.5
		30-60	20.1	20.0	20.4	17.4	16.5
		60-90	8.5	12.7	18.5	17.4	11.0
		90-120	10.6	17.8	18.9	21.5	23.9
	MnPI-9-HF	0-30	50.2	46.3	51.5	50.3	52.5
		30-60	21.8	19.0	11.8	12.9	13.3
		60-90	10.7	16.9	10.1	9.5	9.4
		90-120	17.6	17.8	26.5	37.3	24.8
High Salt	MnPI-9-LF	0-30	41.2	47.8	45.0	52.5	43.4
		30-60	17.0	21.8	16.1	13.9	35.5
		60-90	13.8	21.5	6.8	7.2	21.1
		90-120	28.0	8.9	21.2	26.4	0.0
	MnPI-9-HF	0-30	50.3	53.5	43.0	74.4	39.7
		30-60	16.9	16.5	18.4	24.3	36.4
		60-90	5.3	5.3	16.8	24.5	23.8
		90-120	27.4	24.7	21.8	16.3	0.0

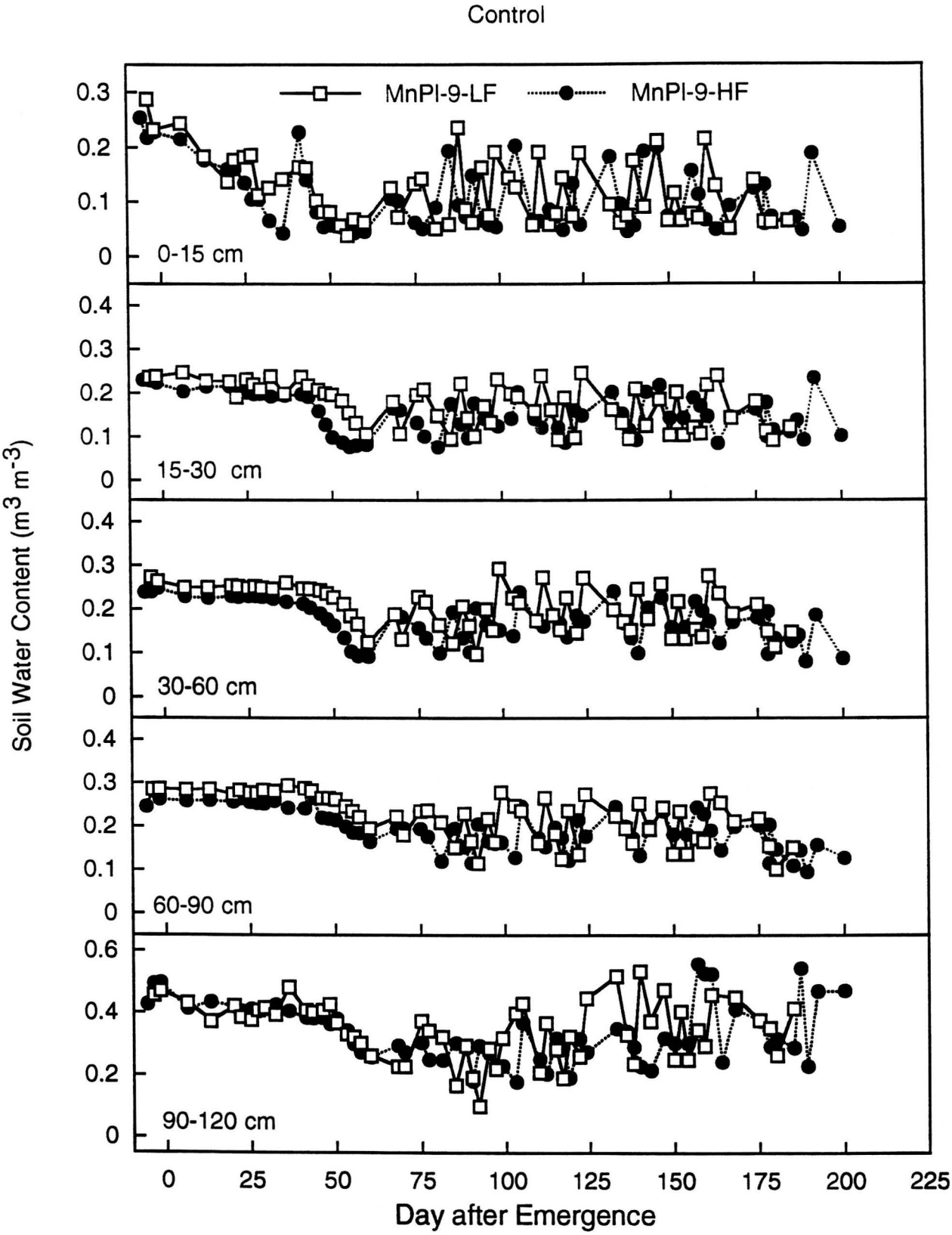


Figure 12. Comparison of soil water content from TDR for five depths for individual cylinders of the low and high fibrous subpopulation of the control.

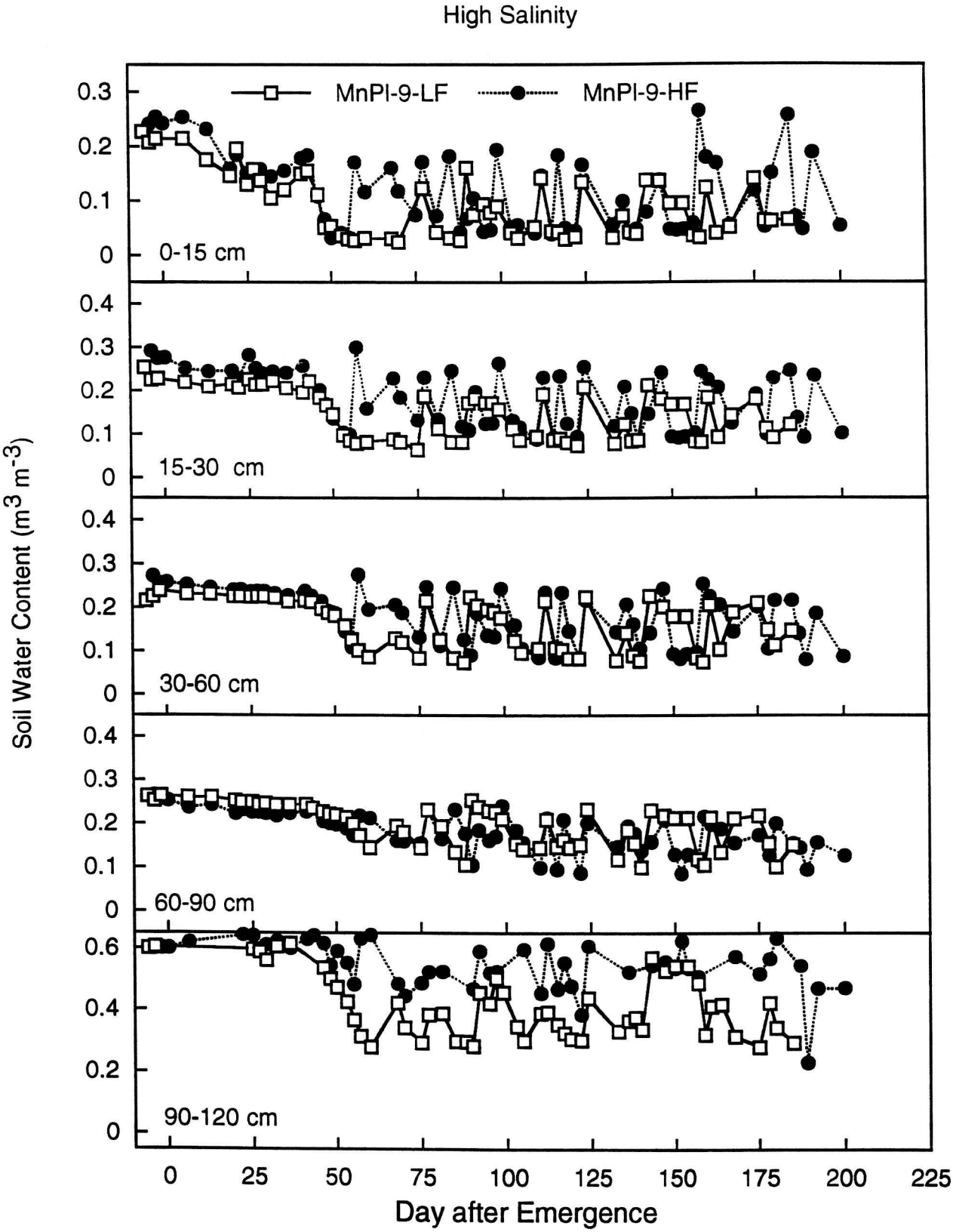


Figure 13. Comparison of soil water content from TDR for five depths for individual cylinders of the low and high fibrous subpopulation of the high salt treatment.

CONCLUSIONS

Our objectives were to study the yield, root growth and water extraction of two subpopulations of alfalfa which differed in their ability to produce fibrous roots under nonuniform saline conditions. We may make several significant conclusions from this study.

First, the moderately saline conditions utilized did not significantly reduce yields for either of the two rooting subpopulations. Thus, alfalfa plants can be grown productively in sulfate salinity with EC_e's that range as high as 23 dS m^{-1} without yield decrement.

Secondly, the high fibrous rooting subpopulation had more root length in the upper, least saline portion of the root zone. This avails the plant of a less negative water potential in the soil solution, and increases its ability to extract water from this region. Therefore, the plant can avoid salts in the root zone through its more plastic root morphology.

Finally, water extraction in the less saline portions of the root zone was higher for the high fibrous root type than for the low fibrous root type, but irrigation frequency was also higher. Therefore, for alfalfa production under minimal leaching or non-leaching conditions, irrigation will need to occur more frequently to maximize production.

REFERENCES

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 Conference, 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.

Bernstein, L., L.E. Francois, R.A. Clark. 1975. Minimal leaching with varying root depths of alfalfa. Soil Sci. Soc. Am. Proc. 39:113-119.

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.

Bower C.A., G. Ogata, and J.M. Tucker. 1969. Rootzone salt profiles and alfalfa growth as influenced by irrigation water salinity and leaching fraction. Ag. J. 61:783-785.

Dudley, L.M., Hanks, R.J., MacAdam, J.W., Mace, R.W., 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. Sta. Res. Rpt. 150.

Eissa, A.M., J.N. Jenkins, and C.E. Vaughan. 1983. Inheritance of seedling root length and relative root weight in cotton. Crop Sci. 23:1107-1111.

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

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.

Johnson, D.W., S.E. Smith, and A.K. Dobrenz. 1992. Selection for increased forage yield in alfalfa at different NaCl levels. Euphyt. 60:27-35.

Jury, W.A., H. Frenkel, H. Fluhler, D. Devitt, and L.H. Stolzy. 1978. Use of saline irrigation waters and minimal leaching for crop production. Hilgardia 46:169-191.

Lonkerd, W.E., C.F. Ehlig, and T.J. Donovan. 1979. Salinity profiles and leaching fractions for slowly permeable irrigated field soils. Soil Sci. Soc. Am. J. 43:287-289.

Lunin, J., and M.H. Gallatin. 1965. Zonal salinization of the root system in relation to plant growth. Soil Sci. Soc. Am. Proc. 29:608-612.

Lynch, J., 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.

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

Munns, R. 1993. Physiological processes limiting plant growth in saline soils: some dogmas and hypotheses. Plant, Cell, Env. 16:15-24.

Noble, C.L., G.M. Halloran and D.W. West. 1984. Identification and selection for salt tolerance in Lucerne. Aust. J. Agric. Res. 35:239-252.

Rhoades, J.D., F.T. Bingham, J. Letey, A.R. Dedrick, M. Bean, G.J. Hoffman, W.J. Alves, R.V. Swain, P.G. Pacheco and R.D. Lemert. 1988. Reuse of drainage water for irrigation: results of Imperial Valley study. Hilgardia 56:1-17.

Rhoades, J.D., F.T. Bingham, J. Letey, G.J. Hoffman, A.R. Dedrick, P.J. Pinter, and J.A. Replogle. 1989. Use of saline drainage water for irrigation: results of Imperial Valley study. Ag. Water Manage. 16:25-36.

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

Smith R.J., and N.H. Hancock. 1986. Leaching requirement of irrigated soils. Agric. Water Manage. 11:13-22.

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

Snapp, S.S., and C.S. Shennan. 1992. Effects of Salinity on Root Growth and Death Dynamics of Tomato, *Lycopersicon esculentum* Mill. New Phyto. 121:71-79.

Thellier, C., K.M. Holtzclaw, J.D. Rhoades, and G. Sposito. 1990. Chemical effects of saline irrigation water on a San Joaquin Valley soil: II. field soil samples. J. Env. Qual. 19: 56-60.

Trimble, M.W., D.K. Barnes, G.H. Heichel, and C.C. Sheaffer. 1987. Forage yield and nitrogen partitioning responses of alfalfa to two cutting regimes and three soil nitrogen regimes. Crop Sci. 27:909-914.

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.

Waisel, Y., and S.W. Breckle. 1987. Differences in responses of various radish roots to salinity. Plant and Soil 104:191-194.

Zartman, R.E., and M. Gichuru. 1984. Saline irrigation water: effects on soil chemical and physical properties. Soil Sci. 138:417-422.

Zhou, M., T.C. Tucker, M. Pessarakli, and J. A. Cepeda. 1992. Nitrogen fixation by alfalfa with two substrate nitrogen levels under sodium chloride stress. Soil Sci. Soc. Am. J. 56:1500-1504.

APPENDIX A. ANOVA TABLES

Table 16. ANOVA for final electrical conductivity of soil saturated paste extracts.

Source of Variation	df	MS	F	P
rep	2	9.22		
salt	3	87.17	42.60	0.001
nitrogen	1	1.15	0.56	0.800
salt x nitrogen	3	9.17	4.48	0.025
error (a)	14	2.05		
root type	1	1.02	0.11	0.900
salt x root type	3	0.69	0.08	0.900
root type x nitrogen	1	2.59	0.28	0.850
salt x nitrogen x root type	3	6.40	0.70	0.750
error (b)	16	9.16		
depth	4	1681.24	539.22	0.000
salt x depth	12	36.54	11.72	0.000
nitrogen x depth	4	7.30	2.34	0.060
salt x nitrogen x depth	12	8.89	2.85	0.002
root type x depth	4	11.69	3.75	0.006
salt x root type x depth	12	5.57	1.79	0.060
nitrogen x root type x depth	4	5.37	1.72	0.150
salt x nitrogen x root type x depth	12	3.50	1.12	0.300
error (c)	128	3.12		
total	239			

Table 17. ANOVA for yield of cumulative harvests.

Source of Variation	df	MS	F	P
rep	2	172.30		
salt	3	37.30	0.70	0.35
nitrogen	1	47.00	0.88	0.99
salt x nitrogen	3	46.60	0.87	0.30
error (a)	14	53.30		
root type	1	361.90	1.36	0.26
salt x root type	3	73.90	0.28	0.84
root type x nitrogen	1	130.80	0.49	0.49
salt x nitrogen x root type	3	104.30	0.39	0.76
error (b)	16	265.40		
total	47			

Table 18. ANOVA for yield of harvest five.

Source of Variation	df	MS	F	P
rep	2	25.28		
salt	3	6.96	1.12	0.60
nitrogen	1	0.35	0.06	0.60
salt x nitrogen	3	0.41	1.36	0.50
error (a)	14	6.19		
root type	1	61.07	2.33	0.15
salt x root type	3	4.19	0.16	0.92
root type x nitrogen	1	14.43	0.55	0.47
salt x nitrogen x root type	3	21.17	0.81	0.51
error (b)	16	26.26		
total	47			

Table 19. ANOVA for the cumulative traced root intensity.

Source of Variation	df	MS	F	P
rep	2	0.53		
nit/rep	3	1.21		
salt	3	0.55	0.96	0.45
error (a)	15	0.57		
root type	1	2.39	3.75	0.08
salt x root type	3	0.73	1.14	0.35
error (b)	20	0.64		
depth	4	10.07	58.45	0.00
salt x depth	12	0.45	2.64	0.00
root type x depth	4	0.22	1.29	0.28
salt x root type x depth	12	0.14	0.83	0.62
error (c)	160	0.17		
total	239			

Table 20. ANOVA for root length density.

Source of Variation	df	MS	F	P
rep	2	77.37		
nit/rep	3	10.82		
salt	3	6.35	0.90	0.50
error (a)	15	7.08		
root type	1	175.00	5.30	0.03
salt x root type	3	19.88	0.60	0.35
error (b)	20	33.04		
depth	4	198.70	48.92	0.00
salt x depth	12	14.23	14.23	0.00
root type x depth	4	19.42	19.42	0.01
salt x root type x depth	12	1.81	1.81	0.94
error (c)	158	4.11		
total	239			

Table 21. ANOVA for the low salt treatment, root length density.

Source of Variation	df	MS	F	P
rep	2	37.59		
nit/rep	3	4.83		
root type	1	145.42	5.890	0.08
error (a)	5	24.67		
depth	4	45.64	15.600	0.05
root type x depth	4	10.15	3.46	0.05
error (b)	38	2.93		
total	59			

Table 22. ANOVA for the medium salt treatment, root length density.

Source of Variation	df	MS	F	P
rep	2	9.93		
nit/rep	3	5.25		
root type	1	65.48	9.70	0.03
error (a)	5	6.76		
depth	4	80.71	37.61	0.00
root type x depth	4	9.07	4.23	0.02
error (b)	40	2.15		
total	59			

Table 23. ANOVA for percent roots concentrated at the PVC face.

Source of Variation	df	MS	F	P
rep	2	944.50		
nit/rep	3	804.30		
salt	3	4764.60	4.88	0.03
error (a)	15	976.84		
root type	1	944.50	0.63	0.45
salt x root type	3	1905.80	1.26	0.30
error (b)	20	1511.16		
depth	4	35223.70	65.60	0.00
salt x depth	12	2572.50	4.79	0.00
root type x depth	4	402.00	0.75	0.55
salt x root type x depth	12	734.50	1.37	0.18
error (c)	160	536.70		
total	239			

Table 24. ANOVA for root dry mass.

Source of Variation	df	MS	F	P
rep	2	0.046		
nit/rep	3	0.014		
salt	3	0.011	1.37	0.30
error (a)	15	0.008		
root type	1	0.063	1.68	0.25
salt x root type	3	0.008	0.22	0.85
error (b)	20	0.037		
depth	4	0.666	168.07	0.00
salt x depth	12	0.009	2.17	0.03
root type x depth	4	0.008	2.14	0.10
salt x root type x depth	12	0.007	1.70	0.10
error (c)	158	0.004		
total	239			

Table 25. ANOVA for specific root length.

Source of Variation	df	MS	F	P
rep	2	1261.80		
nit/rep	3	933.30		
salt	3	777.40	0.48	0.75
error (a)	15	1616.90		
root type	1	1452.90	0.84	0.35
salt x root type	3	566.10	0.33	0.70
error (b)	20	1732.60		
depth	4	68664.10	222.23	0.00
salt x depth	12	270.60	0.88	0.57
root type x depth	4	356.70	1.15	0.33
salt x root type x depth	12	270.90	0.88	0.57
error (c)	158	309.00		
total	239			

Table 26. ANOVA for tap root diameter.

Source of Variation	df	MS	F	P
rep	2	0.038		
salt	3	0.009	0.77	0.45
nitrogen	1	0.003	0.30	0.55
salt x nitrogen	3	0.035	3.08	0.09
error (a)	14	0.011		
root type	1	0.213	6.89	0.02
salt x root type	3	0.016	0.52	0.67
root type x nitrogen	1	0.017	0.56	0.46
salt x nitrogen x root type	3	0.012	0.38	0.77
error (b)	16	0.031		
total	47			

Table 27. ANOVA for number of nodules visible at the PVC window.

Source of Variation	df	MS	F	P
rep	2	339.40		
salt	3	1892.40	3.57	0.05
nitrogen	1	972.00	1.83	0.20
salt x nitrogen	3	706.90	1.33	0.30
error (a)	14	530.40		
root type	1	546.70	1.05	0.32
salt x root type	3	2191.80	1.40	0.28
root type x nitrogen	1	330.80	0.63	0.44
salt x nitrogen x root type	3	4104.40	2.62	0.09
error (b)	16	522.50		
total	47			

Table 28. ANOVA for the root:shoot ratio, utilizing cumulative yield.

Source of Variation	df	MS	F	P
rep	2	0.00116		
salt	3	0.00103	0.77	0.55
nitrogen	1	0.00834	6.21	0.03
salt x nitrogen	3	0.00128	0.95	0.40
error (a)	14	0.0013		
root type	1	0.00069	0.31	0.59
salt x root type	3	0.00033	0.14	0.93
root type x nitrogen	1	0.00027	0.12	0.74
salt x nitrogen x root type	3	0.00245	1.08	0.39
error (b)	16	0.00227		
total	47			

Table 29. ANOVA for the root:shoot ratio, utilizing yield from harvest five.

Source of Variation	df	MS	F	P
rep	2	0.01479		
salt	3	0.10464	1.37	0.30
nitrogen	1	0.16106	2.11	0.20
salt x nitrogen	3	0.01073	0.14	0.90
error (a)	14	0.07617		
root type	1	0.17635	2.63	0.12
salt x root type	3	0.02946	0.44	0.73
root type x nitrogen	1	0.26419	3.95	0.06
salt x nitrogen x root type	3	0.45736	0.83	0.00
error (b)	16	0.06694		
total	47			

Table 30. ANOVA for cumulative ET for five harvest periods.

Source of Variation	df	MS	F	P
rep	2	5.29		
salt	3	11.17	1.46	0.20
nit	1	4.60	0.60	0.55
nit x salt	3	8.50	1.11	0.25
error (a)	14	7.67		
root type	1	40.59	1.25	0.28
salt x root type	3	6.76	0.21	0.89
root type x nitrogen	1	16.73	0.51	0.48
root type x nitrogen x salt	3	11.40	0.35	0.79
error (c)	16	32.52		
total	47			

Table 31. ANOVA for cumulative WUE for five harvest periods.

Source of Variation	df	MS	F	P
rep	2	0.323		
salt	3	0.008	0.25	0.60
nit	1	0.019	0.63	0.50
nit x salt	3	0.031	1.03	0.25
error (a)	14	0.030		
root type	1	0.070	1.35	0.26
salt x root type	3	0.020	0.39	0.76
root type x nitrogen	1	0.050	0.97	0.34
root type x nitrogen x salt	3	0.086	1.66	0.22
error (c)	16	0.021		
total	47			

Table 32. ANOVA for ET from Harvest 4.

Source of Variation	df	MS	F	P
rep	2	0.34		
salt	3	1.50	1.39	0.40
nitrogen	1	0.00	0.00	0.99
salt x nitrogen	3	0.85	0.79	0.50
error (a)	14	1.08		
root type	1	2.59	0.77	0.39
salt x root type	3	0.52	0.15	0.93
root type x nitrogen	1	4.11	1.22	0.29
salt x nitrogen x root type	3	0.90	0.27	0.85
error (b)	16	3.38		
total	47			

Table 33. ANOVA for WUE from Harvest 5.

Source of Variation	df	MS	F	P
rep	2	0.78		
salt	3	0.11	0.40	0.75
nitrogen	1	0.00	0.02	0.90
salt x nitrogen	3	0.19	0.69	0.60
error (a)	14	0.27		
root type	1	1.39	3.83	0.07
salt x root type	3	0.40	1.09	0.38
root type x nitrogen	1	0.01	0.00	0.85
salt x nitrogen x root type	3	0.57	1.57	0.24
error (b)	16	0.36		
total	47			

Table 34. ANOVA for ET for all harvests.

Source of Variation	df	MS	F	P
rep	2	1.08		
nit/rep	3	1.67		
salt	3	2.29	1.53	0.50
error (a)	15	1.49		
root type	1	8.23	1.44	0.55
salt x root type	3	1.39	0.24	0.80
error (b)	20	5.71		
harvest	4	27.33	24.92	0.00
salt x harvest	12	0.52	0.48	0.93
root type x harvest	4	1.33	1.21	0.31
salt x root type x harvest	12	0.67	0.61	0.84
error (c)	158	1.10		
total	239			

Table 35. ANOVA for WUE for all harvests.

Source of Variation	df	MS	F	P
rep	2	1.38		
nit/rep	3	0.03		
salt	3	0.20	1.03	0.65
error (a)	15	0.19		
root type	1	0.03	0.07	0.90
salt x root type	3	0.08	0.22	0.80
error (b)	20	0.37		
harvest	4	1.68	4.98	0.00
salt x harvest	12	0.36	1.08	0.38
root type x harvest	4	0.82	2.43	0.05
salt x root type x harvest	12	0.31	0.92	0.53
error (c)	158	0.34		
total	239			

Table 36. ANOVA for number of irrigations during the fourth harvest period.

Source of Variation	df	MS	F	P
rep	2	0.19		
salt	3	0.83	0.66	0.50
nit	1	1.33	1.06	0.22
nit x salt	3	1.39	1.10	0.20
error (a)	14	1.26		
root type	1	6.75	4.38	0.05
salt x root type	3	0.25	0.16	0.92
root type x nitrogen	1	2.08	1.35	0.26
root type x nitrogen x salt	3	0.58	0.38	0.77
error (c)	16	1.54		
total	47			

Table 37. ANOVA for number of irrigations during the fifth harvest period.

Source of Variation	df	MS	F	P
rep	2	0.021		
salt	3	2.278	3.16	0.06
nit	1	0.000	0.00	0.99
nit x salt	3	0.722	0.95	0.20
error (a)	14	0.759		
root type	1	3.000	1.09	0.31
salt x root type	3	0.167	0.06	0.98
root type x nitrogen	1	1.330	0.48	0.50
root type x nitrogen x salt	3	1.056	0.38	0.77
error (c)	16	2.750		
total	47			

Table 38. ANOVA for water extraction by TDR for both treatments from Harvest 2.

Source of Variation	df	MS	F	P
rep	2	837.00		
salt	1	15743.00	20.47	0.04
error (a)	2	769.00		
root type	1	37888.00	4.11	0.15
salt x root type	1	19495.00	2.11	0.35
error (b)	4	9225.00		
depth	3	43069.00	51.47	0.00
salt x depth	3	3117.00	3.75	0.03
root type x depth	3	5421.00	6.48	0.00
salt x root type x depth	3	3756.00	4.49	0.01
error (c)	24	837.00		
total	47			

Table 39. ANOVA for water extraction by TDR for both treatments from Harvest 3.

Source of Variation	df	MS	F	P
rep	2	10102.00		
salt	1	10869.00	27.9	0.04
error (a)	2	389.00		
root type	1	20282.00	4.00	0.15
salt x root type	1	323.00	0.06	0.09
error (b)	4	5065.00		
depth	3	36123.00	18.41	0.00
salt x depth	3	3106.00	1.58	0.22
root type x depth	3	5125.00	2.61	0.08
salt x root type x depth	3	1851.00	0.94	0.44
error (c)	24	1962.00		
total	47			

APPENDIX B. SOIL WATER CONTENT OF INDIVIDUAL ROOOT CYLINDERS

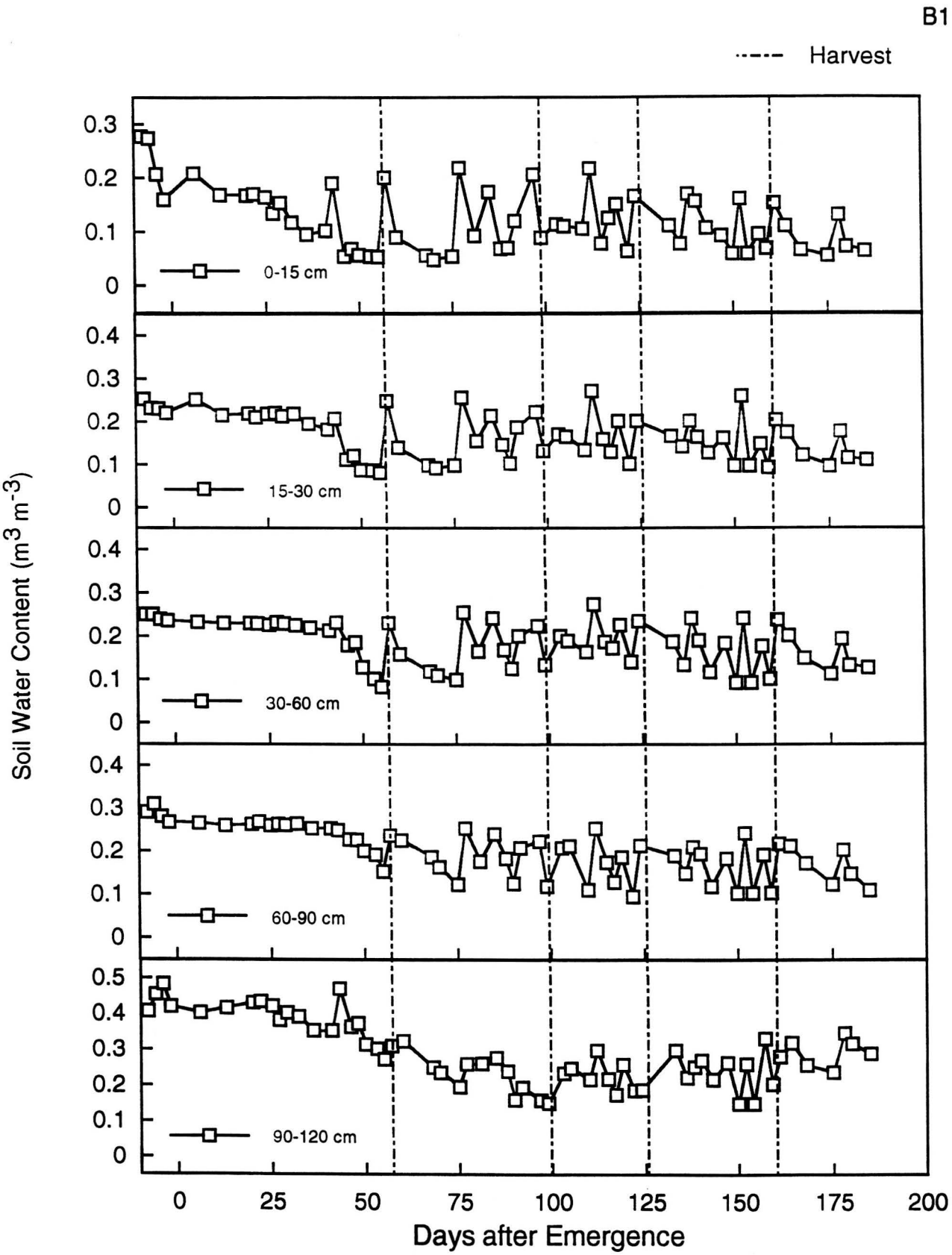


Figure 14. Soil water content from TDR for five depths of a control, low fibrous treatment (tube 4).

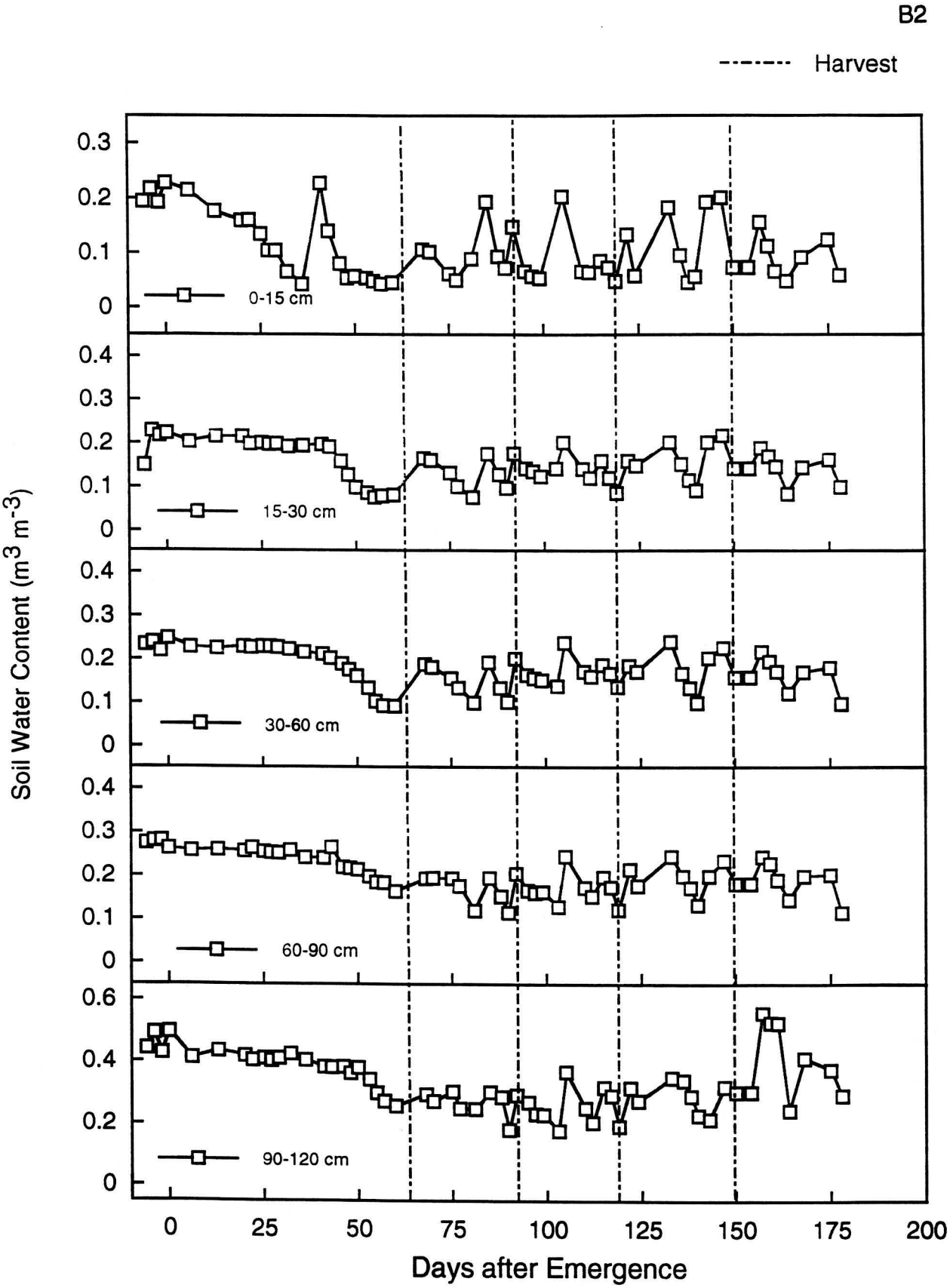


Figure 15. Soil water content for five depths of the control, low fibrous treatment (tube 5).

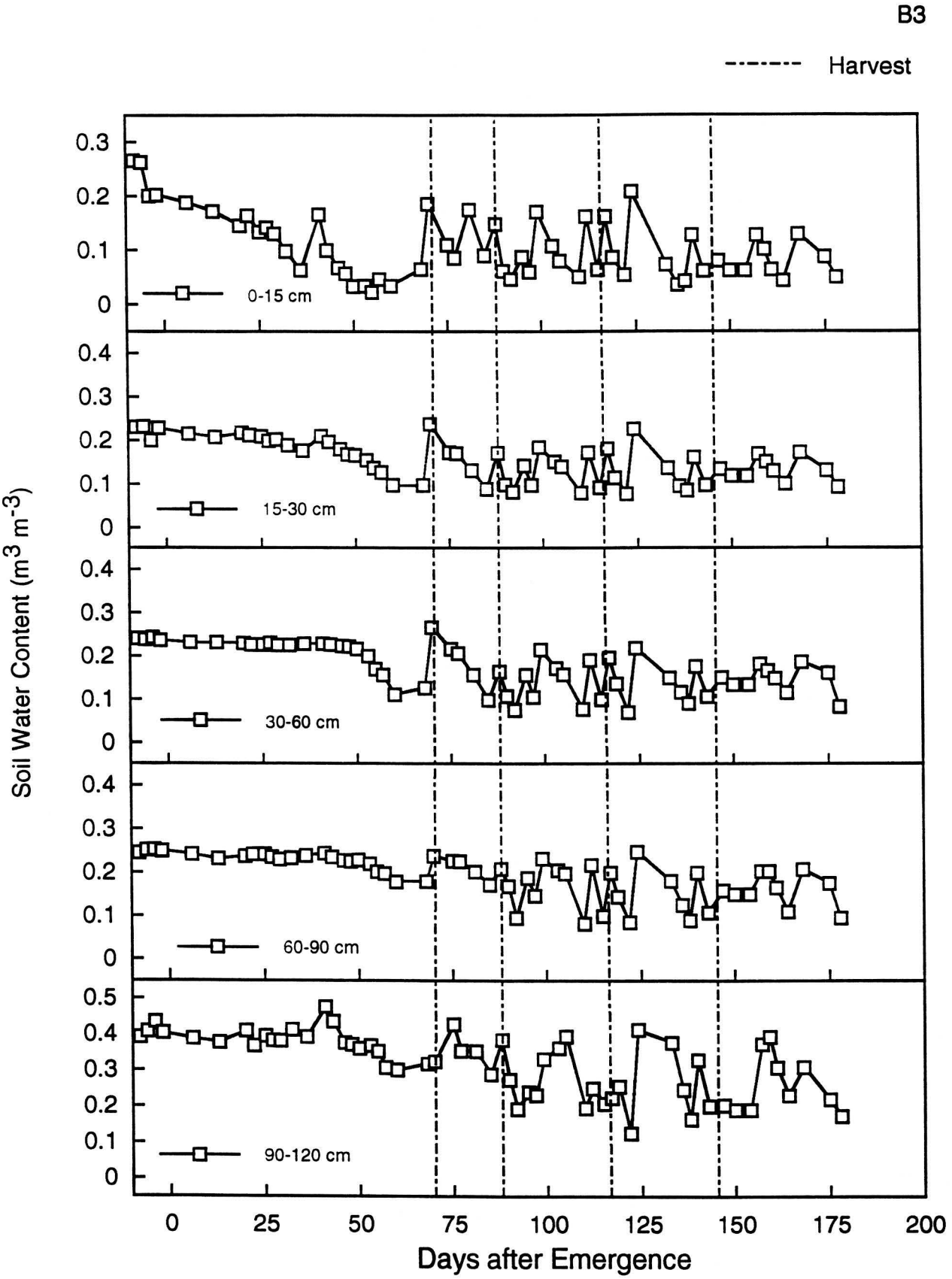


Figure 16. Soil water content for five depths of the control, low fibrous treatment (tube 6).

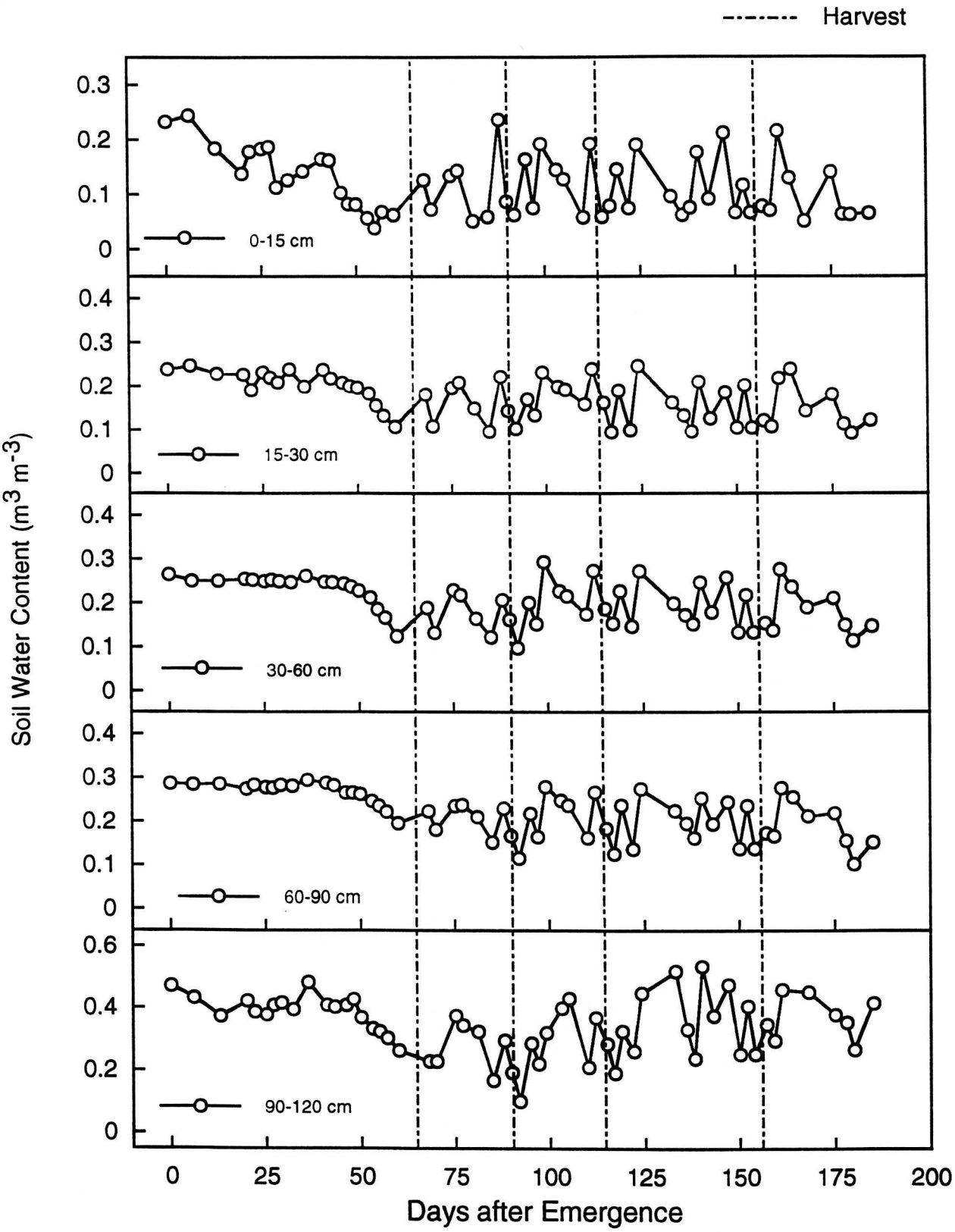


Figure 17. Soil water content for five depths of the control, high fibrous treatment (tube 10).

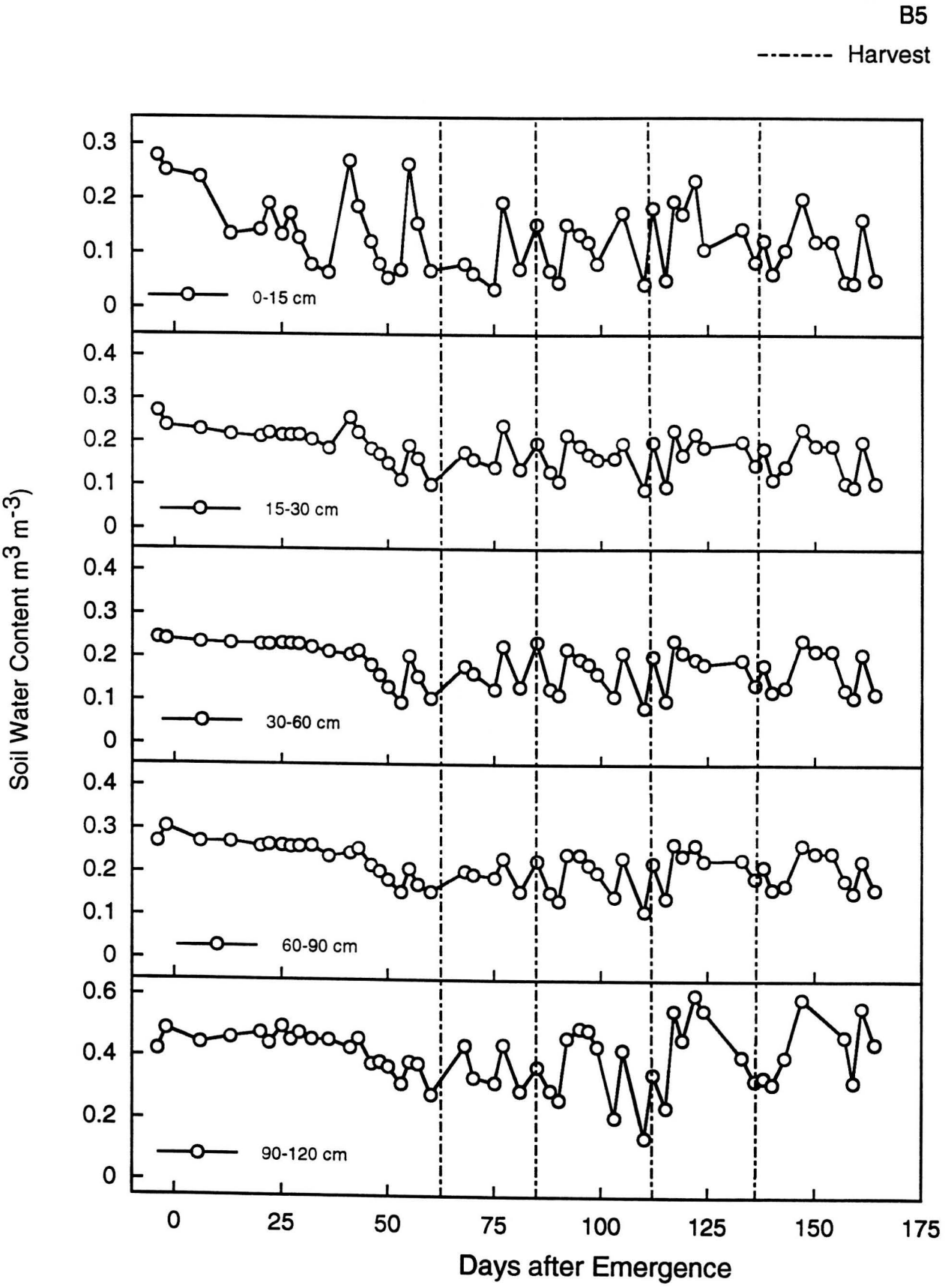


Figure 18. Soil water content for five depths of the control, high fibrous treatment (tube 11).

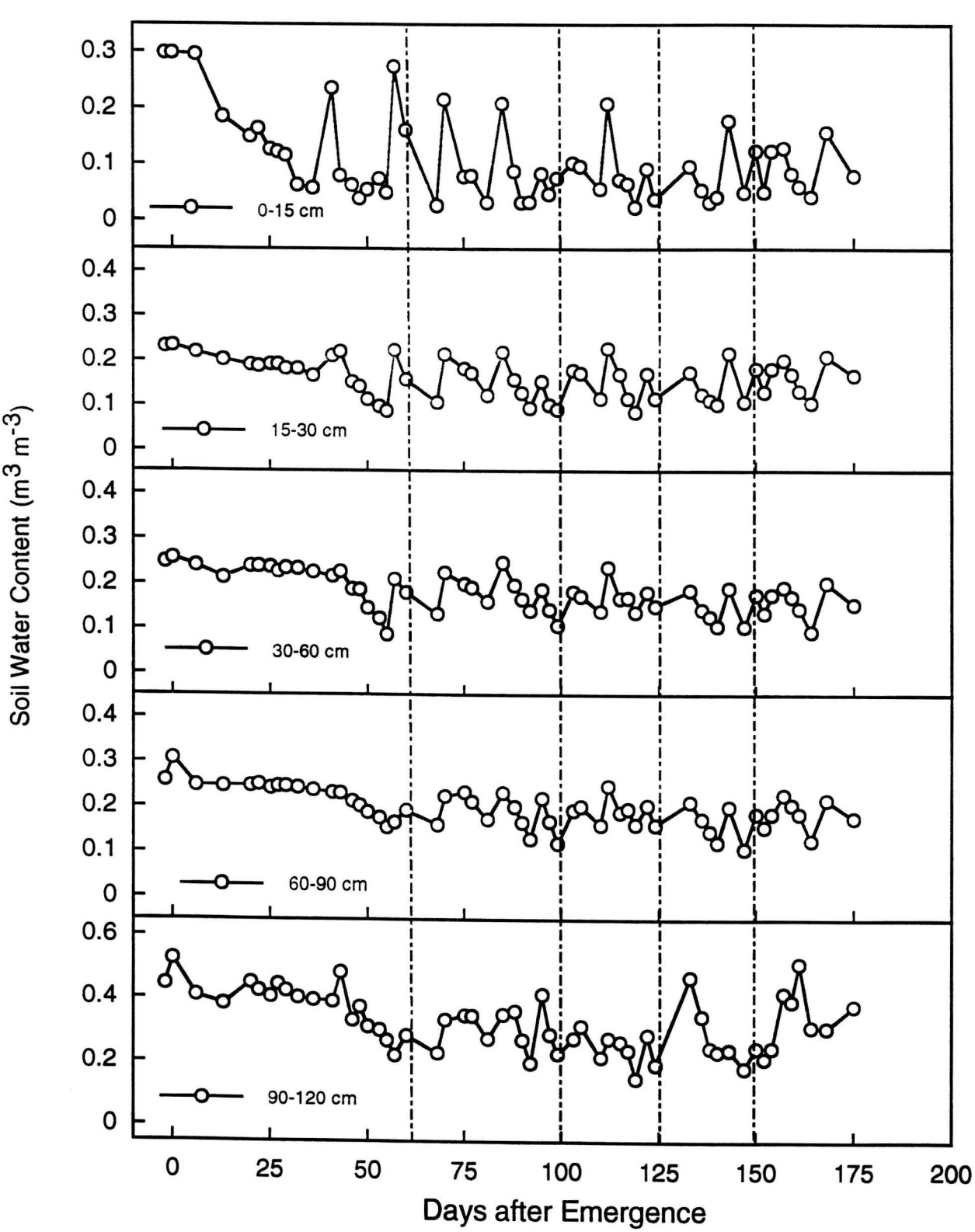


Figure 19. Soil water content for five depths of the control, high fibrous treatment (tube 12).

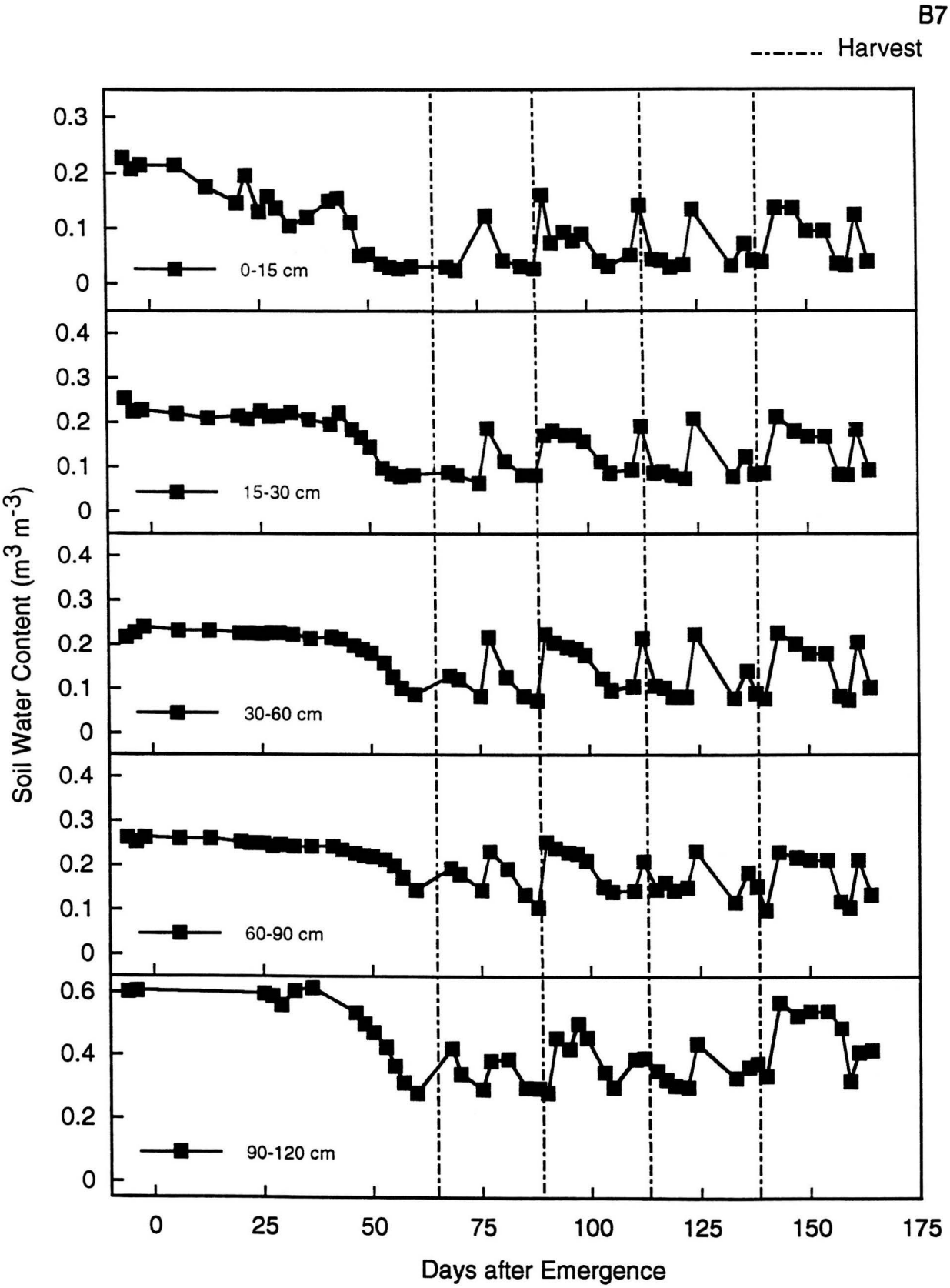


Figure 20. Soil water content for five depths of the high salt, low fibrous treatment (tube 28).

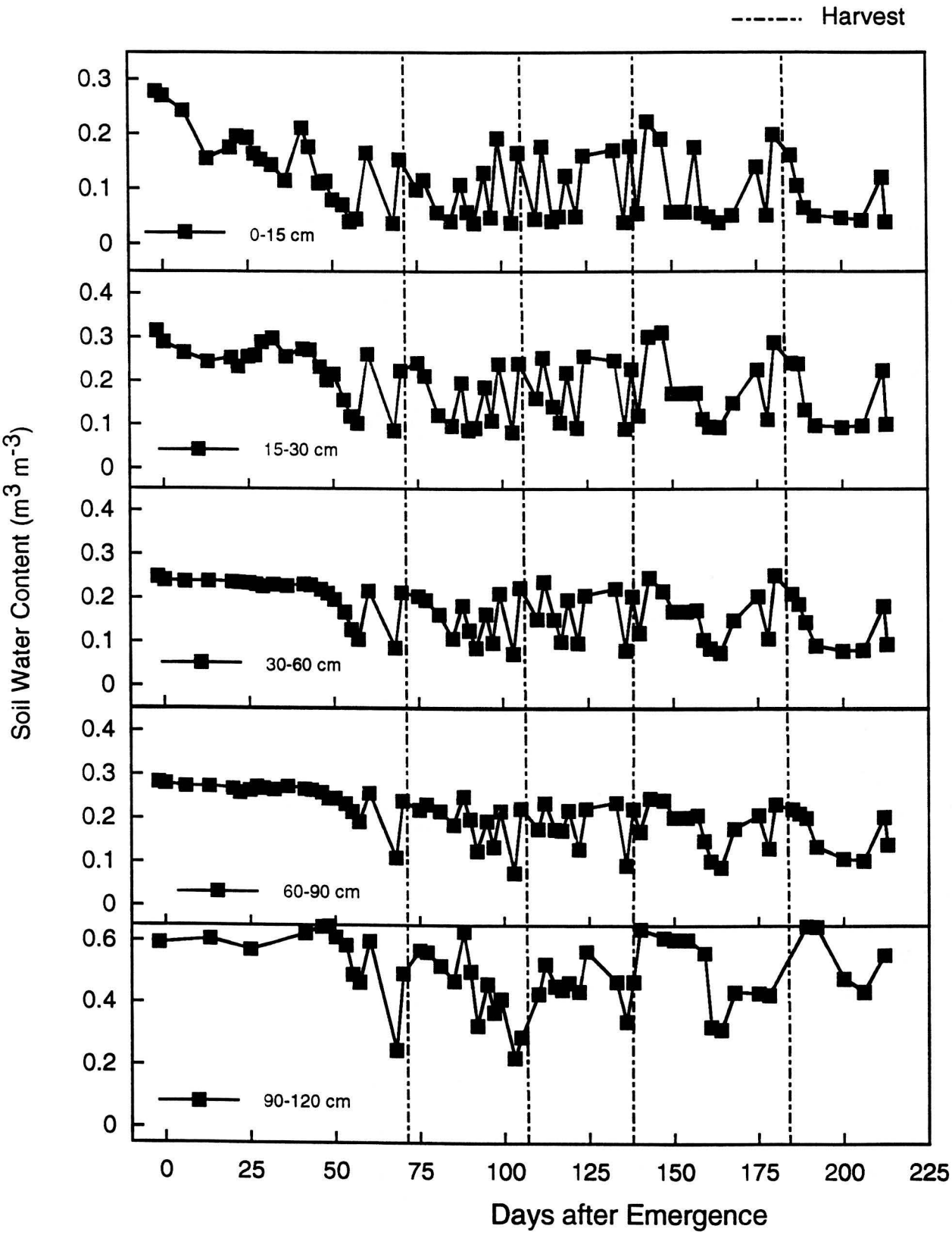


Figure 21. Soil water content for five depths of the high salt, low fibrous treatment (tube 29).

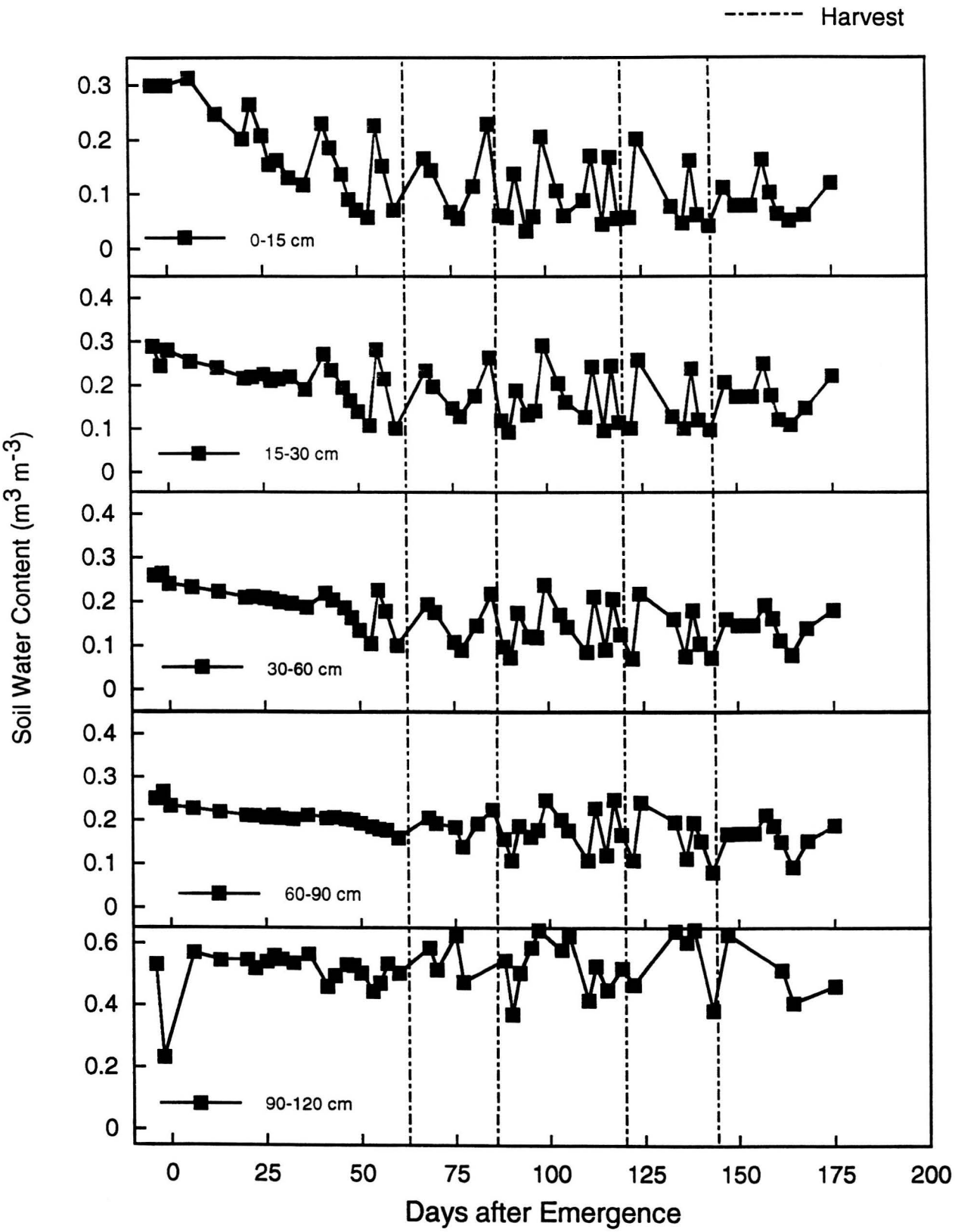


Figure 22. Soil water content for five depths of the high salt, low fibrous treatment (tube 30).

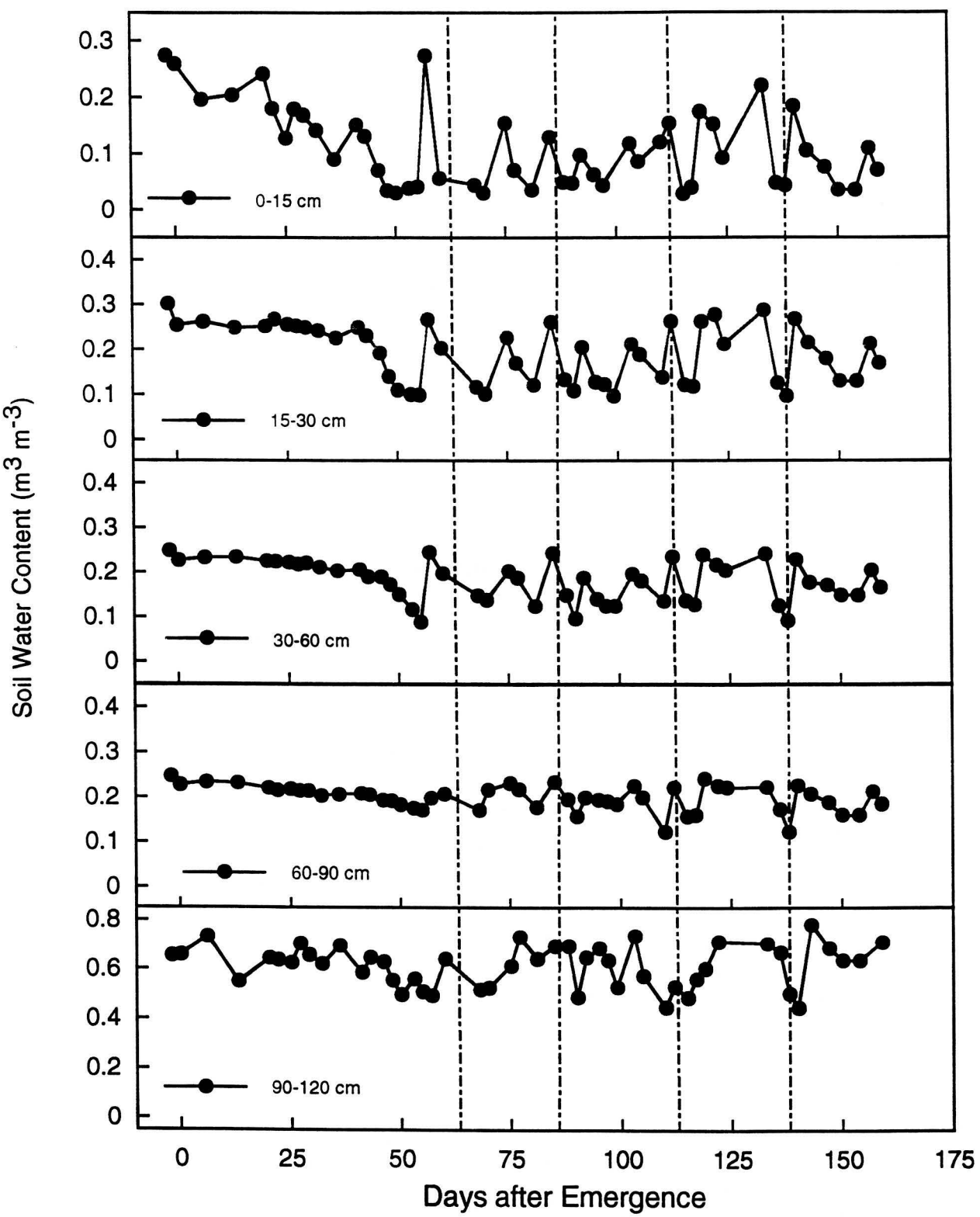


Figure 23. Soil water content for five depths of the high salt, high fibrous treatment (tube 34).

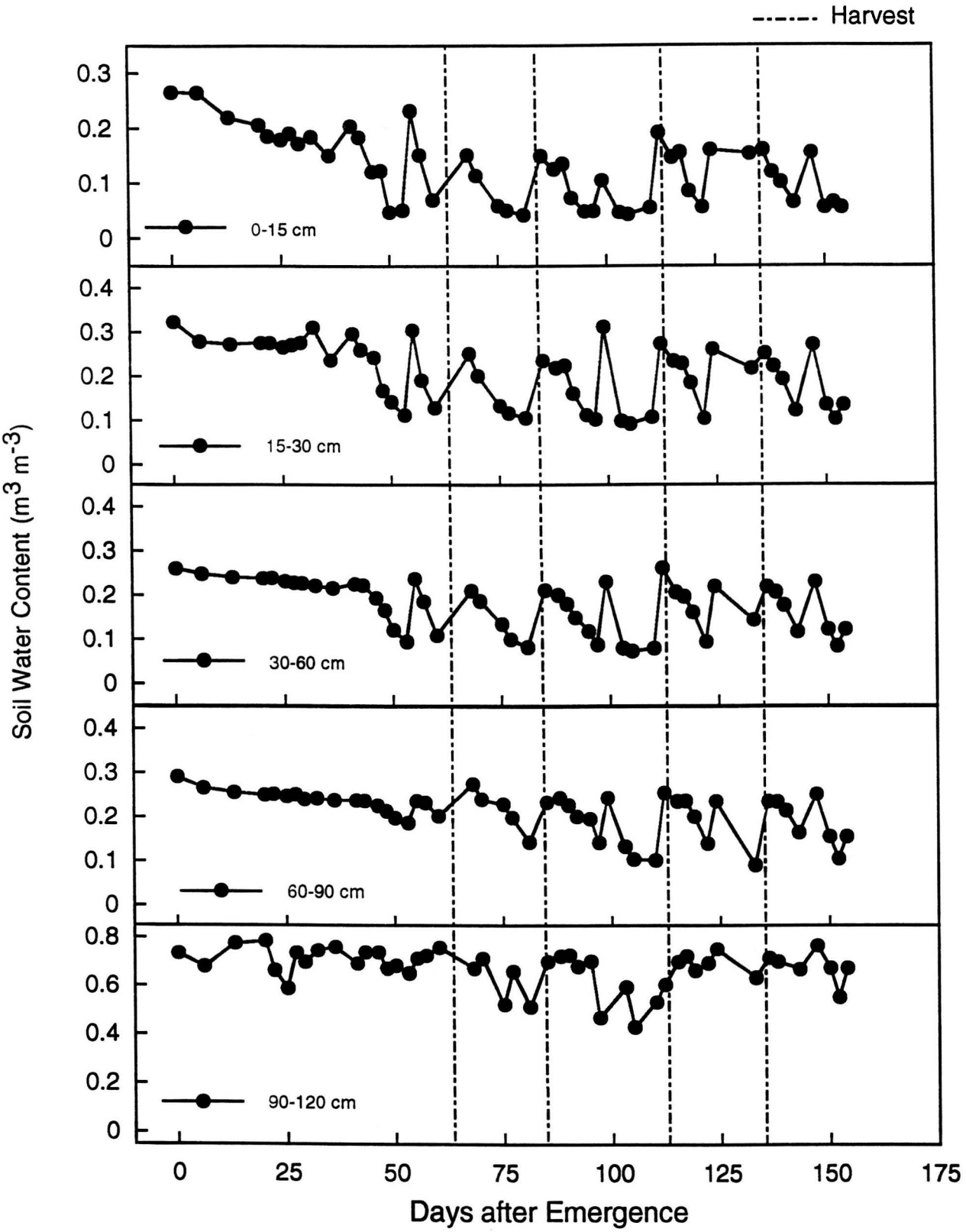


Figure 24. Soil water content for five depths of the high salt, high fibrous treatment (tube 35).

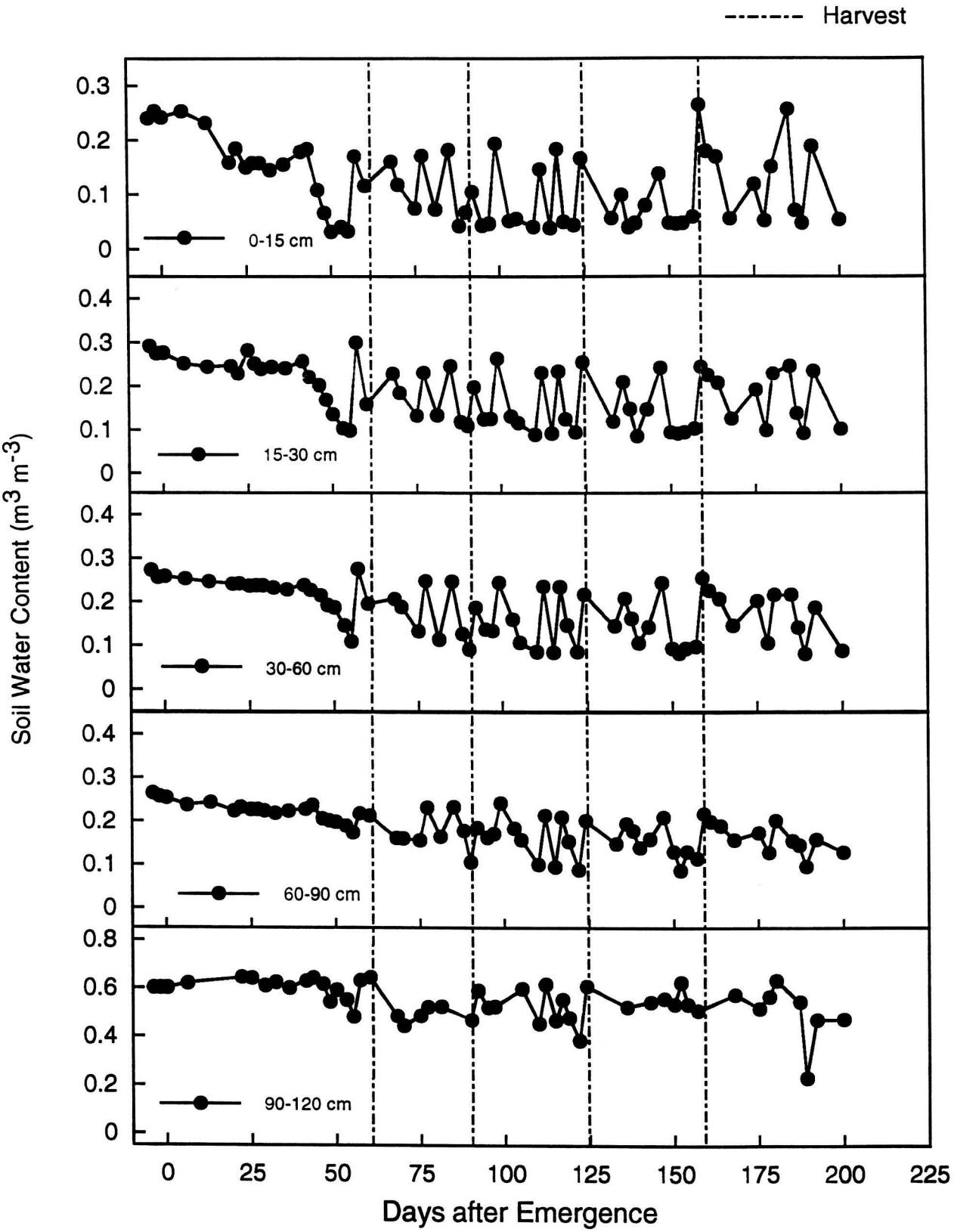


Figure 25. Soil water content for five depths of the high salt, high fibrous treatment (tube 36).