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**RESPONSE OF DRY BEAN ROOTS TO A MANAGEMENT SYSTEM
· DESIGNED TO ALLEVIATE SOIL RELATED STRESSES ·**

By

Rodney Lynn King

A THESIS

**Submitted to
Michigan State University
in partial fulfillment of the requirements
for the degree of**

MASTER OF SCIENCE

Department of Crop and Soil Sciences

1988

ABSTRACT

RESPONSE OF DRY BEAN ROOTS TO A MANAGEMENT SYSTEM DESIGNED TO ALLEVIATE SOIL RELATED STRESSES

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Field and greenhouse studies were conducted to examine the effects of soil compaction alleviation and other management factors on dry bean (*Phaseolus vulgaris*, L.) production. Two crop rotation/tillage systems were studied in combination with irrigation, row spacing, and cultivar variables. Roots were studied by destructive sampling and with minirhizotron observation tubes. A digital image processing system was developed to analyze washed root samples.

Soil bulk density was decreased in the plow layer by a deep rooted legume and deep tillage. Soil moisture and aeration and pore size distribution were improved in the plow layer by the alternative rotation/tillage management system. Shoot dry weights and root length densities were not affected by treatment combinations. Minirhizotron root observations were poorly correlated with the destructive sampling results, and it was concluded that the minirhizotron method is not a valid root study tool on fine textured soils with compaction problems. The image analysis system is slow and overestimated root length by 23% on debris-free samples and 44% on samples with debris.

ACKNOWLEDGEMENTS

Many people have contributed significantly to this thesis and I am pleased to acknowledge several of them.

I want to express gratitude to my guidance committee. Dr. Alvin J. M. Smucker served as my major professor and his guidance throughout my M. S. program has been much appreciated. The assistance of Dr. A. Earl Erickson was most helpful, especially during the weeks of thesis writing and revision. Dr. Ronald L. Perry and Dr. Francis J. Pierce provided valuable assistance in course selection, root analysis techniques, and critical review of my thesis.

John C. Ferguson provided excellent technical assistance related to the minirhizotron and digital image processing work. The daily exchange of ideas with John was both useful and enjoyable. Fellow graduate students Marie-Claude Fortin and Stephanie Schroeder offered many hours of field and laboratory assistance, as well as encouragement and important friendship. I am also indebted to Dr. Amram Eshel and Dr. Juang Wang, both of whom gave freely of their time and expertise.

Finally, and most importantly, I want to express sincere gratitude to my wife, Sandy, who offered constant care, encouragement, and support throughout this program.

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INTRODUCTION

Dry beans (*Phaseolus vulgaris*) are an important crop in the agricultural economy of Michigan. Successful dry bean production depends on the proper management of many interrelated factors. One of the most significant factors is soil, and proper management of this resource is of primary importance if production is to be optimized.

The soil matrix provides a suitable environment for root growth. It must physically support the plant, yet allow for easy root proliferation and penetration. The soil must provide adequate moisture and air for use by the roots and ultimately the whole plant system. It must offer adequate nutrients for root uptake and plant use. The balance between soil moisture and soil air is crucial, and an excess of moisture can mean an inadequate amount of air for plant needs.

Many of the dry beans grown in Michigan are grown on fine textured lake plain soils. These soils are naturally poorly drained and prone to waterlogging, even with artificial drainage. Poor aeration can be an accompanying problem. The soils are subject to compaction by agricultural equipment traffic. Compaction may result in serious mechanical impedance to root growth, and may intensify problems of soil water and air flow and availability to plants.

This thesis reports on a study of problems related to soil stresses which adversely affect dry bean production on fine textured soils. The primary objective was to identify soil conditions which would allow the root system to function in a favorable, reduced stress environment, resulting in a healthy whole plant system capable of maximum yield.

The study included soil and crop management practices designed to minimize environmental stress. The effects on soil physical properties and on the dry bean plants were monitored. The utilization of a deep-rooted legume in the crop rotation was examined for its potential in soil compaction alleviation. Different tillage systems were used to break up existing compaction and to prevent additional compaction. Other management factors included irrigation, row spacing, and cultivar variables. Root studies were conducted to examine the effects of various treatments on the root system and ultimately on whole plant performance.

A greenhouse study was carried out to examine the effects of flood and drought stress on dry bean root and shoot performance. Soil compaction can result in effects similar to those of flood or drought since compaction can alter soil water and air flow and availability to plants.

The study of root systems is hampered by difficulties in quantifying root activities. New methods are needed to allow for faster, more extensive information gathering. Several methods of root study were employed, including two given special attention. The microvideo minirhizotron root

observation method has been developed recently and it was used and tested extensively in this study. Digital image processing of extracted, washed root samples is currently being developed and a system was tested on the roots from this study. A chapter on minirhizotron observation tubes on fine textured soils and a chapter on digital image processing for use in root studies are included.

Soil management to optimize the rooting zone is important not only for dry beans on fine textured soils. The exact nature and the effects of soil stresses vary among soils but the resulting root stress and decreased production is universal. It is hoped that the results of this study will find application to related problems with crop production on similar or on different soils.

CHAPTER 1

LITERATURE REVIEW

SOILS AND CROP PRODUCTION

Successful production of any agronomic crop requires careful management of many interrelated factors. Ultimately, it is the combination of management practices which determines if the production enterprise has been successful.

The soil resource is a basic element in crop production. Proper management of this resource is essential, both for current production and for future use. Improper management of the soil may result in a variety of problems which can have long-term effects. Some of the other factors important in crop production, e.g. weather, are difficult or impossible to manipulate but this is not the case with soil. The soil resource can be and is manipulated with relative ease. Soil tillage is practiced in most agriculture production efforts throughout the world. The benefits are many and have often been realized. But the potential also exists for treating the soil in such a way that the results are counterproductive. There are too many examples worldwide of the misuse of soils which have resulted in erosion, compaction, or other negative effects and decreased productivity.

Soils are quite diverse; therefore, soil management practices must also be widely varied. Soils differ due to parent material and weathering. Depth, horizonation, texture, structure, natural fertility, water and air relations, microbial activity, and organic matter also differ greatly for different soils. The infinite number of combinations of these factors results in soils useful for many different purposes.

Many soils are well suited to one or more particular crops. In other cases crops are grown on soils which may not be especially well suited to the given soil environment but for historical, cultural, economic, or other reasons the growth of a particular crop on a given soil continues. In these cases the management of the soil resource for optimum production and long term conservation becomes even more important.

DRY BEAN PRODUCTION ON THE FINE TEXTURED SOILS OF THE SAGINAW VALLEY IN CENTRAL EASTERN MICHIGAN

Dry edible beans (*Phaseolus vulgaris* L.) have been grown commercially in the Saginaw Valley region of Michigan for at least 100 years (Anderson, 1978). The dry bean is a short season crop which has grown well in the fertile, fine textured lake bed soils. However, in the past two decades dry bean yields in the region have stagnated or even decreased in spite of improved varieties and pest control (Wright, 1978). This trend has been particularly troublesome for producers as

the economic pressures on farmers have increased.

Plant growth and production is highly responsive to the soil environment. As farmers and researchers have noted the trend toward declining dry bean yields, they have begun to look carefully at management practices, particularly soil management. The problem of soil compaction and related effects has been noted as the most limiting factor to increased production (Smucker et al., 1978). Mechanical impedance, soil moisture relations, and aeration are interrelated factors which are often associated with soil compaction and may have a significant effect on plant growth and production (Hillel, 1982).

SOIL COMPACTION

Soil compaction may result from natural or human-induced factors. Among the natural causes are soil formation processes, raindrop impact, wetting and drying cycles with accompanying shrinkage, and root growth (Hillel, 1982; Larson and Allmaras, 1971). The major cause of soil compaction is traffic and tillage operations (Hillel, 1982; Bowen, 1981). As methods of agricultural production change, soil compaction occurs more frequently and is more intense. A primary factor is the increase in the size, weight and power of agricultural tractors and tillage and harvesting machinery. In addition to the physical compaction imposed on the soil, there are numerous related effects on the soil and plant system. Included are changes in the soil moisture relationships, aeration,

root restriction, and disease and pest resistance.

Fountaine (1959) reported that the productivity of compacted soils is altered by increased mechanical impedance, decreased aeration, and altered soil moisture availability. These changes are due to the increase in soil density and accompanying decrease in soil pore space. Most reports show negative effects of increased soil density on productivity across a wide range of crops, soil types, and management systems (Baver, 1944; Douglas and McKyes, 1983; Hakasson, 1985; McKyes et al., 1979). However, Voorhees (1977) reported an increase in soybean yields under moderate compaction in a dry year. This likely indicates an improvement in the water holding capacity of the soil due to additional densification of the soil. It also points out that there can be considerable variation in the effects of compaction and the mechanisms producing those effects.

BULK DENSITY

Bulk density is the most widely reported measure of soil compaction. Cassel (1982) reported that bulk density is easily measured, and is almost always altered by tillage. Cassel further noted that bulk density varies temporally and spatially and sampling must take this into account. In a review of plant response to soil compaction, Rosenberg (1964) noted a parabolic yield response to bulk density. He credited Vomocil (1955) with first describing this response. Vomocil stated that below a critical bulk density, yield variation is

due to factors other than soil physical conditions. Rosenberg concluded that the correlation between yield and bulk density is difficult to quantify due to the numerous factors involved, including soil texture, moisture, aeration, climate, and crop. He further suggested that aeration is often not limiting on compacted soils, especially on coarse and medium textured soils.² Veihmeyer and Hendrickson (1948) suggested that on most soils a bulk density of 1.9 g cm^{-3} will prevent root growth of most common plants and that on clay soils the critical maximum is between 1.5 and 1.7 g cm^{-3} . Zimmerman (1961) found the maximum bulk density for root penetration was about 1.8 and 2.0 g cm^{-3} for a Cherty clay loam and a sandy loam respectively.

McKyes et al. (1979), in a three year study on soil with a high clay content, found that the yield of corn silage decreased significantly as bulk density increased. He also noted that bulk density can be too low for optimum crop yield.

Jones (1983) defined critical bulk density as the soil density at which rooting activity was at a maximum for a given soil water content. He identified critical bulk densities for several crops (cotton, corn, peas, sudangrass, and sugarcane) on soils with differing textural composition and found a strong correlation between critical bulk density and texture, with soils with a high clay content having lower

² Others later disagreed with Rosenberg on this point, as will be discussed later.

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critical densities than those with less clay. He concluded that rooting density above the critical bulk density is affected largely by soil strength rather than soil moisture and aeration which figure more prominently in root growth at lower bulk densities.

MECHANICAL IMPEDANCE AND PLANT RESPONSE

Roots must penetrate the soil profile to physically support the plant and take up water and nutrients necessary for plant growth. If the soil is compacted, the pore size distribution is altered (Bowen, 1981; Hillel, 1982). The volume decrease due to compaction comes from a compression of the macropores and rearrangement of soil particles resulting in an increase in micropores. This leaves fewer macropores through which the roots can grow. Wiersum (1957) and Fountaine (1959) noted that roots have to apply pressure to grow within clods or to deform pores, and that if unable to exert sufficient pressure will be restricted in growth. Gill and Bolt (1956) presented a summary of the classic work by Pfeffer in 1893 on root growth pressure exerted by plants. Pfeffer encased a growing root in plaster of Paris and measured the pressure exerted by the root. He found that roots exerted pressure both radially and axially, up to 10 atm. when pressure was brought to bear on all sides of the root. More pressure was exerted radially, likely due to much larger surface area, and radial pressure was increased when axial growth was constricted. This work provides a basis for

studies on root growth in soil and is important when considering root growth under compacted or constrictive soil conditions.

By growing roots in a precisely defined granular media, Richards and Greacan (1986) found that fine roots are less subject to mechanical impedance than large roots. They also noted that cylindrical expansion behind the growing root tip is at least somewhat effective in relieving soil pressure and enabling the root to elongate.

Roots have also been found to adapt their own morphology in order to grow in compacted or tortuous soil (Taylor and Arkin, 1981). This enables the plant root system to overcome a compacted layer or zone in soil. There are limits to roots' ability to overcome mechanical impedance and there are also extra costs in terms of growth or photoassimilate partitioning. Barley (1961) studied maize roots and found that they continued to grow even when were physically restricted. Growth rate was decreased, however. Increased pressure on the root tip may result in an increase in root diameter due to changes in the internal pressure (Russell, 1977). Russell also suggested that mechanical impedance does not cause roots to become thinner. Rather, mechanical impedance may cause proliferation of lateral roots which can easily be mistaken for root axes of reduced diameter. The study of the response of roots to mechanical stress is essentially the investigation of the effects of the pressures roots must exert to enlarge or create pores.

SOIL MOISTURE, AERATION AND PLANT RESPONSE

Soil compaction can cause or contribute to aeration and soil moisture problems. Warkentin (1971) found that compaction alters the water content and movement in soils by modifying the pore size distribution with the large macropores reduced first. This leads to reduced water movement, especially on fine textured soils. Reduced water movement can cause waterlogging following precipitation as the macropores no longer exist to provide for good drainage, and waterlogging affects plant growth by decreasing or eliminating the necessary soil air.

The volume of soil air depends on both total pore space and water filled pore space. Erickson (1982) suggested that 10% air capacity (% of soil volume) is the lower limit for plant growth for most crops, but that no value should be taken as an absolute minimum due to variations in soil conditions and plant requirements. In addition to the air filled pore space, it is imperative for good plant growth that oxygen diffusion occur rapidly enough to replace that which is taken up by the plant. This includes diffusion through the bulk soil and also through the moisture films surrounding plant roots. The latter is usually the most critical since diffusion through water is approximately 10,000 times slower than through air. Erickson (1982) determined that an oxygen diffusion rate of $< 0.2 \text{ ug cm}^{-2} \text{ min}^{-1}$ is the minimum for oxygen resupply to active roots and $> 0.4 \text{ ug cm}^{-2} \text{ min}^{-1}$ is necessary for normal plant growth. The composition of the

soil atmosphere can vary markedly from the above ground atmosphere. Campbell and Phene (1977) monitored the aeration status of a layered sandy loam in which millet was growing and found that plant growth was unaffected by oxygen concentrations above 15%, but below 15% growth was reduced. In a study by Smucker and Erickson (1977), anaerobic conditions resulted in reduced growth of peas and also contributed to increased exudation of various organic compounds. This increase in exudation indicates a loss of photoassimilates and suggests one way in which compaction can result in decreased yield. Letey et al. (1962) found that roots ceased growing if oxygen content was too low for a short time, and periods of low oxygen supply were more detrimental to young plants than to those with larger root systems. Huck (1970) reported that anaerobiosis can result in cessation of root growth and death of some root tips.

An anaerobic rhizosphere environment most often results from waterlogging (Cannell and Jackson, 1981). This problem is common on compacted soils since compacted layers may prevent adequate drainage. While some plants are more tolerant of a waterlogged environment due to root characteristics such as porosity, length, and the ability to form adventitious roots (Cannell and Jackson, 1981), nearly all plants suffer at least some setback when subjected to too much water and the accompanying lack of oxygen. Root growth was generally retarded (Glinski and Stepniewski, 1985) and shoot growth was affected as well. Jackson and Drew (1984) suggested that

the primary effect of too much water is asphyxiation of the plant as gaseous diffusion almost ceases. Anaerobic conditions predispose the plant root system to attack by pathogens (Stolzy et al., 1965; Miller et al., 1980; Smucker and Erickson, 1987) and cause the plant to exude compounds which may be toxic to the plant (Schwartz, 1980; Smucker and Erickson, 1987).

SOIL COMPACTION AND DROUGHT STRESS

Cortes and Sinclair (1986) studied the water relations of soybeans grown under drought stress. They found that in soybeans the most important mechanism for sustaining growth under limited moisture supply was the maintenance of root growth into deeper portions of the soil profile to tap existing water supplies. Huck et al. (1986) noted that water stress on soybeans resulted in decreased shoot and seed weight but increased total root length, including an increase in rooting depth. Hoogenboom et al., (1987) and Huck (1986) also reported increased rooting depth during periods of drought stress. These studies indicate that a normal response of a plant to moisture stress is a decrease in shoot:root ratio as the plant partitions more of its photoassimilates to the root system in order to sustain growth and access available water. Shank (1945) found the shoot:root ratio of maize changed from 3.4:1 under sufficient (21%) water to 2.5:1 when soil moisture was severely limiting (7.5%). Huck et al., (1986) found that the timing of the moisture

stress was also very important. Their work showed the most crucial growth stage in soybeans is the reproduction stage.

In the case of crops grown on compacted soils or on soils with compacted layers, rooting depth may be restricted due to mechanical impedance (Miller, 1987; Bennie and Botha, 1986; Raghavan et al., 1977; Bertrand and Kohnke, 1957). As long as there is sufficient water available the plant will not suffer. However, during a short term drought, common on many soils, the shallow root system will not be able to access the moisture which may be available in the deeper soil horizons. In addition, the common response of roots growing deeper when soil moisture is limiting will be hampered by the continuing mechanical impedance of the soil.

TILLAGE-INDUCED SOIL COMPACTION

The effects of increased bulk density on root systems have been discussed previously. Increased bulk density often occurs as a result of tillage operations. Phillips and Kirkham (1962) found that a Colo clay soil compacted to various bulk densities by vehicular traffic resulted in reduced stands, maturity, and yield of corn. Raghavan et al. (1979) severely compacted plots on which silage corn was grown by applying 15 passes of tractor traffic. Maximum rooting depth was halved and the depth of dense roots was decreased by one third compared to the noncompacted plots. In a study on a compacted clay soil, Douglas and McKyes (1983) noted a

reduction of silage corn yield of up to 40% compared to on noncompacted soils. They suggested that the plants suffered both from mechanical impedance and poor aeration due to inadequate rainage. Root growth was delayed due to high bulk density levels.

The soil moisture content at the time of tillage operations is important in determining the extent of compaction. According to work done by McKyes (1985), densification can be up to five times as severe on soils tilled at optimum soil moisture when compared to the same soils tilled when quite dry. In laboratory studies Akram and Kemper (1979) found that maximum compaction generally occurred when soils were at or near field capacity for water. Their work was conducted on both fine and coarse textured soils.

Soil texture figures prominently in the extent of compaction on a given soil. McKyes (1985) found an increase of 0.13 g cm^{-3} in bulk density of a clay soil resulted in a 50% decrease in yield of maize. A similar increase in bulk density on a coarse textured soil would likely be insignificant. The difference is due to the differences in macropore/micropore ratios.

SOIL COMPACTION ALLEVIATION

Soil compaction is often caused by tillage operations. Somewhat ironically, tillage may also be used to alleviate compaction. In recent years new tillage practices have become popular and have been shown to be of some value in

ameliorating compacted soil. Subsoiling may be useful in breaking up tillage-induced hardpan layers in the soil, especially moldboard plow layers. Other tillage operations may be carried out to alleviate compaction caused by natural processes or by secondary tillage (Bowen, 1981).

Miller (1987), in a study on the effect of subsoiling and irrigation on dry bean production, found that water stress may develop between irrigations if the plants have not developed a deep enough root system. This study was conducted on a sandy loam soil (85% sand, 2% clay, rigid matrix at 30 cm). Subsoiling allowed deeper root growth and resulted in significantly increased yields. However, in a companion study with the same treatments on a loam soil (45% sand, 9% clay, plowpan at 25 cm), Miller found that subsoiling did not affect yields even though rooting depth and foliage density were increased. He attributed this response to the better water holding capacity of the loam soil compared to the sandy loam. Bennie and Botha (1986) studied rooting depth and water use efficiency for maize and wheat. The work was carried out on irrigated, deep, fine sandy soils. Deep ripping and controlled traffic led to an increase in rooting depth, water use efficiency, and yield (30% in maize and 19% in wheat) compared to conventional tillage.

ALLEVIATION BY NATURAL PROCESSES

Soil compaction can be alleviated to some extent by natural processes (Bowen, 1981; Larson and Allmaras, 1971).

Akram and Kemper (1979) found that wetting and drying cycles improved infiltration rates. The process of freezing and thawing was also found to improve infiltration rates and leave the soil in a generally more friable, less compacted condition. Voorhees (1979, 1983) reported that natural weathering (wetting and drying, freezing and thawing) alleviated some, but not all, traffic-induced compaction on a Nicollet clay loam.

The effect of deep rooted plants or plants with especially hardy root systems has also been studied in relation to compaction alleviation. Radcliffe et al. (1986) examined the effect of a deep-rooted perennial, alfalfa, on subsoil compaction. The combination of alfalfa and the application of gypsum resulted in decreased soil strength as measured by a cone penetrometer. These researchers concluded that the use of a deep rooted legume is effective in loosening compacted soil layers as long as the nutritional status of the subsoil is conducive to root growth in that region. In a study on a Charity clay soil Christenson et al. (1976) found that a crop of alfalfa prior to dry beans resulted in higher dry bean yields than when dry beans were preceded by any other crop. These yield trends are not proof of decreased compaction due to the alfalfa. However, they are strong indicators that this is occurring since the Charity clay is subject to tillage and traffic compaction and the related problems of poor drainage and poor aeration.

Chasse et al. (1967) reported that an extensive root

system near the soil surface (high organic matter) reduced compaction damage due to machinery loads by about 65% compared to bare soil with low organic matter. In a laboratory study comparing three soil textures with varying amounts of organic matter, each at three levels of compaction, Ohu et al. (1985) determined that the presence of organic matter increased the root dry matter and yield of the crop while compaction decreased the same plant parameters. These researchers concluded that high levels of organic matter have the potential to improve the productivity of compacted soils. In a contradictory study using a soil bin as well as laboratory procedures, Gupta et al. (1987) looked at the influence of corn residue on compaction due to wheel traffic and concluded that the presence of this organic matter had little or no effect on soil compaction.

ADDITIONAL MANAGEMENT FACTORS

ROW SPACING

In addition to soil management for the alleviation and prevention of soil compaction, other management factors are important in dry bean production. Row spacing is one such factor.

More work has been reported on soybean yield response to row spacing than dry bean yield response to planting patterns. The response of both crops will be reviewed.

Cooper (1977) documented up to a 25% increase in soybeans grown in narrow (17 cm) vs. wide (50 or 75 cm) rows, all row spacings having a constant seeding rate. Taylor (1982) found that soybeans grown on a silt loam yielded significantly more in 25 or 50 cm rows than when grown in 75 or 100 cm rows when the moisture supply for the crop was good. However, under conditions of limited water there were no yield differences. In a study designed to determine the reasons soybeans generally yield more in narrow vs. wide rows, Bennie et al. (1982) found that there were few differences in nutrient uptake and accumulation due to row spacing (25 cm and 100 cm row spacings were used). In the same study, Mason et al. (1982) noted a 49% increase in root length density under narrow rows, 52% more roots per unit leaf area, and a differential water uptake rate on the wide rows with more water used from the intrarow space than the interrow space. However, overall water use and plant water potentials were not different under irrigation between row spacings and there were no yield differences due to irrigation. These researchers concluded that yield differences between the two row spacings were not due to differential water use which might be expected with large differences in root length densities. Taylor et al. (1982) participated in the same investigation and reported that radiation interception was greater at the narrow row spacing during most of the growing season and especially at the critical reproductive stages. This is due to increased or more

uniform canopy cover. Taylor and associates concluded that this differential radiation interception, rather than differences in plant water status due to rooting differences or differences in nutrient uptake and accumulation, was primarily responsible for increased soybean yield under narrow row conditions.

Atkins (1961) compared red kidney bean production under different row spacings. He found the 23 and 46 cm rows significantly outyielded the conventional 92 cm rows but there were no differences between the 23 and 46 cm row spacing yields. Redden et al. (1987) reported up to a 46% increase in dry bean yields when the crop was planted in 18 cm vs. 107 cm rows. In that study the authors also noted a positive yield response to a tripling of plant population (from 112,500 to 337,500 plants ha^{-1}). Grain yields closely followed the number of pods meter^{-2} , which was positively correlated with ground cover between flowering and the middle of pod fill. Thus the increase in yield was attributed primarily to canopy cover and radiation interception.

SUMMARY

The production of any crop depends on a favorable soil environment. Soil compaction and the related effects of poor water relations, poor aeration, and mechanical impedance are often responsible for weaker and shallower root systems, decreased plant growth, and reduced yields. Dry beans do not naturally have a hardy root system which will tolerate

poor soil structural conditions, poor aeration, and compacted soil layers. The Charity clay soil in the lake plains in the central eastern part of Michigan's lower peninsula is subject to these detrimental conditions. The production of dry beans on this soil is an acute management challenge. The intent of this study was to utilize a combination of factors to overcome the soil management problems of *Phaseolus vulgaris* on these soils. The use of a deep-rooted perennial legume, tillage management to alleviate existing compaction and prevent further compaction, timely irrigation, and the use of narrow row spacing were combined in an effort to achieve a maximum yield of dry beans.

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CHAPTER 2

ALLEVIATION OF SOIL STRESSES ON DRY BEAN ROOT SYSTEMS AS A KEY TO INCREASED DRY BEAN PRODUCTION

INTRODUCTION

The production of dry beans (*Phaseolus vulgaris*) in the Saginaw Valley region of Michigan has for many years been an important component of the farm economy. In recent years the average dry bean yield has stagnated or declined (Wright, 1978). This has occurred in spite of the release and widespread adoption of cultivars which in many geographic areas have proven to be higher yielding than older varieties.

Soil compaction has been suggested as a primary cause of decreased dry bean production (Smucker et al. 1978). *Phaseolus vulgaris* is not a deep rooted plant and does not have a particularly hardy root system. Thus any soil stress is likely to adversely affect root growth and yield.

The soils in the Saginaw Valley are particularly prone to compaction related problems. Some of these soils are fine textured and have a high water holding capacity. They are a Charity clay loam (illitic, calcareous, mesic Aeric Haplaquept) with about 60% clay. The soils are naturally poorly drained but subsurface drainage has been installed on most of the agricultural acreage in the region.

Soil compaction can result from natural forces such as raindrop impact, wind blown particles, and wetting and drying cycles (Bowen, 1981; Cohron, 1971). However, compaction induced by human activity, primarily mechanized agricultural operations, is the primary cause of serious compaction problems (Bowen, 1981; Cohron, 1971). Specifically, compaction caused by current traffic and tillage practices is known to be detrimental to root growth. Russell (1977) pointed out that increased compaction often results in decreased root proliferation and ultimately decreased crop yield. Mechanical impedance can directly affect root growth by restricting root elongation due to high soil strength or a limited number of pores which are of a sufficient size for root penetration (Bowen, 1981; Russell, 1977; Taylor, 1971). Soil water movement is decreased due to compaction (Akram and Kemper, 1979), as is air movement through the soil profile (Grable, 1971). A less extensive root system results in less water and nutrients available to the plant. Slower water movement through the soil profile results in longer periods of anaerobic rhizosphere conditions following a heavy or even a moderate rainfall. It also increases the difficulty in soil management since there are shorter periods of time when soil moisture is at appropriate levels to carry out tillage operations. Oxygen stress, from too much water or too little air, may result in rhizosphere toxicity from ethanol or other organic compound accumulation in the root system and also make the plant more vulnerable to attack by harmful pathogens

(Russell, 1977).

While soil compaction seems likely to be a major cause of limited dry bean yields, other factors may also contribute. Row spacing is one such factor. Dry beans have traditionally been grown in 28 inch (71 cm) rows (Erdmann and Adams, 1978). Narrow row spacing has proven beneficial in soybeans (Cooper, 1977; Bennie et al., 1982; Mason et al., 1982; Taylor et al., 1982). In some dry bean studies narrow rows have resulted in increased yields (Atkins, 1961; Redden et al., 1987) but results were inconclusive in another study (Erdmann and Adams, 1978).

Water stress is often a major cause of limited crop production. The lack of uniform rainfall distribution throughout the growing season may be more acutely noticed on soils which are compacted and do not allow for deep root growth. The compacted soils under the conventional rotation /tillage system in this study would seem to be prime candidates for reduced rooting depth and related water deficit problems. Timely irrigation should alleviate those problems, and the study included irrigation/non-irrigation as a treatment factor to examine that hypothesis.

The objectives of this study were:

- a) to evaluate two rotation/tillage management systems for their effects on soil physical properties, root zone modification, root growth, and whole plant performance;

- b) to evaluate the response of two dry bean cultivars at different row spacings;
- c) to study the effect of irrigation on dry bean performance as related to soil stresses; and
- d) to obtain maximum dry bean yield by incorporating various factors in a management system for soil stress alleviation and optimization of dry bean genetic potential.

MATERIALS AND METHODS

Cultural Practices and Treatments

A field study of management practices for dry bean (*Phaseolus vulgaris*) production was conducted in 1986 at the Saginaw Valley Bean and Beet Research Farm near Saginaw, MI.

Each of four tiers of land, 93 by 20 m, was divided into 24 plots. Treatments were rotation/tillage, irrigation, row spacing, and cultivar. The conventional (CONV) rotation/tillage system included a two year corn-dry beans rotation, moldboard plowing in the fall to a depth of 20 cm, and spring secondary tillage to a depth of 8-10 cm consisting of 3 passes of a Danish S-tyne field cultivator with rolling baskets. The alternative rotation/tillage system was an alfalfa rotation, no secondary tillage (ARNST) system. It included a corn-alfalfa-alfalfa-dry beans rotation, late summer subsoiling to a depth of 40-45 cm, moldboard plowing in the fall to a depth of 20 cm, and no secondary tillage prior to planting. Subsoiling was carried out both in the same direction as the rows and perpendicular to the rows using a V-ripper with shanks spaced at 75 cm.

Two irrigation treatments consisted of no supplemental irrigation and irrigation. Irrigation was applied to half the plots as needed beginning in mid July, or 45 days after

planting. Row spacings evaluated were 18, 36, and 54 cm. The two cultivars were C-20, a white bean developed in Michigan and Black Magic, a black bean.

The study was arranged as a split-split plot with the rotation/tillage factors as the whole plot, irrigation randomly split on rotation/tillage, and row spacing and cultivars arranged as randomized complete blocks in the sub-sub plots. The study was replicated four times.

Fall tillage was carried out in late August and early September of 1985. In the spring of 1986 the CONV plots were tilled on May 1. The ARNST plots were sprayed with glyphosate at a rate of 4.8 l ha^{-1} on May 27. On June 2 two more passes of tillage were applied to the CONV plots. Planting was carried out on the same date using a grain drill (International Harvester, Model 5100 Soybean Special, 10 foot width equipped with adjustable press wheels behind the disc openers). A single row of spring teeth spaced midway between the seed disc openers was mounted under the grain drill hitch and used on the ARNST plots to level the untilled soil sufficiently to create a suitable seed bed. Seeding depth was 3.5-4 cm on the CONV plots and 2.5-5 cm on the ARNST plots, with the greater depth range on the ARNST plots due to the unlevel seed bed. Seeding rate was set to achieve a plant spacing of 7.5 cm ($13.3 \text{ seeds m}^{-1}$) regardless of row spacing. The germination test of the bean seed was low, especially the Black Magic cultivar (73%), so seeding rate was increased to 18 seeds per meter of row. A log chain was dragged behind

the drill on the ARNST plots to complete covering and improve seed-soil contact. The different row spacings were achieved by plugging the appropriate seed drop holes in the drill. Tires on the tractor were spaced at 2.26 m centers on the .30 m tires, thus providing approximately 2.0 m of soil in each plot which was unaffected by wheel traffic. Wheel traffic was controlled following tillage operations.

Fertilizer (22-23-0, with 1% Zn and 4% Mn) was banded at planting at 18 cm spacing regardless of seed row spacing at a rate of 175 kg ha⁻¹. Additional fertilizer (same analysis) was broadcasted at a rate of 200 kg ha⁻¹ on the ARNST plots and 400 kg ha⁻¹ on the CONV plots. The different fertilizer rates were selected because of the differences in soil fertility due to the previous crop.

Disulfoton (Di'syston), a systemic insecticide to control aphids, leafhoppers, mites, Mexican bean beetle, and thrips was banded at planting at the rate of 8 kg ha⁻¹ in 18 cm bands on all row spacings. Weed control was achieved by using a tank mix of chloramben (Amiben) and metolachlor (Dual) (12 l ha⁻¹ and 3.6 l ha⁻¹ respectively in 300 l ha⁻¹ water) sprayed on June 3. Hand weeding was carried out during the growing season as needed.

Soil moisture at planting was good below the top 3 or 4 cm of soil. The tillage on the CONV plots caused a loss of existing moisture in the top several cm of soil. Seed placement was just into the moist soil. Moisture conditions on the ARNST plots were better than on the CONV but seed

placement was less uniform due to the uneven seed bed and imprecise depth control on the drill. Some seeds were placed well into moist soil and others were too shallow to obtain good contact with moist soil. Emergence was uneven and slow due to the uneven seeding pattern resulting from imprecise seed feeding mechanism on the drill, and the lack of rainfall following planting. It was necessary to spot replant about three weeks after planting. This was accomplished by hand and was carried out from June 26 to July 4. Spot thinning of first planting seedlings was accomplished at the same time as replanting to achieve a final seed spacing of 7.5 cm.

Sprinkle irrigation was begun on July 18, 45 days after planting. Irrigation was applied at approximately 2.5 cm per week except when there was sufficient rainfall in a given week.

Benomyl (Benlate) was sprayed three times for white mold (*Sclerotinia sclerotiorum*) control. Applications were at 10% flowering (July 29), at full flowering (August 11), and at late flowering (August 25-primarily second seeding plants). There was little evidence of white mold development in this growing season.

Measurements

Soil physical properties in the top 22.5 cm of soil were measured. Undisturbed soil cores 7.5 cm in diameter and 7.5 cm long were obtained on July 17 and 18 using the Uhland double cylinder hammer method (Blake (1965)). Cores were weighed

in the field on a portable balance (Ainsworth, Denver, CO, Model SC-2000 Electronic Balance) to determine volumetric water content. Ten cores from each rotation/tillage treatment and at each of the three depths (0-7.5 cm, 7.5-15 cm, and 15-22.5 cm) were sampled and taken into the lab for determination of bulk density, soil moisture retention in the 1 to 100 kPa matric suction range, and pore size distribution. Soil cores were saturated by wetting from the bottom for at least 48 hours. Soil water retention in the 1 to 6 kPa range (1, 2, 3, 4, and 6 kPa suction) was determined using a tension table (Leamer and Shaw, 1941). A pressure plate apparatus (Richards, 1965) was employed to determine soil moisture retention at matric suctions of 10, 33.3, and 100 kPa. Cores were oven dried at approximately 104 degrees C for 24 hours and weighed for bulk density determination.

Total porosity of the soil was assumed to be equal to the amount of water loss between saturation and oven drying. Air porosity at each matric suction was determined by subtracting the measured volumetric water content from the total porosity. Pore sizes were determined on the basis of corresponding matric suctions and effective pore size drainage using the capillary rise formula (Vomocil, 1965).

Soil moisture was monitored weekly beginning July 14, 42 days after planting, to a depth of 60 cm. Soil cores (2.5 cm diameter) were removed from the 0-10 and 10-20 cm depths and moisture at each depth determined gravimetrically. Two samples for each treatment combination (two rotation/

tillage x two irrigation variables) were collected. Each of the samples contained ten subsamples which were collected from randomly selected sites in two reps of each treatment area. The soil was dried at 105 degrees C for 24 hours and moisture content calculated as the mass of water relative to the mass of dry soil. Volumetric moisture (%) at 30, 45, and 60 cm was determined using a neutron moisture meter (Campbell Pacific Nuclear, Model 50) in aluminum access tubes inserted perpendicular to the soil surface.

Emergence counts were taken on June 13 and 21. Bean seedlings emerged were counted in 30.5 m of row from the center two rows (15.25 m of each row) of each plot.

Above ground biomass was measured weekly beginning July 14. Ten successive plants in a row were removed (by cutting at the soil surface) from a center row of each plot, dried 48 hours at 70 degrees C, and weighed.

Root growth and distribution was studied by destructive sampling. Destructive root sampling was carried out using the method described by Srivastava et al. (1982). This method involves removal of a soil profile 7.5 cm x 22.5 cm x 45 cm by means of a hammer driven profile sampler mounted on a tractor. Each profile was partitioned into 18 cubes, each of which was 7.5 cm on a side. Profiles were thus divided into a 3 by 6 array of cubes. Profiles were removed at the time of maximum flowering in mid-August. Each profile was taken from the center of the plant to 22.5 cm away from the plant and perpendicular to the row. It was assumed that the

profile represented approximately one half of the root system of one plant. The soil cubes were soaked for 8 to 16 hours in a solution of 5% sodium hexametaphosphate to aid in dispersing the clay and washing out the roots. The soil was washed from the extracted 7.5 cm cubes with a hydropneumatic elutriator (Smucker et al., 1982). Roots were stored in a solution of 20% methyl alcohol until analyzed. Root length was determined by Tennant's line-intersect method (Tennant, 1975), which is a modification of Newman's method (Newman, 1966). A four centimeter square grid was used.

By early September pod fill was nearing completion. Maturity was estimated by visual observation of the percent of plants in a given plot in which greater than or equal to 50% of the leaves had lost their chlorophyll. Maturity estimates were made on September 2, 92 days after planting, and September 8, 98 days after planting.

On September 9 and 10 nearly 12 inches of rain fell and the plots were inundated for 10 days. Plants were completely submerged and at one point as much as 1.3 m of water stood on the plots. A harvest was attempted on September 25 by pulling plants from each plot to obtain yield and yield component estimates. Most of the plots were still under several centimeters of water. The beans plants in 7.6 m of row were pulled from each plot, bagged, and placed in dryers for several days prior to mechanical threshing. Two replications were harvested. Following threshing the beans were cleaned using a shaker cleaning mill and a roller mill and then

weighed and moisture determined. Yield data was corrected to 16.5% moisture. In addition to the 7.6 m of row, five successive plants from a randomly selected location along a row unaffected by wheel traffic were pulled and dried for determination of yield components. Again, only two replications were harvested. Final population counts were made following the flood. The two reps which had not been harvested were used for these determinations. The total number of plants in six meters of row was counted.

All data was analyzed using analysis of variance. Treatment means were compared using least significant difference (LSD) appropriate for split-split-plot design arranged in randomized complete block design (Little and Hill, 1978).

RESULTS AND DISCUSSION

Soil Parameters

Soil bulk density was affected by the rotation/tillage factor as shown in Figure 1. Bulk density was measured to a depth of 22.5 cm, or just below the 20 cm plow layer. There were no significant differences at the 0.05 probability level in the bulk density in either the 0-7.5 cm or 7.5-15 cm depths. However, the conventional management system (CONV) did have a slightly higher bulk density at the 7.5-15 cm depth suggesting a moderate compaction effect due to the three passes of secondary tillage. The difference in bulk density at the 15-22.5 cm depth range was significant with the alfalfa rotation, no secondary tillage (ARNST) soils having a lower bulk density than the CONV soils. This difference can be attributed to the plowpan which existed on the CONV soil but which was broken up by the deep tillage and deep rooted alfalfa on the ARNST soil. Douglas and McKyes (1983) found that bulk density of artificially compacted fine textured soil was not decreased in the 15 to 20 cm depth range by chiseling or plowing to a depth of 25 cm but was decreased by subsoiling to a depth of 45 cm. In another study McKyes et al. (1979) found that chiseling to a depth of 30 cm effectively broke up compacted layers on a fine textured

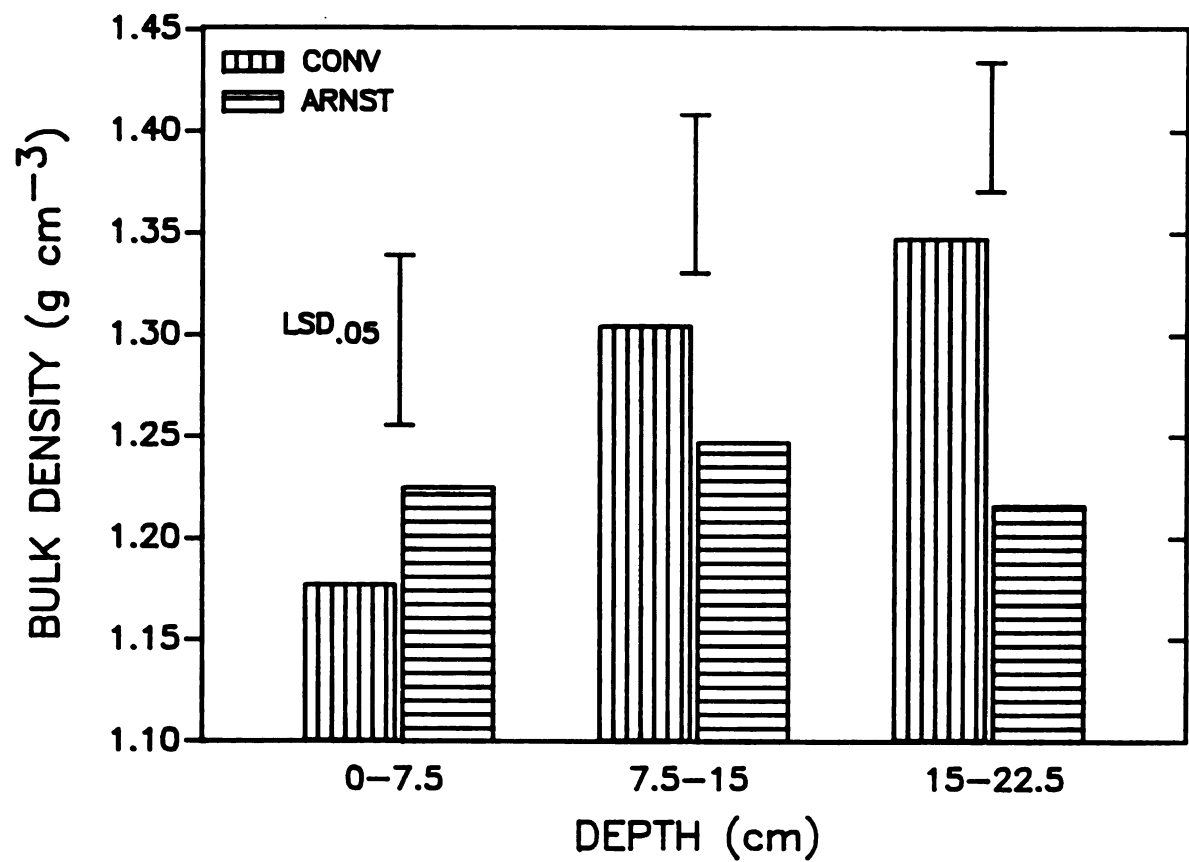


Figure 1. Soil bulk density of Charity clay at three depths as affected by rotation/tillage.

soil. Radcliffe et al. (1986) studied the effect of a deep-rooted legume on soil strength (which is closely correlated to bulk density for a given soil) and found that soil strength was decreased by the action of deep-rooted plants. In the current study the effects of deep tillage and the previous crop of alfalfa were probably combined which resulted in a decreased bulk density on the ARNST plots. From this study it was not possible to conclude that the decreased bulk density was due to either deep tillage or the alfalfa rotation.

Soil bulk density is a good indicator of soil compaction but is limited in predicting detrimental effects on plants since it reflects only changes in total porosity (Voorhees, 1983). Soil moisture and air relations and changes provide a better indication of the potential for reduced plant growth due to compacted soil. Soil moisture retention curves and air-filled porosity relationships for the top 22.5 cm of soil are presented in Figures 2, 3, and 4. Values at saturation are presented at 0.1 kPa matric suction (-0.1 kPa matric potential). The CONV soils had a higher volumetric water content than the ARNST soils at all suctions although the difference is not significant at the 95% level at the surface (Figure 2b). The differences are significant at the 7.5-15 and 15-22.5 cm depths (Figures 3b and 4b).

The differences in drainage due to rotation/tillage are important since the Charity clay soil is prone to waterlogging and related problems of poor aeration and resultant

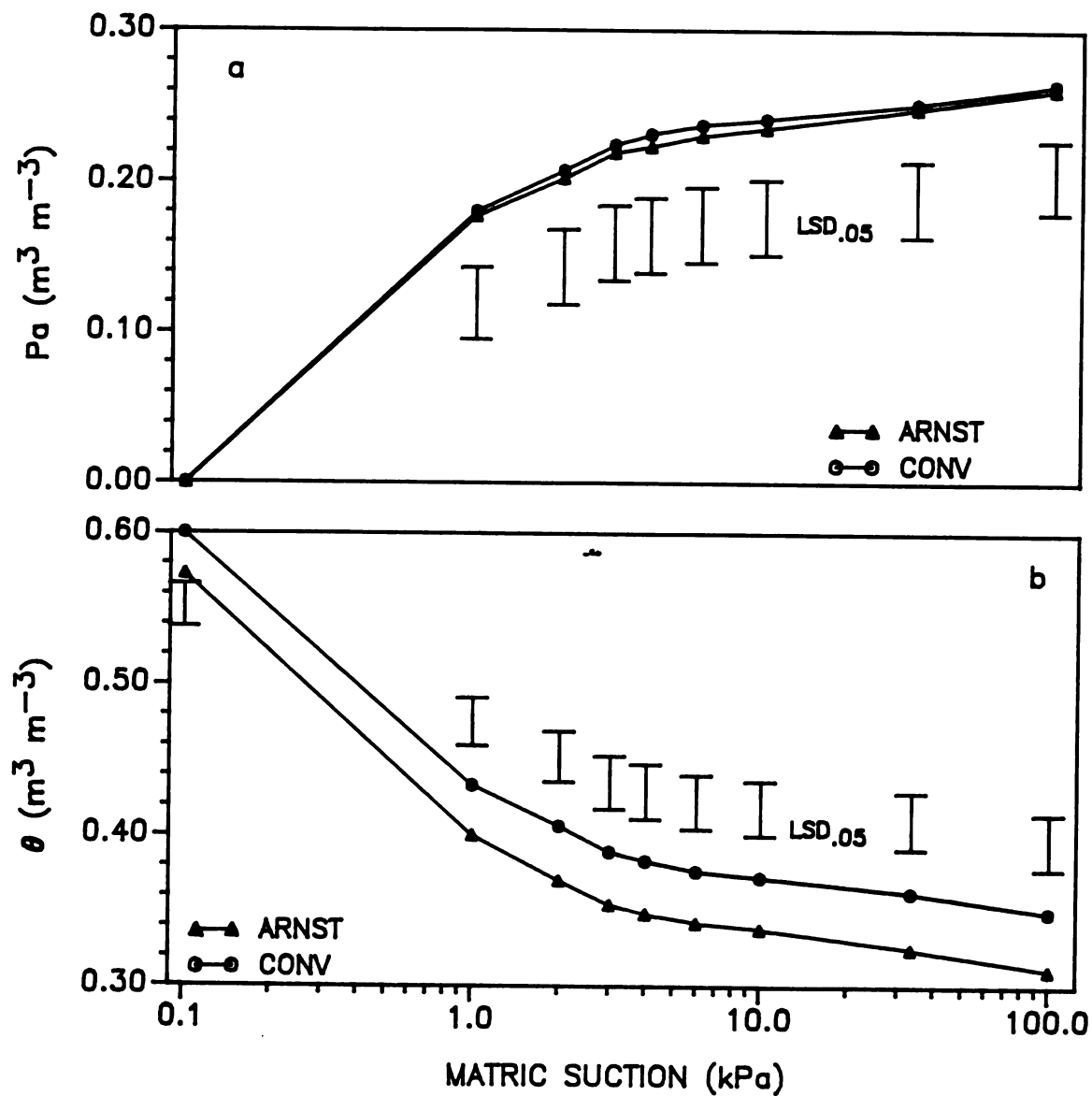


Figure 2. Air-filled porosities (a) and soil moisture retention (b) of Charity clay at different matric suctions and at the 0-7.5 cm depth as affected by rotation/tillage.

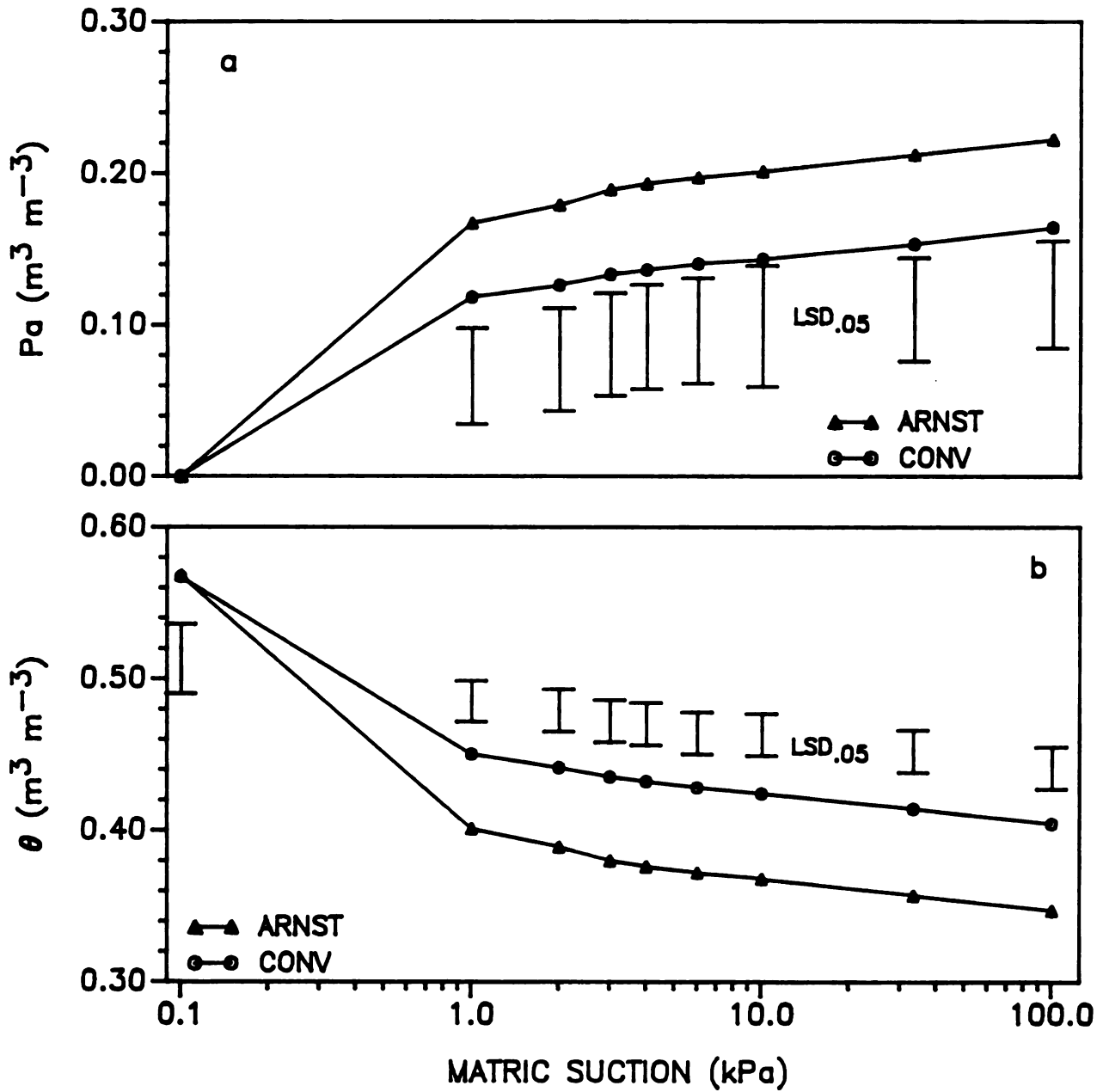


Figure 3. Air-filled porosities (a) and soil moisture retention (b) of Charity clay at different matric suctions and at the 7.5-15 cm depth as affected by rotation/tillage.

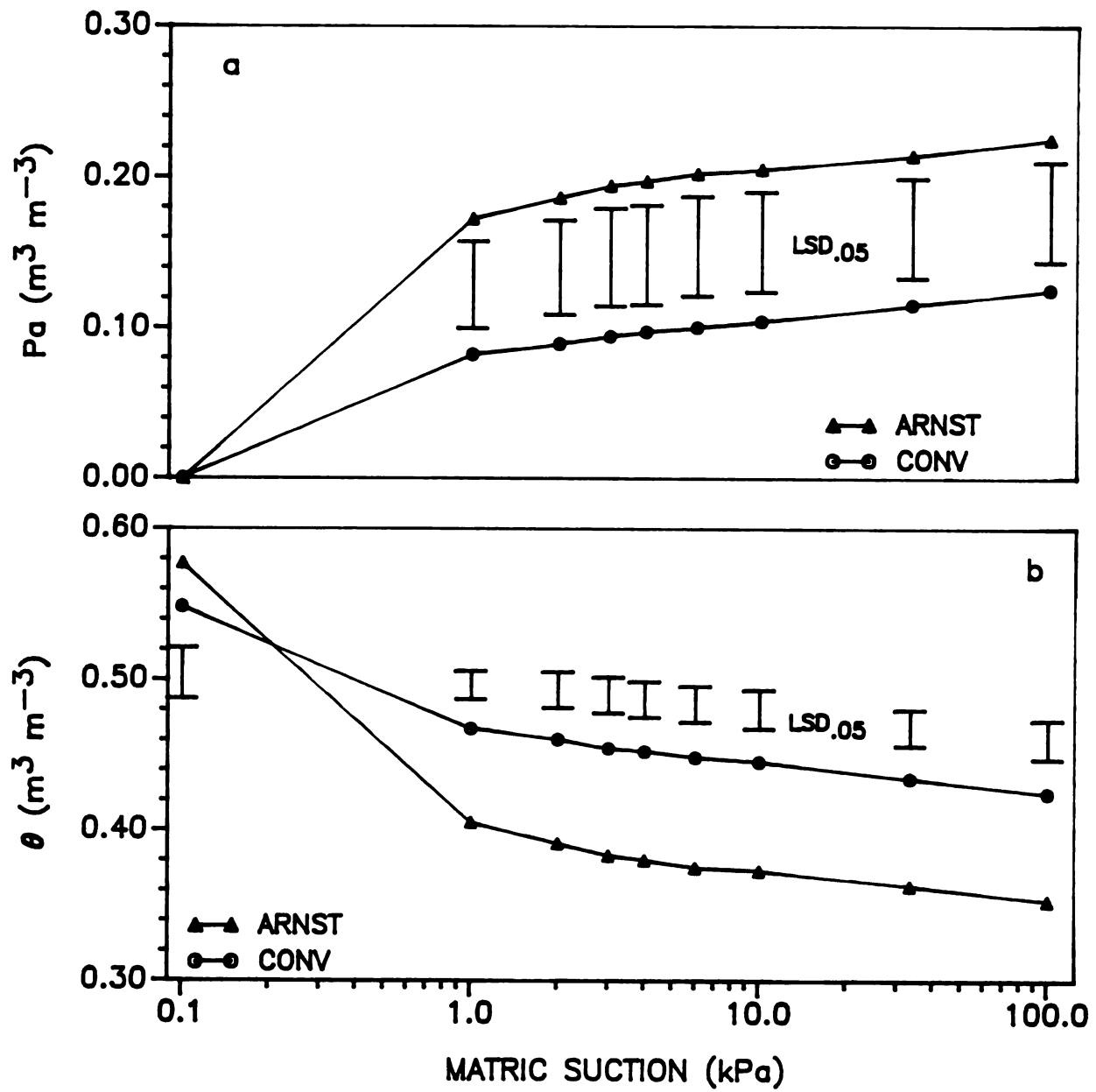


Figure 4. Air-filled porosities (a) and soil moisture retention (b) of Charity clay at different matric suctions and at the 15-22.5 cm depth as affected by rotation/tillage.

anaerobic conditions for the plant root system. Air filled porosity is shown in Figures 2a, 3a, and 4a. As with moisture retention, the differences in the top 7.5 cm are not significant (Figure 2a). Air filled porosity in the 7.5 to 15 cm depth range is higher by up to 42% on the ARNST soil than the CONV soils, although there is no statistical significance at the 95% level. While the air filled porosities at all moisture levels are lower on the CONV soils, they are always above the 10% ($0.10 \text{ m}^3 \text{ m}^{-3}$) level which has been noted as a critical level below which root growth may be affected by the lack of oxygen (Vomocil and Flocker, 1961; Grable, 1971; Erickson, 1982). At the 15 to 22.5 cm depth the differences due to rotation/tillage are more pronounced. The air filled porosity in the CONV soils at this depth are at or below 10% until 10 kPa matric suction, indicating the potential for injurious oxygen deficiencies due to waterlogging. Following a moderate to heavy rainfall or under irrigated conditions the CONV soils will take more time to reach the same air filled porosity as the ARNST soils, thus subjecting the root system to temporary but potentially serious oxygen deficiency. Erickson (1982) pointed out that aeration on most soils is usually not a problem except under heavy rainfall conditions or irrigation, and that in dry years there may be no beneficial effect of a loosened soil due to deep tillage. However, the nature of the Charity clay soil in this study predisposes it to aeration stress due to too much water or too slow drainage.

Pore size distribution for the three measured depths is shown in Figures 5 and 6. Pore size distribution, presented here by pore size radii, is essentially responsible for the air filled porosity differences noted above. Larger pores will drain at lower matric suctions than will smaller pores, as explained by the capillary model (Hillel, 1982). Pores with a radius of ≥ 150 μm will drain at a matric suction of 1 kPa, (10 cm suction) whereas pores with radii < 4.4 μm will retain water at a matric suction of 33 kPa (330 cm). A matric suction of 6 kPa will drain pores with a radius of 25 μm . Differences in pore size distribution due to rotation/tillage in the top 7.5 cm are not significant (Figure 5a). At the second depth (Figure 5b) there is an increase in large pores (radius > 150 μm) due to the ARNST management system. Although not statistically significant, the trend of decreased pore size is evident on the CONV soil and is important when considering the effect of soil compaction. In Figure 5b the increase in pores of radii < 4.4 μm is shown. Thus total pore space may or may not be affected by rotation/tillage and soil compaction factors, but size distribution can be drastically altered with potentially severe consequences for crop growth.

Pore size distribution at the 15 to 22.5 cm depth was significantly affected by rotation/tillage (Figure 6). The ARNST system had more than twice as many large pores. This indicates that the compacted layer had been broken up and soil water can drain more freely. On these soils

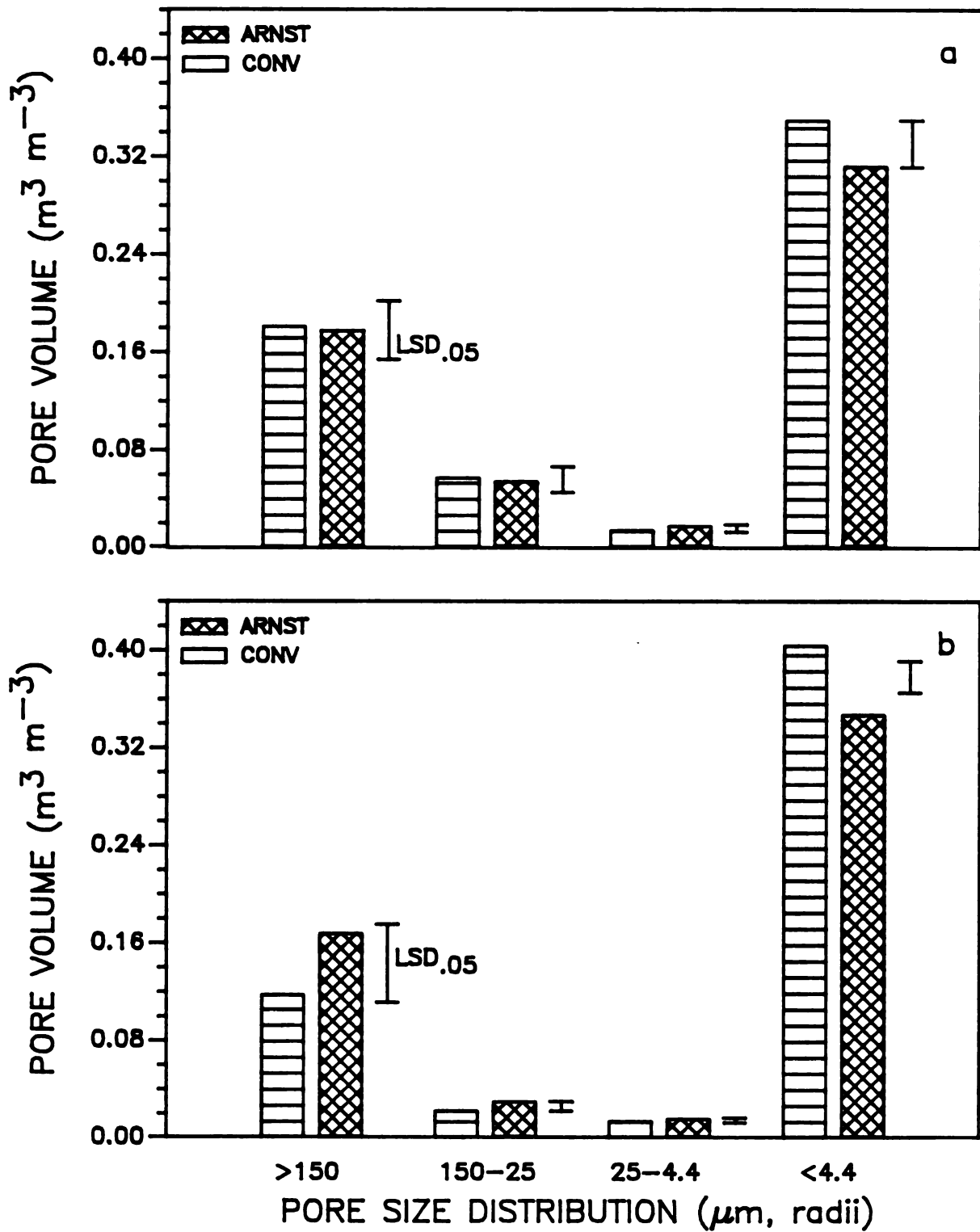


Figure 5. Pore size distribution on Charity clay at the 0-7.5 cm depth (a) and 7.5-15 cm depth (b) as affected by rotation/tillage.

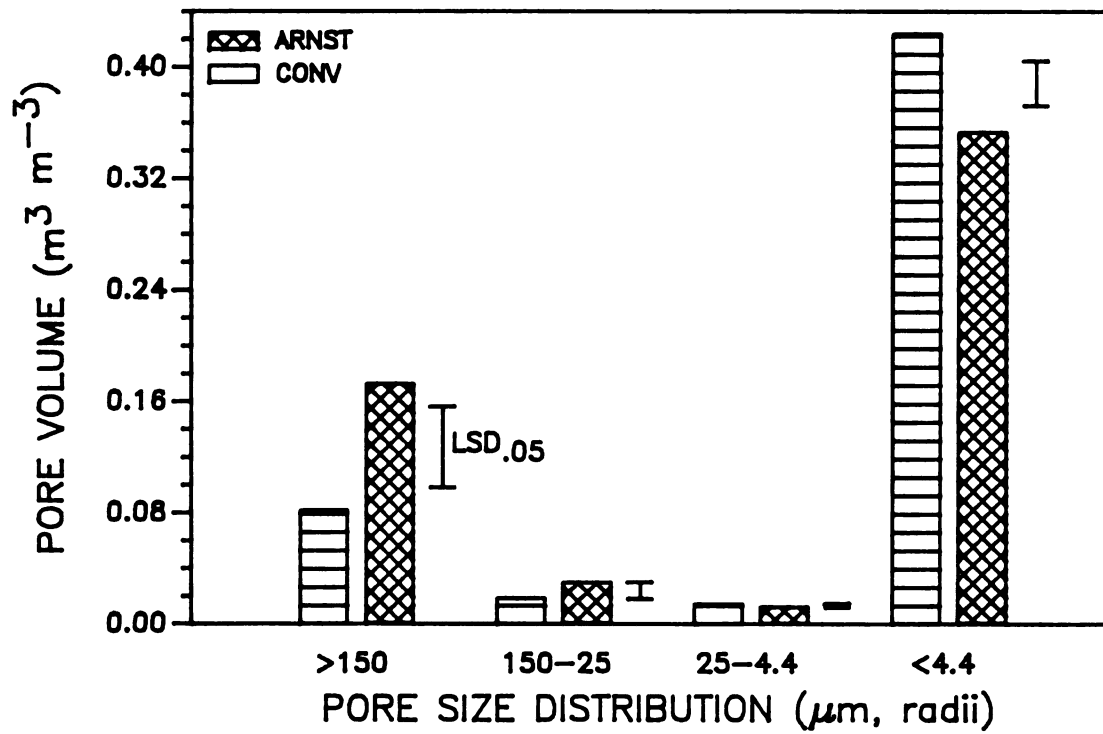


Figure 6. Pore size distribution on Charity clay at the 15-22.5 cm depth as affected by rotation/tillage.

waterlogging following rainfall or irrigation is much less likely than on the CONV soils. Conversely, the CONV soils had significantly more small pores which retain water at higher matric suctions than the ARNST soils but the water in these pores may not be available to plants.

The effects of the ARNST management system in breaking up existing compacted layers were demonstrated in the various soil parameters measured. The physical condition of this fine textured soil was notably improved by the combination of a deep rooted legume in the crop rotation and the careful management of tillage.

Soil Moisture

Further evidence of improved soil physical conditions was found by monitoring soil moisture from 40 to 100 days after planting. Soil moisture as measured gravimetrically was almost always lower on the ARNST soil compared to the CONV soil (Figure 7). On these fine textured soils problems most often arise from too much water in the soil profile and accompanying anaerobiosis, as opposed to many less fine textured soils on which water holding capacity is limited. (It is noted that high soil moisture does not necessarily mean high moisture availability to plants because water retained in fine pores may not be readily available since it is tightly held and roots may not be able to access it via the small pores. The data presented on pore size distribution suggest that the ARNST soils in this study would likely have better

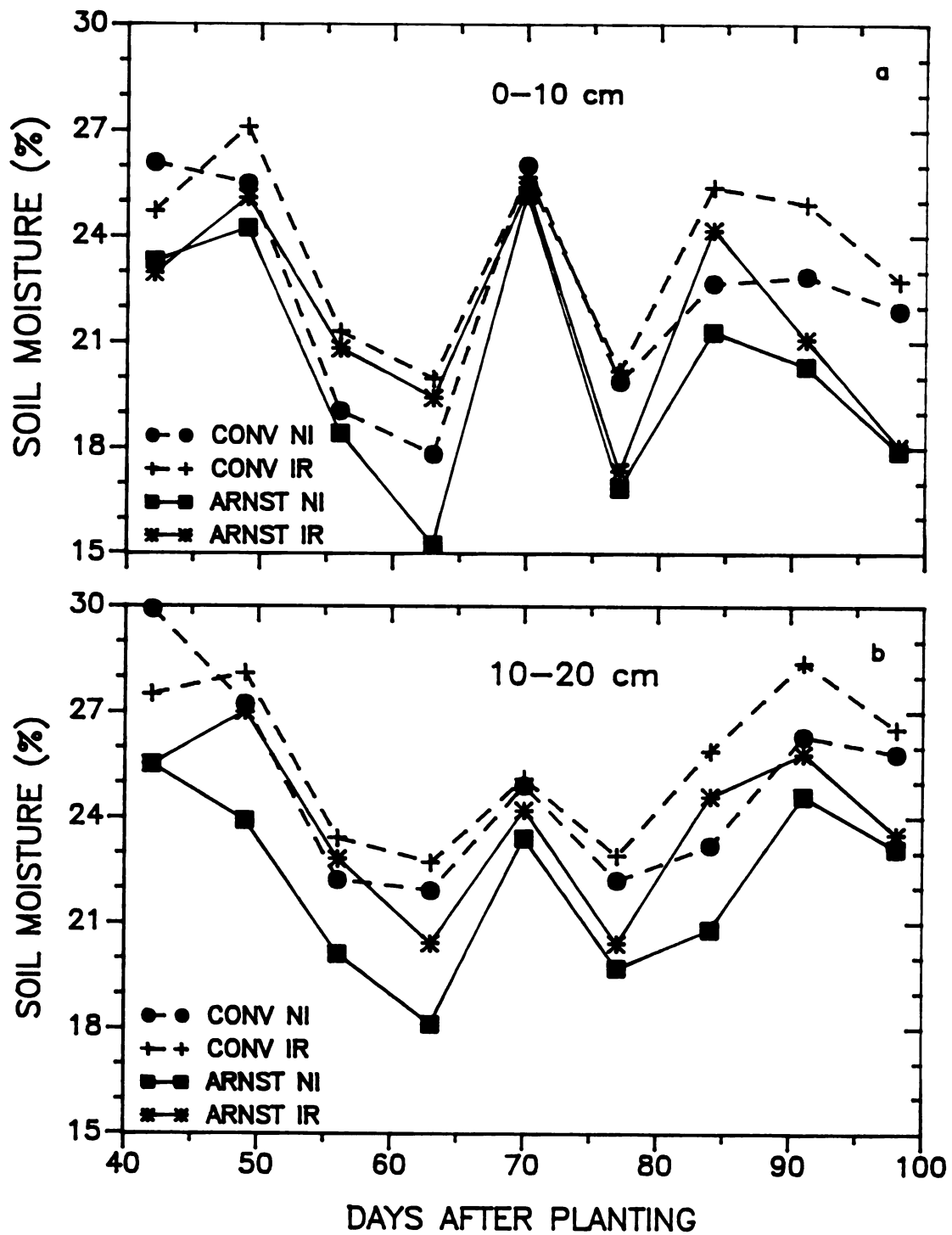


Figure 7. Soil moisture (gravimetric) over time in the 0-10 cm (a) and 10-20 cm (b) depths as affected by rotation/tillage x irrigation treatment combinations.

soil moisture availability to plants since more of the pores on this soil are in the range which would foster rapid drainage of large pores balanced with good water holding capacity. Unger et al. (1981) and Russell (1977) have reviewed this subject in some detail.) Throughout the growing season the moisture content of the ARNST soils was lower than CONV soils both with and without irrigation. In fact, in Figure 7b it can be seen that ARNST irrigated treatment combination had lower soil moisture than the nonirrigated CONV soils seven of nine sampling dates throughout the growing season.

Soil moisture at three depths below the plow layer was monitored using a neutron moisture meter probe. Data for the 30 cm (20-30 cm) and 45 cm (35-55 cm) depths are presented in Figure 8 (volumetric moisture content). Soil moisture at the 60 cm (50-70 cm) depth was also measured but is not reported as it was very similar to that at 45 cm. Below the plow layer the ARNST-treatment, both irrigated and nonirrigated, consistently had lower soil moisture than the CONV soils. The graphs in Figures 7 and 8 show that fluctuations in soil moisture content due to both irrigation and rainfall are similar but the ARNST seems prone to wider swings. This is another indication of the improved drainage due to decreased soil compaction.

Figure 9 compares soil moisture fluctuation with rainfall and irrigation. Irrigation was applied at the rate of approximately 2.5 cm per week. The soil moisture data reflects the rainfall events more prominently than irrigation.

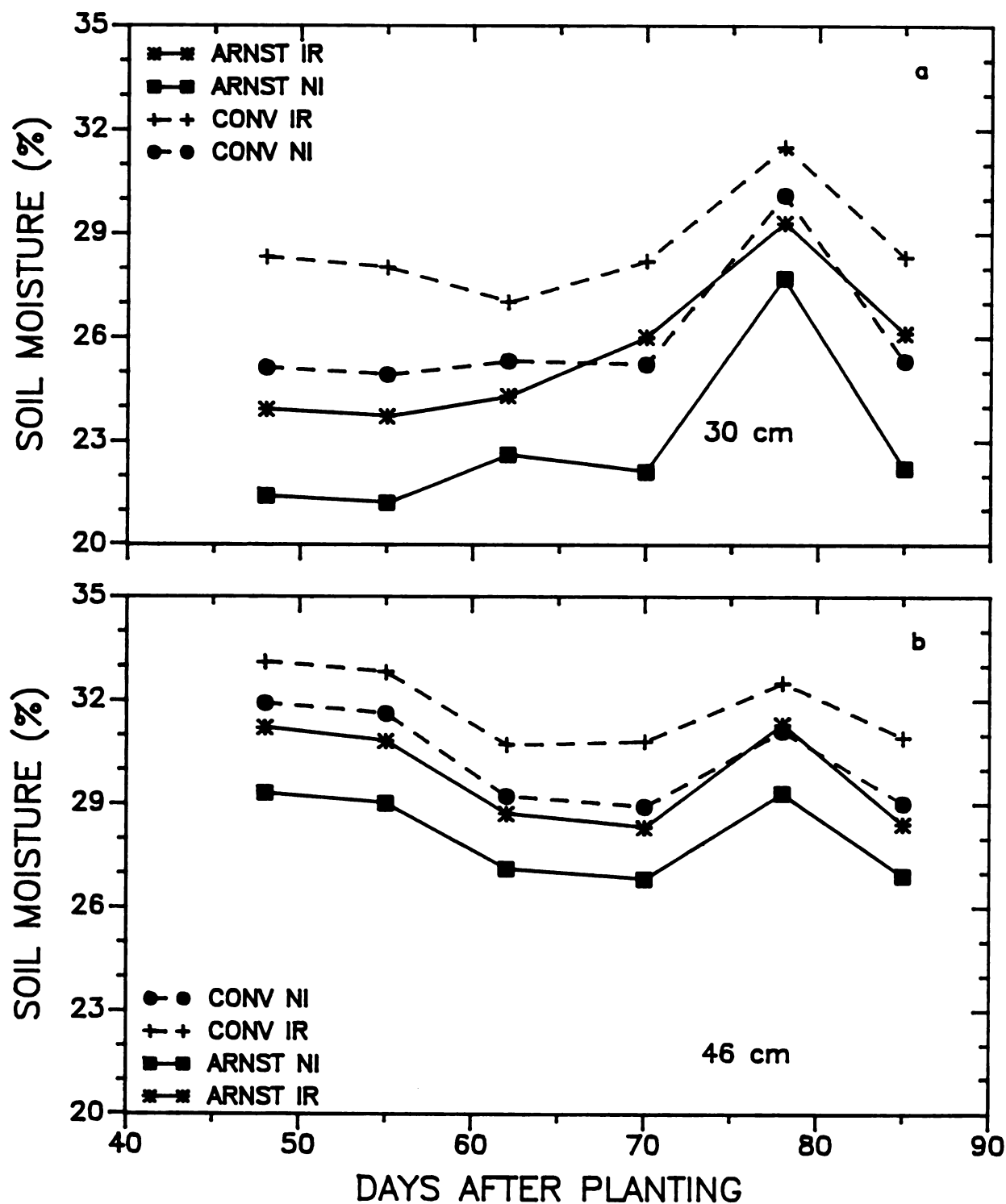


Figure 8. Soil moisture (volumetric) as measured by the neutron moisture meter over time at the 30 cm (a) and 45 cm (b) depth as affected by rotation/tillage x irrigation treatment combinations.

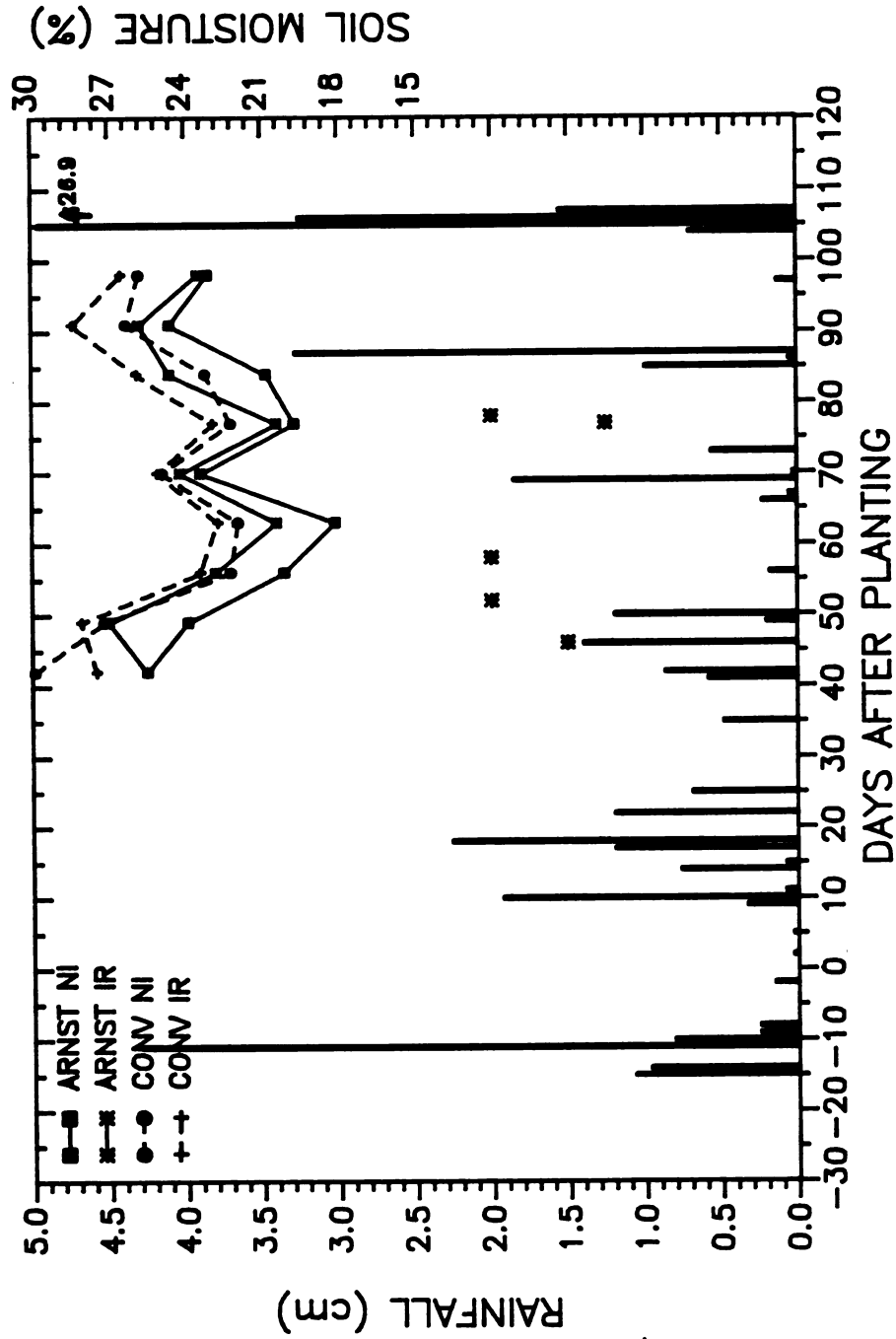


Figure 9. Rainfall throughout the growing season and soil moisture at the 10-20 cm depth from 40 days after planting until harvest as affected by rotation/tillage x irrigation treatment combination. Asterisks represent irrigation dates and amounts.

1986 was a year of good rainfall and irrigation did not have as pronounced an effect as would be expected in a year with less natural precipitation.

In summary, the measured soil physical properties and soil moisture determination support the hypothesis that the soil under the ARNST management system is in a preferred condition for plant root growth. These soils are less dense and have a higher proportion of larger pores which aid in drainage than the conventionally managed soils. Soil moisture measurements confirmed that the ARNST soils drain more readily than the CONV soils. This soil environment should be advantageous for dry bean growth and production.

Plant Parameters

Emergence counts were taken on two dates, 11 and 19 days after planting. The data is presented in Figures 10 and 11. The analysis of variance showed a significant difference in emergence due to row spacing (Figures 10a and 10b). Since the intrarow seed spacing was the same regardless of width between the rows, the emergence differences were unexpected. It seems plausible that the drill dropped seed at different rates on given rows and this could explain the row spacing variations.

Figure 11 presents comparisons of the effects of cultivar and rotation/tillage on emergence. Analysis of variance was carried out by averaging across row spacings. At eleven days after planting the ARNST had 21% higher emergence but

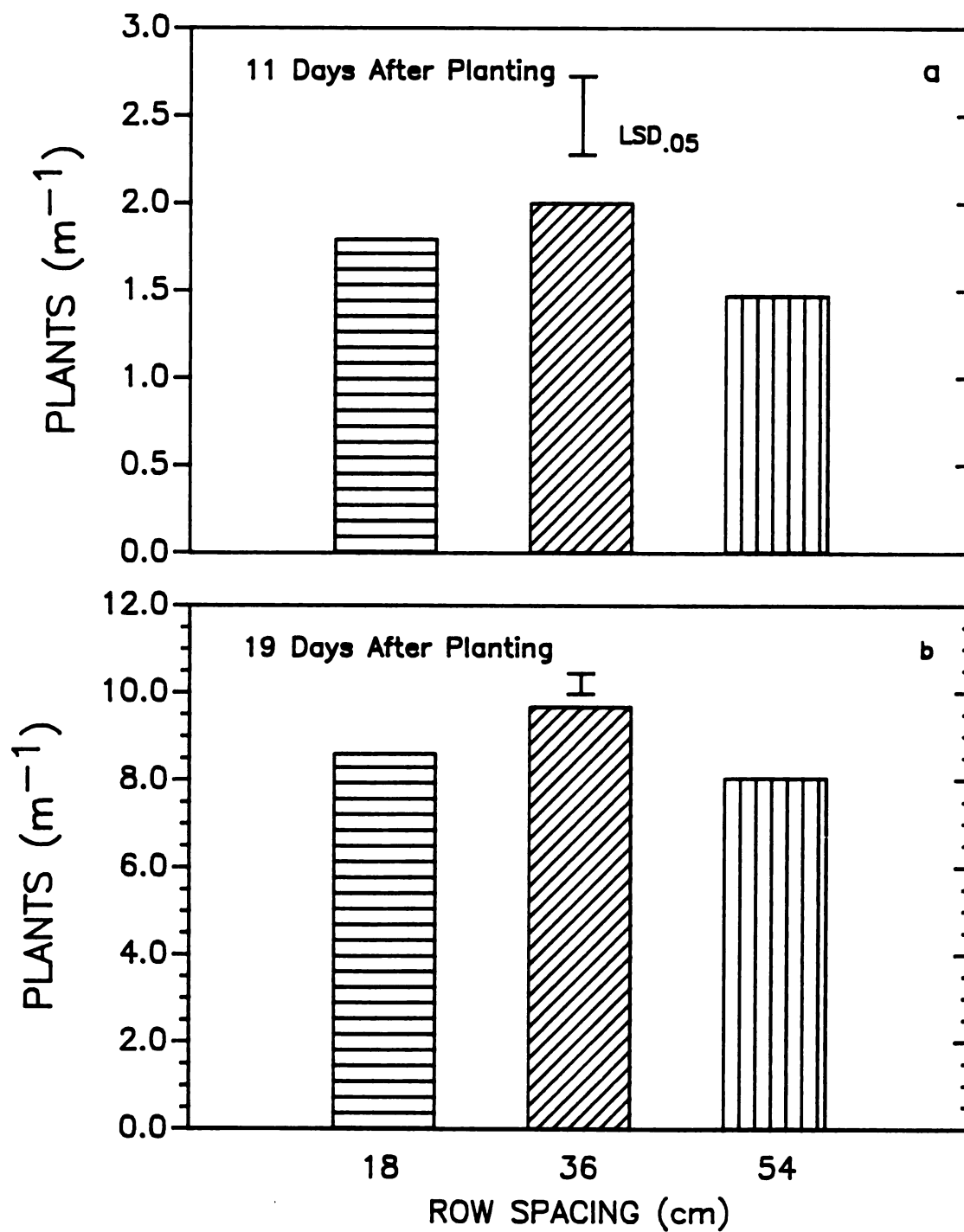


Figure 10. Emergence of dry beans on Charity clay as affected by row spacing at 11 days after planting (a) and 19 days after planting (b).

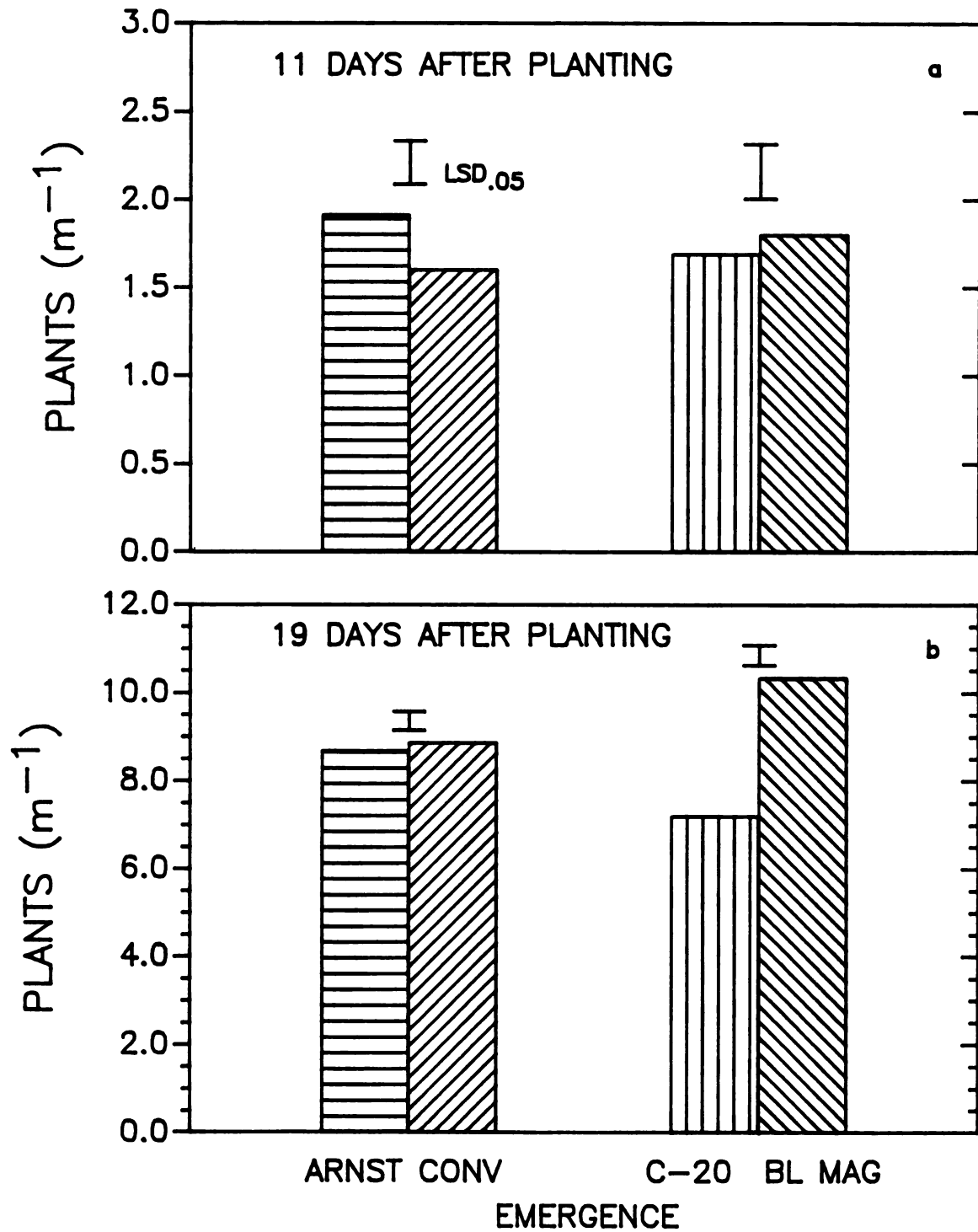


Figure 11. Emergence of dry beans on Charity clay at 11 days after planting (a) and 19 days after planting (b) as affected by rotation/tillage or cultivar.

this difference was not significant (Figure 11a). The emergence counts taken 19 days after planting showed less than one percent difference due to rotation/tillage. Others have reported decreased emergence due to soil crust formation on soils which have been conventionally tilled and are finely pulverized (Phillips and Kirkham, 1962; Bowen, 1981). The conditions in 1986 were such that no serious crust formed on the CONV soils.

Emergence of the plants of the Black Magic cultivar was 43% better than that of the C-20 variety 19 days after planting. This difference was not present 11 days after planting, although Black Magic did exhibit slightly higher emergence than the C-20 on the first counting date. This higher emergence, in spite of lower germination (90% for C-20, 73% for Black Magic) shows that the Black Magic bean is a hardier cultivar than is the C-20.

Above ground biomass was measured approximately weekly. The data for the treatment main effects is shown in Figures 12-15. Biomass data is based on the weight of 10 plants at each sampling and reported on a unit area basis using final population data to determine the number of plants to include in a square meter. There were very few significant differences throughout the growing season. The obvious exception is biomass differences due to row spacing (Figure 12). Since the intrarow seeding rate was the same at all row spacings there were large differences in total plant numbers per unit area and these differences are reflected in biomass per unit

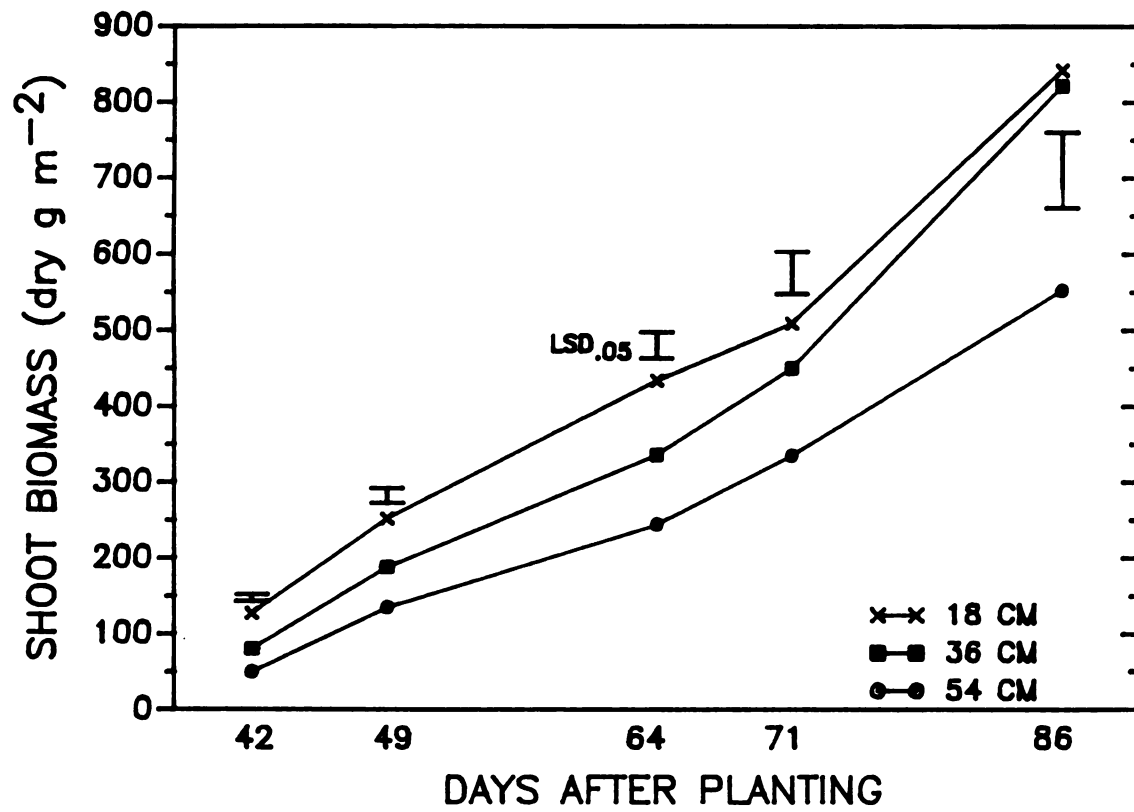


Figure 12. Dry bean shoot biomass on five dates as affected by row spacing.

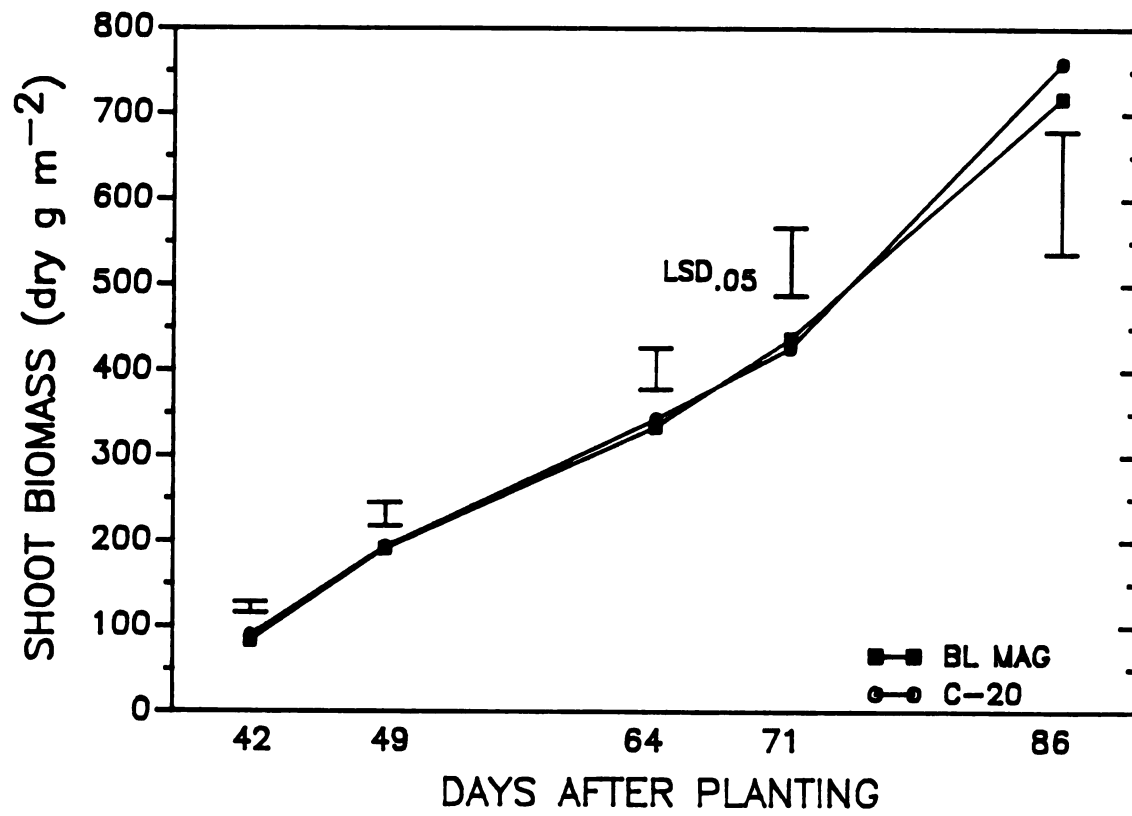


Figure 13. Above ground biomass of two dry bean cultivars on five dates.

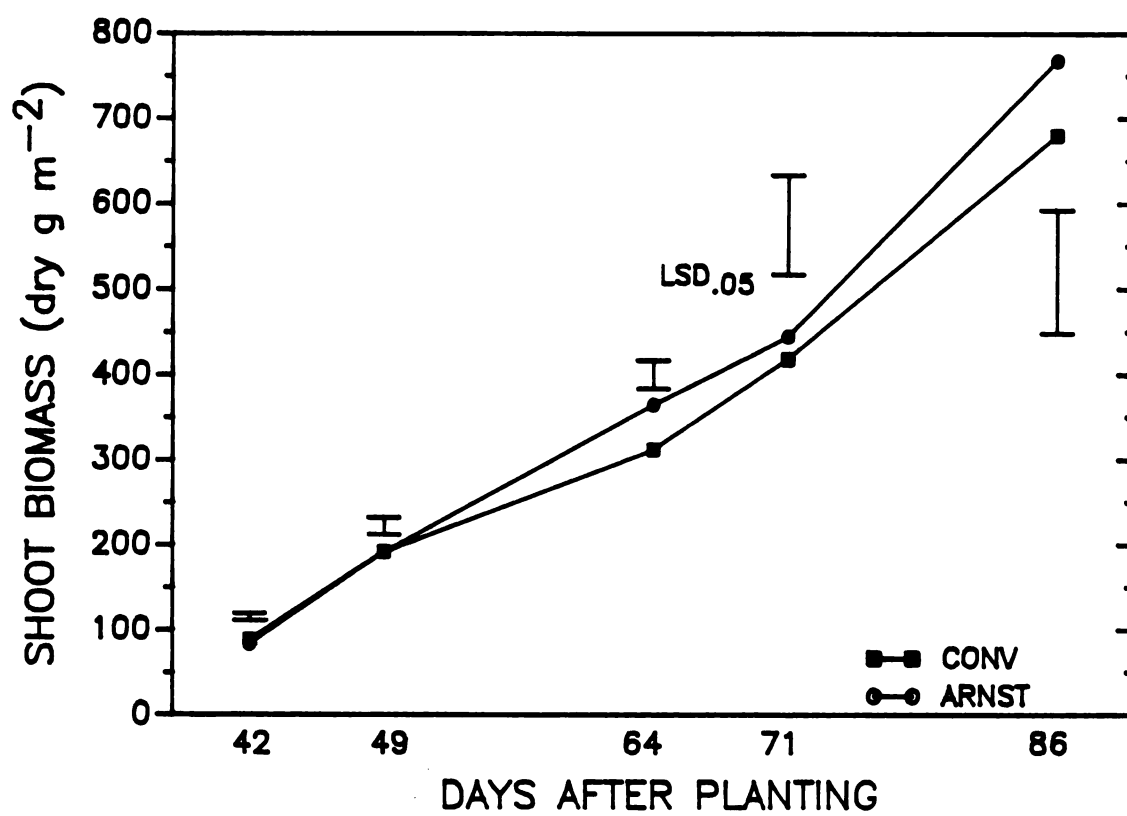


Figure 14. Above ground biomass of two dry bean cultivars on five dates as affected by rotation/tillage.

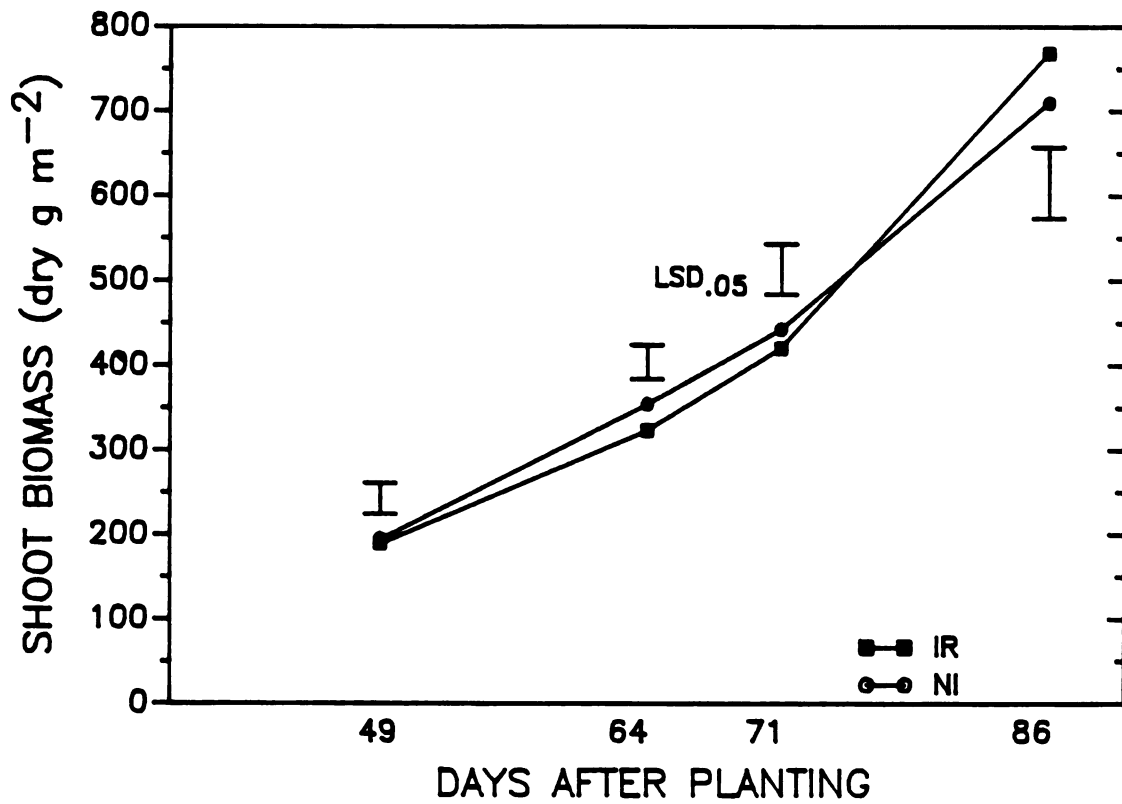


Figure 15. Dry bean shoot biomass on five dates as affected by irrigation.

area, with the narrow rows having higher above ground biomass.

Figure 13 shows that there were no differences in biomass between the two cultivars. There were small differences due to the rotation/tillage system used, as shown in Figure 14. Throughout the growing season the plants on the ARNST soil had higher shoot weight than the CONV soils, although the only statistically significant differences were found on August 4, 64 days after planting. The irrigation data presented in Figure 15 shows that irrigation did not make any difference in shoot growth throughout the season. An examination of the rainfall distribution offers some help (Figure 9). 1986 did not have any prolonged drought periods during the critical stages of flowering and pod fill, although from day 50 to 68 (days after planting) very little rain fell. Apparently there was sufficient soil moisture and root proliferation to sustain growth during that period. Shoot growth was greater under nonirrigated conditions early in the season but the plants that were irrigated caught up and eventually surpassed them in shoot growth. The irrigated beans may have experienced some aeration stress early in the season when additional water was applied and slightly reduced growth resulted compared to the nonirrigated beans.

Differences in shoot growth were small in 1986 and there were no significant differences due to the treatment factors except for row spacing and, on one date, rotation/tillage.

Root Responses

Root responses to the various treatment factors were measured by using minirhizotrons and microvideo recording equipment and by destructive sampling. The results of the data collected from the minirhizotrons were contrary to the results from the destructive sampling. This led to the conclusion that the minirhizotron is not a suitable tool for root study on the fine textured Charity soil. The minirhizotron data is not presented here since it is not germane to the discussion of root responses to the treatments in this study. Chapter 3 is a discussion of the minirhizotron as a root study method and includes the data from this study.

Root growth was studied by destructive sampling at the time of maximum flowering or early podset. Only the 54 cm row spacing samples were analyzed to determine root length and root length density (RLD). Table 1 presents a summary of the root length densities as affected by rotation/tillage, irrigation, and cultivar. The three subsamples at each depth were averaged and analysis of variance carried out to compare the effect of each treatment at each particular depth. There was no significant effect on RLD at any depth due to rotation/tillage.

The differences detected were unanticipated. Since the CONV soil was shown to have a higher bulk density and other positive compaction indicators, it seems logical that root growth would be less than on the non- or less-compacted soil. This should be especially evident below the plowpan

Table 1. Root length densities of dry beans as affected by rotation/tillage, irrigation, and cultivar. Destructive sampling was in mid-August, which was maximum flowering.

Depth (cm)	<u>Rotation/ Tillage</u>		<u>Irrigation</u>		<u>Cultivar</u>	
	ARNST	CONV	IR	NI	C-20	BlMag
	root length density (cm cm ⁻³)					
0-7.5	2.90	3.07	3.18	2.78	2.47	3.49*
7.5-15	2.51	2.19	2.52	2.18*	2.09	2.61*
15-22.5	2.10	1.96	2.01	2.05	1.93	2.13
22.5-30	1.24	1.59	1.41	1.42	1.23	1.60*
30-37.5	0.82	1.19	1.04	0.97	0.85	1.16*
37.5-45	0.80	0.98	0.88	0.91	0.72	1.07*

Averages

0-22.5	2.50	2.41	2.57	2.34	2.16	2.74*
22.5-45	0.95	1.26	1.11	1.10	0.93	1.28*

* Indicates significant differences at the 95% level.

shown to exist in the 15-22.5 cm depth range on the CONV soils. The RLD results are contrary to this supposition. Note the lower three depths all have higher RLDs on the CONV treatment than the ARNST plots, although the differences are not statistically significant at the 5% level.

The "rootmaps" presented in Figure 16 compare the RLDs under the two rotation/tillage systems and averaged across irrigation and cultivar. The CONV rootmap shows a concentration of roots in the top 7.5 cm of soil, whereas the ARNST system has more even root distribution throughout the top 23 cm of soil. This may indicate a secondary tillage compacted layer which restricts root growth at around the 7.5 cm depth.

Irrigation effects were also not significant with the exception of the second depth, 7.5-15 cm, resulting in increased root length density vs nonirrigated plots. This is consistent with the shoot biomass data which also showed no differences due to irrigation.

Root length densities were significantly different for the two cultivars. These differences reflect the hardier, more extensive root system of the Black Magic cultivar. The differences in cultivar RLDs were evident throughout the soil profile. The "rootmaps" in Figures 17 and 18 show the RLDs throughout the extracted profiles for the ARNST nonirrigated by cultivar plots and the CONV irrigated by cultivar treatments. Note the concentration of roots in the top 7.5 cm, especially on the CONV soils (Figure 19), and the much higher RLDs for the Black Magic cultivar on these two rotation/

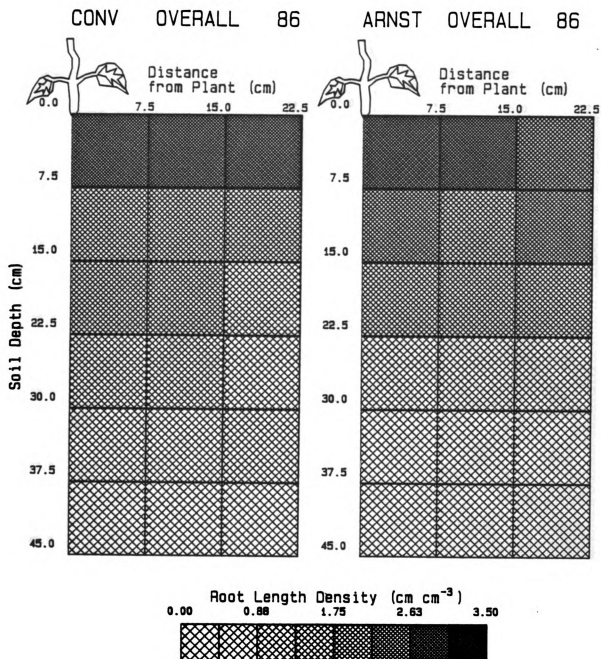


Figure 16. The influence of rotation/tillage on root length densities of dry beans as determined by destructive profile sampling at the time of maximum flowering.

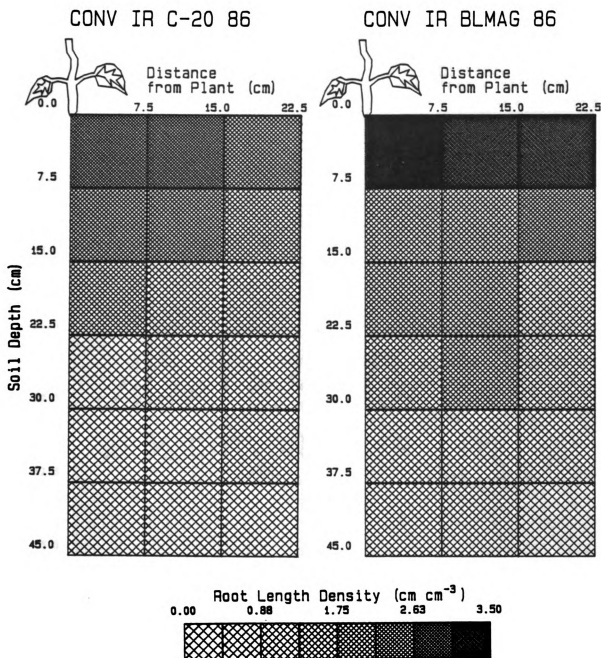


Figure 17. Root length densities of dry beans as determined by destructive sampling at the time of maximum flowering and as affected by cultivar under conventional rotation/tillage and irrigated conditions.

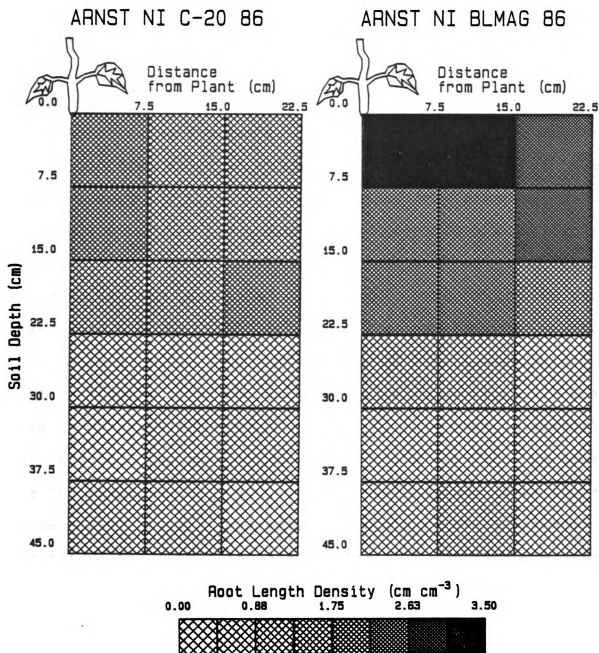


Figure 18. Dry bean root length densities as determined by destructive sampling and as affected by cultivar under an alfalfa rotation/no secondary tillage, nonirrigated management system.

tillage by irrigation treatment combinations.

The root data, like the shoot data, revealed few differences due to the rotation/tillage or irrigation treatments. There were significant differences between the two cultivars.

Harvest Parameters

Maturation of dry beans is important in Michigan because harvest is often affected by adverse weather. Cultivars have been developed to take advantage of the full growing season in the state but the beans must mature rapidly and uniformly to offer the best possibility for a timely harvest.

Maturity was estimated by visual observation on two dates, Sept. 2 and Sept. 8. Maturity was recorded as the percentage of plants with > 50% of the leaves losing chlorophyll. Table 2 shows the data as estimated on Sept. 2. The range of maturity percentages was 0 to 60, with an overall mean of 9.1%. The coefficient of variation was 70%. On this date the beans on the CONV plots were more mature than those on the ARNST plots. Nonirrigated plots were significantly more mature than irrigated plots. The C-20 variety was maturing more rapidly than the Black Magic cultivar, with significant differences at some row spacings (generally the more narrow spacings) and depending on the other treatment combinations.

Maturity on Sept. 8 is presented in Figure 19. There were significant interactions between row spacing and irrigation and between row spacing and rotation/tillage.

Table 2. Influence of rotation/tillage, irrigation, cultivar, and row spacing on maturity of dry beans as estimated on September 2. Percent maturity was visually estimated as the percent of plants in a plot whose leaves had lost 50% or more of their chlorophyll.

<u>Rotation/Tillage</u> ----		ARNST		CONV	
<u>Irrigation</u> ----		IR	NI	IR	NI
<hr/>					
	<u>Row Spacing</u>	maturity (%)			
<u>Cultivar</u>		12.5	20.0	8.8	27.5
C-20	18	12.5	20.0	8.8	27.5
	36	3.8	15.0	7.5	40.0
	54	1.3	5.0	5.0	25.0
BlMag	18	2.5	8.8	0	8.8
	36	0	6.3	0	6.3
	54	0	6.3	0	8.8

LSD.05 - for Rotation/Tillage means at same or different
 Irrigation, Cultivar, or Row Spacing = 10.7.
 - for Irrigation means at same Rotation/Tillage,
 same or different Cultivar or Row Spacing = 8.7.
 - for Cultivar or Row Spacing means at same
 Rotation/Tillage and Irrigation = 9.0.

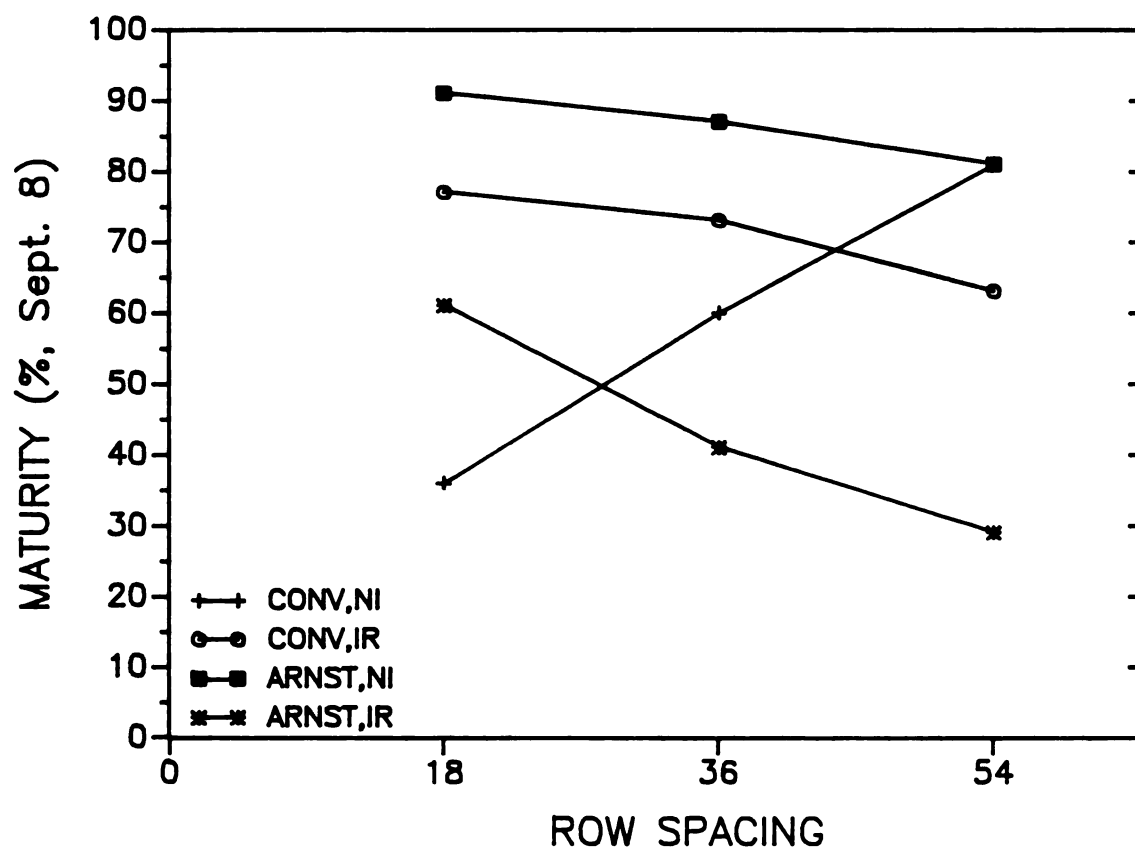


Figure 19. The effects of row spacing, rotation/tillage, and irrigation on dry bean maturity 98 days after planting. Table 3 shows statistical comparison of these treatment interactions.

No significant differences due to cultivar were detected on September 8. The sums of squares of the interactions were partitioned into single degree of freedom components and subjected to F tests to examine more closely the nature of the interaction. The variance components of the interaction are presented in Table 3. The trend analysis in Table 3 shows that the effect of row spacing on maturity was linear for all combinations of rotation/tillage x irrigation, although the differences in maturity due to row spacing were significant only for the ARNST-irrigated and CONV-nonirrigated treatments. Figure 19 shows that the effect of row spacing on maturity was opposite for the CONV-nonirrigated plots (increasing maturity as row spacing increased) compared to the other treatment combinations (decreasing maturity as row spacing increased). The effect of irrigation averaged over the other factors was also significant on Sept. 8, with the irrigated plants maturing more slowly than the nonirrigated plants.

Final population counts were made following the mid-September flood. Only two replications were available for plant counts. Based on this determination the only significant differences in final plant population per square meter were between the row spacings as shown in Figure 20. Final population was 472,000 plants ha^{-1} on the 18 cm row spacing plots, 276,800 plants ha^{-1} on the 36 cm rows, and 177,100 plants ha^{-1} at the widest row spacing. There were slightly higher (13%) populations on the ARNST plots at all three row

Table 3. Trend analysis for components of the irrigation-row spacing-rotation/tillage interactions for plant maturity as estimated on Sept. 8. Each line tests significance of row spacing on maturity under given treatments.

Source of Variation*	df	SS	MS	Observed F	Significant F(.05)
	8	13603			
ARNST, NI					
linear	1	400		.84	
quadratic	1	5		--	
ARNST, IR					
linear	1	4096		8.67	*
quadratic	1	85		--	
CONV, NI					
linear	1	8100		17.16	*
quadratic	1	12		--	
CONV, IR					
linear	1	784		1.66	
quadratic	1				
Error (c)	60	28320	472		

* ARNST = alfalfa rotation, no secondary tillage, CONV = conventional rotation/tillage, NI = nonirrigated, IR = irrigated.

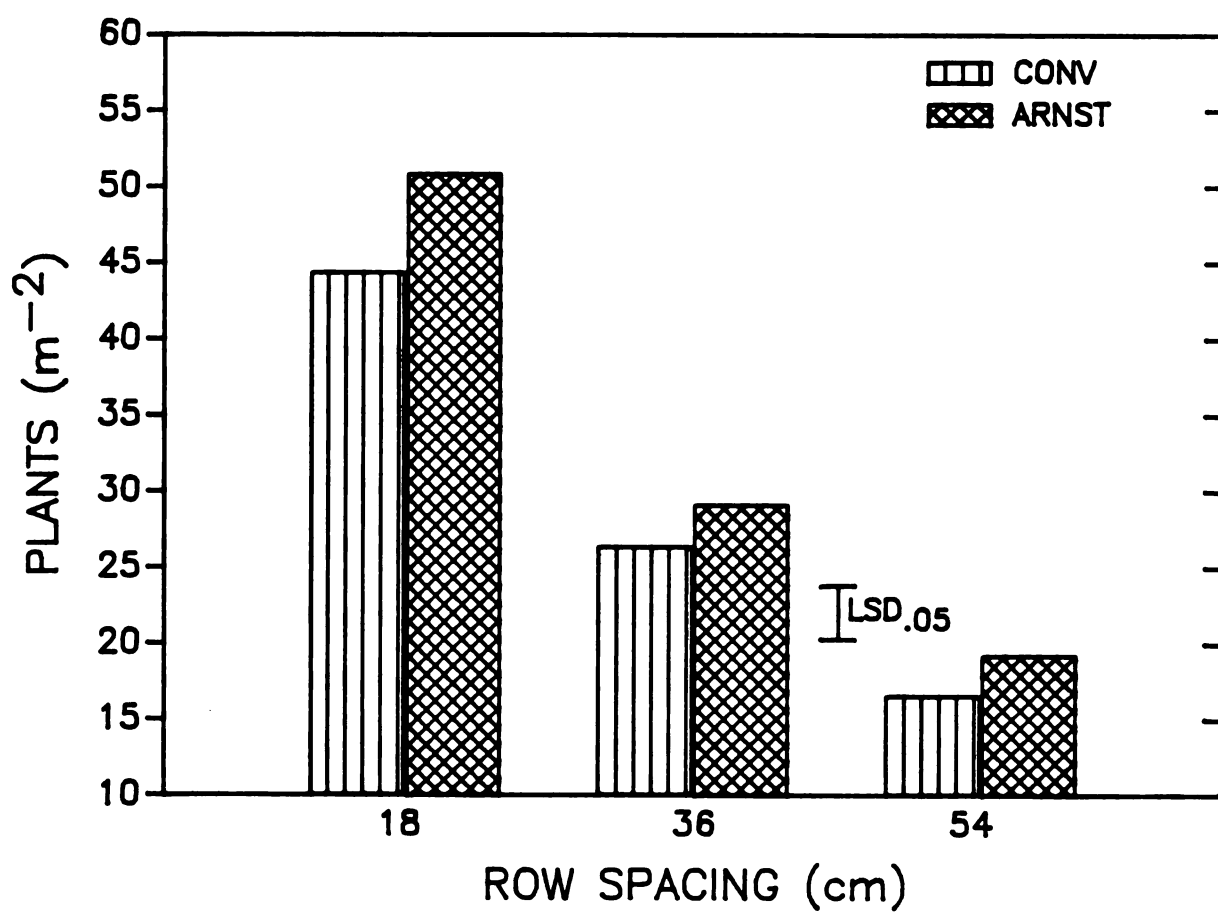


Figure 20. Final population of dry bean plants at three row spacings as influenced by rotation/tillage.

spacings (Figure 20) but these differences were not significant on the basis of the two replications.

Since this was a management study the treatment effects on yield were a key component. Unfortunately, on September 9 it began to rain and about 30 cm (12 inches) of rain fell within 36 hours. The crop was nearing maturity as indicated by the maturity estimates made on Sept. 8. The plots were flooded for over 10 days with as much as 1.2 meters of water. On Sept. 24 an attempt was made to harvest some of the plots in order to obtain yield estimates. Two replications were harvested by pulling 7.6 m of row from each plot.

Yield is reported in Table 4 for all 24 treatment combinations. While there are some large differences in yield, significance was difficult to detect since only two replications were harvested. The only significant treatment effects were cultivar and row spacing, as shown in Table 4 and Figure 21. The Black Magic cultivar outyielded C-20 by 35%. The row spacing differences were also striking with the 36 cm rows yielding 31% more than the 54 cm rows and the 18 cm rows yielding 72% more than the 54 cm rows. While these differences are large it should be remembered that three times as many seeds were planted on the 18 cm rows vs the 54 cm rows. Dry beans will compensate for some differences in plant population but this nearly threefold difference is too great for the wider rows to overcome. The mechanism by which yields are increased when plant rows are closer together is thought to be increased radiation interception at the time of

Table 4. Influence of rotation/tillage, irrigation, cultivar and row spacing on dry bean yield. Figures are the average of two replications.

<u>Rotation/Tillage</u> ----		ARNST		CONV	
<u>Irrigation</u> ----IR		NI		IR	NI
<u>Cultivar</u>	<u>Row Spacing</u>	kg/ha			
	18	1909	2454	2097	2997
C-20	36	985	1696	1971	2258
	54	847	1490	1072	1867
	18	2878	3368	2881	3097
BlMag	36	2045	2639	1987	2999
	54	1223	2251	1969	1921

LSD.05: -for Rotation/Tillage means at same or different Irrigation, Cultivar, and Row Spacing = 2229.
 -for Irrigation means at same Rotation/Tillage & same or different Cultivar and Row Spacing = 770.
 -for Cultivar or Row Spacing means at same Rotation and Irrigation = 785.

Significant (p=.95) Main Effects:

<u>Treatment</u>	<u>Variable</u>	<u>Yield (kg/ha)</u>	<u>LSD.05</u>
Cultivar	C-20	1803	226
	BlMag	2438	
Row Spacing (cm)	18	2710	278
	36	2073	
	54	1580	

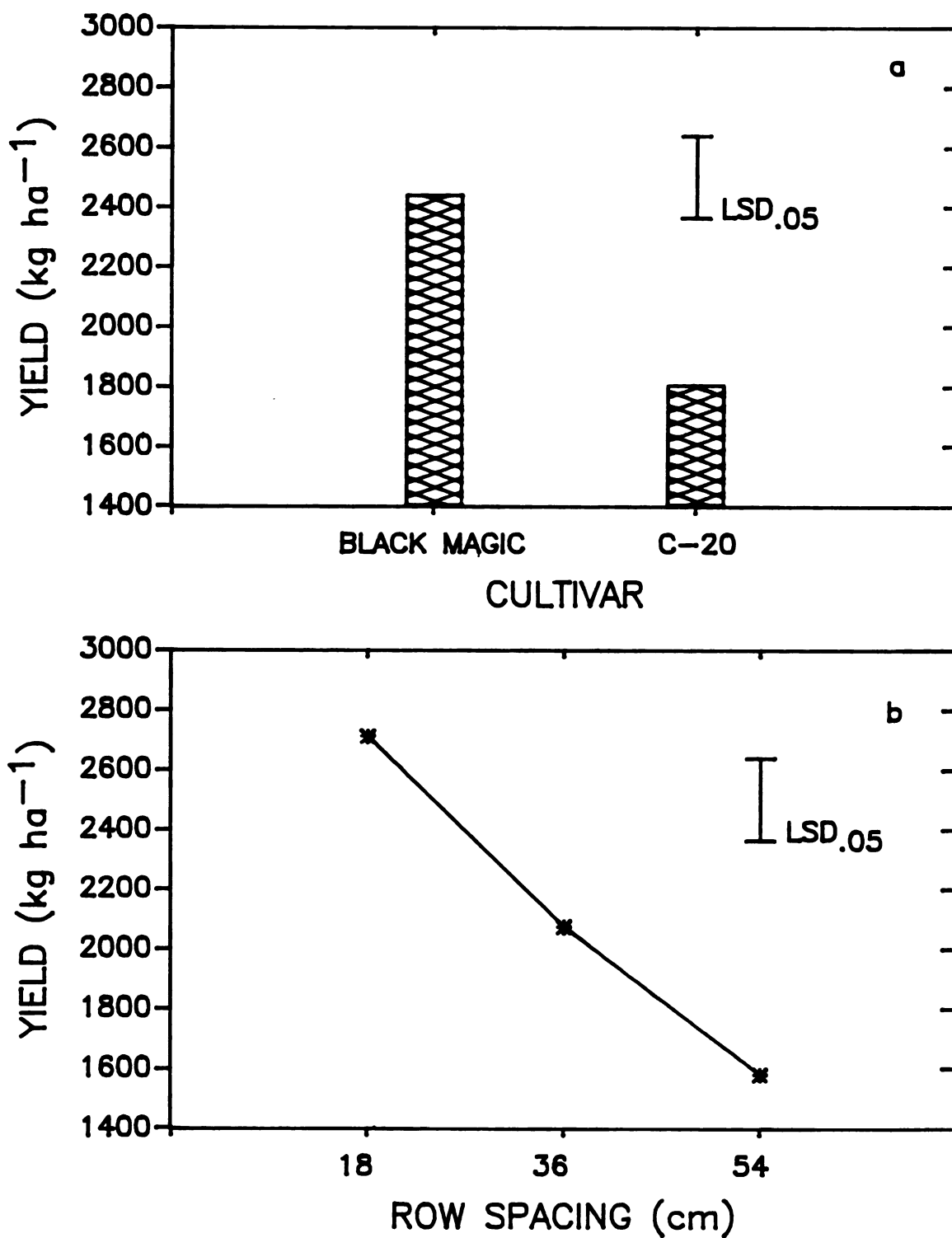


Figure 21. Dry bean yield on Charity clay in 1986 as influenced by cultivar (a) and row spacing (b). Yields were averaged across rotation/tillage and irrigation.

flowering (Taylor, 1982; Bennie et al., 1982).

While the differences are not significant at the 95% level, it is interesting to note the direction and magnitude of the differences due to irrigation and rotation/tillage. The irrigated plot average was 1822 kg ha^{-1} and the nonirrigated 2420 kg ha^{-1} . This 33% difference is significant at the 90% level. The direction of this difference is contrary to that which was expected. There are several possible explanations. Throughout the growing season there were few differences noted in root or shoot parameters due to irrigation. Only late in the season did shoot growth under irrigation surpass that of the nonirrigated plants. In a season with near adequate rainfall the effects of irrigation are few and could even be negative. If there was near adequate soil moisture throughout the growing season then irrigation could actually have a detrimental effect if applied to already moist soil. The additional irrigation moisture could result in short-term anaerobiosis in the rhizosphere. This explanation seems unlikely given the timing and limited amount of water applied but no investigations were made to determine rhizosphere anaerobiosis. The soil moisture data (Figures 7, 8, and 9) suggests that any soil anaerobic conditions should have been evident in the CONV plots rather than ARNST plots since soil moisture was higher on irrigated and nonirrigated CONV plots than on irrigated or nonirrigated ARNST plots.

Perhaps a more plausible explanation is that the beans were at different stages of maturity at the time of the

flood. There was considerable seed damage due to the flood, and beans at different maturity levels may have responded differently to the flooded condition. The nonirrigated plants were more mature than those irrigated. If the more mature seeds withstood a prolonged flood this might be reflected in a set of results favoring the mature seeds.

The average yield on the ARNST plots was 1982 kg ha^{-1} and on the CONV plots 2260 kg ha^{-1} . As indicated above this 14% difference is not significant. Again, however, the working hypothesis for this study suggests that yields should be increased when soil physical conditions are improved. That was not the case this year, as soil physical conditions were improved but yields were not.

Yield components are presented in Table 5. Pods per plant increased as row spacing increased with the exception of the ARNST irrigated C-20s which decreased from 18.2 to 13.5 when row spacing increased from 18 to 36 cm. This difference is not significant. There was no clear effect due to irrigation or rotation/tillage, although there was a significant interaction between these two treatment factors. Pods per plant was higher on the irrigated ARNST plots than the nonirrigated ARNST plots but the opposite occurred on the CONV plots. The C-20s had significantly more pods per plant than did the Black Magics.

Seeds per plant also increased with increasing row spacing. On the CONV soils the irrigated plants had more seeds than did the nonirrigated. However, on the ARNST soils no

Table 5. Influence of rotation/tillage, irrigation, cultivar, and row spacing on dry bean yield components. Data are the average of two replications.

<u>Rotation/Tillage</u> -----		ARNST		CONV	
<u>Irrigation</u> -----		IR	NI	IR	NI
<u>Cultivar</u>	<u>Row Spacing</u>	----- pods/plant -----			
	18	18.2	17.2	15.0	14.1
C-20	36	13.5	20.9	23.7	15.0
	54	25.0	20.1	26.1	22.2
	18	8.3	9.2	9.3	9.2
BlMag	36	13.2	15.8	16.9	15.3
	54	18.5	23.1	16.5	21.9

 LSD.05: - for Rotation/Tillage means at same or different
 - Irrigation, Cultivar, and Row Spacing = 34.2.
 - for Irrigation means at same or different Cultivar
 and Row Spacing = 3.9.
 - for Cultivar or Row Spacing means in the same
 column = 6.0.

		----- seeds/plant -----			
	18	76.7	65.5	60.8	49.7
C-20	36	61.3	80.6	100.6	54.5
	54	116.4	84.6	106.1	87.9
	18	47.8	49.5	52.0	47.5
BlMag	36	68.9	75.0	93.8	66.2
	54	106.5	113.5	93.2	111.5

 LSD.05: - for Rotation/Tillage means at same or different
 Irrigation, Cultivar, and Row Spacing = 164.
 - for Irrigation means at same or different Cultivar
 and Row Spacing = 21.7.
 - for Cultivar or Row Spacing means in the same
 column = 28.3.

trend was obvious as some row spacing by cultivar combinations had more seeds per plant under irrigation while others had more seeds per plant without irrigation. There was a significant difference due to cultivar with C-20 having more seeds per plant than did Black Magic.

An attempt was made to determine seed weight but the seeds had been too badly damaged by the flood and useful data could not be obtained.

Table 6 presents a comparison of meters of root required to produce a gram of seed under the various treatment combinations in the study. According to this data, ARNST system plants required more root length than the CONV plants to produce a gram of seed. The only exception among the treatments examined was the nonirrigated Black Magic combination, in which the ARNST plants produced a gram of seed with 6% less root length.

The largest differences were between the irrigated and nonirrigated treatments. The irrigated treatments required 46% more root length than the nonirrigated (averaged across the other treatments) to produce a gram of seed on both rotation/tillage treatments. It was expected that irrigated plants would be more efficient in seed production per unit length of root due to less stress on the plant system. However, the root length per gram of seed data is based on seed yield reported in Table 4, which showed that the nonirrigated dry beans yielded more than the irrigated plants. It was suggested that the difference in maturity between the

Table 6. Root efficiency in seed production as influenced by rotation/tillage, irrigation, and cultivar. Root length was determined by destructive sampling and is based on the assumption that the extracted profile contained one half of the roots from a single plant.

<u>Rotation/Tillage</u> ---	ARNST		CONV	
<u>Irrigation</u> --IR	NI		IR	NI
<u>Cultivar</u>		meters	root/gram seed	
C-20	49.77	28.55	39.75	24.76
BlMag	38.07	27.98	35.18	29.88

irrigated and nonirrigated plants at the time of the flood was important in the yield data, and that same factor was important in the root efficiency comparisons shown in Table 6.

There was an interaction between the cultivar and irrigation on root length per unit yield. The C-20 cultivar required about the same or less root length per weight of seed produced on the nonirrigated plots than the Black Magic cultivar. However, under irrigated conditions the Black Magic variety appeared to be more efficient in producing seed as evidenced by the lower root length per seed produced than the C-20 cultivar.

SUMMARY AND CONCLUSIONS

The combination of a deep rooted legume and careful tillage management was expected to result in an increase in dry bean yield. Soil compaction is a known problem on the fine

textured Charity clay soil and root growth may be mechanically impeded or slowed due to a lack of aeration, moisture, or other related problems. The soil physical measurements made confirmed the presence of a compacted layer at about the 20 cm, or plowpan, depth. There were also indications of a lesser secondary tillage pan between 6 and 9 cm depth. The study covered only one year, a year that contained some unique problems. In particular the September flood which eliminated the possibility of a harvest made it a difficult year in which to obtain useful data. The variable initial stand of beans and subsequent overplanting resulted in two different sizes of plants throughout the growing season.

The differences in the soil physical properties did not result in many significant differences in the plant parameters. Shoot biomass was largely unaffected by rotation/tillage treatment, as was root length density. No differences were detected in bean yield due to rotation/tillage.

Soil parameters were measured only to a depth of 22.5 cm but root responses were measured to twice that depth. Future studies should include measurement of soil physical properties to a depth of at least 45 cm in order better quantify changes due to deep tillage and the deep rooted legume crop and to interpret root data.

Differences due to row spacing were noted for shoot biomass as well as yield. This one year of data suggests that decreased row spacing will result in additional yield. Data from previous years at the same location support this finding

(Smucker, annual reports from the Saginaw Valley Bean and Beet Farm). In future studies it would be good to decrease intrarow seed spacing on the narrow rows in order to achieve the same population per unit area and thus study response to row spacing without the different populations.

The responses of the two cultivars were consistent. The Black Magic cultivar is known to have a more hardy root system and to be a high yielding variety. In this study the Black Magic variety did have higher root length densities and ultimately a higher yield regardless of the other treatment factors. Since the white navy bean is the bean of preference for most growers in Michigan, further investigation should be conducted to examine the reasons for the superior response of the Black Magic cultivar.

Additional studies should be conducted to investigate the effect of row spacing on emergence to verify if the differences noted in this study were due to planting equipment or to some other factor. The effects of rotation/tillage and its interaction with row spacing and irrigation could also be studied further to answer questions related to physiological maturation.

Root studies on different row spacings would be useful to examine the relationship of root growth and distribution and overall plant response to different row spacings.

One year of field data is insufficient to draw conclusions which can be assumed to be widely applicable. That is true especially of a study in a year of a highly unusual

flood as occurred in 1986. This study, with appropriate modifications, could be profitably repeated in order to verify the results and develop recommendations useful to dry bean producers.

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CHAPTER 3

EVALUATION OF MINIRHIZOTRON OBSERVATION TUBES AS A TOOL FOR ROOT STUDY ON FINE TEXTURED SOILS

INTRODUCTION

Studies of plant root systems are critical to an increased understanding of the plant system and its interaction with the environment. The current status of rhizosphere knowledge lags behind that of the above ground parts of the plant. Because roots grow below the surface of the soil they cannot be studied directly without destroying them and changing the soil matrix. The conventional method of root studies has been destructive sampling. With this method both soil and roots are extracted and separated and only then can the roots be examined. Destructive root studies are important but have the limitation of not allowing study over time. Thus the effects of a given treatment on the root system may be studied at a given point in time but it is impossible to study effects over time on the same root system. In addition, destructive sampling is both time consuming and costly.

Attempts have been made for many years to study root growth *in situ*. Methods have included glass walled rhizotrons of various sizes, glass or clear plastic panels pressed against the sides of trenches dug into the soil, and

more permanent root laboratories. More recently, glass or plastic tubes inserted into the soil have been used for root observation. These tubes and accompanying equipment have become known as minirhizotrons. According to an historical overview by McMichael and Taylor (1987), G. H. Bates in 1937 was the first to use such a tube. Since 1971 the technique has gained widespread acceptance for root studies. The development of improved mirrors, fiber optics, mini or micro video cameras and portable recording equipment has made it possible to more effectively use minirhizotron tubes as in situ root study tools. For a more complete review, see McMichael and Taylor (1987).

Minirhizotron tubes of various sizes are used, ranging from 6 to 150 mm in diameter depending on the boring equipment available (McMichael and Taylor, 1987). The most commonly used size is around 50 mm (Brown and Upchurch, 1987). Most of the tubes used have been round (Upchurch and Ritchie, 1983; Maertens, 1987; Merrill et al., 1987; Levan et al., 1987) but square tubes have also been used (Waddington, 1971). The tubes are installed at various angles, usually from 30 to 45 degrees from vertical (Brown and Upchurch, 1987). Orientation with respect to plants also varies depending on the objectives of the study, personal preference of the researcher, or physical constraints related to installation and root observation. Upchurch and Ritchie (1983) studied four different tube orientations including parallel or perpendicular to the row and within the row or between two

rows. They found few significant differences in root observations due to tube placement. Many different observation and recording devices have been employed, including a series of mirrors, various scopes with appropriate light source, images transferred to and recorded by 35 mm cameras, and video cameras designed specifically for minirhizotron observation and recording. Brown and Upchurch (1987) have reviewed this equipment in some detail.

Minirhizotrons have been used to study roots of various plants, including trees, grasses, grains, and food crops (Brown and Upchurch, 1987). Numerous aspects of rhizosphere dynamics are being studied. Smucker et al. (1987) suggested there may be as many as 35 categories of information available about roots and the soil system from minirhizotron observations. Data collected includes root color, branching, length, depth, density, diameter, and lateral root spread (McMichael and Taylor, 1987). Minirhizotrons are also used to collect data on soil macro and mesofaunal populations, root turnover rates, nodules on leguminous crops, pathogens, and root diseases (Smucker et al., 1987).

Different methods are used for tube installation, including hydraulically driven bores (Upchurch and Ritchie, 1983) or hand operated augers (Merrill et al., 1987). Most researchers agree that proper installation of the tubes is critical if useful results are to be obtained (Upchurch and Ritchie, 1983; Brown and Upchurch, 1987; Smucker et al., 1987; and Maertens, 1987). Data must be representative of root

activity in the surrounding bulk soil. Problems related to installation may affect root growth and thus prejudice data collected. Installation of a tube obviously disturbs the soil and the best that can be hoped for is that disturbance is minimal and will not appreciably affect the growth of roots. If the soil around the tube is compacted, root growth at the tube-soil interface will be restricted and thus not representative of that in the surrounding bulk soil. On the other hand the hole into which the tube is inserted must not be so large as to create unnatural voids at the tube surface as this may favor root proliferation and yield an inaccurate picture of root activity. The ideal installation will result in good contact between the soil and tube without large voids or compacted soil. Factors affecting installation include boring method, cleaning of the hole, and soil conditions at the time of installation. For a review of installation techniques see Brown and Upchurch (1987).

Minirhizotrons have been used on many different soils. Some problems have been encountered on certain types of soil. Such problems may be related to installation techniques or to the physical properties of the soil. Good results have generally been obtained with sandy soils. However, Vos and Groenwold (1987) encountered a problem on a sandy soil with a compacted layer at the 30 cm depth. In their study the installation of minirhizotron tubes resulted in breakup of the compacted layer, creation of larger voids at the tube-soil interface than in bulk soil, and root proliferation at

the tube which was much greater than in the surrounding bulk soil. Fine textured soils have presented more problems. Maertens (1987) concluded that minirhizotrons may be useless on clay soils if not installed very carefully. This is due to the potential for smearing, soil compaction at the tube-soil interface, or creation of unnatural voids which will favor root proliferation. The capacity of many clay soils to shrink and swell make it difficult to maintain a good tube-soil interface. Maertens suggests that these problems can be overcome by boring a hole larger than necessary, loosening the soil wall with a rotating brush, and introducing a tube with a flexible outer membrane. This outer membrane is then inflated and takes the shape of the soil wall. Merrill et al. (1987) has utilized a similar system to counter the unnatural effects of boring a hole for tube insertion.

One of the problems encountered in using minirhizotrons is the poor correlation of data from the top 20-30 cm of the tubes with that of destructive sampling. Upchurch and Ritchie (1983) found that root length densities from the top 20 cm were severely underestimated by the minirhizotron method compared to the results from destructive sampling. Vos and Groenwold (1987), Beyrouthy et al. (1987), and Smucker et al. (1987) noted the same phenomenon. Reasons suggested for this discrepancy include the movement of dry soil away from the tube for an undetermined reason (Upchurch and Ritchie, 1983), the influence of temperature differences at the tube-soil interface (Upchurch and Ritchie, 1983; McMichael and Taylor,

1987) and the introduction of light due to soil disturbance during tube installation or improper sealing of the tube to exclude light (Levan et al., 1987).

Many researchers have found that the number of replications needed to achieve satisfactory results is approximately twice that of destructive sampling. Upchurch and Ritchie (1983) found very little correlation between the results of single tube observations and bulk soil root length densities. They suggest that several tubes must be averaged before meaningful results can be expected. Vos and Groenwold (1987) in a series of experiments used an average of approximately twice as many minirhizotron tubes as destructive samples in order to obtain reliable results.

Data collected has been presented in various forms. Equivalent root length density (Merrill et al., 1987), number of roots observed per unit area (Smucker et al. 1987), root length density (Upchurch and Ritchie, 1983), and root length per area (Vos and Groenwold, 1987) have all been reported. Upchurch (1987) discusses the derivation of root length density from minirhizotron root observations and concludes that this is a valid conversion if certain assumptions and constraints are understood.

Minirhizotron data presents some interesting statistical problems. Upchurch and Ritchie (1983) suggest that since the numbers of root observations per unit area are often quite low and include many zeros the data does not lend itself to classical parametric statistical analysis. They suggest the

need for some transformation so that parametric statistical analysis can be used. Alternatively, nonparametric statistical tools could be used. Glenn et al. (1987) found that the variance of treatment means was positively correlated with the mean, a case which violates the assumptions of the analysis of variance. A transformation was used to reduce this correlation in order to subject the data to analysis of variance. There is need for additional work in the area of statistical analysis of minirhizotron data.

In summary, the minirhizotron is being effectively used for *in situ* root study. Numerous pieces of information can be gathered with this method. The system may be initially expensive but may prove to be much less costly in the long run than conventional destructive sampling. One major advantage is the ability to study root activity over time. However, there are potentially serious problems which may prejudice results. These include the tube-soil interface effect on root growth, changes in temperature or moisture conditions due to the presence of the tube, and a problem of currently undetermined origin with rooting density in the top 30 cm of soil around minirhizotrons. In addition, there are statistical problems associated with handling minirhizotron data which must be addressed.

The current study was undertaken as part of a management study for dry bean (*Phaseolus vulgaris*) production. Root responses of two cultivars to different rotation/tillage management systems and to irrigation were studied both by

destructive sampling and minirhizotron video recordings. By using the two methods it was possible to consider the relative merits of each. The destructive sampling was considered to be the standard against which the minirhizotron method was compared. The objective of this two year study was to evaluate the minirhizotron system for use on fine textured soils and under different soil management practices.

MATERIALS AND METHODS

A two year study of the minirhizotron microvideo method of root study was conducted as part of a management study for dry bean production. The study was conducted at the Saginaw Valley Bean and Beet Research Farm near Saginaw, Michigan in 1985 and 1986. The soil on the farm is a fine textured Charity clay (illitic, calcareous, mesic, Aeric Haplaquept) which is artificially drained. The soil has about 60% clay and is subject to severe compaction by agricultural traffic. Primary and secondary tillage pans are common and can be restrictive to root growth as well as to air and water movement. The management study included two rotation/tillage variables, irrigation vs nonirrigation, and two dry bean cultivars. The rotation/tillage variables were: 1) conventional (CONV), which included a crop rotation of corn-dry beans, fall moldboard plowing, and two to four passes of spring secondary tillage; and 2) alfalfa rotation, no secondary tillage (ARNST) which utilized a four year rotation of corn-alfalfa-alfalfa-dry beans, deep tillage in the fall of the second year of alfalfa followed by moldboard plowing, and no spring secondary tillage. Half of the plots were sprinkle irrigated as needed and the other plots received no irrigation. The two cultivars were C-20, a white navy bean, and Black Magic,

a black soup bean. Both cultivars were studied in 1985 but in 1986 minirhizotron tubes were installed only on the C-20 plots. Data is reported for the C-20 variety. Row spacing was 50 cm in 1985 and 54 cm in 1986.

Minirhizotron tube installation was completed soon after planting and prior to or immediately after crop emergence each year. Clear butyrate plastic tubes 1.83 m long and 51 mm inside diameter with a 3.2 mm wall were installed at a 45 degree angle directly under and parallel to the bean rows. Tube holes were bored using a modified trailer mounted hydraulic soil sampling probe (Giddings Model GSRP-ST) equipped with a cutting bit designed to compact inward rather than outward. After each hole was bored it was cleaned out using a round wire brush. In 1985 the brush was pushed and pulled up and down the hole without rotating it. In 1986 the brush was rotated as it was moved up and down the hole. The change was implemented in 1986 in an effort to decrease the possibility of deep striations or channels resulting from the boring and cleaning operation. Tubes were pushed into the holes by hand or with light tapping. Every attempt was made to insure that the tubes fit snugly. Observations throughout both seasons confirmed that tubes had been inserted without smearing. Tubes were tightly capped at both top and bottom with number 11 rubber stoppers. After insertion the protruding portion of tube (approximately 30 cm) was painted with black paint to exclude light and later with white paint to reduce solar heating of the tube.

Two tubes were installed in each of four replications for a total of eight minirhizotron tubes for each treatment combination.

A microvideo camera (Circon color bore inspection system, Model MV-9011 Agricultural Camera) was inserted into the tube and video images of the roots recorded using a modified hand held Hitachi monitor-viewfinder, portable video cassette recorder (Panasonic Model NV-8420) and portable computer for recording date, depth, and tube identification. Video recording was begun in mid July and was done at approximately one week intervals throughout the summer. Video recording was carried out to the depth of the deepest visible root in each minirhizotron. Video taping was done incrementally with each frame representing an area 1.2 by 1.8 cm, or 2.16 cm². The root images were later counted manually using a 13 inch color monitor and recorded as number of root observations per video frame.

Destructive root sampling was carried out in mid-August of each year (at or near maximum flowering) using the method described by Srivastava et al. (1982). This method involves removal of a soil profile 7.5 cm x 22.5 cm x 45 cm by means of a hammer driven profile sampler mounted on a tractor. Each profile was partitioned into 18 cubes (3 x 6 array), each of which was 7.5 cm on a side. Profiles were removed at the time of maximum flowering which was in mid-August. Each profile was taken from the center of the plant to 22.5 cm away from the plant and perpendicular to the row. It was

assumed that the profile represented approximately one half of the root system of one plant. The soil cubes were soaked for 8 to 16 hours in a solution of 5% sodium hexametaphosphate to aid in dispersing the clay and washing out the roots. The soil was then washed from the extracted 7.5 cm cubes with a hydropneumatic elutriator (Smucker et al., 1982). Roots were stored in a solution of 20% methyl alcohol until laboratory analysis. Root length was determined by Tennant's line-intersect method (Tennant, 1975), which is a modification of Newman's method (Newman, 1966). A four centimeter square grid was used.

RESULTS AND DISCUSSION

Minirhizotron root observations for 1985 and 1986 are presented in Figures 1-4. Root observations were compared on the basis of treatment combination (rotation/tillage by irrigation) and these data are presented in Figures 1 and 3. Root growth was also studied over time and the data from four dates is shown in Figures 2 and 4. Root observations were averaged across the eight replications and for every 10 cm of soil depth to smooth the large variability among individual video frame observations. Upchurch and Ritchie (1983) showed that root observations from individual tubes have very little correlation with bulk soil rooting patterns.

The destructive sampling data from both 1985 and 1986 is presented in Figures 5-8. Each square in the "rootmaps" represents the root length density (RLD) in a cube of soil 7.5 cm on a side. The relative position of the square in the rootmap represents the location of the soil cube in relation to the plant.

Few roots were observed in the minirhizotron tubes in the top 20 cm of soil under any of the treatments. Most roots observed in the tubes were in the 20-60 cm depth range. The rootmaps in Figures 5-8 show that the highest root length density was in the top 7.5 cm of soil, and RLD generally

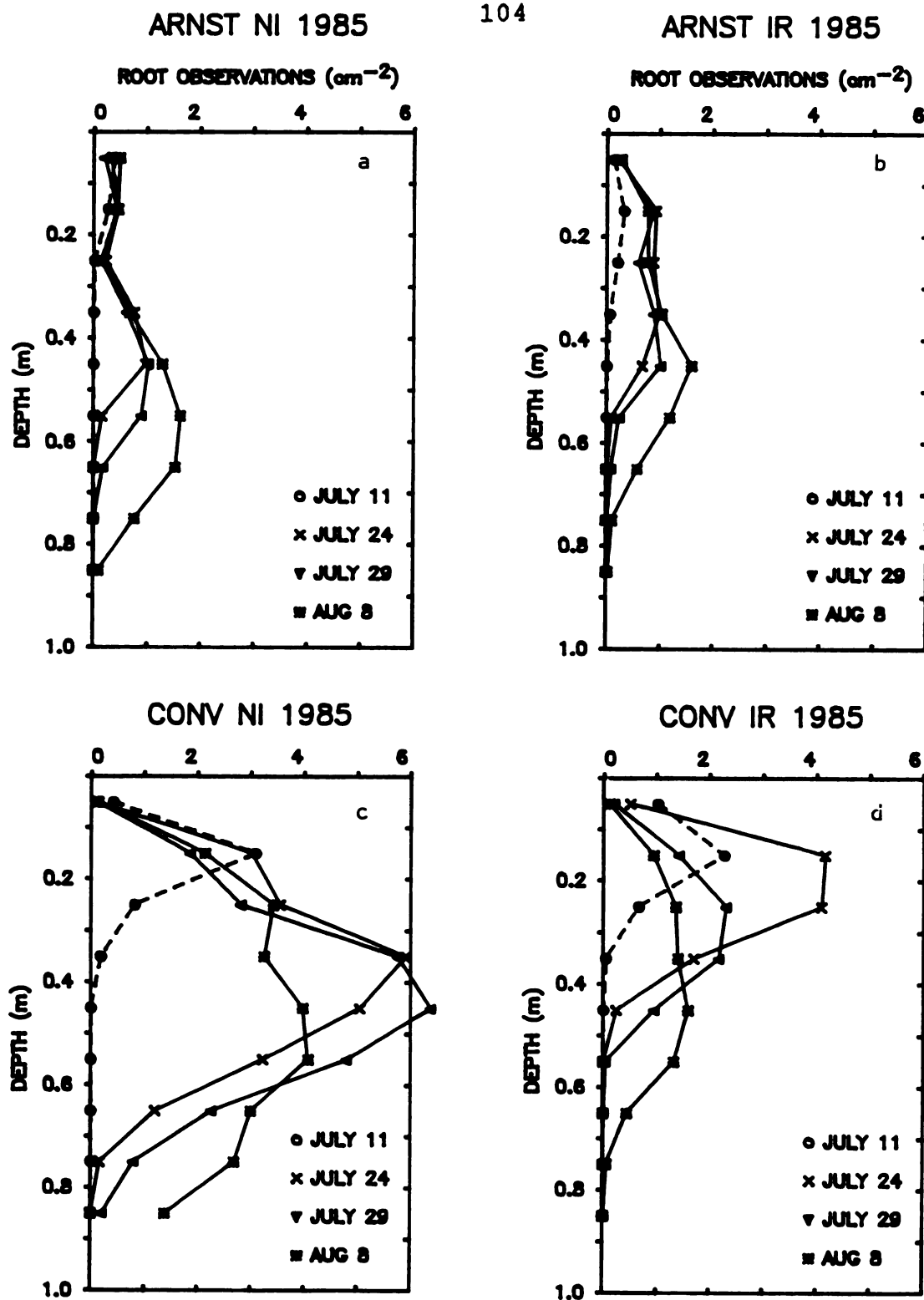


Figure 1. Minirhizotron root observations for four rotation/tillage (ARNST or CONV) and irrigation (IR or NI) treatment combinations in 1985 as affected by date of observation. Data points are the average number of roots observed cm^{-2} by ten cm depth increments.

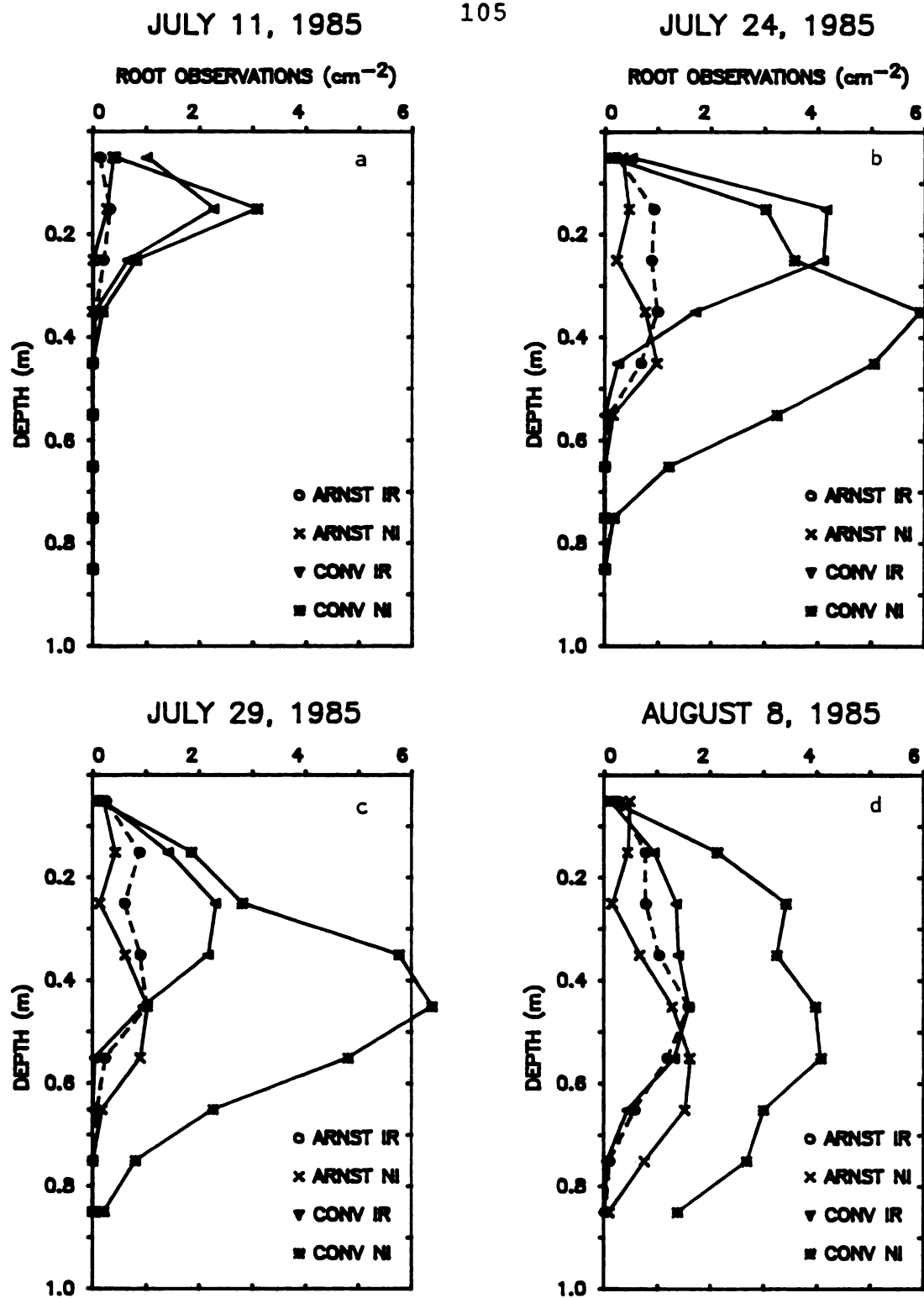


Figure 2. Minirhizotron root observations on four dates in 1985 as affected by rotation/tillage (ARNST or CONV) and irrigation (IR or NI) treatment combinations. Root observations are reported as the average number of roots observed cm^{-2} in ten cm depth increments.

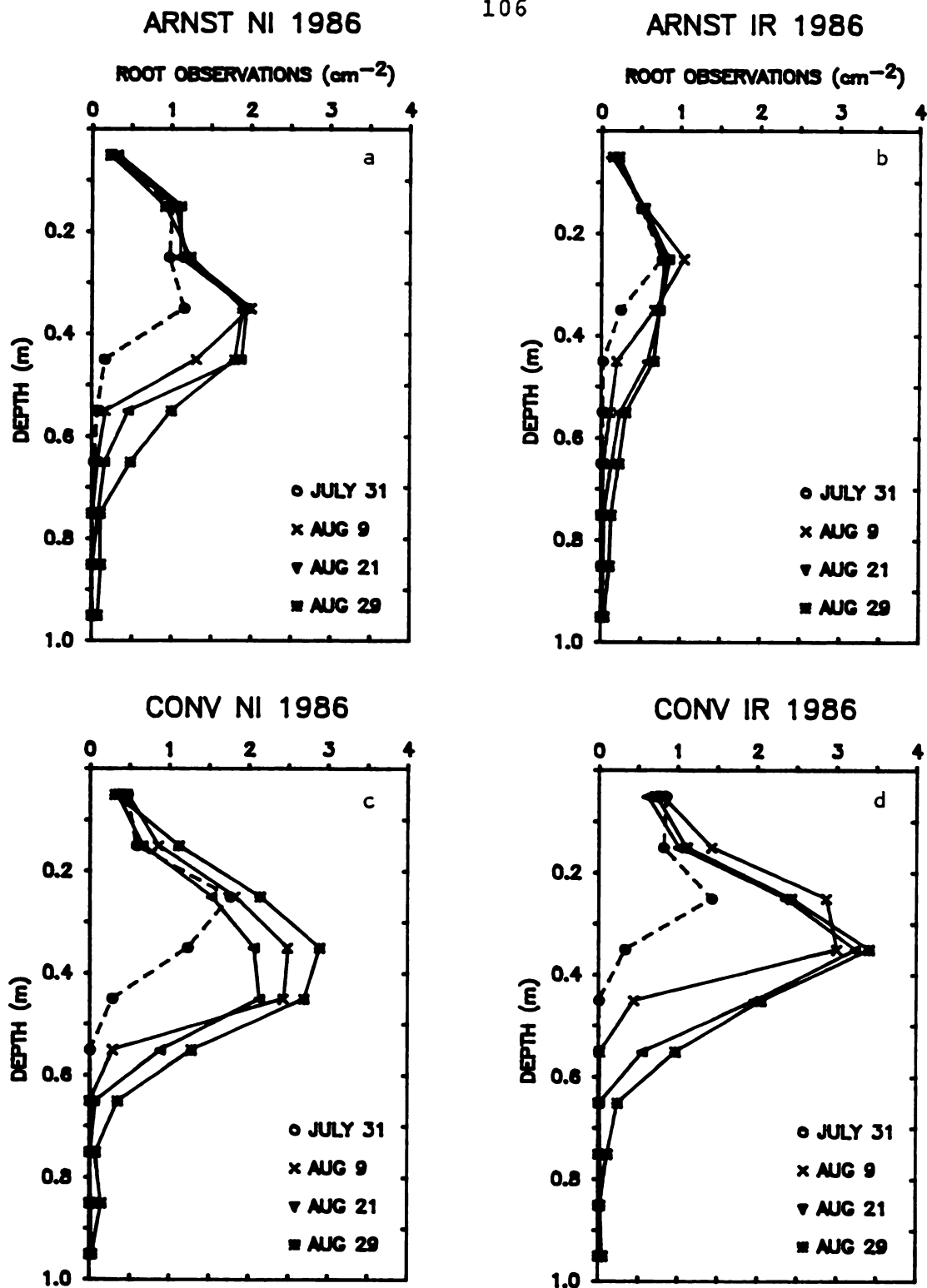


Figure 3. Minirhizotron root observations for four rotation/tillage (ARNST or CONV) and irrigation (IR or NI) treatment combinations in 1986 as affected by date of observation. Data points are the average number of roots observed cm⁻² by ten cm depth increments.

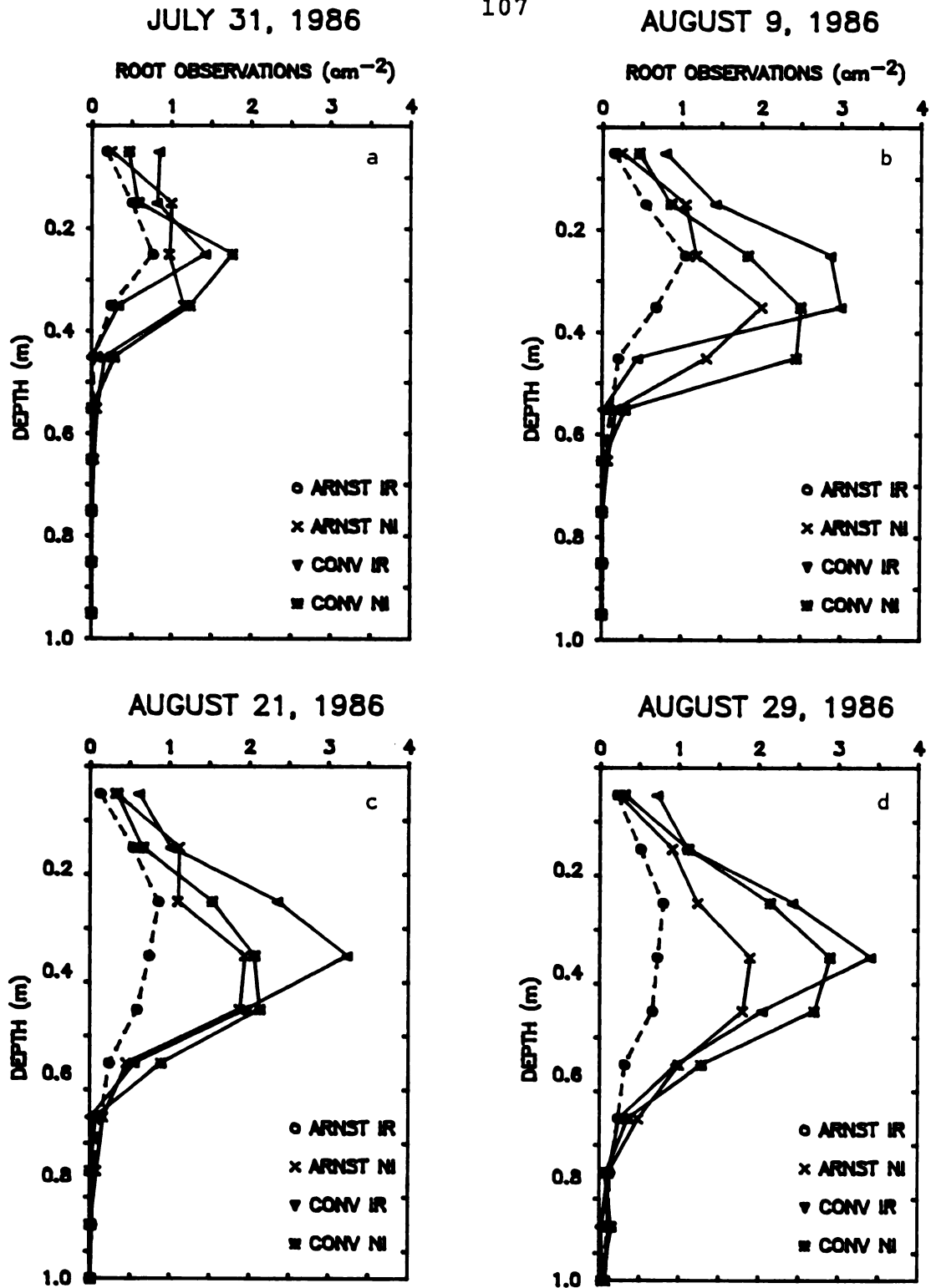


Figure 4. Minirhizotron root observations on four dates in 1986 as affected by rotation/tillage (ARNST or CONV) and irrigation (IR or NI) treatment combinations. Root observations are reported as the average number of roots observed cm^{-2} in ten cm depth increments.

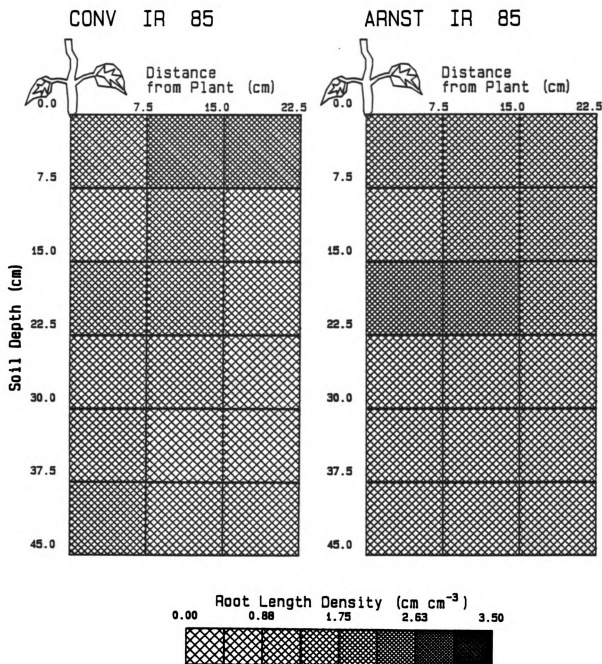


Figure 5. Root length density as determined by destructive sampling in mid-August, 1985 and as influenced by rotation/tillage under irrigated conditions.

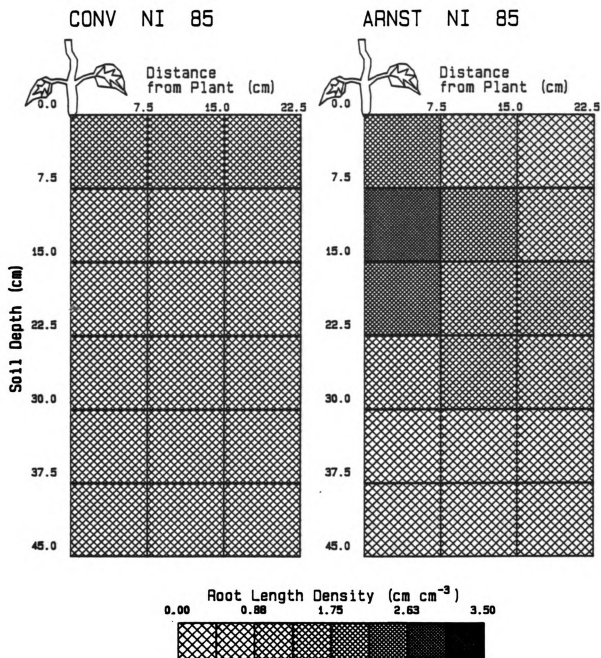


Figure 6. Root length density as determined by destructive sampling in mid-August of 1985. The effects of rotation/tillage under nonirrigated conditions are shown.

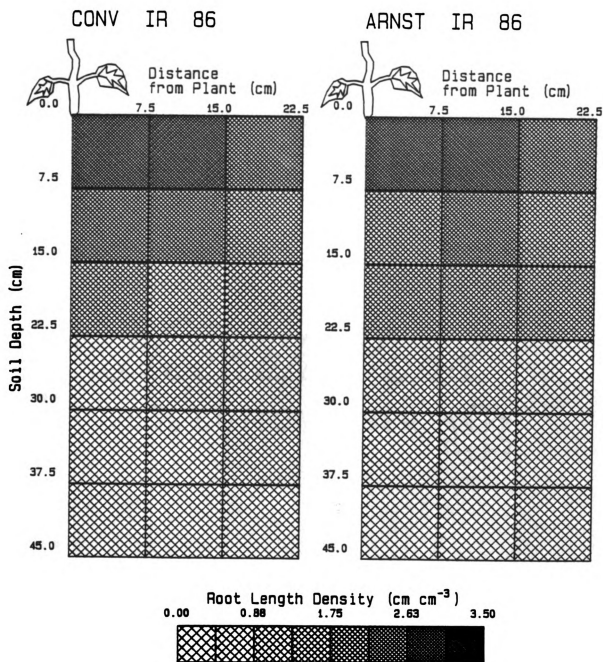


Figure 7. Root length density as determined by destructive sampling in mid-August, 1986 and as influenced by rotation/tillage under irrigated conditions.

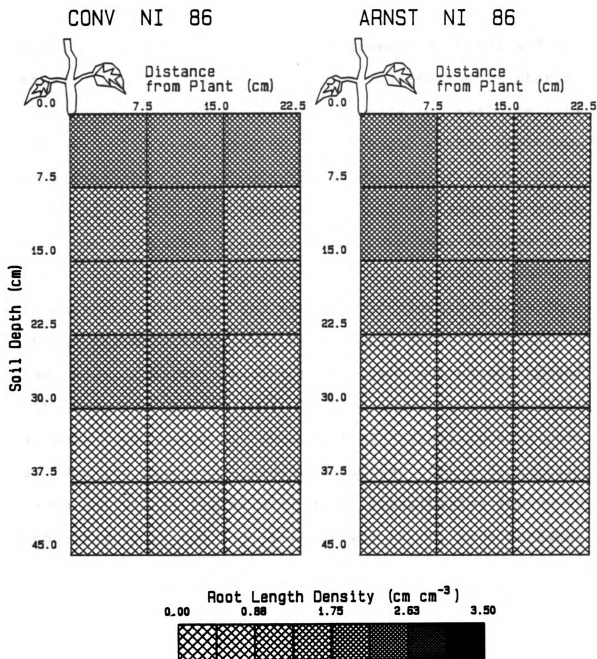


Figure 8. Root length density as determined by destructive sampling in mid-August of 1986. The effects of rotation/tillage under nonirrigated conditions are shown.

decreased with soil depth. This discrepancy between the root observations in the minirhizotron tubes and rooting patterns in the bulk soil is consistent with reports of other studies (Upchurch and Ritchie, 1983; Vos and Groenwold, 1987; and Levan et al., 1987). The exact cause or causes of this phenomenon are not known. Every effort was made to seal the tube from light. It seems unlikely that light within the tubes caused a decrease in root growth as can occur according to Levan et al. (1987). A temperature effect cannot be ruled out but the protruding part of each tube was painted with white paint to minimize excessive heating. Another possibility is that the soil near the surface was disturbed more during tube installation than the deeper parts of the soil. This may have occurred when the wire brush was moved up and down the hole to clean it prior to tube insertion. The additional soil disturbance may have altered normal rooting patterns. Vos and Groenwold (1987) speculated that a disruption to the normal water regime may be a contributing factor to the observed decrease in root growth at the tube surface. The soil disturbance from tube insertion procedures would have altered the normal water regime.

The observed effects of rotation/tillage on root growth patterns are quite different between the minirhizotron tubes and the destructive sampling method. As discussed above, that difference is not unexpected in the top 20 cm of the soil profile. However, the minirhizotron tubes showed many more roots cm^{-2} in the lower portions of the profile (20-60

cm depth) under the CONV management system than under the ARNST system. This difference was evident both years, under both irrigated and nonirrigated conditions (Figures 1 and 3,) and throughout the growing season (Figures 2 and 4). The differences were more pronounced when the crop was not irrigated in 1985 (compare, for example, Figures 1a and 1c with 1b and 1d), but the opposite was observed in 1986 (Figures 3a and 3c vs 3b and 3d). Despite this observed difference due to irrigation, the differences due to rotation/tillage are striking.

The direction of the differences described above was unanticipated. The CONV rotation/tillage treatment had evidence of a primary, and possibly a secondary, tillage pan. (See Chapter 2, soil physical measurements.) The ARNST treatment included deep tillage and two years of a deep-rooted legume prior to the dry bean crop. These plots had lower bulk density and more large pores than the CONV soils, which is an indication that the previously existing tillage pans had been broken up. It was expected that this would result in deeper root growth on the ARNST soils than on the CONV soils. However, the minirhizotron root observations from both years showed much more prolific root growth in the deeper horizons of the profile on the CONV soils.

The destructive sampling data did not show many differences between root length density due to rotation/tillage. When analyzed by depth there were almost no differences which were statistically different.

The large increase in minirhizotron root observations on the CONV vs the ARNST soils is not substantiated by the destructive sampling results. This finding raises serious questions about the validity of the minirhizotron method on fine textured soils such as the Charity clay.

The proliferation of roots under the CONV system as seen in the minirhizotron tubes must be attributed to an effect of the tubes. Upchurch and Ritchie (1983), McMichael and Taylor (1987), and Vos and Groenwold (1987) have pointed out some of the possible effects of the tube on root growth. The most likely explanation of the effect of the tube on root growth in the current study is that roots on the CONV soils are restricted in downward growth by the compacted soil layers except at the tube. The number of roots which intersect the tube is increased as some of the restricted roots grow laterally. The roots at the tube-soil interface then proliferate due to less physical restriction since the compacted layers were broken up during tube insertion. There are sufficient voids at the tube surface to allow roots to proliferate. Additionally, there may be an effect due to improved water and aeration conditions along the tube.

The individual video frames of the roots support the suggestion that the tube effect is significant in allowing for greater root proliferation. Figure 9 shows minirhizotron observation images representative of each soil management system. The ARNST images show few bundled roots but there are many such images from the CONV tubes. These bundles are



Figure 9. Minirhizotron observation images representative of those seen under the two crop/soil management systems: upper: CONV management, compacted layer at 20 cm depth; and lower: ARNST, compacted layer broken up by alfalfa roots and deep tillage.

masses of intertwined roots which follow the tube for some length, often 8-10 frames (10-12 cm along the tube) or more. Normal root growth would not include this bundling effect, nor would roots normally grow in a straight path for this distance.

The minirhizotron root observations do present a good picture of downward growth over time (Figures 2 and 4). This downward growth was observed on both rotation/tillage treatments. Maertens (1987) states that one of the useful applications of the minirhizotron method is to study rooting depth. The experience of the current study suggests that this may be possible even on fine textured soils. However, the possibility of a tube effect on rooting depth must be examined in order to develop confidence in the minirhizotron method. Since the destructive sampling was done only once, it was not possible to verify the minirhizotron results to insure that there were no tube effects on rooting depth.

SUMMARY

The minirhizotron root observation system has been found to be useful and accurate in many situations. However, this study showed that the system does not work well on fine textured soils with compacted layers. The data obtained was opposite of that expected and was negatively correlated with that from destructive sampling. The results from the

minirhizotron observations showed very few roots in the top 20 cm of soil and a large proliferation of roots under the CONV system vs the ARNST system in the 20-60 cm range. The data suggest that minirhizotrons may be useful for studying rooting depth on fine textured soils.

If minirhizotrons are to be useful on this soil, methods will need to be developed to insure that root observations in the tubes are representative of the roots in the bulk soil. This may include changes in installation techniques to obtain better tube-soil contact. Another option might be the use of an inflatable sleeve over the tube which would conform to the shape of the surrounding soil. Such a system, even if it could be perfected for use on fine textured soils, may prove to be too unwieldy for field studies with many tubes.

Despite the high cost of destructive sampling, it is still a superior method to the minirhizotron for general root studies on fine textured soils.

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CHAPTER 4

ROOT LENGTH AND WIDTH DETERMINATION BY DIGITAL IMAGE PROCESSING

INTRODUCTION

Root studies have been hampered by the physical difficulties of examining the root system. The processes of root excavation, separation from soil, and analysis of the root system are very laborious. Weaver (1926) and Dittmer (1937) reported detailed studies of root systems. These studies required massive amounts of time and effort. Few researchers since then have been willing to expend a similar amount of energy to study a single root system. The known variability of root systems (Russell, 1977) also frustrates efforts to examine the roots of only a few plants and extrapolate to general root understandings. Different rhizosphere environments also result in different root responses.

Root dry weights have been widely reported due to the relative ease of collecting dry weight data. Dry weights have the limitation of not being well correlated to root activity, especially nutrient and water uptake. Root surface area is a better parameter to measure to gain an understanding of total root activity. However, root surface area is almost impossible to measure directly because of the size,

shape, and number of roots of any single plant.

Root length has become the standard parameter for root studies in the past few decades, and has been widely accepted in lieu of surface area. Direct measures of root length are almost impossible to obtain. Fortunately, estimation procedures have been developed and shown to be quite accurate. Newman (1966) developed a method to estimate root length by counting the number of root intersections with randomly placed lines in a tray of well dispersed root sections. Marsh (1971) and Tennant (1975) modified Newman's method. Tennant's modified line intersect method has become the current standard. In this method, roots are evenly spread in a tray and a square grid is placed under the tray. The number of intersections of roots with the grid lines is then counted. The number of intersections is converted to root length by a simple mathematical formula. While it has allowed for significant advances in knowledge of root function and activity, this method does have several limitations. It is known to overestimate total root length by 5-15% depending on the grid size used. The line-intersect method is time consuming and tedious. It is also prone to investigator error due to fatigue as well as differences resulting from different persons carrying out the procedure.

New methods are needed to improve the accuracy of root length determination, the efficiency with which roots are studied, and to provide investigators with information in addition to root length. Recent developments in computers,

specifically image processing, have led to the possibility of greatly enhancing plant root studies through the use of this technology (Smucker et al., 1987).

Computer image analysis has several potential benefits. These include:

- a) decreased labor input required;
- b) increased accuracy of root data;
- c) determination of root diameter and surface area;
- d) determination of branching characteristics such as frequency and angle;
- e) increased number of samples examined, which would aid in determining statistically significant differences between or among treatments.

There are also numerous problems to be addressed in computer image analysis. These include:

- a) initial cost of image processing equipment;
- b) preparation and capture of the root image to be analyzed;
- c) calibration of the system;
- d) development of appropriate algorithms to gain the desired information from the image.

This study summarizes efforts to date in developing a system to analyze roots by using a digital image processor. The system included root sample extraction, washing the roots free of soil and other material, staining the roots, video taping root images by a computer-driven high resolution video

camera, and developing the algorithms to enable the digital image processor to analyze the root video images.

The objective of the system development effort was to incorporate current image processing knowledge and technology into a system to more quickly and accurately analyze washed root samples to obtain information on root length. An additional objective was the determination of root width which would allow for calculation of an estimate of total surface area. Total surface area would be calculated on the assumption that roots are round and that root width as determined by the image analysis computer from the two dimensional image is equal to root diameter. A third objective for this system was to obtain information on root branching frequency, branching angle, and other characterization of root branching.

MATERIALS AND METHODS

Root Extraction and Preparation

Dry beans (*Phaseolus vulgaris*) were grown in washed sand in the greenhouse or in Charity clay soil in the field. Roots from the greenhouse study were extracted and washed gently in a water bath by hand. Since these plants were grown in sand there was no organic or mineral debris remaining after the sand had been washed away. Roots were patted dry and frozen until analyzed. Field roots were extracted at the time of maximum flowering, assumed to represent maximum root growth. The root profile sampler method (Srivastava et al., 1982) was used to extract field roots. Root-soil cubes 7.5 cm on a side were extracted by this method. These cubes were soaked in a 5% solution of sodium hexametaphosphate for 8-16 hours and then washed using a hydropneumatic elutriation chamber (Smucker et al., 1982). The separated roots were stored at 4 degrees C in a 20% methyl alcohol solution until analyzed. These root samples contained varying amounts of organic and mineral debris which was not washed out in the elutriation chamber.

Roots were stained with a 5% malachite green solution prior to video taping or hand counting.

Root length was determined by Tennant's modified line

intersect method (Tennant, 1975). A four cm grid was used. This method was considered the standard against which the image processing would be compared.

Root Image Recording

Root images were recorded on video tape for future processing. The process and equipment for this are described below.

The roots from one cube of soil (field roots) were considered one sample. Greenhouse roots were divided into subsamples to achieve the desired amount of roots per tray for counting or video taping. After staining, one sample of roots was placed in a custom made glass tray 43 x 43 cm with 3 cm high glass sides. 750-800 ml of water was added to the tray and the roots were spread evenly throughout the tray. This was accomplished by teasing the roots apart using two small forceps. This procedure is the same for manually counting root-line intersections or video recording the roots for image processing. (For samples containing few roots, a smaller tray, 21.5 x 21.5 cm was used with about 200 ml water. Tray size is discussed more fully in "RESULTS AND DISCUSSION, Tray Size".)

An automated system was developed for video recording the roots. This system consisted of three parts: 1) system control, 2) x y motion (in a horizontal plane), and 3) video acquisition and storage. The system was controlled by an IBM-XT computer with external digital to analog and stepper

motor controllers. The x y motion mechanism included two stepper motors that provide power to move a digital video camera (Javelin, MOS) over the root tray. The video acquisition system utilized the video camera and a video recorder (Panasonic, Model AG-6300) to store the images. Images were stored on standard 1/2 inch VHS format video tape.

The procedure was as follows.

The tray of stained and spread roots were placed on a table backlit with the diffused light of six fluorescent bulbs and positioned to an exact location. The video camera was suspended above the table on an x y scanner. The scanner consisted of a frame of two aluminum tables which move in either the x or y direction along four 1/2 inch polished steel shafts fastened to a framework resting on the floor. The backlit table is also attached to this frame. The camera is moved along the steel shafts to exact locations by two stepper motors. The camera is moved to one corner of the tray of roots and an image is recorded. The camera is then moved to the adjacent image in the y direction and another image is recorded. The whole tray is ultimately recorded in 64 images (an 8 x 8 grid pattern). Each image represents a 5.4 cm x 5.4 cm square of the tray.

The system is fully automated. A computer program written in Microsoft Basica is utilized to operate the various pieces of equipment needed to produce the video images. A bar code is recorded for each tray of roots prior to any root images. This bar code is entered manually and is used to

identify each tray. The bar code is later read by the image processing computer and included with the output for a tray of roots. An audio tone, generated by the computer, is also recorded during each image, including the bar code image. This tone is later used to signal the image processing computer to capture (digitize) the image for processing.

Approximately 19 trays of roots, or 1216 images, can be recorded on one two-hour video tape. Six minutes and 4 seconds are required to actually video one tray. Total time to prepare (after the roots have been collected and stored) and video one tray is dependent on the time necessary to spread the roots, and was found to vary from 8-25 minutes.

Image Analysis

The video tapes of root images are played back and the images captured in succession and analyzed by a digital image processor (DIP). The hardware used includes the video cassette recorder, a time base corrector (Fortel, Model CCDHP), an analog to digital converter (Quasitronix, Model Q-3024), and a digital image processor (Vicom, Model 1800). The Vicom 1800 is a stand alone image processing computer with a Motorola 68010 central processing unit. Images are stored in a 512 x 512 pixel array with 16 bits per pixel. The DIP also includes an image digitizer, hard disc storage, computer terminal, and image display system with monitor.

The system is fully automated and is controlled by the DIP. Images from the VHS tapes are first passed through the

time base corrector to synchronize the timing with the image digitizer in the DIP. Time base correction is required because video tape is subject to stretching during playback. This stretching can result in inconsistencies in timing between frames, which, if not corrected, can lead to improperly digitized images. The image is then digitized and stored by the digital image processing computer for processing.

The software is divided into five sections. These are control, preprocessing and thresholding, skeleton width encoding, debris extraction, and measurement.

The control section of the program controls the video tape, digitizes each image, and deciphers the bar code which is used for identification. Preprocessing involves enhancing the image in order to accentuate the roots and eliminate as much noise as possible. Thresholds are then chosen to identify objects in the image. Thresholding results in a binary image ready for further processing.

Binary images are thinned to one pixel in the skeleton width encoding operation. The remaining centerline can be measured for length. The thinning process also allows for width determination by counting the pixels that are thinned away.

A debris extraction routine is available for root samples which contain any non-root objects. Most samples will fall into this category unless they have been grown in a medium with no organic matter or it is possible to remove any debris. Object shape is used to distinguish between roots

and debris. Any object with a length:width ratio less than approximately 3:1 is considered to be debris.

Following the above steps, length and width measurements can be made. Length per width class is recorded in pixels. There are five width classes, 1-2, 3-4, 5-6, 7-8, and 9-10 pixels.

RESULTS AND DISCUSSION

Imaging System

The automated video recording system worked well for imaging roots. Approximately 200,000 images have been recorded with no significant mechanical or operational problems. Time per tray varied considerably but the video recording system did not require as much labor as the line intersect counting method. The time required to spread the roots was the same for the two systems. With the automated video system, one tray could be prepared while another was being recorded, but with the line intersect method additional time was required to actually count the intersections. It is estimated that the video procedure requires 5-10 minutes per tray less than the line intersect method.

Image Processing

The first step in image processing was to test the system, including the hardware, synchronization, image capture, and analysis algorithm. Approximately 8,750 images of field roots and 5,750 from greenhouse plants (washed sand medium) images were analyzed by the DIP. This represents a total of about 450 trays. All of these trays were also hand counted using the line intersect method. Additionally, several trays

of different gauge wire and several trays of string were used for initial testing and calibration.

Initial Calibration

It was necessary to calibrate the DIP in order to convert the output in pixels to root length. This was done using string of a known length. Several trays of string were used, each with a different total length of string. Different thicknesses of string were used and pieces were randomly cut into lengths from 0.5 cm to 3 cm to simulate roots. String thicknesses varied from 0.25 to 1.0 mm and length per tray varied from 2.25 m to 27 m. This wide range of string length was used to simulate the wide variability in root length among root samples. The string trays were video taped with the same procedure as actual roots and images were processed by the DIP both with and without the debris extraction algorithm. Each tray of strings was video recorded, mixed and re-spread, and video taped again. In this way the repeatability of the DIP analysis could be tested since there were two trays of exactly the same total string length.

Data are presented in Table 1. String length (cm) was per tray. The "Total Pixels" columns represent the total number of pixels found by the DIP. The values in the "Pixels/cm" columns were calculated by dividing the total pixels by the total string length. These are the figures applicable to actual root image analysis.

In Table 1, note that the pixels/cm is a function of

Table 1. Calibration of digital image processor using trays of strings. Each line represents the same tray of strings analyzed with or without the debris extraction algorithm engaged. Each length was analyzed twice, with the strings mixed and redistributed between analyses.

Length of String (cm)	Without Debris Extraction Algorithm		With Debris Extraction Algorithm	
	Total Pixels	Pixels /cm	Total Pixels	Pixels /cm
225	26763	118.95	23111	102.72
225	26457	117.59	22957	102.03
450	52666	117.04	45995	102.21
450	51724	114.94	42935	95.41
675	80604	119.41	68281	101.16
675	80051	118.59	68577	101.60
900	88855	98.70	80312	89.24
900	90210	100.20	78840	87.60
1800	174965	97.20	152337	84.63
1800	175263	97.40	151310	84.06
2700	257561	95.40	218738	81.01
2700	256600	95.00	218765	81.02

density of the string. As more string was added to the images, the DIP results showed fewer pixels per actual length. This is thought to be due to the effect of overlapping pieces of string. Obviously, as more string is added to the tray, there will be more overlapping. The DIP identifies only one length of string where there may actually be two or more. Thus the pixels/cm figure used to convert the DIP output from root images will vary with density of the sample, or the accuracy will vary with density of the roots. This problem has not been solved to date.

Secondly, note that the DIP results are similar for two trays with the same total length of roots. The largest discrepancy is in the 450 cm string trays with debris extraction. There are approximately 7% more pixels reported for one tray than the other. Reasons for any discrepancy include differences in overlapping of strings, noise on the video tape, differences in the analog to digital conversion at the time the image is fed into the DIP, and general electronic noise due to current fluctuation.

Finally, a comparison of the results between analysis with or without debris extraction shows large differences. The difference is approximately 14%, and is consistent across the range of string lengths/tray used. Apparently the DIP identifies some "debris" in these trays of string. Since the debris extraction algorithm uses a length:width ratio to distinguish between debris and roots there must be some non-string characters appearing in the image. The most likely

cause of the "debris" detected is video noise, which is electronic noise recorded on the video tape or picked up during the analog to digital conversion of the image. Another possible source of the "debris" is surface roughness of the strings which is enhanced by the processing algorithms and appears as small branches. This roughness characteristic may be a problem on roots as well. Some of the root roughness may be related to root hairs or branching of very fine roots and thus should be counted as root length. However, the current resolution is not sufficiently high to detect root hairs so it would be more accurate to discard any length due to root surface roughness if that were possible. At present that distinction cannot be made.

Additional calibration was carried out using several images with different total lengths and gauges of wire. These analyses were used to test the width determination part of the algorithm. As described previously, the DIP analysis is carried out on a pixel basis and it was necessary to determine the relationship of a pixel width to actual width. The current algorithm was designed to divide the roots into 5 width categories, 1-2, 3-4, 5-6, 7-8, and 9-10 pixels wide.

Two sets of tests were conducted. In the first, images used were composed of pieces of wire cut into 1 or 2 cm lengths. Six diameters of wire were used, including 0.1, 0.2, 0.25, 0.6, 1.1, and 1.6 mm. 38 images were recorded using various combinations of wires and various orientations within the images (e.g. x oriented, y oriented, crossed,

diagonal, random, systematic, etc.). Data is presented in Table 2.

There are several images (rows of data) for which no pixels were found by the DIP. One probable reason is that the wires in these images are too fine to have sufficient contrast in the image for the DIP to find the objects in the image. This could possibly be corrected by setting a different threshold. However, a lower threshold would allow additional "noise" to be picked up along with lower contrast objects in the image. The choice of threshold level was made after careful testing. The threshold is reset (automatically) for each image and this may explain why the wires are picked up in some images while in other images the same size and number of wires are not found. This inconsistency raises questions about the reliability of the current system in accurately analyzing fine roots.

The data in Table 2 show that the separation of wires into different width classes was not conclusive in this test. This is true at all width classes, but especially at the 1-2 and 3-4 pixel width classes.

The results of a second series of tests for width class determination are presented in Table 3. Each image contained eight 1 cm pieces of wire. The wires in each image were the same width. Fourteen different widths of wire, ranging from 0.1 to 1.4 mm, were tested. The images were video taped, the wires within each image randomly rearranged, and the image video taped again. This was done three times. The results

Table 2. Width and length calibration data from digital image processor. Each row represents one image. Wire pieces were 1 cm long except for the 1.6 mm width, which was 2 cm long.

Total Length	Wire Width	Width Classes (pixels)					Total Length	Pixels /cm
		1-2	3-4	5-6	7-8	9-10		
(cm)	(mm)	length (pixels)					(pixels)	
1	0.1	0	0	0	0	0	0	0
1	0.1	0	0	0	0	0	0	0
1	0.1	0	0	0	0	0	0	0
3	0.1	160	122	0	0	0	281	93
6	0.1	608	210	0	0	0	818	136
6	0.1	0	0	0	0	0	0	0
8	0.1	523	305	0	0	0	828	104
8	0.1	478	327	0	0	0	805	101
1	0.2	83	26	0	0	0	109	109
1	0.2	0	0	0	0	0	0	0
1	0.2	0	0	0	0	0	0	0
8	0.2	340	453	0	0	0	793	99
1	0.25	58	40	0	0	0	98	98
1	0.25	210	90	0	0	0	300	300
1	0.25	30	84	0	0	0	114	114
8	0.25	223	559	0	0	0	782	98
1	0.6	21	82	18	0	0	121	121
1	0.6	7	97	6	0	0	109	109
1	0.6	0	84	13	0	0	97	97
8	0.6	17	725	51	0	0	792	99
1	1.1	9	9	7	52	37	114	114
1	1.1	17	13	9	69	18	126	126
1	1.1	12	6	5	35	53	111	111
8	1.1	23	48	32	550	135	788	99
2	1.6	50	8	5	7	4	73	37
2	1.6	21	16	5	11	5	57	29
2	1.6	7	12	3	11	5	38	19
8	1.6	25	27	25	32	26	134	17
7	*	215	181	37	41	58	531	76
7	*	102	291	14	80	13	500	71
7	*	79	279	65	75	31	529	76
7	*	106	306	45	84	20	561	80
7	*	104	289	24	9	5	430	61
7	*	133	242	22	12	7	416	59
7	*	134	245	5	6	5	395	56
7	*	119	213	40	12	7	392	56
24	**	593	1642	91	1	0	2327	97
24	**	536	1632	176	0	0	2344	98

* 1 cm length of each of the 5 smallest width classes, 2 cm of 1.6 mm width.

** 8 cm length of each 0.2, 0.25, and 0.6 mm width.

Table 3. Results of width class calibration. Each row represents one image analyzed three times. Figures reported are the average of the three analyses. Each image contained eight 1 cm pieces of wire of the given width. Breaks between rows of data indicate break between width classes as determined by digital image processor.

Wire Width (mm)	Width Classes (pixels)					Total Length (pixels)	Pixels /cm
	1-2	3-4	5-6	7-8	9-10		
----- length (pixels) -----							
0.10	278.5	528.6	0.0	0.0	0.0	807.1	101
0.20	234.4	571.3	0.0	0.0	0.0	805.7	101
0.25	246.2	582.4	0.3	0.0	0.0	828.9	104
0.35	135.1	667.8	2.2	0.0	0.0	805.1	101
0.45	36.6	749.9	3.5	0.0	0.0	790.0	99
0.50	32.6	780.0	5.0	0.0	0.0	817.5	102
0.60	25.9	746.4	26.6	0.0	0.0	798.8	100
0.75	30.4	82.6	702.3	27.6	0.3	843.2	105
0.85	154.3	59.9	371.3	365.7	0.4	951.5	119
0.90	164.0	77.9	246.8	471.1	0.0	959.8	120
1.05	342.7	74.5	45.3	599.6	107.9	1170.0	146
1.10	197.2	89.9	51.7	466.5	268.2	1073.5	134
1.25	231.9	85.1	50.9	225.3	508.7	1101.9	138
1.40	181.9	100.6	60.2	66.6	54.3	463.7	58

presented in Table 3 are the averages of the three images for each width class. The repeatability of the DIP analysis was found to be quite good as the variability among the three analyses was small.

The results of this test were more conclusive than the first width class calibration. While the distinctions between the width classes are not precise, the general trends are defined. The division between the two smallest classes, 1-2 and 3-4 pixels, is ambiguous but wires from 0.1 to 0.35 mm are split between the 1-2 and 3-4 pixels width classes, while wires 0.45 to 0.6 mm appear primarily in the 3-4 pixel width class. The 5-6 pixel wide group includes wires between 0.75 and 0.9 mm. Wires 1.05 and 1.1 mm in diameter are predominantly in the 7-8 mm class. 1.25 mm wires are in the widest class, 9-10 mm, and wires 1.4 mm in diameter were too large to be included in the given width classes.

The fact that the distinctions are not precise between or within width classes is due to electronic noise, bounce or shadows which may appear as separate images, or some thresholding differences. The bounce problem is most evident in the wider groups. Note that the total pixels (last column, Table 3) reported for wires in each width class is relatively constant until wire width exceeds 0.75 mm diameter. The total pixels then increases appreciably. There is a more pronounced bounce effect with larger wires since the shadows are larger. These bounces are above the threshold and are counted as objects.

In summary, current width class distinction is operational in a general way but is not yet sufficiently precise for broad application. The problems appear to be due to image recording, transfer, and capture rather than the analysis algorithm.

Tables 2 and 3 provide additional data on pixel length calibration. In Table 2 the last column is pixel/cm determined by dividing total pixels by the known length of wire in the image. Values range from 33.5 to 300 pixels/cm but most of them fall between 60 and 110. As discussed relative to Table 1, pixels/cm is a function of density. In the images with 8 cm total length the pixels/cm is constant at around 100. Images with less than 8 cm are much more varied. The images in Table 3 all contained 8 cm of wire. Again, the last column is pixels per centimeter and is relatively constant at around 100 until wire width increases to the point that the bounce problem is more significant.

Time

One of the objectives for the development of image processing for use in root studies is to increase the speed by which root samples can be analyzed. Time for video recording the roots was discussed previously and, while still requiring much time, it is a slight improvement over the conventional line intersect method. DIP processing time, however, adds significantly to the total time necessary for data collection. The current algorithm used to analyze clean root

samples (no debris extraction) requires approximately 1.2 minutes per image. Since each sample of roots is spread out in a tray which is recorded in 64 video images, 75 minutes are required to analyze one root sample. The system is automated so that all of the images on a two hour video tape can be analyzed without an operator present. However, the total number of samples analyzed in one 24 hour period is limited by processor speed to a maximum of 18 or 19.

The processing of root samples with debris is considerably more time consuming. The operation of the debris extraction routine approximately triples the time required to process an image which means that about 6 or 7 samples can be processed in one 24 hour time period. Since nearly all field root and many greenhouse root samples contain debris, the extraction routine is a critical part of the analysis. A comparison of processing time required per root sample for different root applications is presented in Figure 1.

Processing speed must be increased if the system is to be useful in analyzing large numbers of root samples. Options for higher speed include an improved algorithm or a faster computer. Another option would be subsampling, or analyzing only a fraction of the total images. Tests were run to examine the possibility of subsampling and to determine what fraction of the images in a tray would need to be analyzed to achieve acceptable results. Figure 2 summarizes test results. Figure 2 was developed using output from DIP analysis of a tray of roots. Total pixels of length for each

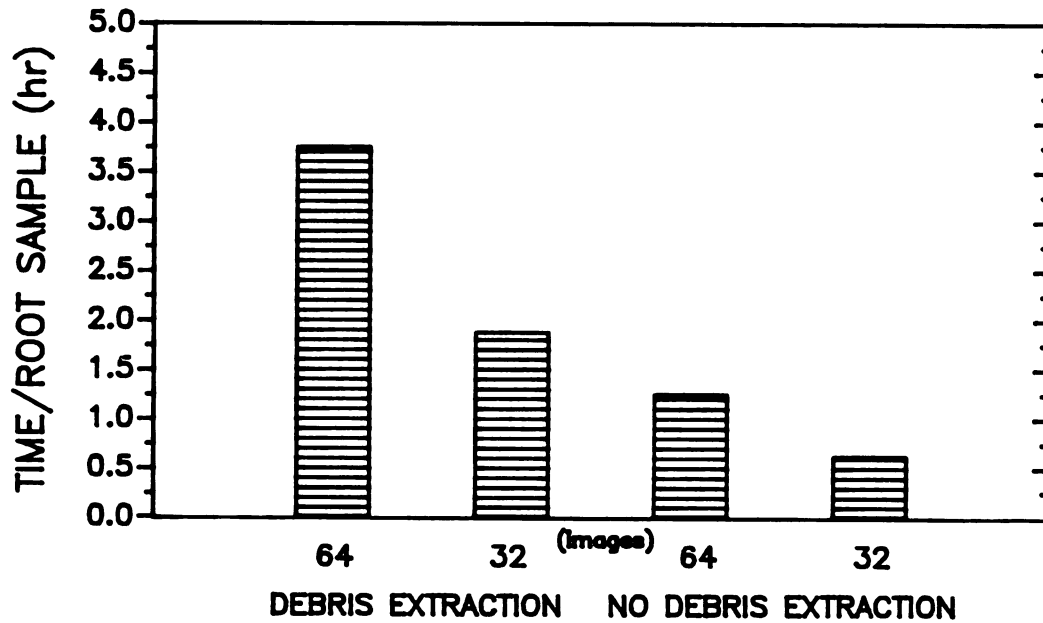


Figure 1. Time required for image processing as affected by the use of the debris extraction algorithm and the number of images processed.

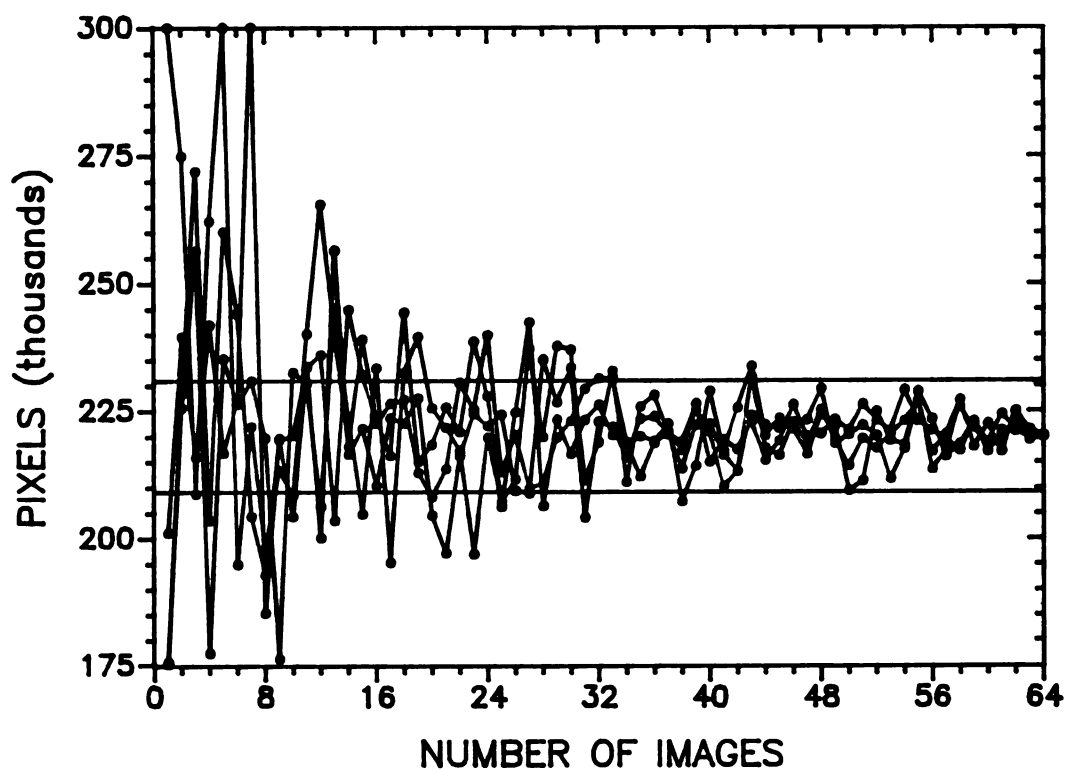


Figure 2. Results of tests to determine the fraction of images from a tray of roots which must be processed in order to obtain acceptable estimates of root length. Horizontal lines represent $\pm 5\%$ of the actual (DIP) analysis total. Each data point represents the pixel sum (y) of the randomly selected images (x) multiplied by $(64/\text{number of images})$.

of 64 images in a tray were compiled. A given numeral of the 64 was randomly selected and multiplied by the fraction $64/1$ to yield an estimate of the total pixels in the tray if each image contained the same number of pixels as the selected image. This same procedure was carried out for 2, 3, 4, etc., to 64 images. (For example, the sum of pixel lengths of 23 randomly selected images was multiplied by the fraction $64/23$.) The results of several runs of this procedure are plotted in Figure 2. The horizontal lines represent plus and minus 5% of the total number of pixels as determined by summing the pixels lengths from each of the 64 images. This procedure was carried out on several trays of roots and the results were very similar to those presented in Figure 2. These results led to the conclusion that analyzing one half of the images in a tray and multiplying their sum by two would yield total length estimates which would be acceptable since they are almost always within 5% of the total. Reductions below 32 in the number of images analyzed would decrease the likelihood of obtaining results within the $\pm 5\%$ range selected as acceptable error. Analyzing 32 of the images cuts the time in half which is obviously a major step in increasing processing speed without seriously decreasing accuracy of the results.

Tray Size

Most of the testing and root analysis carried out has been done using the 43 x 43 cm trays for root video taping.

However, since some root samples contain very few roots, a smaller tray was developed for use. The smaller tray was used in order to concentrate the roots sufficiently to eliminate analysis problems due to limited density. When appropriate, the small tray also has the advantage of saving time both in video recording (1 min., 47 sec. to video the small tray vs 6 min., 4 sec. for the large tray) and in DIP analysis time (16 images in the small tray vs 64 images in the large tray). Comparison of results of analysis using the two tray sizes indicated that the two sizes could be used interchangeably with only a slight difference in results. The large tray resulted in slightly more pixel length recorded, probably due to the previously described noise problem.

Analysis of Debris-Free Roots

Roots of dry beans (*Phaseolus vulgaris*) grown in washed sand in a greenhouse were analyzed by the DIP. The debris extraction routine was not used in processing these images since there was no debris in the root samples. Roots were cut into 1-3 cm pieces and evenly spread and separated prior to counting by the line intersect method or video taping. A comparison of the line intersect and DIP methods is presented in Figure 3. The pixel length conversion factor used was 100 pixels = 1 cm root length (determined from calibration testing as reported in Table 1). The results show that the DIP overestimates root length compared to the line intersect method. The overestimation on the 169 trays of roots compared

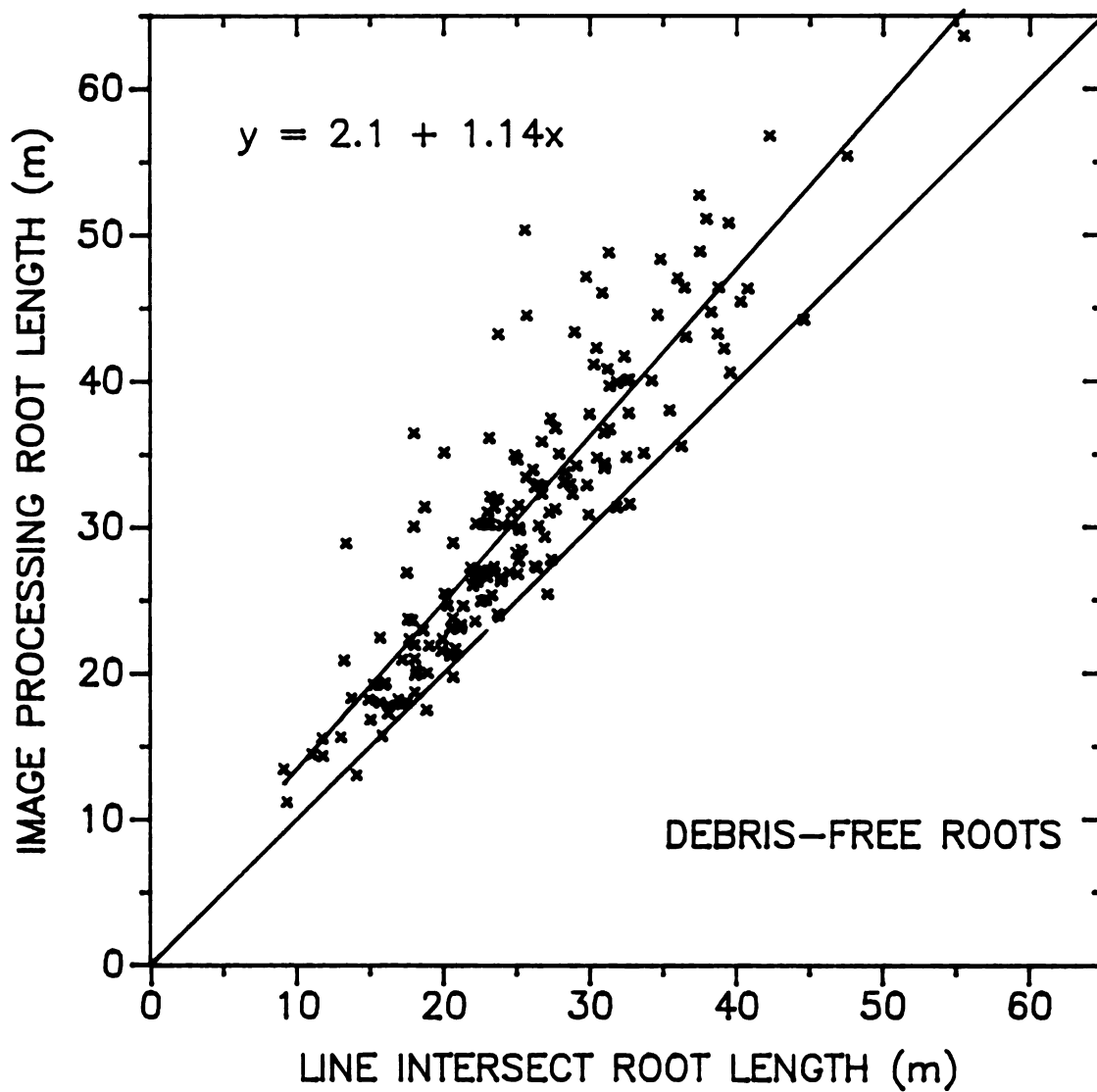


Figure 3. Comparison of root length determinations by image processing and line intersect methods. Each point represents one tray of roots, or 64 images. Diagonal line represents 1:1 correlation; top line is regression line.

averaged 23%, with a range of -6 to 125%. However, the overestimation is nearly linear in the range of lengths examined, as shown by the regression line (top line) and the 1:1 ratio line in Figure 3. The overestimation is due to electronic noise, video tape imperfections, bounce, and imperfections in the image transfer from digital to analog and back to digital again. These causes are discussed in other sections of this report.

Analysis of Field Roots

The DIP was used to analyze a set of field roots and the results compared to those obtained by the conventional line intersect method. Field roots of dry beans (*Phaseolus vulgaris*) were extracted and processed as described in Chapter 2. A summary of results from the analysis is presented in Table 4. In this table root length densities (RLD) are presented for the DIP analysis and compared to the line intersect method on a percentage basis. Four treatment combinations are presented: ARNST is alfalfa rotation, no secondary tillage; CONV is conventional rotation/tillage; IR is irrigated and NI is nonirrigated. The first column of numbers, 1-18, identifies the location of the 7.5 cm cube of soil in the extracted soil profile. Cubes 1-3 are from the center of the plant away from the plant and from 0-7.5 cm depth. 4-6 represent the next three cubes, from 7.5-15.0 cm depth, and so on down the profile.

Each figure in Table 4 is the average of four

Table 4. Comparison of root length density (RLD, cm cm⁻³) determination by the digital image processor (DIP) and the line intersect (LIN) methods. Root length densities reported are calculated from the DIP length determinations. % LIN is the DIP RLD divided by the line intersect RLD. The 18 cases represent the RLDs from 18 cubes of soil, 7.5 cm on a side, from a soil-root profile 7.5cm thick x 22.5 cm wide x 45 cm deep.

Soil Cube #	ARNST IR RLD	% LIN	ARNST NI RLD	% LIN	CONV IR RLD	% LIN	CONV NI RLD	% LIN
1	3.79	112	5.60	145	4.74	115	4.89	143
2	4.10	133	4.71	127	4.47	126	5.68	168
3	4.99	146	3.72	128	4.48	129	5.73	162
4	6.25	176	3.88	151	6.03	280	3.57	154
5	4.11	164	3.55	132	6.58	287	4.17	180
6	5.64	169	4.58	153	6.26	260	4.49	206
7	3.27	145	4.34	162	4.39	222	3.41	157
8	2.91	141	3.62	150	3.72	184	3.66	173
9	2.47	121	2.73	131	2.80	150	3.51	184
10	1.57	104	1.15	76	2.28	136	2.53	126
11	1.54	107	1.81	122	2.79	134	2.66	138
12	1.43	102	1.25	115	2.27	134	1.88	137
13	1.08	114	0.83	116	1.93	132	1.67	132
14	0.87	101	1.30	129	2.54	169	2.04	123
15	1.53	118	0.89	119	1.58	128	1.35	113
16	0.84	106	1.24	122	1.82	141	1.77	158
17	1.10	112	1.34	119	1.41	127	1.92	132
18	1.10	117	0.93	125	1.39	143	1.61	163

replications. Thus each RLD as determined by the DIP includes the results from 256 images (4 replications or trays of roots x 64 images per tray). The results show that the DIP overestimates the root length by a large margin. The range is 76% to 280%, with only one point below 100%. Averages for the rotation/tillage treatments at each depth are presented in Table 5, along with the overall averages. The overall average is 144% of the line intersect method. Conversion factor for the pixel lengths determined by the DIP was 100 pixels/cm.

In addition to the general overestimation problem, there is a problem with the debris extraction routine. The field root samples contain varying amounts of debris, primarily organic debris from previous years' crops. The CONV treatments contain more debris than the ARNST treatments because corn was grown the previous year on the CONV and alfalfa on the ARNST plots. The cornstalks were plowed down following grain harvest. This is indicated in the DIP RLD data in soil cubes 4-9, which represent the 7.5-22.5 cm depths of soil. It is in this depth range that the majority of the decomposing corn stalks would be expected. The CONV treatments at this depth show the greatest overestimation of RLD by the DIP. These root samples had large amounts of organic debris but did not have appreciably more roots than the ARNST samples at the same depths (line intersect data). Figure 4 shows the overestimation of RLD by the DIP for all depths. The points in the higher RLD ranges primarily represent the samples from

Table 5. Summary of digital image processing (DIP) root length density (RLD) results compared to line intersect results. Figures represent DIP RLDs/line intersect RLDs x 100. Roots are field grown dry beans, ARNST is alfalfa rotation, no secondary tillage and CONV is conventional management.

DEPTH (cm)	ARNST (ave)	CONV (ave)	OVERALL (ave)
	----- % of line intersect -----		
0-7.5	131.7	140.7	136.2
7.5-15.0	157.3	227.9	192.6
15.0-22.5	141.7	178.2	160.0
top 22.5 cm	143.6	182.3	162.9
22.5-30.0	104.3	134.2	119.2
30.0-37.5	116.2	132.8	124.5
37.5-45.0	116.8	144.0	130.4
22.5-45.0 cm	112.4	137.0	124.7
Overall Averages			
0-45.0 cm	128.0	160.0	143.8

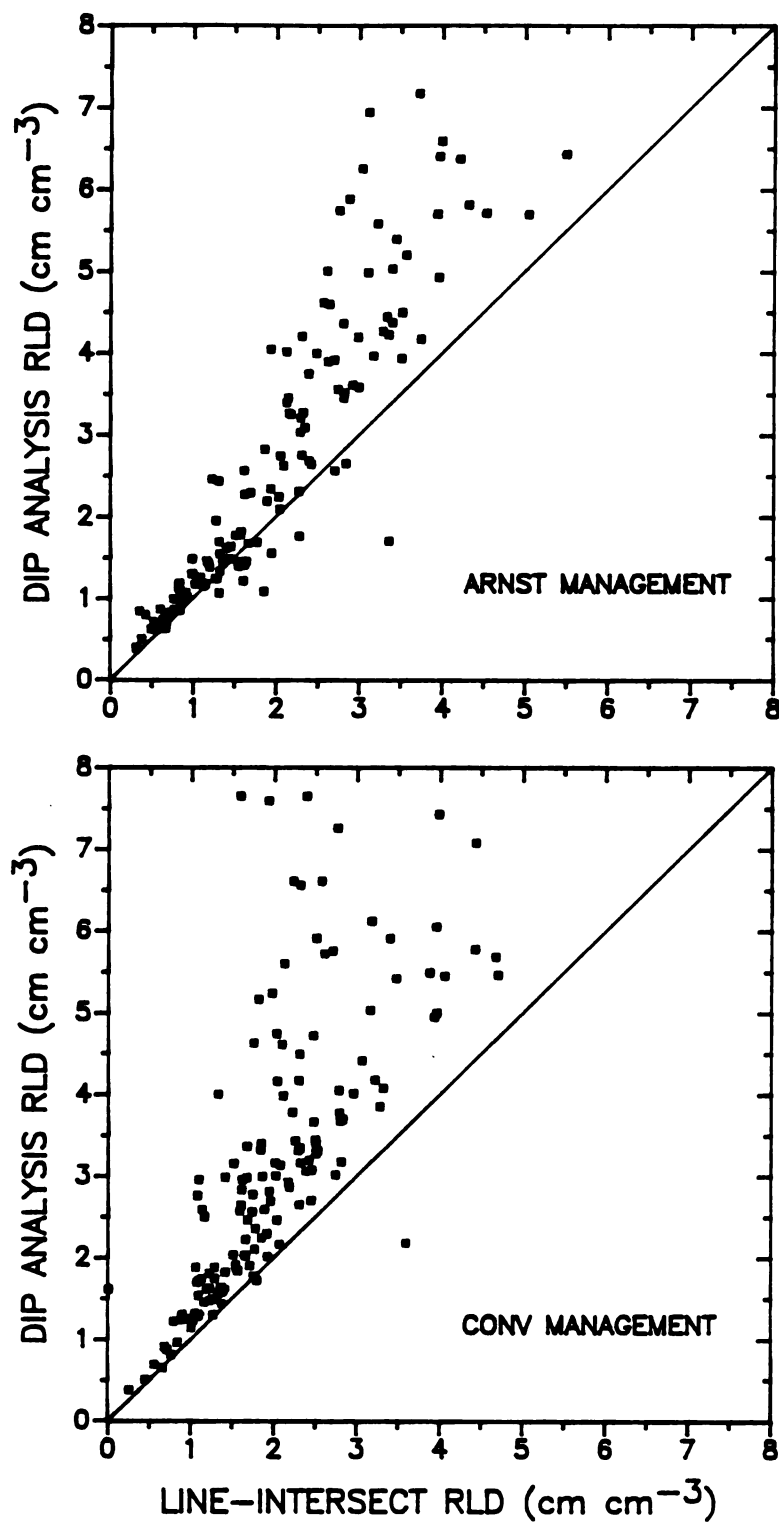


Figure 4. Comparison of root length density as determined by the digital image processor and the line intersect method for dry bean roots under two crop and soil management systems. Each point is the average of 4 replications.

the top 22 cm of soil. Also, those points further from the 1:1 correlation line represent samples with greater amounts of debris.

SUMMARY

Numerous tests have been run on the root imaging and digital image processing systems. These tests have provided a comprehensive look at the system and indicate the positive aspects as well as various problems to be dealt with prior to using the system to collect useful root data.

The imaging system works well. The hardware and software are well integrated and automated. Given the technology used, the system produces high quality root images.

The digital image processing hardware is also well integrated and functions well. Occasional breakdowns in processing occurred for unknown reasons. Electrical current fluctuations are suspected as the primary cause.

Many of the problems encountered seem to be related to image resolution. At the current resolution many of the roots are only one or two pixels wide. Much of the "noise" in the images is one, two or three pixels wide and thus is picked up by the DIP and included as roots. A higher resolution would allow for elimination of the video noise by thresholding or other image enhancement techniques. The problem of debris extraction is critical to the success of any

root analysis procedure. The current resolution and algorithm do not adequately separate the debris from the roots in the images. If resolution were increased, the debris problem could more easily be handled since the distinctions between roots and debris would be magnified. However, this problem will not easily be solved since some of the debris is organic matter which is almost exactly like roots in appearance. Additional developments in software may enhance the possibilities for successfully distinguishing between roots and debris. Finally, some samples with excessive amounts of debris may need to be separated into two or more subsamples to decrease root/debris crossover which increases the difficulty of accurately analyzing the sample.

Many of the problems encountered are related to the image storage mechanism, video tape. There are several related problems which suggest this technology may be inherently flawed for this application. The electronic and magnetic tape "noise" is a primary source of error. The images must be converted from digital (video camera) to analog (video tape) and back to digital (for processing by the DIP). The conversion process is not precise and introduces potentially significant error. In the long run it will be advantageous to develop the system around a different image storage technology, for example, laser disc.

Increased resolution has been suggested as offering significant improvement in the system. However, increased resolution would introduce yet another problem. The time

required to video record the images would be increased. Even more importantly, the time required for processing the images with the current digital image processor would be increased from its already prohibitive level. A doubling or quadrupling of the number of images would require an equal increase in processing time.

Image processing has much potential for greatly enhancing root studies. However, there are numerous problems to be solved before the system is widely applicable to root study. The system development and testing reported here are an important step in the direction of root study by digital image processing.

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CHAPTER 5

EFFECTS OF FLOOD OR DROUGHT STRESS ON DRY BEAN ROOT AND SHOOT GROWTH

INTRODUCTION

Field grown crops are often subject to stress from too little or too much moisture. The most common cause of stress is insufficient or excessive precipitation. The effects of less than optimum precipitation may be compounded by soil conditions, as will be discussed later. Temporary or permanent plant injury from flooding or drought can result in costly damage to the crop.

Schwartz (1980) has pointed out that too little soil water can damage plants due to the unavailability of water for plant roots, the accumulation of toxic ions, stomatal closure which results in restricted CO₂ uptake, and temporary or permanent plant wilt. Laude (1971) noted that stomatal closure results in reduced photosynthetic activity per unit leaf area and an overall decrease in leaf area. A reduction in plant size is typical of plants growing without sufficient moisture.

Plants subjected to a mild drought may recover quickly and production may not be greatly affected. Plants have been found to grow more rapidly for a short time following

rewatering than those which experienced no drought (Laude, 1971). However, plants subjected to a more prolonged drought may suffer irreversible damage. Plants may be damaged to the extent that they are incapable of resuming growth when the drought ends.

Flooding can be equally detrimental to plant growth. Schwartz (1980) noted that flooding may leach nutrients essential to plant growth, reduce O_2 content, induce plant chlorosis, and lead to accumulation of toxic byproducts from anaerobic metabolism. Jackson and Drew (1984) suggest that the primary effect of flooding is asphyxiation of the plant due to decreased gaseous diffusion. Root growth is generally retarded and root survival time varies greatly with species (Glinski and Stepniewski, 1985). Jackson and Drew (1984) also point out that leaf growth is extremely sensitive to flooding and root anoxia.

Soil conditions, for example soil compaction, may exacerbate the problems related to too much or too little water. A compacted soil or compacted layers in a soil may increase the effects of either problem. A compacted horizon can prevent adequate drainage following rainfall or irrigation. The resulting waterlogging will quickly lead to an anaerobic rhizosphere environment (Cannell and Jackson, 1981). Anaerobic conditions have been reported as the cause of decreased plant growth rate (Smucker and Erickson, 1987), cessation of root growth (Letey et al., 1962; Glinski and Stepniewski, 1985), and death of some root tips (Huck, 1970).

Root systems have been found to grow deeper in a moisture shortage situation (Klepper et al., 1973; Cortes and Sinclair, 1986; Huck, 1986; and Hoogenboom et al., 1987). However, downward root growth can be slowed due to compaction related mechanical impedance (Bertrand and Kohnke, 1957; Raghavan, 1977; and Bennie and Botha, 1986). When this occurs the root system may be prevented from tapping into the available moisture deeper in the soil profile.

Shoot:root ratios are usually altered by environmental stress. In general, stress on the root system such as flooding or drought leads to a decreased shoot:root ratio as the plant partitions more of its photoassimilate to the root system (Shank, 1945; Taylor, 1981; Hoogenboom et al., 1987). As this occurs it results in decreased shoot growth which can lead to decreased yield, especially when the stress occurs during a critical growth stage such as reproduction (Huck et al., 1986).

The objectives of this study were: a) to examine the effects of drought or flood stress on growth parameters of dry beans (*Phaseolus vulgaris*); and b) to gain some indication of expected dry bean response to conditions of less than optimum moisture which may occur when the crop is grown on a soil with compaction problems. The study was conducted in a greenhouse in order to control environmental conditions. Both shoot and root responses were monitored. The response and recovery of the plant to the stress was measured at three times following the stress application.

MATERIALS AND METHODS

Description of Experiment

A greenhouse study of the effects of drought and flood on dry bean shoot and root parameters was conducted in the spring of 1986. Dry beans (cultivar C-20) were planted in PVC tubes sealed at the bottom with the exception of a drain hole approximately 1 cm in diameter. The tubes were 70 cm long and 7.6 cm inside diameter. Planting medium was washed silica sand (0.3-0.6 mm). Four seeds were planted in each pot. Soon after emergence each pot was thinned to two uniform seedlings. Plants were irrigated with half strength Hoaglund's nutrient solution. Drip irrigation was carried out as needed to maintain good moisture throughout the sand profile. Irrigation varied from 4 to 6 times per day for 15 or 30 minutes per irrigation. Differences in irrigation were due to weather conditions and plant size.

The first seedlings emerged 4 days after planting and emergence was approximately 90% by the following day. Fluorescent lighting was used to supplement available sunlight and to extend the photoperiod to 16 hrs day⁻¹. This photoperiod was maintained throughout the study.

Stress treatments were applied 42 days after planting. Plants were at maximum flowering at this time. One third of

the plants received drought stress as follows. A vacuum pump was used to apply suction to the bottom of the tubes. Suction was applied for 3-4 minutes per pot to draw off free water. The drought stress was continued for 7 days with only enough solution added to maintain plant viability.

The flood treatment was also applied 42 days after planting. Irrigation solution was pumped into the tubes from the bottom via the drainage port until it overflowed the top. The drainage outlet at the bottom of the tubes was then closed off and flooding maintained for 7 days. The remaining one third of the plants, designated the control, were irrigated normally.

Ambient weather prior to and during stress treatments was cloudy and cool. Without sunlight the stress effects were less pronounced in a given period of time. The 7 day stress period was chosen in order to subject the plant systems to a severe stress.

Following the stress period the drainage ports of the flooded tubes were opened and all plants received normal irrigation.

The experimental design was a randomized complete block with four replications.

Measurements

Whole plants were harvested on three dates. Shoots were cut off at the soil surface. Roots and sand were pushed out of the tubes using air pressure. The sand-root

cores were cut into three sections by depth (0-20 cm, 20-40 cm, and 40-70 cm). The sand was washed away from the roots by gentle motion in a water tub. Roots were patted dry and frozen for future analysis.

The first harvest was immediately post-stress or 49 days after planting. Measurements included stem and leaf fresh and dry weights, and leaf area determined by a Licor Leaf Area Meter. Roots were harvested as outlined above.

The second harvest was carried out 12 days after the first or 61 days after planting and measurements were the same.

The third harvest was at maturity (90 days after planting, 41 days after termination of stress). Parameters measured included the roots, fresh and dry weights of stems, pods, and beans, and number of beans and pods per plant.

Root length was determined using Tennant's line intersect method (Tennant, 1975) or by video image analysis as described in Chapter 4. Roots were stained with 5% malachite green prior to counting or video taping. A four cm square grid was used for the line intersect method.

RESULTS AND DISCUSSION

Shoot parameters, including both fresh and dry weights, for the three harvest dates are presented in Table 1. The effect of the imposed stress was evident soon after the plants were subjected to either flood or drought. The drought stressed plants showed a greater decrease in growth than did the flooded plants relative to the control plants. Dry weight of the shoots subjected to drought was significantly lower than the control shoots. The flooded plants had shoot dry weights 15% lower than the control plants but the difference was not significant at the 95% level. Decreased growth was seen in both the stems and leaves.

The effects of the environmental stresses were evident throughout the remainder of the study. At 12 days post stress the dry weights of both the flood and drought stressed plant shoots were significantly lower than the control. The shoots subjected to drought weighed 23% less than the flooded plant shoots at this harvest. However, at final harvest the drought stressed plants had surpassed the flooded plants in shoot fresh and dry weights. The recovery of growth of dry beans subjected to drought was greater than the recovery of the flooded beans when measured over a time period of several weeks. At final harvest the shoot dry weights of both

Table 1. Dry bean shoot parameters on three dates following stress as affected by flood and drought stress.

	Immed Post Stress			12 Days Post Stress			41 Days Post Stress		
	Plant	Stems	Leaves	Plant	Stems &Pods	Leaves	Plant	Stems	Stems
----- dry weight (g) -----									
Control	15.9	7.8	8.2	38.2	27.9	10.2	44.1	9.7	
Flood	13.4	7.2	6.1	24.6	18.6	6.0	20.8	5.0	
Drought	9.2	4.4	4.8	19.0	12.3	6.8	25.9	7.2	
LSD.05	5.7	3.4 (ns)	2.5	8.9	7.1	2.1	13.8	5.4 (ns)	
----- fresh weight (g) -----									
Control	126.2	61.3	65.0	257.6	173.0	84.7	71.9	31.4	
Flood	97.8	51.0	46.8	168.7	111.2	57.5	27.7	9.8	
Drought	68.0	35.6	32.5	144.9	77.3	67.7	41.2	18.9	
LSD.05	34.6	20.1	16.7	67.3	9.0	25.5 (ns)	36.6	24.4 (ns)	

flooded and drought stressed plants were significantly lower than the dry weight of the control plants.

Leaf area is reported in Table 2. The effects of the stress included a statistically significant decrease in total leaf area per plant and area per leaf. The total number of leaves per plant also decreased due to the imposed stress. The plants subjected to drought suffered a greater decrease in leaf number and area than did the flooded plants relative to the control plants. Laude (1971) suggested that drought causes stomatal closure which results in decreased photosynthesis and activity per unit leaf area and an overall reduction in leaf area. However, as noted previously in the shoot weight data, the recovery of the drought stressed plants was greater than the flooded plants. Laude (1971) also noted that plants subjected to drought may recover quickly and for a short time even grow more rapidly than plants not subjected to insufficient moisture conditions. The drought stressed plants apparently began adding new leaves soon after the end of the stress period as they had 20% fewer leaves than the flooded plants immediately post stress but twelve days later the plants subjected to drought had 18% more leaves than the flooded plants. Area per leaf was less for the drought stressed plants but leaf area per plant was slightly greater compared to the flooded plants, indicating that many of the leaves on the plants subjected to drought were new, smaller leaves.

Root length for the three treatments and three harvests

Table 2. Leaf area and number of leaves immediately post stress and 12 days after stress as affected by flood and drought stress.

	Immed. Post Stress			12 Days Post Stress		
	Lf Area Plnt ⁻¹	# Leaves	Lf Area Lf ⁻¹	Lf Area Plnt ⁻¹	# Leaves	Lf Area Lf ⁻¹
	cm ²		cm ²	cm ²		cm ²
Control	3170	80	39.8	4243	116	38.3
Flood	2527	76	33.3	2889	83	34.7
Drought	1852	61	30.2	2995	101	29.9
LSD.05	804	21 (ns)	3.6	964	35 (ns)	9.9 (ns)

are reported in Table 3. Total root length was significantly decreased by the flood and drought stresses relative to the control plants, and the roots of the stressed plants remained smaller throughout the study. Rooting depth was also adversely affected by flood and drought.

Immediately post stress, the drought stressed plants had the smallest total length of roots. Root distribution from top to bottom of the tube-pots was most even in the control with 68% of the roots in the top 20 cm of sand, 19% in the 20-40 cm depth range, and 13% in the bottom 30 cm of the pots. The flooded plants had a much higher concentration (80%) of roots in the top 20 cm. Only 5% of the total root length on the flooded treatment was in the lower 30 cm of the pot. This was probably due to increased oxygen availability near the surface. Jackson and Drew (1984) pointed out that

Table 3. Total root length of dry beans by depth for three dates following flood and drought stress.

Immediately Post Stress*				
Treatment	0-20	Depth (cm)		Total
		20-40	40-70	
		-----	root length (m)	-----
Control	198.1	55.4	36.1	289.6
Flood	172.2	33.3	10.2	215.7
Drought	147.9	37.9	16.1	201.9
LSD.05	67.2	28.0	24.4	

12 Days Post Stress**				
		-----	root length (m)	-----
Control	286.9	111.3	63.8	462.0
Flood	176.8	80.0	47.3	304.1
Drought	234.7	55.4	33.4	323.5

* One replication only

41 Days Post Stress*				
		-----	root length (m)	-----
Control	231.3	67.5	65.0	363.8
Flood	98.5	13.1	5.4	117.0
Drought	127.6	46.6	28.8	203.0
LSD.05	90.8	27.0	32.2	

* One replication only

* Average of three replications

** One replication

flooding generally retards root growth and that root survival time varies from a few minutes to several days. Since the flood stress in this study was applied over a seven day period, it is probable that some root death occurred. The plants subjected to drought were intermediate, with 73% of their roots in the top 20 cm of the profile, 19% in the 20-40 cm depth range, and 8% in the lower 30 cm of the pots.

12 days after the stress the plants subjected to drought had recovered more than the flooded plants as evidenced by the greater total root length measured at this harvest. This higher rate of recovery by the water deficit plants was also seen in the leaf data, (Table 2) and, by the third harvest, the shoot data (Table 1). Apparently the rate of damage or root kill was less on the drought stressed plants than on the flooded beans and this allowed the plant system to recover more rapidly.

By the final harvest the plants were mature and total root length had decreased for each treatment from the two earlier harvests. At this harvest the drought stressed plants had greater total root length as well as more even distribution of roots than the flooded plants. 63% of the roots of the plants subjected to drought were in the top 20 cm of soil, 23% in the 20-40 cm range, and 14% of the roots in the lower part of the profile. On the flooded plants the root length distribution by depth from top to bottom was 84, 11, and 5%. The control root systems had penetrated well throughout the profile, with 64, 19, and 18% of their root

length respectively from top to bottom.

Shoot dry weight:root length ratios are presented in Figure 1. Shoot:root ratios increase with time as expected (Russell, 1977). However, the higher shoot:root ratios expected on the control plants compared to the stressed plants were not evident. Taylor (1981) suggested that a plant will favor the stressed part of the plant, i.e. if the root is stressed it will receive an increased share of photoassimilate with a resulting decrease in shoot:root ratio. In this study the effects of the stresses were such that root length was severely restricted and the expected decrease in shoot:root ratio was not evident.

Final harvest parameters are presented in Table 4. The effects of the flood and drought stresses are clearly reflected in the yield parameters. The control plants yielded significantly more beans, both in number of beans as well as fresh and dry weight, than did the stressed plants. The yield response of the two stress treatments were similar. The drought stressed plants outyielded the flooded plants by 16%, (dry weight of beans per plant) although the differences were not significant at the 95% level. The yield advantage of the drought stressed plants over the flooded plants is another indication that the plants subjected to drought recovered better from the stress than did the flooded plants.

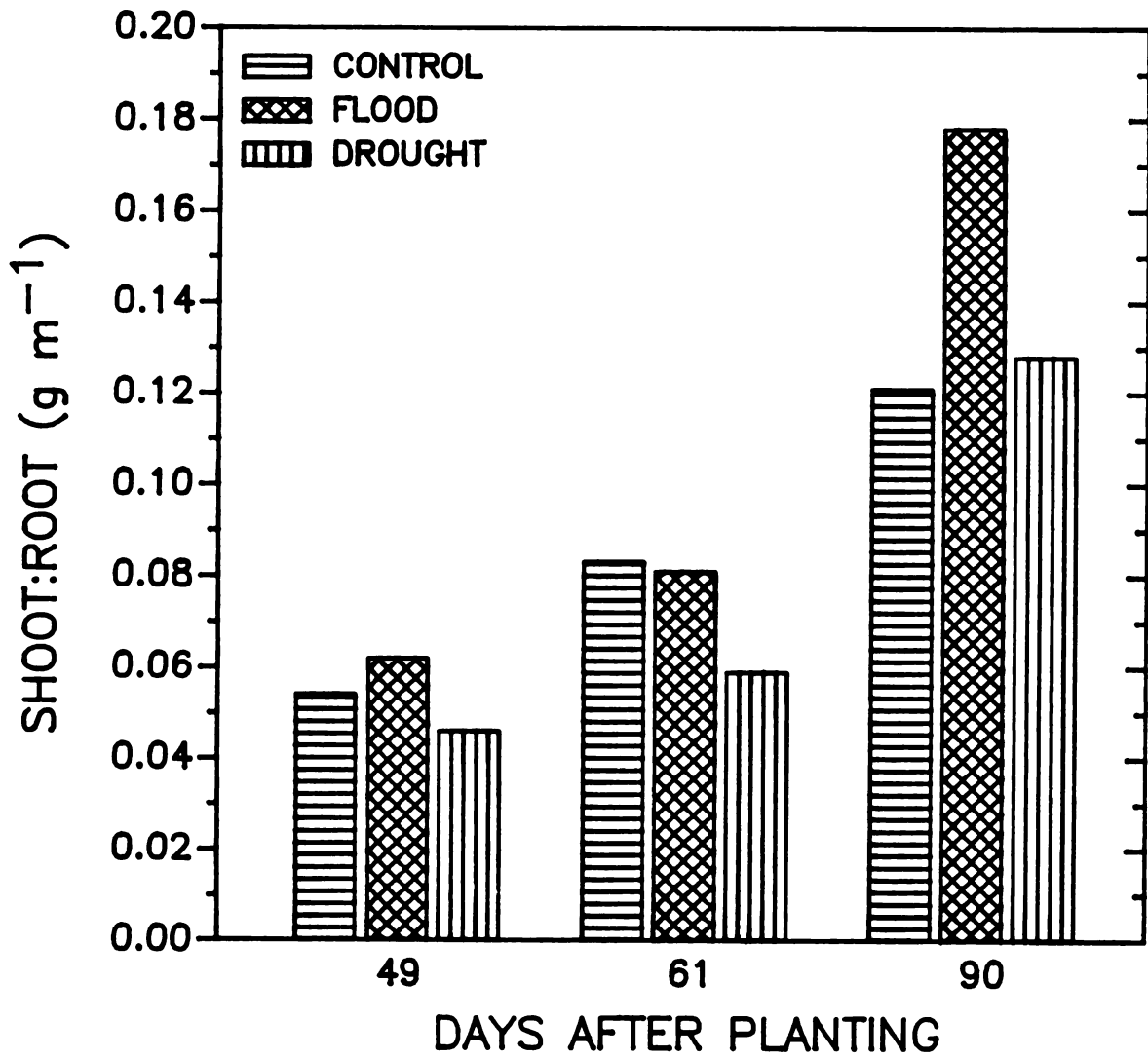


Figure 1. Shoot:root ratios (dry weight:length) of dry beans as affected by flood or drought stress and time following stress.

Table 4. Dry bean yield components as affected by flood and drought stress.

	Fresh Wt		Dry Wt		#	#	Beans
	Pods	Beans	Pods	Beans	Pods	Beans	/Pod
	----- grams plant ⁻¹ -----						
Control	9.5	31.1	7.5	27.5	34.1	141.0	4.18
Flood	4.1	13.8	3.5	12.3	19.0	84.3	4.45
Drought	8.4	13.9	4.0	14.7	18.9	81.8	4.45
LSD.05	5.7	10.0	2.9	5.9	11.8	30.8	1.00
	(ns)						(ns)

SUMMARY

In summary, the effects of drought and flood were detrimental to plant growth at the time of the stress. The effects continued on well after the stress had been alleviated. Shoot growth, leaf area, and root growth were all significantly less for plants subjected to flood or drought stress. The injury to the stressed plants was too great, and final yield was significantly decreased relative to non-stressed plants. Growth of the drought stressed plants was more negatively affected than the flooded plants at the time of the stress. However, the drought stressed plants responded better following the stress than did the flooded plants as evidenced by shoot, root, and yield parameters over time. The flooded root systems sustained more permanent damage and failed to recover to the same extent as plants with too little soil moisture.

The effects of flood and drought stress are clear. The results provide some indication as to what might be expected under field conditions. If a crop is subjected to stress due to too much or too little moisture, an adverse affect on yield can be expected. It is expected that the effects of such environmental stresses which often occur naturally will be increased when soil problems such as compaction are present.

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CHAPTER 6

CONCLUSIONS

Each of the preceding chapters included the primary conclusions from the study detailed in that chapter. This chapter summarizes those conclusions.

Chapter 2: Alleviation of Soil Stresses on Dry Bean Root Systems As a Key to Increased Dry Bean Production

A) Soil physical properties of Charity clay were improved by the use of a deep-rooted legume, deep tillage, and no secondary tillage.

- Bulk density was significantly less at the 15-22.5 cm depth range on the ARNST soil compared to the CONV soil.

- Soil moisture and aeration relations were improved by the ARNST management system.

- Pore size distribution was more favorable on the ARNST soils, with more larger pores for improved drainage and fewer smaller pores which would inhibit drainage, air flow, and possibly root penetration.

- Hydraulic conductivity was significantly improved on the ARNST soils at the 15-22.5 cm depth.

- B) Plant growth was largely unaffected by the different rotation/tillage and irrigation treatments. Differences were detected due to row spacing and cultivar.
- Shoot biomass and root length density did not differ significantly due to rotation/tillage and irrigation.
 - Narrow rows did have higher biomass per unit area.
 - Emergence of the Black Magic cultivar was better than that of the C-20 cultivar.
- C) Yield was not determined because of extensive flooding. Yield estimates showed no significant differences due to rotation/tillage or irrigation. The narrow rows had higher yields than the wider rows. The Black Magic cultivar outyielded the C-20 cultivar.

In the one year of this study no advantages to plant growth, root length density, or yield were detected due to the ARNST management system in spite of apparent improvements in soil physical conditions.

Chapter 3: Evaluation of Minirhizotron Observation Tubes as a Tool for Root Study on Fine Textured Soils

- A) The minirhizotron observation tubes did not present an accurate picture of root activity in the top 20-30 cm of soil under either rotation/tillage system in either 1985

or 1986. Destructive sampling revealed greater numbers of roots than did the minirhizotron tubes in this soil horizon.

- B) The minirhizotron method results showed much greater root activity in the 30-60 cm depth range under the CONV soils than on the ARNST soils. This data was contrary to that collected by destructive sampling.

The results indicate that the minirhizotron method of root observation is not a useful tool on fine textured soils with compacted zones. Roots apparently proliferate at the tube-soil interface, especially on soils which are compacted.

Chapter 4: Root Length and Width Determination by Digital Image Processing

- A) Washed root samples can be easily video recorded using the system described.
- B) The digital image processing system was not entirely successful in determining root length. Major problems were related to debris in the sample and the noise in the system from the video recording and image processing steps.

- C) Root width determination is only generally possible with the current system.
- D) The current system is time consuming.

The use of digital image processing for root studies holds much potential. The current system shows that the technology is available to carry out the task. However, much work remains to be accomplished prior to broad scale application to root length and width determination.

Chapter 5: The Effects of Flood and Drought on Dry Bean Root and Shoot Growth

- A) Flood and drought stress are detrimental to plant growth. Dry bean plants were not able to recover completely following subjection to flood or drought.
- B) Dry beans recovered more quickly following drought than flood.

The effects of these two environmental stresses are harmful to plant growth and are long lasting to the extent that yield is significantly impaired.