THE INFLUENCE OF BACTERIAL AND FUNGAL ISOLATES FROM THE RHIZOSPHERE OF TAMCOT CAMD-E ON HOST RESPONSE TO PHYMATOTRICHUM ROOT ROT OF COTTON

A Thesis

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ABSTRACT

The Influence of Bacterial and Fungal Isolates
From the Rhizosphere of Tamcot CAMD-E on Host Response
To Phymatotrichum Root Rot of Cotton (August 1984)
Gerard Raymond Lazo, B.S., Texas A&M University
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The Multi-Adversity Resistance (MAR) genetic improvement program has made significant progress in developing cotton cultivars with resistance to diseases and insects. Phymatotrichum root rot is one disease to which resistance is being gained. One hypothesis within the program is that host selected rhizosphere microorganisms play a role in resistance to soilborne pathogens. Cumulative results from several model studies imply that exudate properties of MAR cotton root surfaces have an influence in controlling the quality and quantity of microorganisms in the rhizosphere. Resistance to *Phymatotrichum omnivorum* appears to be influenced by such a mechanism. Investigations have been conducted over a three year period in greenhouse and field experiments to determine what effect microorganisms from MAR cottons have on resistance to root rot. Bacteria and fungi which were predominant in the seedling rhizosphere of the MAR variety Tamcot CAMD-E were isolated and used as treatments for cotton grown in *P. omnivorum* infested soils. The treatments were applied as a root drench prior to expected natural occurrence of symptoms and the number of dead plants was recorded weekly. Cotton in test plots were also harvested for yield. The microorganisms used in treatments involved four bacterial isolates all characterized in part as being rod shaped, asporogenous, aerobic, and non-fermenting. Three fungal isolates were also included. One was identified as *Fusarium solani* and the other two were identified as *F. oxysporum*. Several interactions appeared to have occurred, however few statistical differences were noted between treatments. Interactions occurred over experiments, time, location, cultivars, and treatments for both disease incidence and yield. Interpretations were based on the observance of consistent trends for certain treatments over experiments. The treatment that appeared most consistent in reducing the incidence of Phymatotrichum root rot was a combination consisting of a mixture of the bacterial isolate designated WA-E and the *Fusarium* species. This treatment was effective in both greenhouse and field experiments. The overall results suggest that key beneficial microorganisms appear cultivar dependent and may be under genetic influence of the host, and in this case, can influence the severity of Phymatotrichum root rot of cotton.

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INTRODUCTION

Phymatotrichum omnivorum (Shear) Duggar is a soilborne fungus that parasitizes cotton. In addition, it attacks 2,000 other dicotyledonous plants in the Southwestern U.S. and Mexico (74). In areas where Phymatotrichum root rot occurs, damage to cotton is costly. Although research efforts have focused on this problem since the late 1800's, presently there is no practical, effective means of control that is consistent. Attempts at control have involved fungicides, crop rotation, deep plowing, burial of organic matter, and escape (5,6). To date, no economically feasible fungicide has been developed, and current crop rotation strategies with a monocot are inconsistent in reducing the incidence of Phymatotrichum root rot. The cost of deep plowing, which can reduce the pathogen population in the plowed profile, limits its use. In addition the pathogen persists in zones too deep to reach with plows. The production and burial of organic matter is costly, but has been effective in increasing soil saprophytes which actively compete with the root rot pathogen, a persistent, but poor competitor with other soil microorganisms. The early planting of fast maturing cotton varieties is promising as an escape measure, allowing cotton bolls to mature before the the plant is parasitized and killed. Even though the above mentioned practices help to some degree, the problem of cotton root rot remains a burden to many cotton growers.

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In the study of interactions between plants and soilborne pathogens, several factors must be considered. The soil, especially that in close proximity to the plant root, is a dynamic system which in part is influenced by root exudates and microorganisms that occur there. Numerous accounts have associated the exudate and microbial biology to host plant resistance or susceptibility, to soil-inhabiting pathogens (18,20,31,63,72,75,76,77,86). Similarly, microorganisms are important in defining disease suppressing and disease inciting soils (1,15,51,53,55,68,70,71). It is considerations such as these that involve judgments concerning host response to environment and in management practices. In the case of Phymatotrichum root rot, studies suggest disease suppression is related to the presence of high microbial populations.

The multi-adversity resistance (MAR) cotton research program considers resistance to Phymatotrichum root rot among the many adversities for which resistance is being obtained (12). Adversities include fungal and bacterial pathogens, nematodes, insects, and environmental stresses. The assumptions that justify investigating a basis for broad spectrum MAR protection is that genetic change influences plant exudate chemistry, conditioning MAR cottons to selectively favor the predominance of symbiotic microorganisms which cause the host to resist damage caused by plant pathogens and insects. The term 'symbiotic' is used to refer to those microorganisms that are in and closely associated with the host which may be beneficial to the plant. Cause and effect models have been constructed which support the assumptions for root rot resistance (3,27,84,85). Additionally, results from preliminary field experiments, in which microorganisms from MAR cottons were applied as treatments, support this assumption.

The role of the cotton plant, as the host, has been given relatively little attention in research for controlling the root rot disease. In view of the importance of non-pathogenic microorganisms in disease interactions, it is important to investigate the potential role of the host in influencing a microbiological protective system that aids the cotton plant to resist damage. The purpose of this research is to determine if microorganisms isolated from MAR cotton root rhizospheres do influence host resistance to cotton root rot, and if so, to identify the more effective ones.

LITERATURE REVIEW

The Pathogen

The existence of P. omnivorum is restricted to a certain geographical domain dictated by soil type and environment. The fungus is found in soils that are generally alkaline, calcareous, montmorillite clays, and where optimum environmental conditions involve high temperatures and high soil moisture (52,74). This fungus is persistent in the soil, but it is not present in numbers as high as other common soil pathogens such as species of Rhizoctonia, Pythium, Verticillium, and Fusarium. P. omnivorum overwinters in the soil as sclerotia which germinate during optimal conditions, growing as hyphal strands through the soil and entwining descending cotton roots. After cotton fruit initiation, the fungus growth reaches the top of the root and kills the periderm, colonizing the inner root, occluding vascular flow, and causing rapid wilting and death of the plant (87). Observation of such root tissue reveals the presence of white to buff colored mycelial growth over the tap root surface.

The Nature of Disease

Several investigations have been conducted to resolve the nature and control of Phymatotrichum root rot. Evidence indicates a strong relationship between carbohydrate levels of the *plant* and microorganisms associated with the roots. Variation in carbohydrate content of cotton roots during host non-fruiting and fruiting stages and the existence of wet or dry conditions was shown to determine whether the plant was more resistant or susceptible to parasitism by P. omnivorum (33). A high amount of carbohydrate is demanded by the plant as it starts fruiting, reducing levels of carbohydrate in the root bark and subsequently altering the microbial ecology about the root. Higher concentrations of root carbohydrate with shifts in numbers of microorganisms on the roots have been considered in explaining why the host is more resistant during non-fruiting and during dry conditions. P. omnivorum is a poor competitor with other soil microorganisms, soon reappearing in their absence, and is generally nonspecific in its mode of parasitism, as demonstrated by its wide host range.

In contrast, young cotton plants containing low carbohydrate concentrations associated with the root system and grown in non-competitive, low microbial populated soils, were demonstrated to resist infection by P. omnivorum, where additions of carbohydrate to the plants increased susceptibility (16). Also infections of cotton seedlings, usually unaffected by P. omnivorum at that stage, were induced by germinating cotton seed on agar dishes containing high levels of carbohydrate.

Aside from nutritional studies, the presence of high microbial populations have been implicated with reductions of Phymatotrichum root rot. Microbes were demonstrated to strongly influence pathogen activity in tests where 'resistant' corn plants showed disease symptoms when grown in sterilized soil inoculated with P. omnivorum (15,81). Normally this pathogen has not been observed to parasitize monocotyledonous plants, which have commonly been grown side-by-side to susceptible carrot, okra, and cotton plants under non-sterile conditions (88). With soil amendments of green manures, notable increases in bacteria, actinomycete, and saprophytic fungi, were attributed to successful competition against the pathogen (44). It is believed the observed reductions in root rot, although slight, are the result of premature germination of overwintering propagules, which make hyphae susceptible to chemical decomposition and microbial competition (28).

In genetic studies, differences among cotton cultivars suggest that some resistance to P. omnivorum can be gained. After evaluating more than 30 cultivars, Goldsmith and Moore (39) reported variations in susceptibility to cotton root rot. By carrying forward the better cultivars and making crosses among them, improved levels of resistance were obtained. The controls maintained high levels of disease, emphasizing that genetic improvement was possible. However, the improvement was relatively slight and the basis of resistance was not determined.

The Role of Microorganisms Relative to Disease

Studies on different soybean cultivars have demonstrated differences in resistance to Pythium spp. and Fusarium spp., were dependent on the presence of higher phenolic levels in the resistant cultivars (49). In comparing species of plants resistant to P. omnivorum, the presence of alkaloids has been correlated with resistance, but the evidence was inconclusive (40). The nature of the rhizosphere environment influenced by the host may also favor colonization by certain microbes rather than inhibiting them. In the rhizosphere of nodulating and non-nodulating soybean lines, differences in total numbers of bacteria and differences in nutritional requirements of microbial isolates were found (34). Resistant lines of spring wheat may have a characteristic rhizosphere flora possessing a higher number of cellulolytic, amylolytic, and pectinolytic microorganisms than in susceptible lines (58,59). It has also been suggested that certain gene activating mechanisms may result in the selection of certain plant associated microorganisms which may influence a plant's susceptibility to disease. This selection may be determined by the action of root exudates upon saprophytic microflora (41,50).

In the rhizosphere, where soil is subject to influence by the root, complex and obvious microbial differences occur in root-free and root influenced soils (29,31,60,66,82,83). Application of exudates from paddy, wheat and moong crops, to remote soils, significantly increased bacterial and fungal populations in which the increase in fungi was less pronounced (57). Such work has suggested that certain kinds and types of microorganisms may predominate in the host rhizosphere, and are influenced by the particular host and its environment.

Soils can contain characteristic microbial populations that may play a role either in disease suppression or disease incitement. Examples of soil suppression of pathogens can be associated with 'antagonistic' effects of certain soil microbes; some commonly mentioned ones are species of Arthrobacter spp., Pseudomonas spp., and

Chromobacterium spp. (61). Experiments with Fusarium-wilt suppressing soils suggested that competition between saprophytic and pathogenic members of the soil microflora can play a role in disease suppression (64). Heat treatments of the suppressive soils resulted in a breakdown in the suppressing effect which was believed correlated with reductions in number of soil fungi, especially those of saprophytic Fusarium species. In nature Fusarium spp. may be related to a cross-protection mechanism, where non-pathogenic forms of the fungus can act to suppress disease incidence caused by the pathogenic forms (25,30). In experiments where alfalfa was inoculated with bacterial and fungal pathogens, non-pathogenic bacteria were influential in determining host reaction to pathogens, suggesting that non-pathogenic bacteria can be involved in disease suppression, similar to cross-protection by fungi (69).

Microbial interactions are important in that they may exploit antagonistic relationships between microflora symbiotic with the host and soilborne pathogens. In experiments on cotton seedling disease, certain fungicide treatments, in addition to reducing disease, increased bacterial populations of the soil (7,8,14). It was additionally observed that bacteria isolated from fungicide treated soils could be used as a treatment in place of a fungicide and provide reductions in seedling disease which were comparable to reductions caused by certain fungicide treatments. Conclusions from these data suggested that certain microbes were effective in reducing disease incidence by Rhizoctonia solani Kuehn, Pythium spp., and Thielaviopsis basicola (Berk. & Br.) Ferr..

It has been hypothesized that the rhizosphere of most plants, possess the microfloral members that can act antagonistically to any given pathogen (43,46,56). Seed treated with Pseudomonas fluorescens previously isolated from cotton rhizosphere, had a 50% reduction in damping-off caused by Rhizoctonia solani (42). In another series of experiments bacterial treatments of carnation were found to reduce the incidence of Fusarium wilt (54). Other modes of protection, such as growth regulation, induction, or competition have been shown to be involved in improved plant growth (2,17,20,35,48). Many of these concepts have been devised through bacterization studies (21,23,36,65,79,89). The addition of rhizobacteria, termed 'plant growth promoting bacteria' (PGPB) has resulted in better yields when applied to potato crops (26,46). Little is known about the mode of action and several theories have been postulated (45,46). The isolation of deleterious non-pathogenic bacteria has also been reported (78,80).

The MAR Hypothesis

The MAR program has been involved in developing cottons resistant to several adversities. Multi-adversity resistant varieties can be characterized as cold tolerant, fast maturing strains which have resistance to several diseases and insects. Methodology in genetic improvement has involved screening techniques designed to identify genes which account for levels of resistance to two or more pathogens (11). At present, management practices have used resistance and escape to reduce losses caused by Phymatotrichum root rot, with cold tolerant seedlings having resistance to seedling diseases which can reach boll maturity before Phymatotrichum can kill the plant. This program is now at the stage where increased resistance to Phymatotrichum is being attained. This resistance is related to a reduction in percentage of cotton plants killed. Several models, using multiple regression variable selection, have been constructed which predict how spermoplane, rhizosplane and rhizosphere components can be involved in multi-adversity resistance in cotton. Root exudates of cotton were found to influence rhizosphere bacteria and actinomycete populations, accounting for variability in disease severity (84,85). Further work demonstrated influences on bacterial and actinomycete populations at the rhizosphere-rhizoplane interface which occurred over the life-time of the plant (27). Resistance to Phymatotrichum was associated with higher eubacterial and lower actinomycete bacterial levels on the rhizoplane of 55 day old plants. Studies of ionic chemistry associated with cotton seed suggest that certain ionic metabolites are heritable among cottons, supporting the hypothesis that exudate composition can influence micoorganisms associated with the cotton plant (62).

Additional researchers have isolated symbiotic bacteria from phyllosphere plant tissue of improved MAR cottons. Certain of the bacterial isolates when applied as treatments have protected susceptible cottons from bacterial blight, boll weevil, Heliothis species, and Phymatotrichum root rot (4,13). These preliminary studies have prompted further research to determine if similar associations can be present in the rhizosphere, affecting resistance to soilborne pathogens.

Assuming that root exudate constituents of MAR cottons are under genetic control, and that types and kinds of microflora are influenced by this control, it is hypothesized that the rhizosphere-rhizoplane environment can be microbially altered to benefit the plant, causing a resistance to pathogen challenge. In an attempt to expand understandings related to the MAR hypothesis, this study will attempt to detect microbial interactions that will benefit the plant.

MATERIALS AND METHODS

The Host

Two MAR cotton cultivars, Tamcot CAMD-E and Tamcot SP21S, were used in field and greenhouse experiments (9,10). Both cultivars were developed in the multi-adversity resistance program and possess fast fruiting traits as well as resistance to several cotton adversities. Resistance to Phymatotrichum root rot is reported as ranging from partial to intermediate for the two cultivars.

The Pathogen

Sclerotia of *Phymatotrichum omnivorum* (Shear) Duggar were used as inoculum in greenhouse experiments. The sclerotia were grown in autoclaved soil culture, with some modifications to the original method used by Dunlap (32). Jars containing 250 g sterile Houston Black Clay soil were layered with wet, autoclaved sorghum seed (25 g) and 45 ml of sterile distilled water was added. Each soil was inoculated with a mycelial plug of *P. omnivorum* from which sclerotia formed after several weeks of incubation in the dark at a room temperature of 22 C. Sclerotia were collected by sieving the soil through a 1 mm mesh screen and separating them from soil particles and debris by suspending the mixture in a sucrose solution (1.25 Kg/L) and collecting the floating sclerotia. Sclerotia were 'surface sterilized' by rinsing with sodium hypochlorite (5.25 % in water), followed by a rinse with sterile distilled water.

In field experiments, the inoculum source consisted of naturally occurring propagules of P. omnivorum known to be present in the soil. Field experiments were conducted at two locations, Temple and McGregor Texas. They were carried out over a period of three years.

The Symbionts

Microorganisms isolated from a MAR cotton cultivar, Tamcot CAMD-E, were used. Bacteria were isolated from the rhizosphere of nine-day-old cotton seedlings. Isolations were made on soil extract agar containing in g/L: 0.5 K_2HPO_4, 1.0 glucose, 15.0 agar, 100.0 soil extract (filtrate from 1 Kg soil in 1 L water), 900.0 distilled water, and 0.0336 of cyclohexamide. A soil dilution technique was used for bacterial isolations. Determination of the predominant microorganisms was based on total numbers of colonies with similar color, morphology, and texture. The isolates were maintained in culture on potato carrot dextrose agar (PCDA: 0.2 g $CaCO_3$, 0.3 g $MgSO_4$, 0.5 g yeast extract, 2.5 g peptone, 10.0 g agar, 40.0 g potato dextrose agar, 15.0 ml commercial carrot juice, in 1000.0 ml distilled water). The bacteria were given tentative laboratory designations based on colony appearance on soil extract agar. The fungi collected were commonly isolated from seedling root tissue plated on water agar.

Characterization

Characterization of the bacterial and fungal isolates was performed using several standard microbiological techniques (24,37,38,73). Tentative identifications of the cotton rhizosphere microbes were assigned based on information obtained.

Treatments

All experiments in the greenhouse and field consisted of at least ten treatments using the four bacteria and three fungi isolated from Tamcot CAMD-E seedlings. The three fungi were included in a treatment as a combined mixture. Each bacterial isolate and fungal mixture was applied singularly as a treatment and as bacterial-fungal mixtures. Microbial treatments consisted of distilled water suspensions of five-day-old bacterial cultures and seven-day-old fungal mycelial and spore mats grown on PCDA. Fifteen milliliter aliquots of the suspensions were added as treatments for plants. The concentrations of the bacteria and fungi were 10^7 to 10^9 bacteria/ml and 10^6 propagules/ml, respectively. Concentrations were determined using a cell counting chamber. A water-only control was included as a treatment in each experiment.

Greenhouse Experiments

The cotton cultivar, Tamcot CAMD-E, was grown as the host plant and the pathogen consisted of P. omnivorum sclerotia. The bacterial and fungal treatments were applied through tubes positioned in the soil next to each plant.

The experiments were set up with two plants in each greenhouse pot. Treatments were applied for each plant by adding microbial suspensions into 2 cm wide poly-vinyl chloride (PVC) tubes positioned next to each plant. Each 20 cm long tube was buried 16 cm deep and had drilled openings directed toward the plant roots. The tubes were stoppered at the top 4 cm from the soil line and closed at the bottom. In the center of each pot a 1.8 x 15 cm test tube was buried 8 cm deep. To inoculate each pot the test tube was removed; sclerotia of P. omnivorum were dropped down the opening and the holes were then filled with soil.

The treatments were applied in 15 ml amounts to each tube situated next to each plant. Plants were watered daily, supplimented with Hoagland's nutrient solution weekly, and sprayed for insects as needed. Each experiment consisted of 5-6 replications organized in a randomized block design on greenhouse benches. Data collection consisted of recording disease progression by plant death as a function of time and determining seed cotton yield.

Experiment One

Three applications were made to cotton plants utilizing the ten microbial treatments descibed. Applications of treatments were made at the time of planting, and 10 days and 46 days after planting. Six randomized replications were used for each of the ten treatments. In this experiment, the soil substrate consisted of a 50% mixture of Houston Black clay in sand, which was inoculated with 0.2-0.3 g of P. omnivorum sclerotia on the day of planting. Rainwater was used for watering the plants which were harvested 140 days after planting. Control of spider mites was implimented with use of Orthene.

Experiment Two

Results obtained with treatments of the previous greenhouse experiment and one field experiment justified additional treatments being incorporated into this greenhouse test. The treatments used involved the ten original ones and fourteen additional treatments involving several combinations of the four bacteria and fungal isolates. Treatments were applied to plants in greenhouse pots containing 100% Houston Black clay, conducive to the growth of P. omnivorum. Only two treatment applications were employed, one 15 days and the other 36 days after planting. An initial treatment at the day of planting was excluded because previous evidence suggested low seedling tolerance to high microbial concentrations at that stage of development. The soil was inoculated with 0.2-0.3 g sclerotia 15 days after planting and heating coils were placed amongst the greenhouse pots to stabilize temperatures around 28 C. A grading system was used for the measurement of disease progression in the greenhouse. Symptoms were scored as 0) no symptoms, 1) leaf discoloration, 2) drooping of the top leaves, 3) drooping of remaining leaves, 4) initiation of dryness in leaves, and 6) a dead plant. The recording of symptoms were obtained over several time intervals in the course of the experiment. Plants were watered with rainwater and distilled water and harvested 120 days after planting. Orthene and Phlectan were used to control spider mites in the greenhouse.

Field Experiments

Cotton was grown under non-irrigated field conditions in Phymatotrichum root rot nurseries at Temple and McGregor, Texas. Each test site had a history of cotton root rot and had been used successfully for root rot studies in previous years. Experiments were conducted in a randomized block design with six replications for each treatment at each location. Each 27.4 m cotton plot was thinned to allow 38 to 46 cm between each plant. The treatments used in the study again utilized the bacterial and fungal isolates obtained from Tamcot CAMD-E seedlings, and included a water-only control.

Treatments were administered by a root drench technique using a 15 ml microbial suspension applied with a hand-held sprayer to the plant base. This treatment effectively drenched the upper root portions situated between cracks in the soil. Measurment of disease incidence was obtained by recording counts of dead plants over a seven week period. Final lint yield per acre was obtained by making two stratified harvests.

Experiment One

Two MAR cottons, Tamcot CAMD-E and Tamcot SP21S, were planted during the 1980 growing season. Ten treatments were used, each was applied 51 and 73 days after planting. Stratified harvests were obtained at the Temple nursery 134 and 150 days after planting. Only one harvest was obtained at the McGregor location 150 days after planting.

Experiment Two

Several cultivar interactions occurred in Experiment One. However, insufficient information was obtained to justify an analysis of cultivar-treatment interactions. During the 1981 growing season one cultivar, Tamcot CAMD-E, was used to measure the influence of the microbial isolates. Treatments were applied 68 and 97 days after planting. Harvest dates were 146 and 161 days after planting at the Temple location and 161 days at the McGregor location.

Experiment Three

Results from previous experiments appeared to indicate that certain bacteria and fungi acted favorably to reduce root rot of cotton when applied as a drench treatment. To test the extent of this effect, an investigation was conducted to study cultivar-microbe interaction. From previous experiments, one bacterial isolate (WA-E) and one fungal isolate (Fu-10) showed promise in controlling root rot. Another bacterial isolate (SWO) had been used successfully in simulating host resistance to cotton adversities, such as root rot, in previous experiments. These microorganisms were used in treatments applied to cotton in this experiment (4,13). Treatments consisted of various combinations of the two bacteria and one fungal isolate. A water-only control was included. Cotton cultivars, Tamcot CAMD-E and Tamcot SP21S, were used. Treatments were applied 61 and 82 days after planting and harvested 119 and 139 days after planting at Temple and 138 days at McGregor.

RESULTS

The Symbionts

Several microbial isolates were obtained from the rhizosphere of a MAR cotton, Tamcot CAMD-E. The isolates were used as biological control treatments in greenhouse and field experiments and each isolate used occurred frequently in root-soil dilutions plated on soil extract agar. The microbial isolates selected for use as treatments were based on their predominant occurrence on soil extract agar as identified by color and texture of the colonies. The representative microbial isolates from Tamcot CAMD-E consisted of four bacteria and three fungi. The designations for the bacteria were RW-E (rough white), SW-E (smooth white), T-E (translucent), and WA-E (waxy), and the fungal designations were Fu-2, Fu-5, and Fu-10.

Bacterial Characterizations

Several standard microbiological techniques and biochemical tests were used in an attempt to identify the bacterial isolates (Table 1). Based on the results, no conclusive identity could be assigned to any of the bacterial isolates. Characteristically all isolates were asporogenous, aerobic, non-fermenting bacteria. The SW-E, T-E and WA-E isolates were Gram negative and RW-E was Gram positive.

Table 1. Characterization of the bacterial isolates from the rhizosphere of Tamcot CAMD-E seedlings.

Test	Bacterial observations[1]			
	WA-E	T-E	RW-E	SW-E
Gram stain	-	-	+	-
Shape	rod	rod	rod	rod
Length	3-7 µm	3-5 µm	2.5-5 µm	1.5-2 µm
Diameter	0.4 µm	0.6 µm	0.5 µm	0.5 µm
Colony description	beige waxy	translucent mucoid	beige rough	white mucoid
Oxidase	-	-	+	+
Catalase	+	+	-	-
Simmon's Citrate	-	-	-	-
Methyl red, 22 C	-	-	-	-
Vogues-Proskauer, 22 C	-	-	-	-
$NO_3 \rightarrow NO_2$	-	-	-	-
$NO_2 \rightarrow N_2$	-	-	-	-
OF glucose	nr	nr	nr	nr
OF sucrose	nr	nr	nr	nr
OF arabinose	nr	nr	nr	nr
OF mannitol	nr	nr	nr	nr
OF lactose	nr	nr	nr	nr
TSIA glucose	nr	nr	nr	nr
TSIA lactose, sucrose	nr	nr	nr	nr
TSIA H_2S	-	-	-	-
TSIA gas	-	-	-	-
Gelatin hydrolysis	-	-	-	-
Starch hydrolysis	w	+	-	-
MIO motility	-	+	-	+
MIO indole	-	-	-	-
MIO ornithine decarboxylase	-	-	-	-
Lysine decarboxylase	-	-	-	-
Tryptophan deaminase	-	-	-	-
Arginine dehydrolase	-	-	-	-
Urease	w	-	-	+
DNase	+	-	-	-
ONPG	+	+	+	+
YEA-congo red-mannitol	red	no uptake	maroon	slow uptake
Rhizobium medium	ng	ng	ng	heavy growth
Utilization:				
Arabinose or Inositol	+	+	+	+
Glucose or Sucrose	+	+	-	-
Ethanol or Sorbitol	+	-	-	-
Melibiose or Amygdalin	+	+	-	+
Rhamnose	-	+	-	-

[1] Abbreviations: nr = no reaction, ng = no growth, w = weak reaction, + = positive reaction, - = negative reaction.

RW-E Bacterium

The RW-E bacterium grew slowly on PCDA, forming small cream colored, rough textured colonies. Short irregular rods, sometimes curved, were formed and variable Gram stains were sometimes encountered. Based on information obtained, this isolate is similar to that of the Coryneform group of bacteria, belonging to the section of non-pathogenic Corynebacteria as described in Bergy's manual (24). The non-pathogenic section contains many of these microbes which are insufficiently characterized to permit differentiation to species. The isolate did conform to some Coryneform descriptions, but did not fit into any well documented group.

SW-E Bacterium

The SW-E bacterium grew well on nutrient rich PCDA medium, forming a mucoid slime in copious amounts over time. The bacterium grew as circular colonies which were smooth and white in texture and color. After one week incubation at room temperature, some color clearing was observed in the colonies, a characteristic similar to that of some Rhizobium species. The isolate was observed to be motile and grew well on selective medium for Rhizobium. When grown on a mannitol yeast extract-congo red agar and incubated in darkness, little uptake of congo red was observed. The rate of growth of the bacterium on a mannitol medium would place the microorganism among the 'fast growing' group. Because this isolate was not obtained from root nodules, it would be difficult to characterize it as Rhizobium, for several leguminous hosts would need to be screened to detect any nodulating ability. However, this bacterium did fit some characteristics related to this genus.

T-E Bacterium

Growth of the T-E bacterium on PCDA was a translucent, mucoid slime that formed sparse irregularly shaped colonies. The bacterial colonies tended to dry out on the media when kept more than a week, and was difficult to maintain in culture. The bacterium was motile as determined by the hanging drop tehnique and use of motility medium. Motility appeared to be of the gliding type. Attempts to demonstrate the presence of flagella by staining were unsuccessful. The T-E bacterium had rather large cylindrical cells with blunt rounded edges, and cells sometimes appeared in pairs. The colony characteristics fit some descriptions of colonies formed by the gliding bacteria and that of the Azotobacteraceae. No observable fruiting structures or bright colored fluorescent pigment was observed, common to that of some gliding bacteria. The bacterium grew well on nitrogen deficient medium, but no cyst formation associated with Azotobacteraceae was observed, and many of the characteristics did not conform to the genera descriptions. This bacterium is very similar to some microbes described for the gliding bacteria group or Flavobacterium, but this microbe does not produce any of the characteristic color pigments as described for either of these groups.

WA-E Bacterium

Grown on PCDA, WA-E formed smooth, circular beige to tan colonies. As colony growth progressed, notable browning of the medium occurred. Staining and microscopic observation of colonies on semi-solid, nutrient poor medium demonstrated chains of rods within filamentous structures, having occasional branching. A sheath was observed to be present, although it was not common. The noted sheath and the discoloration seen in the glucose-peptone based medium may conform to some characteristics described for the genera _Leptothrix_ or _Sphaerotilus_. No motility or incrustation by ferric and manganic oxides were observed, suggesting the microbe may be related to _Sphaerotilus_.

SWO Bacterium

This bacterium was previously isolated from cotton leaves of Tamcot CAMD-E and has been implicated as being a beneficial 'symbiont' in simulating resistance to _Xanthomonas_ _campestris_ pv. _malvacearum_ (Smith) Dye, seedling disease, root rot, and boll weevil (4,13). Grown on PCDA, good colony growth was seen after 24 hours. Colonies were smooth and white, being convex with entire edges. Gram stains demonstrated Gram positive short plump bacilli (3.0 x 1.1 μm). Endospores were also visible. The SWO bacterium was oxidase negative, catalase positive, could hydrolyze gelatin, and had weak urease activity. This bacterial isolate is believed to be an aerobic _Bacillus_.

Fungal Characterization

The three fungal isolates were identified as belonging to the genus Fusarium. The isolates Fu-2 and Fu-5 were identified as F. oxysporum (Schlecht) Snyd.&Hans. and Fu-10 was identified as F. solani (Mart.)(Appel&Wr.) Snyd.&Hans.. These designations were verified by R. G. Davis, plant pathologist at Mississippi Agricultural Experiment Station, Stoneville, Mississippi, C. E. Windel, scientist at the University of Minnesota, and R. A. Taber, research scientist at the Texas Agricultural Experiment Station, College Station.

Greenhouse Experiments

Experiment One

Phymatotrichum root rot disease symptoms first appeared 105 days after inoculation, occurring later than expected and when greenhouse temperatures were higher than optimal conditions. The incidence of root rot averaged 23 percent, with the water-only treated plants averaging a higher disease incidence compared to other treated plants. The treatment causing the lowest incidence of root rot was RW-E+Fu-E, with only one plant killed. The occurrence of root rot appeared to be delayed by treatments treated with T-E, Fu-E and T-E+Fu-E (Table 2). Plants with the water-only treatment had the highest incidence of disease, and also had the lowest seed cotton yield; but the RW-E+Fu-E treated plants had the lowest incidence of root rot and one of the lowest yields (Table 3). Trends apparent in this experiment suggested that the microbial treatments were effective in reducing the incidence of root rot. However, yield and disease incidence were not closely associated. Based on the rate and occurrence of disease symptoms, the soil-sand mixture used was the probable cause of a low occurrence of disease.

Table 2. The effect of treatments with symbiotic microorganisms on the incidence of dead plants for greenhouse Experiment One.

Treatment number and microorganism(s)	Number of dead plants for days after inoculation[1]					
	105	110	115	120	125	130
	#	#	#	#	#	#
1 Control	0	3	3	4	4	5
2 T-E	0	0	0	0	2	3
3 SW-E	1	1	1	2	2	2
4 WA-E	1	1	1	3	3	3
5 RW-E	0	1	1	1	2	3
6 Fu-E	0	0	0	0	1	3
7 T-E+Fu-E	0	0	0	2	3	3
8 SW-E+Fu-E	0	1	1	1	2	2
9 WA-E+Fu-E	2	2	2	2	3	3
10 RW-E+Fu-E	0	1	1	1	1	1

[1] A total of 12 plants were inoculated for each treatment. The experiment had an average of 23 % incidence of dead plants.

Experiment Two

A 66 percent incidence of root rot occurred for Experiment Two. The added treatments included in this experiment allowed for the observation of additional microbial interactions. The treatments

Table 3. Dead plants and seed cotton yield for Experiment One in the greenhouse testing the effects of microbial treatments on Phymatotrichum root rot.

Treatment number and microorganism(s)	Dead plants[1]		Seed cotton yield	
	%		g	
5 RW-E	25.0	ab[2]	5.80	a[2]
4 WA-E	25.0	ab	5.57	ab
8 SW-E+Fu-E	16.7	ab	5.56	ab
3 SW-E	16.7	ab	5.22	abc
7 T-E+Fu-E	25.0	ab	5.01	abc
9 WA-E+Fu-E	25.0	ab	5.01	abc
6 Fu-E	25.0	ab	4.91	abc
10 RW-E+Fu-E	8.3	a	4.54	bc
2 T-E	25.0	ab	4.52	bc
1 Control	41.6	b	4.31	c
Average	23.3		5.04	
C.V.%	214.6		28.60	

[1] A total of 12 plants were included in each treatment.

[2] Averages not followed by the same letter are different according to Duncan's test for the 10 % level of significance.

which appeared most effective in reducing disease (Table 4), without reducing yield (Table 5) were the WA-E+Fu-E and T-E treatments for which no plants were killed. Disease symptoms did occur in plants with these treatments late in the experiment, based on the grading scale used (Table 6). The treatments associated with death of most plants appeared to involve combinations of the SW-E and RW-E bacteria and also the WA-E+RW-E+Fu-E treatment. Plants treated with water-only were intermediate for disease and yield, suggesting some treatments may have caused the host to become more susceptible. Some of the treatments appeared to be more effective in reducing root rot and improving plant yield. Some of the favorable treatments from Experiment One consisted of WA-E+Fu-E, SW-E, WA-E and T-E.

Table 4. Dead plants over time for Experiment Two in the greenhouse testing the effects of microbial treatments on Phymatotrichum root rot.

Treatment number and microorganism(s)	Number of dead plants for days after inoculation[1]					
	50	60	70	80	90	100
	#	#	#	#	#	#
1 Control	0	0	1	2	2	3
2 T-E	0	0	0	0	0	0
3 SW-E	0	0	0	0	0	1
4 WA-E	0	0	0	0	0	0
5 RW-E	0	2	2	4	4	4
6 Fu-E	0	0	0	1	1	1
7 T-E+Fu-E	0	1	2	4	5	5
8 SW-E+Fu-E	0	0	2	3	4	4
9 WA-E+Fu-E	0	0	0	0	0	0
10 RW-E+Fu-E	0	0	2	2	4	4
11 T-E +SW-E	0	0	1	2	4	5
12 T-E +WA-E	1	1	2	2	2	2
13 T-E +RW-E	0	0	0	0	0	0
14 SW-E+WA-E	0	2	4	4	4	4
15 SW-E+RW-E	1	5	6	6	7	8
16 WA-E+RW-E	0	0	1	2	2	2
17 T-E +SW-E+Fu-E	0	0	1	2	2	2
18 T-E +WA-E+Fu-E	0	1	2	2	2	2
19 T-E +RW-E+Fu-E	0	3	3	3	3	3
20 SW-E+WA-E+Fu-E	0	4	4	4	4	4
21 SW-E+RW-E+Fu-E	0	2	4	5	6	7
22 WA-E+RW-E+Fu-E	1	4	7	7	8	10
23 T-E +SW-E+WA-E+RW-E	2	3	3	4	4	4
24 T-E +SW-E+WA-E+RW-E+Fu-E	0	0	1	2	2	3

[1] A total of 10 plants were inoculated for each treatment. The experiment had an average of 66 % incidence of plant death.

The treatments applied in greenhouse Experiment One demonstrated a trend of lowering disease incidence and increasing yield. This did not hold true for all treatments in this experiment. The application

Table 5. Yield for Experiment Two in the greenhouse testing effects of microbial treatments on Phymatotrichum root rot.

Treatment number and microorganism(s)	Stratified yield of seed cotton[1]		
	1st harvest	2nd harvest	Total
	g	g	g
13 T-E +RW-E	44.5	3.0	47.5
9 WA-E+Fu-E	28.0	19.0	47.0
4 WA-E	28.0	10.5	38.5
24 T-E +SW-E+WA-E+RW-E+Fu-E	35.0	0.0	35.0
2 T-E	20.5	14.5	35.0
17 T-E +SW-E+Fu-E	29.0	5.0	34.0
16 WA-E+RW-E	21.0	12.0	33.0
3 SW-E	30.5	0.0	30.5
18 T-E +WA-E+Fu-E	30.0	0.0	30.0
14 SW-E+WA-E	25.0	3.0	28.0
1 Control	15.0	13.0	28.0
19 T-E +RW-E+Fu-E	21.0	6.0	27.0
12 T-E +WA-E	19.5	7.0	26.5
6 Fu-E	9.0	17.0	26.0
11 T-E +SW-E	21.0	3.5	24.5
7 T-E+Fu-E	17.5	6.0	23.5
10 RW-E+Fu-E	15.5	6.5	22.0
5 RW-E	13.0	9.0	22.0
8 SW-E+Fu-E	12.5	9.5	22.0
23 T-E +SW-E+WA-E+RW-E	14.5	5.0	19.5
20 SW-E+WA-E+Fu-E	11.0	6.5	17.5
22 WA-E+RW-E+Fu-E	12.0	0.0	12.0
21 SW-E+RW-E+Fu-E	11.5	0.0	11.5
15 SW-E+RW-E	6.0	4.0	10.0
Average	20.4	6.7	27.1

[1] Total weight is from 10 plants for each treatment. No analysis of variance was done because seed cotton weights are pooled.

of treatments T-E+Fu-E, RW-E+Fu-E, RW-E, and SW-E+Fu-E did not result in a reduction of cotton root rot or higher yield compared to the water-only treatment (Tables 4 and 5). Although plants treated with Fu-E had less disease than the water-only treatment, the yield was also less.

Table 6. Grades for cotton root rot severity in greenhouse Experiment Two for cotton plants treated with rhizosphere microbial treatments.

Treatment number and microorganism(s)	Disease grades by days after inoculation[1]			
	75	82	89	97
9 WA-E+Fu-E	0.0 a[2]	0.0 a[2]	0.0 a[2]	0.0 a[2]
2 T-E	0.0 a	0.0 a	0.0 a	0.0 a
4 WA-E	0.0 a	0.0 a	0.0 a	0.4 ab
3 SW-E	0.0 a	0.0 a	0.0 a	0.5 ab
13 T-E +RW-E	0.0 a	0.0 a	0.0 a	0.8 b
6 Fu-E	0.5 ab	0.5 ab	0.8 b	1.0 b
17 T-E +SW-E+Fu-E	1.0 bc	1.0 b	1.0 b	1.0 b
16 WA-E+RW-E	1.0 bc	1.0 b	1.0 b	1.0 b
18 T-E +WA-E+Fu-E	1.0 bc	1.0 b	1.0 b	1.0 b
12 T-E +WA-E	1.0 bc	1.0 b	1.0 b	1.0 b
24 T-E +SW-E+WA-E+RW-E+Fu-E	1.0 bc	1.0 b	1.2 b	1.8 c
19 T-E +RW-E+Fu-E	1.9 d	1.9 c	1.9 cd	1.9 c
1 Control	0.5 ab	0.5 ab	1.4 bc	2.0 c
8 SW-E+Fu-E	1.5 cd	1.8 c	2.0 cd	2.0 c
5 RW-E	1.9 d	2.0 c	2.0 cd	2.0 c
14 SW-E+WA-E	2.0 d	2.0 c	2.0 cd	2.0 c
20 SW-E+WA-E+Fu-E	2.0 d	2.0 c	2.0 cd	2.0 c
23 T-E +SW-E+WA-E+RW-E	2.0 d	2.0 c	2.0 cd	2.0 c
10 RW-E+Fu-E	1.8 d	2.4 c	2.4 d	2.4 c
11 T-E +SW-E	1.4 cd	2.0 c	2.0 cd	2.5 c
7 T-E+Fu-E	2.0 d	2.4 c	2.5 d	2.5 c
21 SW-E+RW-E+Fu-E	3.0 e	3.6 d	3.7 e	3.9 d
15 SW-E+RW-E	3.3 e	3.5 d	3.5 e	4.0 d
22 WA-E+RW-E+Fu-E	3.7 e	4.0 d	4.3 f	5.0 e
Average	1.4	1.5	1.6	1.8
C.V.%	68.9	57.0	48.7	46.2

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[1] Grading system: 0 = no symptoms, 1 = leaf discoloration, 2 = drooping of top leaves, 3 = drooping of all leaves, 4 = leaves begin to dry, 5 = dead plant. A total of 10 plants were inoculated for each treatment. The experiment averaged a 66 % incidence of plant death.

[2] Averages not followed by the same letter are different according to Duncan's test for the 10 % level of significance.

Field Experiments

Experiment One

Statistically significant differences between treatments for percentage root rot appeared to occur during the first few weeks after application of treatments which was during the early developmental period of disease expression. Plants at the McGregor location had a higher incidence of root rot than at Temple, averaging 13 percent for all treatments in the experiment. None of the treatment interactions were significant over locations and cultivars for dead plants, but some of the bacterial and/or fungal treatments were associated with less root rot, based on the final average of diseased plants and in comparison to the water-only treatment.

At the Temple location the treatments on Tamcot CAMD-E which resulted in less root rot over time were WA-E+Fu-E and T-E+Fu-E (Table 7). The treatments WA-E+Fu-E and T-E+Fu-E were consistent in allowing less dead plants than the control for the entire period of disease development. However, all of the treated plants had less disease than the control during the first week. The plants treated with T-E, SW-E, and SW-E+Fu-E were intermediate in disease incidence for the seventh week of disease development. Treatments that appeared to give initial reductions in plant disease relative to the control were Fu-E, RW-E, and WA-E, but over time the effect diminished. The RW-E+Fu-E treated plants had more dead plants than the control every week after initial appearance of root rot in the nursery.

Table 7. Plants with Phymatotrichum root rot in field Experiment One at Temple for microbial treatments applied to Tamcot CAMD-E.

Treatment number and microorganism(s)	Dead plants by days after planting and weeks after initial root rot symptoms[1]						
	79 1	86 2	93 3	100 4	106 5	114 6	121 7
	%	%	%	%	%	%	%
9 WA-E+Fu-E	*0.0	*2.2	*2.2	*2.2	*2.2	*5.6	*5.6
7 T-E+Fu-E	*1.5	*1.5	*1.5	*2.6	*4.8	*7.0	*7.0
2 T-E	*3.3	4.4	*4.4	*5.5	*5.5	*5.5	*6.6
8 SW-E+Fu-E	*1.1	4.4	5.6	*5.6	*5.6	*6.6	*6.6
1 Control	3.6	3.6	4.6	5.7	5.7	9.3	9.3
10 RW-E+Fu-E	*1.2	5.8	5.8	5.8	10.6	11.8	11.8
3 SW-E	*2.1	4.3	*4.3	6.7	7.8	*7.8	*8.9
6 Fu-E	*0.0	*0.9	*4.1	7.2	10.0	10.9	10.9
4 WA-E	*0.0	*1.3	*3.6	7.7	9.9	12.2	12.2
5 RW-E	*1.0	*2.1	*4.0	8.2	9.3	11.4	11.4
Average	1.4	3.1	4.0	5.7	7.1	8.8	9.0
C.V.%	284.7	194.6	148.7	107.6	111.1	100.5	102.4

[1] None of the averages are different according to Duncan's test at the 10 % level of significance. Values preceeded by an asterisk (*) signify a reduced occurrence of root rot with respect to the control.

At McGregor the treatments on Tamcot CAMD-E plants which tended to reduce root rot were not effective until the second or third week after the appearance of initial disease symptoms (Table 8). In this test, once a reducing trend began, it remained so for the duration of disease development. Cotton plants treated with WA-E, Fu-E, and WA-E+Fu-E were involved with low occurrences of disease, while those treated with the SW-E bacterium had the higher levels of disease. Again, as at Temple, plants treated with WA-E+Fu-E and T-E+Fu-E resulted in reduced root rot at McGregor.

Table 8. Plants with Phymatotrichum root rot in field Experiment One at McGregor for microbial treatments applied to Tamcot CAMD-E.

Treatment number and micoorganism(s)	Dead plants by days after planting and weeks after initial root rot symptoms[1]						
	79	86	93	100	106	114	121
	1	2	3	4	5	6	7
	%	%	%	%	%	%	%
4 WA-E	0.0 a	*3.5	*3.5	*8.2	*9.2	*11.1	*11.1
6 Fu-E	1.0 ab	*1.0	*5.7	*10.4	*10.4	*10.4	*10.4
7 T-E+Fu-E	1.2 ab	8.3	*9.5	*10.7	*10.7	*11.9	*13.1
9 WA-E+Fu-E	4.5 ab	5.6	*9.2	*10.8	*11.9	*11.9	*11.9
10 RW-E+Fu-E	0.0 a	7.1	*8.1	*11.4	*11.4	*11.4	*12.6
1 Control	0.0 a	5.2	9.6	14.8	17.4	17.4	17.4
5 RW-E	1.1 ab	7.0	15.7	19.4	21.6	21.6	21.6
8 SW-E+Fu-E	1.4 ab	8.3	16.7	19.7	22.9	24.1	24.1
3 SW-E	0.0 a	9.3	16.8	19.7	19.7	20.6	21.6
2 T-E	4.4 b	6.6	17.9	21.2	23.4	25.6	25.6
Average	1.4	6.2	11.3	14.6	15.9	16.6	16.9
C.V.%	239.5	134.7	117.0	107.1	101.9	98.1	98.5

[1] Averages not followed by the same letter are different according to Duncan's test for the 10 % level of significance. Values preceeded by an asterisk (*) signify a reduced occurrence of root rot with respect to the control.

On Tamcot SP21S, significant differences occurred among treatments during the first week of disease counts at both locations. The treatment RW-E+Fu-E was effective in reducing the incidence of root rot at both locations. At Temple the treatments that were most effective on Tamcot SP21S were RW-E and RW-E+Fu-E, with the latter treatment also being effective at McGregor (Tables 9 and 10). At McGregor, the better treatments consisted of WA-E and WA-E+Fu-E. The plants treated with SW-E and SW-E+Fu-E had a low occurrence of root rot over time at McGregor, but had reduced incidence of root rot only during the second to fourth weeks at Temple. Most of the treatments had a beneficial effect on Tamcot SP21S at Temple, but at McGregor there appeared a division into effective and non-effective treatments. At McGregor, non-effective treatments resulted in reductions in disease only during the first few weeks. The treatment RW-E did not reduce root rot during any week of the test at McGregor.

Table 9. Plants with Phymatotrichum root rot in field Experiment One at Temple for microbial treatments applied to Tamcot SP21S.

Treatment number and microorganism(s)	Dead plants by days after planting and weeks after initial root rot symptoms[1]						
	79 1	86 2	93 3	100 4	106 5	114 6	121 7
	%	%	%	%	%	%	%
10 RW-E+Fu-E	0.0 a	*2.1 a	*2.1	*3.2	*3.2	*4.2	*4.2
5 RW-E	2.4 ab	*3.5 ab	*3.5	*3.5	*3.5	*4.6	*4.6
6 Fu-E	0.9 ab	*2.0 a	*2.9	*3.7	*5.5	*8.9	*11.0
8 SW-E+Fu-E	2.4 ab	*2.4 a	*2.4	*5.0	*7.7	14.7	14.7
2 T-E	2.2 ab	4.8 ab	*6.1	*6.1	*7.0	*9.2	*9.2
4 WA-E	1.2 ab	*3.5 ab	*3.5	*6.8	*6.8	*10.2	*10.2
7 T-E+Fu-E	2.0 ab	*3.0 ab	*4.0	*6.8	*6.8	*8.8	*8.8
3 SW-E	2.2 ab	*3.4 ab	*5.7	*8.0	*9.2	12.6	12.6
1 Control	0.0 a	4.5 ab	7.8	8.9	9.9	11.2	12.3
9 WA-E+Fu-E	4.7 b	8.1 b	8.1	9.4	*9.4	*9.4	*9.4
Average	1.8	3.7	4.6	6.1	6.9	9.4	9.7
C.V.%	195.3	132.3	136.9	128.2	129.8	125.9	124.5

[1] Averages not followed by the same letter are different according to Duncan's test for the 10 % level of significance. Values preceeded by an asterisk (*) signify a reduced occurrence of root rot with respect to the control.

The only significant interaction in the test was that of cultivar-location for lint yield per acre. Plants of the cultivar Tamcot SP21S had a higher lint yield at both locations in comparison with those of Tamcot CAMD-E. At McGregor for both varieties, there

Table 10. Plants with Phymatotrichum root rot in field Experiment One at McGregor for microbial treatments applied to Tamcot SP21S.

Treatment number and microorganism(s)	Dead plants by days after planting and weeks after initial root rot symptoms[1]						
	79	86	93	100	106	114	121
	1	2	3	4	5	6	7
	%	%	%	%	%	%	%
8 SW-E+Fu-E	1.4 ab	*1.4	*4.1	*5.4	*8.7	*8.7	*10.1
4 WA-E	*0.0 a	*1.3	*6.1	*6.1	*7.4	*8.6	*8.6
9 WA-E+Fu-E	*0.9 ab	*2.3	*4.4	*7.0	*7.0	*7.0	*7.0
10 RW-E+Fu-E	*0.0 a	*1.1	*7.7	*9.9	*9.9	*13.1	*13.1
3 SW-E	*0.0 a	*1.5	*6.4	*10.9	*11.8	*12.7	*13.7
1 Control	1.4 ab	6.1	14.5	14.5	15.5	15.5	15.5
2 T-E	*0.0 a	*4.9	*12.0	17.0	17.0	17.0	18.0
7 T-E+Fu-E	2.3 b	*4.2	*14.4	17.6	19.7	19.7	20.6
6 Fu-E	*0.0 a	11.1	*13.8	19.4	19.4	19.4	19.4
5 RW-E	*0.0 a	8.3	16.5	21.9	24.5	24.5	24.5
Average	0.6	4.2	10.0	13.0	14.1	14.6	15.0
C.V.%	333.2	207.7	120.2	115.2	110.3	110.1	109.4

[1] Averages not followed by the same letter are different according to Duncan's test for the 10 % level of significance. Values preceeded by an asterisk (*) signify a reduced occurrence of root rot with respect to the control.

were significant differences for yield among treated plants (Table 11). The T-E treatment resulted in higher yields for SP21S plants at both locations. Tamcot CAMD-E plants treated with WA-E averaged significantly higher lint yields at McGregor than those of other treatments. At Temple, all treatments were effective in improving yield for CAMD-E. However, only plants treated with RW-E+Fu-E, T-E, and Fu-E had higher yields than the control for SP21S (Table 11). Only plants treated with WA-E+Fu-E were consistent in having reduced disease at both locations on both cultivars. However, this led to no advantage in yield.

Table 11. Lint yield for field Experiment One for testing the effects of microbial treatments on Phymatotrichum root rot.

Treatment number and microorganism(s)	Lint yield per acre[1]				
	Temple		McGregor		
	CAMD-E	SP21S	CAMD-E	SP21S	Average
	lbs.	lbs.	lbs.	lbs.	lbs.
2 T-E	198.3	287.7	87.1 bc	117.1 a	172.7
10 RW-E+Fu-E	175.2	288.4	101.8 abc	107.4 ab	168.2
7 T-E+Fu-E	196.9	234.4	98.9 abc	108.2 ab	159.6
3 SW-E	187.5	234.8	107.5 ab	104.5 ab	158.7
4 WA-E	193.4	189.1	122.8 a	97.0 ab	150.6
6 Fu-E	158.0	281.7	93.4 abc	63.9 b	149.2
5 RW-E	215.9	225.9	73.5 c	76.1 ab	147.8
9 WA-E+Fu-E	195.0	185.7	98.2 abc	110.4 ab	145.8
1 Control	148.3	266.6	107.5 ab	98.2 ab	145.1
10 SW-E+Fu-E	197.2	144.2	75.6 bc	99.1 ab	129.0
Average	186.6	229.8	96.0	98.2	152.7
C.V.%	38.6	43.3	29.5	42.3	46.5

[1] Averages not followed by the same letter are different according to Duncan's test at the 10% level of significance.

Experiment Two

The incidence of Phymatotrichum root rot averaged 31 percent at Temple compared to 15 percent the previous year (Table 12). At Temple, a majority of the treatments applied to Tamcot CAMD-E reduced the occurrence of root rot compared with the control. The Fu-E and T-E treated plants had a reduced incidence of root rot every week except the last, but a reduction of root rot for plants treated with SW-E did not occur until the fourth week. Significant differences in dead plants occurred for treatments during the first, second, and fourth weeks. At Temple, the RW-E and RW-E+Fu-E treatments resulted in plants with the lowest incidence of root rot during the last week, while the WA-E and T-E+Fu-E treatments caused significantly less disease during the fourth week.

Table 12. Plants with Phymatotrichum root rot in field Experiment Two at Temple for microbial treatments applied to Tamcot CAMD-E.

Treatment number and microorganism(s)	Dead plants by days after planting and weeks after initial root rot symptoms[1]						
	83 1	89 2	97 3	104 4	110 5	118 6	125 7
	%	%	%	%	%	%	%
4 WA-E	*0.0 a	*0.0 a	*0.0	*1.6 a	*6.7	*15.9	*34.0
7 T-E+Fu-E	*0.0 a	*0.0 a	*0.9	*1.8 a	*13.4	*19.4	*31.9
10 RW-E+Fu-E	*0.0 a	*0.0 a	*0.9	*2.8 ab	*7.5	*14.2	*19.1
5 RW-E	*0.0 a	*0.0 a	*1.5	*3.0 ab	*6.4	*10.6	*24.5
6 Fu-E	*0.0 a	*0.0 a	*0.7	*3.4 ab	*15.4	*23.8	41.0
2 T-E	*0.8 ab	*0.8 ab	*1.6	*4.2 ab	*15.9	*22.0	36.0
8 SW-E+Fu-E	*0.0 a	*0.8 ab	*0.8	*4.4 ab	*7.2	*14.1	*26.8
9 WA-E+Fu-E	*0.0 a	1.7 ab	*1.7	*5.8 ab	*16.4	*23.1	*34.4
3 SW-E	2.6 b	3.5 b	3.5	*6.3 ab	*12.4	*20.2	*26.7
1 Control	1.4 ab	1.4 ab	3.1	10.0 b	21.8	25.2	35.7
Averages	0.5	0.8	1.5	4.3	12.3	18.9	31.0
C.V.%	383.9	343.7	357.7	164.0	123.2	98.6	74.4

[1] Averages not followed by the same letter are different according to Duncan's test for the 10 % level of significance. Values preceeded by an asterisk (*) signify a reduced occurrence of root rot with respect to the control.

At McGregor, all applied treatments, with exception of RW-E+Fu-E, resulted in plants having a lower incidence of disease for the last week. Most of the treatments which reduced root rot were not effective until the fourth week after development of initial disease symptoms. The treatments which consistently reduced disease compared to the control were Fu-E and SW-E+Fu-E (Table 13). Cotton treated with WA-E+Fu-E had the only plants having less to slightly less disease than the control over Experiments One and Two. This was also one of the better treatments in the greenhouse experiments.

Table 13. Plants with Phymatotrichum root rot in field Experiment Two at McGregor for microbial treatments applied to Tamcot CAMD-E.

Treatment number and microorganism(s)	Dead plants by days after planting and weeks after initial root rot symptoms[1]						
	83	89	97	104	110	118	125
	1	2	3	4	5	6	7
	%	%	%	%	%	%	%
6 Fu-E	3.4	*4.3	*8.6	*15.5	*24.1	*33.7	*47.7
3 SW-E	*2.7	5.4	11.7	*18.0	*35.2	*45.4	*51.8
4 WA-E	4.4	6.9	10.5	*18.8	*25.6	*38.2	*45.8
8 SW-E+Fu-E	*0.9	*5.1	*7.7	*19.6	*30.6	*41.7	*52.3
5 RW-E	*1.8	*4.5	11.4	*20.3	*30.1	*41.5	*54.0
9 WA-E+Fu-E	*1.8	*4.5	14.0	*20.4	*30.4	*40.7	*59.1
2 T-E	*0.0	7.9	15.9	*21.3	*37.2	*39.2	*51.0
7 T-E+Fu-E	*2.8	5.2	16.3	*23.4	*37.0	*50.6	*60.1
1 Control	3.2	5.2	9.2	24.9	40.3	51.7	61.3
10 RW-E+Fu-E	*3.1	6.9	12.6	28.4	41.1	52.4	*54.3
Average	2.4	5.6	11.8	21.0	33.2	43.5	53.7
C.V.%	164.3	91.8	81.2	60.2	45.1	38.8	31.2

[1] None of the averages are different according to Duncan's test at the 10 % level of significance. Values preceeded by an asterisk (*) signify a reduced occurrence of root rot with respect to the control.

As in field Experiment One, significant differences among treatments for yield occurred only at McGregor. Plants with the WA-E treatment resulted in a significantly higher yield than the control at McGregor, which had the lowest yield (Table 14). Contrary to the yield results at McGregor, the plants treated with WA-E at Temple had the lowest lint yield, whereas plants with other microbial treatments had higher yields in comparison with the control. With the exception of WA-E at Temple, all microbial treatments resulted in plants with higher yields than the control. Over Experiments One and Two all microbial treatments, with the exception of SW-E+Fu-E, tended to have average cotton yields slightly higher to higher than the control.

Table 14. Seed cotton yield for Experiment Two in field experiments for testing the effects of microbial treatments on Phymatotrichum root rot.

Treatment number and microorganism(s)	Lint yield per acre Temple	McGregor	Average
	lbs.	lbs.	lbs.
7 T-E+Fu-E	211.7	85.8 ab	148.3
5 RW-E	193.2	100.0 ab	145.6
9 WA-E+Fu-E	185.7	97.6 ab	142.6
4 WA-E	145.3	109.0 a	142.6
3 SW-E	172.5	102.6 ab	142.5
8 SW-E+Fu-E	196.7	103.0 ab	140.6
2 T-E	157.0	89.4 ab	132.9
10 RW-E+Fu-E	176.7	71.2 ab	131.2
6 Fu-E	172.9	77.0 ab	125.4
1 Control	150.6	57.4 b	115.9
Average	175.2	89.3	136.8
C.V.%	38.8	49.6	42.9

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[1] Averages not followed by the same letter are different according to Duncan's test at the 10% level of significance.

Experiment Three

There was considerable difference in the occurrence of root rot between the Temple and McGregor sites. The averages of dead plants were 33 percent at Temple and 5 percent at McGregor. Due to the extremely low occurrence of root rot at McGregor, these results will not be presented.

The more effective treatments on Tamcot CAMD-E were WA-E, SWO+Fu-10, SWO, SWO+WA-E, and Fu-10 ,with the former two being most effective (Table 15). On Tamcot SP21S, plants treated with SWO+WA-E, SWO+WA-E+Fu-10, and WA-E had reductions in root rot more so than those of other treatments and the control (Table 16). Some significant differences occurred for the percentage root rot between treatments on Tamcot SP21S. The treatments common in reducing disease for both cultivars were WA-E and SWO+WA-E.

Table 15. Tamcot CAMD-E plants with Phymatotrichum root rot in Experiment Three at Temple.

	Dead plants by days after planting and weeks after initial root rot symptoms[1]						
	70	77	84	91	98	107	114
Treatment	1	2	3	4	5	6	7
	%	%	%	%	%	%	%
Fu-10	*1.8	6.0	6.0	*10.5	*23.9	34.1	*35.0
SWO+Fu-10	*0.0	*1.6	*4.8	*11.4	*17.8	*22.5	*26.8
SWO	2.6	4.6	5.8	*11.5	*23.8	*30.4	*33.1
SWO+WA-E+Fu-10	*1.6	5.8	11.2	*13.8	31.4	36.7	37.4
WA-E	*0.0	*0.0	7.8	*14.8	*18.8	*21.5	*24.2
WA-E+Fu-10	*0.9	*1.8	*3.7	*15.6	*29.2	35.1	37.0
Control	2.5	2.5	5.0	16.7	31.0	33.7	36.3
SWO+WA-E	*0.7	2.6	8.0	17.2	*25.7	*33.4	*34.4
Average	1.2	3.1	6.5	13.9	25.2	30.9	33.0
C.V.%	202.4	167.0	122.1	83.0	61.6	58.0	56.0

[1] Averages were not significant for the Duncan's test at the 5% probability level. Values preceeded by an asterisk (*) signify a reduced occurrence of root rot with respect to the control.

All of the treatments, on both cultivars, caused a reduction in root rot compared to the control during at least one week of

Table 16. Tamcot SP21S plants with Phymatotrichum root rot in Experiment Three at Temple.

	Dead plants by days after planting and weeks after initial root rot symptoms[1]						
	70	77	84	91	98	107	114
Treatment	1	2	3	4	5	6	7
	%	%	%	%	%	%	%
Fu-10	*0.0 a	*0.0 a	*2.6	*4.6	*21.6	31.1	31.9 ab
SWO+WA-E+Fu-10	*0.0 a	*0.8 a	*2.5	*6.9	*13.8	*21.5	*23.2 ab
SWO+WA-E	*0.0 a	2.7 ab	*6.5	*11.3	*16.0	*17.2	*19.1 a
WA-E+Fu-10	2.5 ab	2.5 ab	*5.8	*11.8	27.8	37.1	42.8 b
SWO	*1.2 ab	2.9 ab	8.3	*11.8	29.2	36.7	42.6 b
WA-E	*0.7 ab	*2.3 ab	*5.4	*12.3	*23.3	*27.1	*28.7 ab
Control	1.5 ab	2.4 ab	7.4	13.8	25.2	31.1	31.1 ab
SWO+Fu-10	3.4 b	5.0 b	9.0	15.8	27.5	35.9	39.2 ab
Average	1.2	2.3	5.9	11.0	23.0	29.7	32.3
C.V.%	195.7	122.0	105.4	87.4	64.4	51.4	48.8

[1] Averages not followed by the same letter are significantly different according to Duncan's test for the 5% probability level. Values preceeded by an asterisk (*) signify a reduced occurrence of root rot with respect to the control.

accumulated dead plants, with the exception of SWO+Fu-10 applied to SP21S. The only significant interpretation from these results suggests disease reduction is influenced the most by a combination of the *rhizosphere* isolate (WA-E), and the other experimental phyllosphere isolate of CAMD-E (SWO), and possibly involving F. solani.

Considering yield, very few of treated plants had much higher yields than the control plots. Only plants treated with SWO+WA-E were consistent in having higher yields on both of the cultivars (Table 17). The other treatment which caused a higher lint yield than the control was SWO+Fu-10 applied to Tamcot CAMD-E. The WA-E+Fu-10 treatment was not as effective as the WA-E+Fu-E treatment used in previous experiments.

Table 17. Seed cotton yield for entries in field Experiment Three for testing effects of microbial treatments on Phymatotrichum root rot at Temple.

Treatment	Lint yield per acre[1]		
	CAMD-E	SP21S	Average
	lbs.	lbs.	lbs.
SWO+WA-E	215.6	254.9	235.2
Control	196.4	252.0	224.2
WA-E	192.4	234.2	213.3
SWO+Fu-10	216.4	205.9	211.2
Fu-10	193.6	224.3	209.0
SWO+WA-E+Fu-10	171.2	231.2	201.2
SWO	178.5	210.0	194.2
WA-E+Fu-10	182.5	201.5	192.0
Average	193.3	266.7	230.0
C.V.%	29.4	25.3	

[1] None of the averages were significantly different according to Duncan's test at the 10% level of significance.

Combined Results

Treatments being most consistent in reducing root rot over cultivars, locations, and years were WA-E, Fu-E, and WA-E+Fu-E. Summarized results for CAMD-E over Experiments One and Two indicates these treatments, as well as T-E+Fu-E, caused an average of less than 10 percent root rot the fourth week after development of initial symptoms (Table 18). The averages also suggest that each of the treatments independently had some effect in reducing root rot because more dead plants were present in the control plots. There were 13.8 and 30.9 percent dead plants for the fourth and seventh week after initial symptoms, respectively.

The WA-E treatment, when applied to CAMD-E in field Experiments One, Two, and Three (5 tests, 3 years), demonstrated rather consistent reductions in root rot with respect to controls and other treatments; Experiment One at Temple was the only exception (Tables 7, 8, 12, 13, and 15). The WA-E treatment was also effective on cultivar SP21S, being consistent over years and locations to reduce root rot, especially at McGregor (Tables 9, 10, and 16).

The Fu-E treatment also reduced root rot with respect to the control and other treatments. The results for treatment Fu-E were consistent over years and locations on Tamcot CAMD-E to reduce disease, especially at McGregor (Tables 7, 8, 12, 13, and 15). An interesting exception occurred in Experiment One at Temple where the Fu-E treatment did not reduce root rot, contrary to the effect seen in the other experiments. On SP21S, reductions in the occurrence of root rot were present only at Temple (Tables 9 and 16).

The WA-E bacterium and _Fusarium_ spp. mixture in combination as the WA-E+Fu-E treatment, gave some protection from root rot on Tamcot CAMD-E, and appeared consistently more effective than the WA-E, or the Fu-E treatment alone (Tables 2, 4, 7, 8, 12, 13, and 15). This treatment was consistent in having reduced disease incidence in both greenhouse and field experiments. Other treatments did not appear to be as consistent in reducing root rot.

Table 18. Combined results over years and locations showing plants with Phymatotrichum root rot for microbial treatments applied to Tamcot CAMD-E.

Treatment number and microorganism(s)	Dead plants for weeks after initial symptoms[1] 1	2	3	4	5	6	7
	%	%	%	%	%	%	%
4 WA-E	*1.1	*2.9 ab	*4.4	*9.0	*12.8	*19.4	*25.8
6 Fu-E	*1.1	*1.6 a	*4.8	*9.1	*15.0	*19.7	*27.5
7 T-E+Fu-E	*1.4	3.8 ab	7.0	*9.6	*16.5	*22.2	*28.0
9 WA-E+Fu-E	*1.6	*3.5 ab	6.8	*9.8	*15.2	*20.3	*27.8
10 RW-E+Fu-E	*1.1	4.9 ab	6.8	*12.1	*17.6	*22.5	*24.4
8 SW-E+Fu-E	*0.8	4.7 ab	7.7	*12.3	*16.6	*21.6	*27.4
3 SW-E	*1.8	5.6 b	9.1	*12.7	*18.8	*23.5	*27.2
5 RW-E	*1.0	*3.4 ab	8.2	*12.7	*16.8	*21.3	*27.9
2 T-E	2.1	5.0 ab	10.0	*13.0	*20.5	*23.1	*29.8
1 Control	2.0	3.8 ab	6.6	13.8	21.3	25.9	30.9
Average	1.4	3.9	7.1	11.4	17.1	21.9	27.7
C.V.%	245.2	164.1	144.3	118.2	100.2	90.6	77.1

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[1] Averages not followed by the same letter are different according to Duncan's test for the 10 % level of significance. Values preceeded by an asterisk (*) signify a reduced occurrence of root rot with respect to the control.

Data from Experiments One and Two were combined to get the average effect each microbe as a treatment, or part of a treatment, had on reducing root rot (Table 19). In comparing each microbe with the combined effects of other microbes, only the WA-E and the Fu-E treatments were consistent in slightly reducing root rot over time for cultivar Tamcot CAMD-E. Only treatments with the WA-E, SW-E, and T-E bacteria were associated with improved yields in comparison with the other microbes used in treatments. The treatments including RW-E and Fu-E were not better in yield compared to the other treatments, but did yield higher than the control. Based on these comparisons, an improved yield was not associated with a reduction in disease, as shown with Fu-E. But those plants receiving microbial treatments tended to have higher yields than those treated with water-only.

Table 19. A sequential comparison of each microbe with the average of all other microbe treatments on Tamcot CAMD-E. The results are combined over field experiments One and Two.

	Percent dead plants by weeks after first symptoms[1]							
Treatment	1	2	3	4	5	6	7	Yield
	%	%	%	%	%	%	%	lbs/ac
2,7 T-E	1.8	4.4	8.5	11.3	18.5	22.6	28.9	140.6
Non T-E	1.2	3.8	6.8	11.1	16.1	21.8	26.9	138.7
3,8 SW-E	1.4	5.2	8.4	12.5	17.7	22.6	27.4	141.6
Non SW-E	1.3	3.6	6.9	10.8	16.4	21.2	27.3	138.4
4,9 WA-E	1.3	*3.2	*5.6	*9.4	*14.0	*19.9	*26.8	142.7
Non WA-E	1.3	4.1	7.7	11.7	17.4	22.0	27.5	138.1
5,10 RW-E	*1.0	4.2	7.5	12.4	17.2	21.9	*26.2	138.4
Non RW-E	1.4	3.9	7.1	10.8	16.5	21.4	27.7	139.3
6-10 Fu-E	*1.2	*3.7	*6.6	*10.6	*16.2	*21.3	*27.0	137.6
Non Fu-E	1.5	4.2	7.9	11.9	17.2	21.8	27.7	140.9
Control	2.0	3.8	6.6	13.9	21.3	25.9	30.9	115.9
C.V.%	245.2	164.1	144.3	118.2	100.2	90.6	77.1	42.9

[1] None of the comparisons were significantly different according to Duncan's test at the 10 % level of significance. Values preceeded by an asterisk (*) signify a reduced occurrence of root rot with respect to the other microbial treatments.

There were several instances where treatments were associated with improvements in yield relative to the control. The interactions between locations and cultivars appeared important, but were not consistent over experiments. In Experiments One and Two, almost all of the treated plants averaged higher yield with respect to the control (Tables 11 and 14). But in considering each cultivar at each location differences were apparent. However, results from Experiment Three were in contrast to previous expectations showing only one treatment, SWO+Fu-E, to result in improved yield over cultivars in comparison with six other microbial treatments used in that test (Table 17). In previous field experiments, several of the microbial amendments resulted in improved yields for cotton plants.

An attempt was made to analyze the influence of the ten treatments on yield of Tamcot CAMD-E by using combined data of field Experiments One and Two. Total yield was used as the dependent variable in a stepwise multiple regression analysis to determine which of the cumulative percentage of dead plants by weeks, as independent variables, would best explain variation in yield. The variables in the optimum selection step for each treatment, the one having the lowest coefficient of variability and the highest R^2 value, were chosen as best for explaining variability in yield. The sign of the partial regression coefficient for the week or weeks of accumulative dead plants reflects the direction of the influence. This was done within each treatment in order to estimate which was more effective in preventing death of plants in a manner to affect yield (Table 20). More detailed results are given in Appendix tables 23 through 30. Results for the overall field experiments indicate the fourth week was in most cases negatively related to yield, meaning cumulative dead plants through the fourth week had the greater impact on decreasing yield.

Table 20. Sequential selection by stepwise multiple regression showing which week(s) of accumulated dead plants had the greatest influence on explaining variability in yield for Tamcot CAMD-E in Experiments One and Two.

Treatment	R²	Sign of the partial regression coefficient for each week after initial plant death[1] 1	2	3	4	5	6	7	C.V.%
1 Control	0.355				−				40.8
2 T-E	0.222				−				59.8
3 SW-E	0.586	+			−		−	+	30.2
4 WA-E	0.497				−	+		−	29.0
5 RW-E	0.348			−					44.6
6 Fu-E	0.291					−			53.4
7 T-E Fu-E	0.454				−				41.4
8 SW-E+Fu-E	0.416					−			40.8
9 WA-E+Fu-E	0.446	−	+	+	−				44.9
10 RW-E+Fu-E	0.442	+					−		43.4

[1] All models were significant at the 10 percent level. The sign of the partial regression coefficient indicates the week or weeks which were important.

For the treatments T-E, RW-E, Fu-E, T-E+Fu-E, SW-E+Fu-E and the control there was only one week during the course of disease development that cumulative dead plants had an impact in reducing yield. The treatments T-E and T-E+Fu-E had a negative association, or an adverse affect, on yield up to the fourth week, being identical with the control. For treatments Fu-E and SW-E+Fu-E, there was a negative association with yield the fifth week suggesting it took that long for enough dead plants to accumulate before a negative effects on yield occurred. For RW-E, there were enough cumulative dead plants by the third week to have a negative effect on yield.

In other treatments there were varied associations with yield over the course of disease development. It is believed that the effect of root rot on competition for soil moisture and/or the presence of root rot causing less attractiveness to insects can cause such changes in the relationship of cumulative dead plants over time with yield. Such relationships may have occurred with treatments SW-E, WA-E, WA-E+Fu-E, and RW-E+Fu-E.

For treatment SW-E, an initial accumulation of dead plants helped to increase yield, but by the fourth and sixth weeks there were enough dead plants to reduce yield. By the seventh week those plants left were associated with an increase in yield. For treatment RW-E+Fu-E, like SW-E, there was an initial occurrence of root rot that had a beneficial effect on yield, but in this case the accumulation of dead plants was not sufficiently high enough to reduce yield until the sixth week. With the delayed negative effect, the plants in this treatment may have had one to three weeks longer to develop cotton before plant death affected yield.

Treatments WA-E and WA-E+Fu-E also caused, or were associated with a change in yield relationships over time. In treatment WA-E the initial negative association with yield occurred during the fourth week, and was followed by an accumulation of dead plants to give it a positive association, which was again followed with a negative association by the seventh week. The WA-E+Fu-E treatment followed the same general trend of yield relationships, but it occurred earlier at the first week of dead plant accumulation. By the fourth week the number of dead plants was sufficient to reduce yield.

Thus in comparison with the control, the most effective treatments were those that delayed the negative associations with yield over time. The most effective were RW-E+Fu-E, SW-E+Fu-E, Fu-E, and WA-E, respectively. The SW-E treatment may have been beneficial since it had a positive association for yield at the seventh week. The cotton crop was basically mature by the seventh week.

DISCUSSION

The complexity of the root-environment and other environmental factors greatly influenced the results and interpretations. Statistically significant differences between variables were not common. Even so, there was enough consistency in the results that agreed with the hypothesis to permit having some confidence in the results. In studying the host rhizosphere-microbe interactions, an attempt was made to determine the impact of a few representative microbes commonly found in association with the host by adding them back to the rhizosphere. This was done without knowing the quality and quantity of the microorganisms present in the natural state. The microbes used in this study did not fit into any well documented groups of bacteria, indicating that further research is needed to investigate the properties of root associated microorganisms. There are many microorganisms of the soil which remain to be isolated and characterized. However, the rhizosphere bacteria used in this study were found to be asporogenous and aerobic in nature, and did appear to have some influence on the host's disposition to environmental stresses, mainly that of Phymatotrichum root rot. Of the fungi, Fusarium spp. were prevalent which were known to be typical inhabitants of the soil and in many cases have demonstrated a close association with the rhizosphere of cotton plants.

Valuable information concerned with rhizosphere-microbe studies involves the recognition of interactions between the host, microbe, and the pathogen. Probably the most recognized interaction between microorganisms is antagonism of a microbe to a pathogen. Of the microbial isolates used in this study, only the T-E bacterium exhibited inhibition of P. omnivorum growth when plated against each other on PCDA. The overall ineffectivity of the T-E bacterium as a treatment in field experiments suggests that other microbial interactions are present, or more influential in the suppression of disease.

In bacterization research emphasis is placed on rhizosphere microorganisms that demonstrate, in screening experiments, an ability to influence plant growth and health (21,47,65,72). With root rot of cotton it is believed that the composition of the root exudate of MAR cottons causes selective colonization by the favorable microorganisms. The isolates used were from roots of cotton seedlings, and it should be noted that these microbes were primarily used to treat cotton cultivars from which they came. Results suggest the bacterial treatments did have a suppressing affect on disease. Re-isolation of bacteria from root tissues of the host revealed lower colony forming units per gram of tissue on the treated roots than on uninfected plants of the control (not shown). If this is the case, the applied bacteria established themselves on the treated roots at the expense of other natural occurring forms. The influence of the Fusarium spp. was not accounted for.

In studies on wheat infected by Gaeumannnomyces graminis var. tritici, electron microscope studies revealed several asporogenous bacteria associated closely with infected wheat tissue in disease suppressive soil, as compared to few numbers of bacteria found on healthy plant tissue. These asporogenous bacteria have been hypothesized as being site-specific in suppressing disease, having the ability to prevent further hyphal germination, or causing other dysfunction of the pathogen. Evidence for the presence of disease suppressing, site-specific bacteria is found in experiments by Vijinovic, where diseased wheat tissue was more antagonistic to the pathogen, than untreated soil, or healthy plant tissue residue (in 67). If the bacteria from cotton acted in such a manner, they could be effective when applied to plants already colonized by P. omnivorum, increasing the population of site-specific bacteria already associated with the cotton roots. Based on overall results, the WA-E bacterium, because it was most effective as a treatment would be the most likely canidate for this mode of action. The experimental design for this investigation, however, was not for differentiating treatments on the basis of colonized and non-colonized plants.

The major factor in failing to obtain statistically significant differences among treatments concerns the interactions involved with the complex soil environment. Based on the results for final disease counts and yield, interactions accounting for high variations in data involved location differences, inoculum potential, environment, cultivars, and treatments. Although P. omnivorum is restricted mostly to a clay soil, differences are present between the Temple and McGregor locations, each having Houston Black Clay and San Saba Clay soils, respectively. The inoculum potential differed over years, because the pathogen's ability to thrive is influenced primarily by soil moisture content and soil temperature (90). Plant development at both locations was also influenced by insects. These were the factors which made interpretations difficult, and with attempts to alleviate these problems by greenhouse experiments, the conditions are not always representative of that in the field. Under the conditions of so many uncontrolled variables, trends which were consistent over locations and years offer the only evidence for reliability of conclusions.

The results suggest Tamcot CAMD-E microbial isolates can be effective in reducing the incidence of Phymatotrichum root rot, supporting the MAR hypothesis, which states that the host can selectively colonize itself with microorganisms which benefit the plant's disposition to adversities. With all interactions considered, the microorganisms that appeared most beneficial for reducing root rot were the WA-E bacterium, and the Fu-E fungal mixture, not necessarily as single treatments. Each of these microbes was involved with treatments responsible for reduced disease. The WA-E and Fu-E microorganisms appeared most effective at McGregor over both years, but did not cause as much protection at Temple. In combination as WA-E+Fu-E, this treatment was consistent over both years and both locations in reducing root rot.

At Temple, there was a general tendancy for the Fu-E mixture in conjunction with a bacterial treatment to reduce root rot incidence. At McGregor, where a higher inoculum potential was present, the Fu-E mixture had little effect in reducing root rot. The varying results from treatments over the three years was probably brought about by differences in inoculum potential. At a low inoculum potential during Experiment One, differences were larger among treatments, although not statistically so. During field Experiment Two, with a high inoculum potential, differences were not detectable, but all of the microbial treatments averaged lower disease and better yields. This influence may indicate that the microbial effect is present even at a high level of disease.

In most cases, increased yield was related to the treatment applications. Considering all the treatments for the experiment as a whole, the WA-E and Fu-E components appeared most beneficial in causing delay or suppressing onset of disease (Table 19). This was most apparent by the consistent involvement of the WA-E+Fu-E treatment with reduced disease incidence in both greenhouse and field experiments. The T-E+Fu-E treatment in some cases was as effective as the WA-E and/or Fu-E treatments, and it was primarily effective on Tamcot CAMD-E only.

The first year of field testing indicated interactions among cultivars and treatments. The differences in disease incidence between the two cultivars used in Experiment One showed that plants of Tamcot SP21S died at a slower rate than those of Tamcot CAMD-E. This can be associated with inherent cultivar differences, but it is assumed that certain microbial types may not behave the same on every cultivar. Concerning the interactions, the SW-E and SW-E+Fu-E treatments worked well on Tamcot SP21S at McGregor, but were not as effective on Tamcot CAMD-E. Additionally, the RW-E and RW-E+Fu-E treatments acted well in reducing disease at Temple on Tamcot SP21S, but were not as effective on Tamcot CAMD-E. The above two examples indicate differences between locations, where the RW-E bacterium was favored at the Temple location and SW-E performed best at McGregor on the cultivar Tamcot SP21S. It is hoped that in future experiments involving microorganisms isolated from improved cotton strains, a broadly effective beneficial organism can be found that is a plant symbiont and does not interact over different cultivars.

The final field experiment gave results supporting the assumption that a certain quality and kind of microorganism may complement its natural host to suppress disease. The experiment utilized three microbial isolates, two bacteria (WA-E and SWO) and one fungal isolate (Fu-10). The WA-E bacterium is that mentioned in the Materials and Methods section, including Fu-10, which was used because in other experiments it appeared to be more beneficial than the other _Fusarium_ spp. in reducing cotton seedling disease. The SWO bacterium is a phyllosphere isolate from Tamcot CAMD-E which was effective in earlier experiments in protecting cotton from root rot and it was also effective in other studies for reducing damage caused by bacterial blight, boll weevil, _Heliothis_ species, and seedling pathogens. Cultivars Tamcot SP21S and Tamcot CAMD-E were both used in tests planted at Temple and McGregor, Texas. The SWO+Fu-10 treatment was favorable for disease reduction on Tamcot CAMD-E, but not on SP21S. The WA-E treatment; however, had lower disease incidence than the control over time for both cultivars. The results again illustrate the point that certain treatments will favorably complement one host, but not another.

CONCLUSIONS

Those bacteria found representative in the rhizosphere of Tamcot CAMD-E comprised mostly asporogenous aerobic bacteria and the fungal species comprised the genus Fusarium. A balance in the quantity and quality of these microorganisms appear to play a role in the host's response to Phymatotrichum root rot, which may vary depending on the cultivar involved.

Results obtained from this study demonstrate that some control of *Phymatotrichum root* rot of cotton can be implimented with use of microbial isolates from partially resistant cultivars. Because microorganisms were from a cotton variety demonstrating a partial resistance to the pathogen, this supports the assumption that a certain quality of microorganism may be associated with the host plant to create the beneficial effect. The evidence that all treatments were not beneficial to different cultivars supports the assumption that certain kinds of microbes must be present.

In the case of Tamcot CAMD-E, the WA-E and Fu-E components used in experiments appeared to play the major role in reducing disease severity. These microbial components were considered most effective because of their consistency over years and locations in reducing root rot and improving yield. Their effects were partially broadened with some effect on Tamcot SP21S. Depending on the cultivar-treatment and/or location-treatment interactions, other treatments appeared to have an influence on disease severity. Thus, the hypothesis that the host may have genetic influence on its root environment causing it to be selectively colonized by certain beneficial microorganisms to reduce disease severity is supported.

LITERATURE CITED

1. Alabouvette, C. 1978. Fusarium wilt-suppressive soils. 3 - comparison of germination of Fusarium oxysporum chlamydospores in conducive and suppressive soils. (Abstr.) Page 171 in: Third International Congress of Plant Pathology, Munchen, Germany. 435 pp.

2. Ayers, W. A., and Zentmyer, G. A. 1971. Effect on soil solution and two soil pseudomonads on sporangium production by Phytophthora cinnamomi. Phytopathology 61:1188-1193.

3. Batson, W. E., Jr. 1971. Interrelations among resistance to five major diseases and seed, seedling, and plant characters in cotton. Ph. D. thesis. Texas A&M University, College Station. 126 pp.

4. Benedict, J. H., Bird, L. S., and Liverman, C. 1979. Bacterial flora of MAR cottons as a boll weevil resistant character. Proc. Beltwide Cotton Prod. Res. Conf., Cotton Disease Council 39:228-230.

5. Bird, L. S. 1964. Cotton disease control in Texas. Cot. Gin Oil Mill Press, Dallas, Texas. 2:7,34.

6. Bird, L. S. 1970. Cotton varieties which resist and escape multiple diseases. (Abstr.) Phytopathology 60:581-582.

7. Bird, L. S. 1974. Integrated control of cotton seedling disease. Pages 51-54 in: The Relation of Soil Microorganisms to Soilborne Plant Pathogens.,G. C. Papavizas, ed. Va. Agric. Exp. Stn. So. Coop. Ser. Bull. No. 183, 98 pp.

8. Bird, L. S. 1974. The dynamics of cotton seedling disease. Pages 75-79 in: The Relation of Soil Microorganisms to Soilborne Plant Pathogens. Va. Agric. Exp. Stn. So. Coop. Ser. Bull. No. 183, 98 pp.

9. Bird, L. S. 1978. Tamcot CAMD-E, a multi-adversity resistant cotton variety. Texas Agric. Exp. Stn. L-1720.

10. Bird, L. S. 1978. Tamcot SP21S, a multi-adversity resistant cotton variety. Texas Agric. Exp. Stn. L-1721.

11. Bird, L. S. 1982. The MAR (multi-adversity resistance) system for genetic improvement of cotton. Plant Dis. 66:172-176.

12. Bird, L. S., Bush, D. L., Percy, R. G., Bourland, F. M., and Hood, J. E. 1977. Resistance to Phymatotrichum root rot in cotton. Proc. Beltwide Cotton Prod. Res. Conf., Cotton Disease Council. 37:229-232.

13. Bird, L. S., Liverman, C., Thaxton, P., and Percy, R. G. 1980. Evidence that microorganisms in and on tissue have a role in a mechanism of multi-adversity resistance in cotton. Proc. Beltwide Cotton Prod. Res. Conf., Cotton Disease Council 40:283-285.

14. Bird, L. S., and Reyes, A. A. 1966. Seedling diseases of cotton controlled by indirect and direct action of fungicides. (Abstr.) Phytopathology 56:871.

15. Black, H. S. 1968. Influence of the microflora on monocot resistance to <u>Phymatotrichum</u> <u>omnivorum</u>. Bioscience 39:161-179.

16. Blank, L. M. 1940. The susceptibility of cotton seedlings to <u>Phymatotrichum</u> <u>omnivorum</u>. Phytopathology 30:1033-1041.

17. Blondeau, R. 1975. Les syntheses bacteriennes de substance

trophiques vitaminiques et phytohormonales dans la rhizosphere. (Abstr.) Bull. Soc. Bot. Fr. 122:145-155.

18. Bloss, H. E. and Gries, G. A. 1967. Physiologic responses of resistant and susceptible root tissues infected with Phymatotrichum omnivorum. Phytopathology 57:380-384.

19. Bowen, G. D., and Rovira, A. D. 1976. Microbial colonization of plant roots. Annu. Rev. Phytopathol. 14:121-144.

20. Bowen, G. D., and Theodorou, C. 1979. Interactions between bacteria and ectomycorrhizal fungi. Soil Biol. Biochem. 11:119-126.

21. Broadbent, P., Baker, K. F., Franks, N., and Holland, J. 1977. Effect of Bacillus spp. on increased growth of seedlings in steamed and in nontreated soil. Phytopathology 67:1027-1034.

22. Broadbent, P., Baker, K. F., and Waterworth, Y. 1971. Bacteria and actinomycetes antagonistic to fungal root pathogens in Australian soils. Aust. J. Bio. Sci. 24:924-944.

23. Brown, M. E. 1974. Seed and root bacterization. Annu. Rev. Phytopathol. 12:181-197.

24. Buchanan, R. E. and Gibbons, N. E. (editors). 1974. Bergy's Manual of Determinative Bacteriology. Eighth edition. Williams & Wilkins Company, Baltimore. 1268 pp.

25. Burgess, L. W. 1981. General ecology of the fusaria. Pages 225-235 in: Fusarium: Diseases, Biology, and Taxonomy, P. E. Nelson, T. A. Tousoun, and R. J. Cook, eds. Penn State University Press, University Park. 457 pp.

26. Burr, T. J., Schroth, M. N., and Suslow, T. 1978. Increased

potato yields by treatment of seedpieces with specific strains of Pseudomonas fluorescens and P. putida. Phytopathology 68:1377-1383.

27. Bush, D. L. 1980. Variation in root leachate and rhizosphere-rhizoplane microflora among cultivars representing different levels of multi-adversity resistance in cotton. Ph. D. thesis. Texas A&M University, College Station. 271 pp.

28. Chavez, H. B., Bloss, H. E., Boyle, A. M., and Gries, G. A. 1976. Effects of crop residues in soil on Phymatotrichum root rot of cotton. Mycopathologia 58:1-7.

29. Curl, E. A. 1982. The rhizosphere: relation to pathogen behavior and root disease. Plant Dis. 66:624-630.

30. Damicone, J. P. and Manning, W. J. 1982. Avirulent strains of Fusarium oxysporum protect asparagus seedlings from crown rot. Can. J. Plant Pathol. 4:143-146.

31. Diem, H. G., and Mangenot, F. 1975. Influence dela rhizosphere sur les interactions microbiennes dans le sol. (Abstr.) Bull. Soc. Bot. Fr. 122:203-212.

32. Dunlap, A. A. 1941. A convienient soil culture method for obtaining sclerotia of the cotton root fungus. Amer. J. Bot. 28:945-947.

33. Eaton, F. M., and Rigler, N. E. 1946. Influence of carbohydrate levels and root surface microfloras on Phymatotrichum root rot in cotton and maize plants. J. Agric. Res. 72:137-161.

34. Elkan, G. H. 1962. Comparison of rhizosphere microorganisms of genetically related nodulating and non-nodulating soybean lines.

Can. J. Microbiol. 8:79-87.

35. Ford, E. J., Gold, A. H., and Snyder, W. C. 1970. Induction of chlamydospore formation in Fusarium solani by soil bacteria. Phytopathology 60:479-484.

36. Gemrich, E. G. II., and Vanderstreek, E. 1980. Effectiveness of Bacillus uniflagellatus in controlling plant diseases. Plant Dis. 64:382-384.

37. Gibbs, B. M. and Shapton, D. A. (editors). 1968. Identification Methods for Microbiologists, Part B. Academic Press, New York. 212 pp.

38. Gibbs, B. M. and Skinner, F. A. (editors). 1966. Identification Methods for Microbiologists, Part A. Academic Press, New York. 145 pp.

39. Goldsmith, G. W. and Moore, E. J. 1941. Field tests of the resistance of cotton to Phymatotrichum omnivorum. Phytopathology 31:452-463.

40. Greathouse, G. A. 1938. Suggested role of alkaloids in plants resistant to Phymatotrichum omnivorum. Phytopathology 28:592-593.

41. Hadwiger, L. A. and Schwochau, M. E. 1969. Host resistant responses - an induction hypothesis. Phytopathology 59:223-227.

42. Howell, C. R., and Stipanovic, R. D. 1979. Control of Rhizoctonia solani on cotton seedlings with Pseudomonas fluorescens and with an antibiotic produced by the bacterium. Phytopathology 69:480-482.

43. Hutchins, A. S. 1980. In vitro inhibition of root rot pathogens Phellinus weirii, Armillariella mellea, Fomes annosus, and

Phytophthora cinnamomi by a newly isolated Bacillus sp.. Microb. Ecol. 6:253-259.

44. King, C. J., Hope, D., and Eaton, E. D. 1934. Some microbiological activities affected in manurial control of cotton root rot. J. Agric. Res. 49:1093-1107.

45. Kloepper, J. W., Loong, J., Teintze, M., and Schroth, M. N. 1980. Enhanced plant growth by siderophores produced by plant growth promoting rhizobacteria. Nature 286:885-886.

46. Kloepper, J. W. and Schroth, M. N. 1981. Relationship of in vitro antibiosis of plant growth promoting rhizobacteria to plant growth and the displacement of root microflora. Phytopathology 71:1020-1024.

47. Kloepper, J. W., Schroth, M. N., and Miller, T. D. 1980. Effects of rhizosphere colonization by plant growth promoting rhizobacteria on potato development and yield. Phytopathology 70:1078-1082.

48. Koltin, Y., and Chorin-Kirsch, I. 1971. Alteration of fungal morphology induced by a substance from Bacillus cereus. J. Gen. Microbiol. 66:145-151.

49. Kraft, J. M. 1974. The influence of seedling exudates on the resistance of peas to Fusarium and Pythium root rot. Phytopathology 64:190-193.

50. Lochhead, A. G. 1940. Qualitative studies of soil microorganisms. III - Influence of plant growth on the character of the bacterial flora. Canad. J. Res. Sect. C., Bot. Sci. 18:42-53.

51. Louvet, J. 1978. Fusarium wilt-supressive soils. 1 - transfer of

the suppresive effect and prospect of biological control. Page 170 in: Third International Congress of Plant Pathology, Munchen, Germany. 435 pp.

52. Lyda, S. D. 1978. Ecology of Phymatotrichum. Annu. Rev. Phytopathol. 16:193-209.

53. Marshall, K. C. and Alexander, M. 1960. Competition between soil bacteria and Fusarium. Plant Soil 12:143-153.

54. Michael, A. H., and Nelson, P. E. 1972. Antagonistic effects of soil bacteria on Fusarium roseum culmorum from carnation. Phytopathology 62:1052-1056.

55. Mitchell, R. B., Hooton, D. R., and Clark, F. E. 1941. Soil bacteriological studies on the control of Phymatotrichum root rot of cotton. J. Agric. Res. 63:535-547.

56. Moore-Landecker, E., and Stotzky, G. 1973. Morphological abnormalities of fungi induced by volatile microbial metabolites. Mycologia 65:519-530.

57. Mukherjee, R., and Chattopadhyay, S. B. 1977. Effect of root exudates of different crops on population of bacteria and fungi in rhizosphere and remote soil. (Abstr.) Indian Ag. 21:201-206.

58. Neal, J. L., Jr., Atkinson, T. G., and Larson, R. I. 1970. Changes in the rhizosphere microflora of spring wheat induced by disomic substitution of a chromosome. Can. J. Micro. 16:153-158.

59. Neal, J. L., Jr., Larson, R. I., and Atkinson, T. G. 1973. Changes in the rhizosphere populations of selected physiological groups of bacteria related to substitution of specific pairs of chromosomes of spring wheat. Plant Soil 39:209-212.

60. Newman, E. I., and Bowen, H. J. 1974. Patterns of distribution of bacteria on root surfaces. Soil Biol. Biochem. 6:205-209.

61. Papavisas, G. C., and Lumsden, R. D. 1980. Biological control of soilborne fungal propagules. Annu. Rev. Phytopathol. 18:389-413.

62. Percy, R. G. 1979. Inheritance of chemical constituents in leachate from the seed coat of cotton and their interrelations with several seed characters. M. S. thesis. Texas A&M University Library, College Station. 78 pp.

63. Prikryl, Z., and Vancura, V. 1980. Root exudates of plants. VI. Wheat root exudation as a dependent on growth, concetration gradient of exudates and the presence of bacteria. Plant Soil 57:69-83.

64. Rouxel, F. 1978. Fusarium wilt-suppressive soils. 2 - Role of the native Fusarium. (Abstr.) Page 171 in: Third International Congress of Plant Pathology, Munchen, Germany. 435 pp.

65. Rovira, A. D. 1963. Microbial inoculation of plants. I. Establishment of free-living nitrogen-fixing bacteria in the rhizosphere and their effects on maize, tomato, and wheat. Plant Soil 19:304-314.

66. Rovira, A. D. and Davey, L. 1974. Biology of the rhizosphere. Pages 155-204 in: The Plant Root and its Environment. Univ. Press. Charlotesville.

67. Rovira, A. D. and Wildermuth, G. B. 1981. The nature and mechanism of suppression. Pages 385-415 in: Biology and Control of Take-all, M. J. C. Asher and P. J. Shipton, eds. Academic Press, New York. 538 pp.

68. Scher, F. M. and Baker, R. 1980. Mechanism of biological control in a Fusarium-supressive soil. Phytopathology 70:412-417.

69. Schinde, P. A., and Lukezic, F. L. 1974. Interactions of Pseudomonas marginalis var. alfalfa, Erwinia amylovora var. alfalfae, and an unidentified bacteria (WB-3) with certain root pathogens of alfalfa. Phytopathology 64:1169-1173.

70. Schroth, M. N., and Hancock, J. G. 1981. Selected topics in biological control. Annu. Rev. Microbiol. 35:453-476.

71. Schroth, M. N., and Hancock, J. G. 1982. Disease-suppresive soils and root-colonizing bacteria. Science 216:1376-1381.

72. Schroth, M. N., and Hildebrand, P. C. 1964. Influence of plant exudates on root infecting fungi. Annu. Rev. Phytopathol. 2:101-132.

73. Skerman, V. B. D. (editor). 1969. Abstracts of Microbiological Methods. John Wiley & Sons, Inc., New York. 883 pp.

74. Streets, R. B., and Bloss, H. E. 1973. Phymatotrichum Root Rot. Amer. Phytopathol. Soc. Monograph. No. 8. 38 pp.

75. Sulochana, C. B. 1962. Amino acids in root exudates of cotton. Plant Soil 16:312-326.

76. Sulochana, C. B. 1962. B-vitamins in root exudates of cotton. Plant Soil 16:327-334.

77. Sulochana, C. B. 1962. Cotton roots and vitamin-requiring and amino acid-requiring bacteria. Plant Soil 16:335-346.

78. Suslow, T. V. 1982. Role of deleterious rhizobacteria as minor pathogens in reducing crop growth. Phytopathology 72:111-115.

79. Suslow, T., Kloepper, W., Schroth, M. N., and Burr, T. J. 1979.

Beneficial bacteria enhance plant growth. Calif. Agric. 33:15-17.

80. Suslow, T. V., and Schroth, M. N. 1980. Interactions of growth promoting rhizobacteria with deleterious rhizosphere bacteria and fungi. Phytopathology 71:259.

81. Taubenhaus, J. J., and Ezekiel, W. N. 1932. Resistance of monocotyledons to Phymatotrichum root rot. Phytopathology 22:443-452.

82. Taylor, G. S., and Parkinson, D. 1961. The growth of saprophytic fungi on root surfaces. Plant Soil 15:261-267.

83. Thom, C., and Humfeld, H. 1932. Notes on the association of microorganisms and roots. Soil Science 34:29-36.

84. Tsai, H. L. 1974. Biology of spermoplane and seedling rhizoplane in relation to disease resistance in cotton. M. S. thesis. Texas A&M University. College Station. 122 pp.

85. Tsai, H. L., and Bird, L. S. 1975. Microbiology of host-pathogen interactions for resistance to seedling disease and multi-adversity resistance in cotton. Proc. Beltwide Cotton Prod. Res. Conf., Cotton Disease Council. 35:39-45.

86. Tuckey, H. B. 1966. Leaching of metabolites from above-ground plant parts and its implications. Bull. Torrey Bot. Club. 93:385-401.

87. Watkins, G. M. 1938. Histology of Phymatotrichum root rot of field grown cotton. Phytopathology 28:195-202.

88. Watkins, G. M., and Watkins, M. O. 1940. Experimental Phymatotrichum root rot of retama and corn. Torrey Bot. Club Bull. 67:489-501.

89. Weller, D. M. and Cook, R. J. 1981. Control of take-all of wheat with fluorescent pseudomonads. Phytopathology 71:1007.

90. Wheeler, J. E., and Hine, R. B. 1972. Influence of soil temperature and moisture on survival and growth of strands of *Phymatotrichum omnivorum*. Phytopathology 62:828-832.

APPENDIX

Table 21. Sequential selection by stepwise multiple regression showing which week(s) of accumulated dead plants had the greatest influence on explaining variability in yield for Tamcot CAMD-E treated with a water-only control (1).

Data sets and selected steps	Sign of the partial regression coefficient for each week after initial plant death[1]								
	R^2	1	2	3	4	5	6	7	C.V.%
Over Experiments One and Two									
1	0.355				–				40.8
2	0.372	–			–				41.2
3	0.378		–	+		–			42.0
4	0.381		–	+	–	–			43.0
5	0.387		–	+	–	–	+		43.9
6	0.392		–	+	–	–	+	–	45.0
7	0.392	–	–	+	–	–	+	–	46.4
Over McGregor only, Experiments One and Two									
1	0.301					–			46.4
2	0.313		–					–	48.5
3	0.381		–		+		–		48.8
4	0.398	+	–		+		–		51.5
5	0.399	+	–	–	+		–		55.5
6	0.672		–	–	+	+	–	+	44.9
7	0.828	–	–	–	+	+	–	+	36.4
Over Temple only, Experiments One and Two									
1	0.476				–				25.8
2	0.546				–		+		25.3
3	0.554				–		+	–	26.6
4	0.578	–		+	–		+		27.7
5	0.584	–		+	–		+	–	29.7
6	0.584	–		+	–	–	+	–	32.5
7	no further improvement								
Over Experiment One at Temple and McGregor									
1	0.644		–						24.4
2	0.657	+	–						25.2
3	0.796	+	–				+		20.6
4	0.805	+	–		–		+		21.5
5	0.869	+	–		–	+	+		19.1
6	0.899	+	–	+	–	+	+		18.4
7	no further improvement								
Over Experiment Two at Temple and McGregor									
1	0.238				–				56.6
2	0.245				–			+	59.4
3	0.290			–			–	+	61.1
4	0.362		+	–			–	+	61.9
5	0.544		+	–		+	–	+	56.5
6	0.550		+	–	–	+	–	+	61.5
7	0.565	–	+	–	–	+	–	+	67.6

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[1] All models were significant at the 10 percent level.

Table 22. Sequential selection by stepwise multiple regression showing which week(s) of accumulated dead plants had the greatest influence on explaining variability in yield for Tamcot CAMD-E treated with T-E (2).

Data sets and selected	Sign of the partial regression coefficient for each week after initial plant death[1]								
steps	R^2	1	2	3	4	5	6	7	C.V.%
Over Experiments One and Two									
1	0.222				−				59.8
2	0.255		+		−				59.9
3	0.257		+		−			−	61.3
4	0.267		+		−	+		−	62.4
5	0.268	+	+		−	+		−	64.1
6	0.268	+	+		−	+	+	−	66.0
7	0.268	+	+	−	−	+	+	−	68.0
Over McGregor only, Experiments One and Two									
1	0.560				−				39.9
2	0.622		+		−				38.9
3	0.637		+		−			+	40.5
4	0.688		+		−	−		+	40.1
5	0.699		+	+	−	−		+	42.6
6	0.701	+	+	+	−	−		+	46.5
7	0.704	+	+	+	−	−	+	+	51.7
Over Temple only, Experiments One and Two									
1	0.029				+				56.2
2	0.253					+		−	52.0
3	0.315	−				+		−	52.8
4	0.453	−	+			+		−	50.4
5	0.459	−	+		−	+		−	54.2
6	0.464	−	+	−	−	+		−	59.1
7	0.464	−	+	−	−	+	−	−	66.1
Over Experiment One at Temple and McGregor									
1	0.212			−					65.3
2	0.237		+	−					67.8
3	0.481		−				−	+	59.3
4	0.494		−	−			−	+	62.6
5	0.545		−	−	+		−	+	64.1
6	0.545	−	−	−	+		−	+	70.2
7	no further improvement								
Over Experiment Two at Temple and McGregor									
1	0.280				−				56.0
2	0.407	+			−				53.6
3	0.528			+	−	+			50.7
4	0.545	+		+	−		+		53.3
5	0.566	+	−	+	−		+		56.2
6	0.579	+	−	+	−	+		+	60.6
7	0.580	+	−	+	−	+	−	+	67.7

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[1] All models were significant at the 10 percent level.

Table 23. Sequential selection by stepwise multiple regression showing which week(s) of accumulated dead plants had the greatest influence on explaining variability in yield for Tamcot CAMD-E treated with SW-E (3).

Data sets and selected	Sign of the partial regression coefficient for each week after initial plant death[1]								
steps	R^2	1	2	3	4	5	6	7	C.V.%
Over Experiments One and Two									
1	0.530					–			29.9
2	0.554				–		–		29.9
3	0.581	+			–		–		29.6
4	0.586	+			–		–	+	30.2
5	0.593	+			–	+	–	+	30.8
6	0.593	+	–		–	+	–	+	31.7
7	0.593	+	–	+	–	+	–	+	32.7
Over McGregor only, Experiments One and Two									
1	0.533				–				28.6
2	0.584	–			–				28.5
3	0.689		–				+	–	26.1
4	0.713		–			–	+	–	26.9
5	0.726	+	–			–	+	–	28.3
6	0.740	+	–	+		–	+	–	30.2
7	0.757	+	–	+	–	–	+	–	32.6
Over Temple only, Experiments One and Two									
1	0.495					–			22.4
2	0.677	+				–			18.9
3	0.806	+			–		–		15.5
4	0.822	+			–		–	+	15.9
5	0.828	+	+		–		–	+	16.8
6	0.830	+	+		–	+	–	+	18.3
7	no further improvement								
Over Experiment One at Temple and McGregor									
1	0.409							–	34.2
2	0.612	+						–	29.2
3	0.636	+			+	–			30.0
4	0.649	+			+	–		–	31.5
5	0.649	+			+	–	+	–	34.0
6	0.650	+	+	–	+	–		–	37.2
7	0.650	+	+	–	+	–	+	–	41.6
Over Experiment Two at Temple and McGregor									
1	0.762						–		22.9
2	0.776						–	+	23.4
3	0.793			–	+		–		23.9
4	0.811			–	+		–	+	24.4
5	0.829			–	+	+	–	+	25.0
6	0.853	+	–	–	+		–	+	25.5
7	0.892	+	–	–	+	+	–	+	24.3

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[1] All models were significant at the 10 percent level.

Table 24. Sequential selection by stepwise multiple regression showing which week(s) of accumulated dead plants had the greatest influence on explaining variability in yield for Tamcot CAMD-E treated with WA-E (4).

Data sets and selected	Sign of the partial regression coefficient for each week after initial plant death[1]								
steps	R^2	1	2	3	4	5	6	7	C.V.%
Over Experiments One and Two									
1	0.401							-	30.2
2	0.453				-			-	29.5
3	0.497				-	+		-	29.0
4	0.506		-	+	-			-	29.5
5	0.556		-	+	-	+		-	28.7
6	0.561	-	-	+	-	+		-	29.4
7	0.564	-	-	+	-	+	+	-	30.2
Over McGregor only, Experiments One and Two									
1	0.633					-			24.5
2	0.674		+			-			24.3
3	0.709		+		-	-			24.4
4	0.735		+		-	-	+		24.9
5	0.756	+	+	-	-	-			25.7
6	0.761	+	+	-	-		-	+	27.9
7	0.761	+	+	-	-	-	-	+	31.2
Over Temple only, Experiments One and Two									
1	0.399							-	24.6
2	0.519					+		-	23.2
3	0.681				-	+		-	20.1
4	0.732				-	-	+	-	19.6
5	0.734		-		-	+	+	-	21.1
6	0.735		-	+	-	+	+	-	23.1
7	no further improvement								
Over Experiment One at Temple and McGregor									
1	0.129				-				30.6
2	0.174			+	-				31.4
3	0.440		-	+		-			27.5
4	0.448		-	+	+	-			29.2
5	no further improvement								
6									
7									
Over Experiment Two at Temple and McGregor									
1	0.604							-	29.4
2	0.663				-			-	28.6
3	0.708				-	+		-	28.2
4	0.735	+			-	+		-	28.8
5	0.818	+		-	-	+		-	25.7
6	0.831	+	+	-	-	+		-	27.2
7	0.832	+	+	-	-	+	-	-	30.3

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[1] All models were significant at the 10 percent level.

Table 25. Sequential selection by stepwise multiple regression showing which week(s) of accumulated dead plants had the greatest influence on explaining variability in yield for Tamcot CAMD-E treated with RW-E (5).

Data sets and selected steps	Sign of the partial regression coefficient for each week after initial plant death[1]								
	R^2	1	2	3	4	5	6	7	C.V.%
Over Experiments One and Two									
1	0.348			-					44.6
2	0.374			-				-	44.8
3	0.392	+		-		-			45.2
4	0.430	+		-	+	-			44.9
5	0.437	+		-	+	-	+		45.8
6	3441	+	-	-	+	-	+		47.0
7	0.441	+	-	-	+	-	+	-	48.4
Over McGregor only, Experiments One and Two									
1	0.380		-						39.7
2	0.413	+	-						40.7
3	0.519		-				-	+	39.1
4	0.554		-	+			-	+	40.2
5	0.600		-	+	-		-	+	41.1
6	0.618	+	-	+	-		-	+	44.0
7	0.619	+	-	+	-	-	-	+	49.2
Over Temple only, Experiments One and Two									
1	0.386							-	24.3
2	0.414				+		-		25.0
3	0.445			-	+		-		25.8
4	0.458	+		-	+		-		27.3
5	0.464	+		-	+	+	-		29.3
6	0.466	+		-	+	+	-	-	32.0
7	no further improvement								
Over Experiment One at Temple and McGregor									
1	0.409					-			51.4
2	0.441	+				-			52.7
3	0.531			-	+	-			51.2
4	0.647	+		-	+	-			47.5
5	0.653	+	+	-	+	-			50.9
6	0.653	+	+	-	+	-	-		55.7
7	no further improvement								
Over Experiment Two at Temple and McGregor									
1	0.368							-	37.9
2	0.419			+			-		38.4
3	0.507		-	+			-		37.5
4	0.603		-	+		+	-		35.9
5	0.734		-		+	+	-	+	31.8
6	0.865	+	-		+	+	-	+	24.8
7	0.877	+	-	+	+	+	-	+	26.4

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[1] All models were significant at the 10 percent level.

Table 26. Sequential selection by stepwise multiple regression showing which week(s) of accumulated dead plants had the greatest influence on explaining variability in yield for Tamcot CAMD-E treated with Fu-E (6).

Data sets and selected steps	Sign of the partial regression coefficient for each week after initial plant death[1]								
	R^2	1	2	3	4	5	6	7	C.V.%
Over Experiments One and Two									
1	0.291					-			53.4
2	0.306		+			-			54.1
3	0.338			+	-	-			54.1
4	0.346		+	+	-	-			55.2
5	0.383			+	-	-	+	-	55.1
6	0.386		+	+	-	-	+	-	56.6
7	0.386	+	+	+	-	-	+	-	58.3
Over McGregor only, Experiments One and Two									
1	0.341					-			38.7
2	0.441					-	+		37.5
3	0.583					-	+	-	34.4
4	0.650		+			-	+	-	33.7
5	0.662		+	+		-	+	-	35.8
6	0.697	-	+	+		-	+	-	37.1
7	0.699	-	+	+	+	-	+	-	41.3
Over Temple only, Experiments One and Two									
1	0.426					-			41.9
2	0.512				-			-	40.7
3	0.590			+	-		-		39.7
4	0.624			+	-		-	+	40.6
5	0.674		+	+	-		-	+	40.8
6	0.674		+	+	-	+	-	+	44.7
7	no further improvement								
Over Experiment One at Temple and McGregor									
1	0.410				-				48.4
2	0.654			+	-				39.0
3	0.660			+	-	+			41.0
4	0.660		-	+	-	+			43.9
5	no further improvement								
6									
7									
Over Experiment Two at Temple and McGregor									
1	0.424							-	53.1
2	0.435					-		-	55.4
3	0.466				+	-		-	57.1
4	0.487	+	-		+			-	59.9
5	0.489	+	-		+	-		-	64.5
6	0.491	+	-	-		-	+	-	70.6
7	0.493	+	-	-	+	-	+	-	78.8

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[1] All models were significant at the 10 percent level.

Table 27. Sequential selection by stepwise multiple regression showing which week(s) of accumulated dead plants had the greatest influence on explaining variability in yield for Tamcot CAMD-E treated with T-E+Fu-E (7).

Data sets and selected steps	Sign of the partial regression coefficient for each week after initial plant death[1]								C.V.%
	R^2	1	2	3	4	5	6	7	
Over Experiments One and Two									
1	0.454				-				41.4
2	0.473			+	-				41.6
3	0.502			+	-			+	41.5
4	0.513			+	-	-		+	42.1
5	0.527	+	-	+	-			+	42.6
6	0.542	+	-	+	-	-		+	43.2
7	0.542	+	-	+	-	-	-	+	44.5
Over McGregor only, Experiments One and Two									
1	0.469					-			43.3
2	0.597					-	+		39.8
3	0.611	+				-	+		41.5
4	0.624	+		+		-	+		43.6
5	0.697	+	-	+		-		+	42.2
6	0.731	+	-	+		-	-	+	43.5
7	0.744	+	-	+	+	-	-	+	47.5
Over Temple only, Experiments One and Two									
1	0.548				-				22.5
2	0.568				-			+	23.2
3	0.604				-	-		+	23.5
4	0.675	-		+	-	-			22.8
5	0.731	-		+	-	-	+		22.4
6	0.747	-		+	-	-	+	-	23.8
7	no further improvement								
Over Experiment One at Temple and McGregor									
1	0.433				-				41.0
2	0.483				-	+			41.2
3	0.533		-			+	-		41.6
4	0.551		-			+	-	+	43.5
5	0.553	+	-			+	-	+	47.0
6	0.554	+	-	+		+	-	+	51.4
7	no further improvement								
Over Experiment Two at Temple and McGregor									
1	0.583					-			40.6
2	0.606			+	-				41.6
3	0.712	+		+	-				37.7
4	0.806	+	-	+	-				33.0
5	0.822	+	-	+	-	-			34.2
6	0.858	+	-	+	-	-	+		33.5
7	0.876	+	-	+	-	-	+	+	35.0

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[1] All models were significant at the 10 percent level.

Table 28. Sequential selection by stepwise multiple regression showing which week(s) of accumulated dead plants had the greatest influence on explaining variability in yield for Tamcot CAMD-E treated with SW-E+Fu-E (8).

Data sets and selected steps	Sign of the partial regression coefficient for each week after initial plant death[1]								
	R^2	1	2	3	4	5	6	7	C.V.%
Over Experiments One and Two									
1	0.416					−			40.8
2	0.430		−				−		41.2
3	0.443	+	−			−			41.8
4	0.446	+	−	−			−		42.7
5	0.449	+	−	−	+		−		43.8
6	0.450	+	−	−	+	−	−		45.0
7	0.451	+	−	−	+	−	−	+	46.4
Over McGregor only, Experiments One and Two									
1	0.446				−				36.3
2	0.518	−			−				35.7
3	0.556	−			−	+			36.3
4	0.568	−			−	+		−	38.3
5	0.574	−	−		−	+	−		41.1
6	0.574	−	−	−	−	+	−		45.0
7	0.574	−	−	−	−	+	−	−	50.3
Over Temple only, Experiments One and Two									
1	0.659				−				19.6
2	0.797				−			−	16.0
3	0.814	+			−			−	16.2
4	0.829	+	−		−			−	16.6
5	0.838	+	−		−	+		−	17.5
6	0.839	+	−		−	+	+	−	19.1
7	0.839	+	−	−	−	+	+	−	21.3
Over Experiment One at Temple and McGregor									
1	0.400						−		50.4
2	0.515	+					−		47.8
3	0.552	+	−				−		48.7
4	0.581	+	−		+		−		50.3
5	0.582	+	−	−	+		−		54.3
6	0.584	+	−	−	+	+	−		59.4
7	no further improvement								
Over Experiment Two at Temple and McGregor									
1	0.562							−	30.5
2	0.720	−						−	25.7
3	0.743	−					+	−	26.2
4	0.782	−			−		+	−	25.8
5	0.788	−	+		−		+	−	27.5
6	0.796	−		+	−	−	+	−	29.5
7	0.796	−	+	+	−	−	+	−	33.0

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[1] All models were significant at the 10 percent level.

Table 29. Sequential selection by stepwise multiple regression showing which week(s) of accumulated dead plants had the greatest influence on explaining variability in yield for Tamcot CAMD-E treated with WA-E+Fu-E (9).

Data sets and selected	Sign of the partial regression coefficient for each week after initial plant death[1]								
steps	R^2	1	2	3	4	5	6	7	C.V.%
Over Experiments One and Two									
1	0.282				-				47.5
2	0.319	-				-			47.3
3	0.395	-		+	-				45.7
4	0.446	-	+	+	-				44.9
5	0.446	-	+	+	-		+		46.1
6	0.453	-	+	+	-		+	-	47.2
7	0.454	-	+	+	-	-	+	-	48.6
Over McGregor only, Experiments One and Two									
1	0.398				-				39.0
2	0.419		-		-				40.4
3	0.433		-	+	-				42.3
4	0.449		-	+	-		+		44.6
5	0.485			+	-	-	+	-	46.6
6	0.485		-	+	-	-	+	-	51.0
7	0.485	+	-	+	-	-	+	-	57.1
Over Temple only, Experiments One and Two									
1	0.214							-	37.0
2	0.344		+		-				35.6
3	0.345		+		-	+			37.8
4	0.348		+		-	+		-	40.3
5	0.348		+		-	+	+	-	43.5
6	no further improvement								
7									
Over Experiment One at Temple and McGregor									
1	0.225	-							52.2
2	0.355	-	+						50.2
3	0.618	+			-		+		41.0
4	0.620	+	-		-		+		43.7
5	0.621	+	-	+	-		+		47.1
6	no further improvement								
7									
Over Experiment Two at Temple and McGregor									
1	0.587						-		37.4
2	0.616				-			-	38.0
3	0.619				-	+		-	40.1
4	0.627	+			-	+		-	42.4
5	0.653	+	-	-		+		-	44.2
6	0.655	+	-	-	+	+		-	48.3
7	0.655	+	-	-	+	+	+	-	54.0

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[1] All models were significant at the 10 percent level.

Table 30. Sequential selection by stepwise multiple regression showing which week(s) of accumulated dead plants had the greatest influence on explaining variability in yield for Tamcot CAMD-E treated with RW-E+Fu-E (10).

Data sets and selected steps	Sign of the partial regression coefficient for each week after initial plant death[1]								
	R^2	1	2	3	4	5	6	7	C.V.%
Over Experiments One and Two									
1	0.370				-				45.1
2	0.442	+					-		43.4
3	0.456	+			-		-		43.9
4	0.464	+			-	+	-		44.8
5	0.471	+			-	+	-	+	45.7
6	0.499	+	-	+	-		-	+	45.7
7	0.503	+	-	+	-	+	-	+	46.9
Over McGregor only, Experiments One and Two									
1	0.430				-				51.0
2	0.516		-					-	49.5
3	0.664		-				+	-	43.8
4	0.689		-		+		+	-	45.0
5	0.765		-		+	-	+	-	42.3
6	0.787	-	-		+	-	+	-	44.1
7	0.801	-	-	+	+	-	+	-	47.6
Over Temple only, Experiments One and Two									
1	0.262						-		30.9
2	0.424		+				-		28.8
3	0.573			+			-	+	26.3
4	0.636		+			+	-	+	26.0
5	0.665		+		-	+	-	+	26.9
6	0.665	+	+		-	+	-	+	29.5
7	no further improvement								
Over Experiment One at Temple and McGregor									
1	0.235				-				41.4
2	0.421	+			-				37.9
3	0.559		+	-	-				35.1
4	0.683		+	-			+	-	31.8
5	0.690		+	-		+	+	-	34.0
6	0.691	-	+	-		+	+	-	37.2
7	no further improvement								
Over Experiment Two at Temple and McGregor									
1	0.558						-		47.4
2	0.601	+					-		47.5
3	0.628				+	-	-		48.6
4	0.688		-		+	-		-	47.6
5	0.798	+	-	+		-		-	41.4
6	0.821	+	-	+	+	-		-	42.6
7	0.822	+	-	+	+	-	-	-	47.6

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[1] All models were significant at the 10 percent level.

VITA

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