

FACTORS INFLUENCING DEVELOPMENT AND CONTROL OF ROOT  
ROT OF BEAN, PHASEOLUS VULGARIS L., IN THE WILLAMETTE  
VALLEY OF OREGON

by

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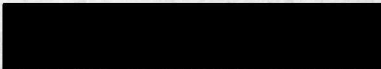
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FACTORS INFLUENCING DEVELOPMENT AND CONTROL OF ROOT ROT  
OF BEAN, PHASEOLUS VULGARIS L., IN THE WILLAMETTE  
VALLEY OF OREGON

INTRODUCTION

The snap bean is the most important vegetable grown for processing in the Pacific Northwest, and in Oregon is the major processing crop west of the Cascades. The value of the crop after processing approximates the combined values of all market garden crops and of other vegetables grown for processing.

In Oregon, which is rated first in snap bean production, the acreage has increased more than 200 per cent in the last ten years. In the last two years the acreage has reached a high of ten thousand acres. With the exception of two or three hundred acres, all of these are grown in nine counties of the Willamette Valley.

In the past, rotation of other crops with beans was practiced mainly because the processing concerns allotted acreages of beans among the growers. In the last decade the rapid growth of frozen food processing has resulted in an increased demand for snap beans. As a result, beans are being grown year after year in the same field with little attempt at rotation. In many fields in the Willamette Valley, where yields in the past have often exceeded ten tons per acre, yields of seven tons or less are frequently obtained.

Staff members of the Oregon Agricultural Experiment Station and field men for the processing concerns have for years observed the almost universal occurrence of root rot in beans. It is the general opinion of most of these people that root rot has become

increasingly severe during the last few years.

Concern over bean root rot is not limited to this area, but is wide-spread throughout the western states and in other bean growing areas of the United States. At present root rot investigations are being carried out in most of the western states and in 1953 a regional project was activated for the study of virus diseases and root rots of dry and snap beans in the western region.

While root rots are commonly found throughout the bean fields of the Willamette Valley, their presence and importance is largely unrecognized by the growers. Only in those instances where severe reductions in yield have been sustained are the growers becoming concerned with the root rot problem.

The investigations reported in this thesis were designed to:

- (a) determine what organisms are responsible for root rot of beans in the Willamette Valley, (b) study the factors associated with disease development, (c) obtain an estimate of the damage caused by root rot in terms of reduced yields, and (d) study possible control measures for the disease.



## SYMPTOMS OF THE DISEASE IN THE WILLAMETTE VALLEY

The first symptoms of root rot can be observed in the field before the bean plants emerge. If the germinating seeds are removed from the soil, reddish pin-point discolorations can be observed on the developing tap root and hypocotyl. The number of these discolorations varies from one or two per plant to at least a hundred, probably in direct relation to the amount of inoculum in the soil.

A month after germination the pin-point discolorations have spread and coalesced to a stage where almost the entire tap root and underground part of the hypocotyl may be covered. Occasionally the discolorations do not coalesce but spread linearly. In such a case part of the tap root and hypocotyl may be covered by a solid discoloration and the other part by streaks.

In cases of severe infection the lower portion of the tap root may be entirely destroyed six weeks after germination. At this stage little growth of the bean plant can be observed. For two or three weeks the plants remain in a stunted condition, during which time a cluster of adventitious roots develop from the hypocotyl area just above the lesions and below or at the soil line. After the development of the adventitious roots, normal growth of the bean plant is resumed.

When the infection is moderate, the reddish discoloration becomes brown or black in color six weeks after germination and

longitudinal fissures can be observed on the tap root and hypocotyl. As these plants develop the lower portion of the hypocotyl may become pithy.

At the end of the growing season a dark-red discoloration of the pith of the bean plant is frequently encountered. This discoloration, which may extend to three or four inches above the soil line, is not always associated with a severe infection as it has not been found in several fields where infection was quite severe.

At harvest time a reddish-brown surface discoloration of the hypocotyl is frequently encountered. This discoloration may extend above the soil line for two or three inches.

With the exception of a temporary stunted condition, no symptoms appear on the stem or foliage of the bean plant at any time during the growing season. Wilted plants or plants with yellow leaves have been found which showed severe root rot infection, however, upon close examination it was found that symphilids or wire worms were feeding on the roots.

Infection appears to be fairly uniform throughout the bean fields in the Willamette Valley, although in some first or second year bean fields only scattered infections have been observed.

## LITERATURE REVIEW

Six root rot diseases of bean (Phaseolus vulgaris L.) have been reported in the literature. The diseases that are more widespread in occurrence will be reviewed first.

### Dry Root Rot

In 1916, Burkholder (4) reported the occurrence of a root disease of beans in New York which affected all commercial varieties. The disease was found in 90 per cent of the fields of the six major bean producing areas of the state and had been observed by growers for at least twenty-five years prior to Burkholder's report.

The first symptoms (5, pp. 1004-1006) 12, p. 20) of the disease on beans are reddish discolorations, on the tap root which are evident about ten days after the plants appear above ground. The discolorations may occur in streaks or cover almost the entire tap root without any definite margins. Later the diseased areas turn brown and longitudinal fissures develop in the cortex. As the disease progresses the lateral roots and the tap root become dry and pithy. Other roots produced above the lesions may replace the tap root.

The above ground symptoms of the disease are not conspicuous. The plants are frequently stunted early in the spring, but as the season progresses this is less noticeable. At podding time the



symptoms of the disease become more evident. Few pods are formed and these frequently drop before maturing. The leaves occasionally turn yellow and fall. Usually there is no wilting of the plant.

Burkholder (5) isolated the causal organism and described it as Fusarium martii phaseoli n. form. The pathogen is now designated as Fusarium solani f. phaseoli (Burk) Snyder and Hansen in the classification (27) of the genus Fusarium now in common use.

Burkholder (5, p. 1019) reported that the following species are susceptible to infection by F. solani f. phaseoli: Phaseolus acutifolius var. latifolius Freeman., P. coccineus L., P. angularis Willd., P. acutifolius Jacq., lunatus f. macrocarpus (Benth) Van Ess., Vigna sinensis (L.) Endl., and Dolichos biflorus L. as well as P. vulgaris. Infection was light on P. coccineus and P. lunatus f. macrocarpus. Harter and Zaumeyer (13, p. 23) added Pueraria thunbergiana (Sieb. & Zucc.) Benth. to the host range. Reinking (22) isolated a strain of the dry root rot organism which proved to be a virulent pathogen of Pisum sativum L. He also demonstrated that strains of Fusarium solani f. pisi (F. R. Jones) Snyder and Hansen, which caused severe root infections on peas, were capable of infecting beans but produced only mild infections.

Burkholder (5, p. 1019) also reported that the following species and varieties are non-susceptible to infection by F. solani f. phaseoli: Pisum sativum L., Trifolium pratense L., T. hybridum L., Vicia sp., Three varieties of Soja max Piper, Zea mays L., Solanum tuberosum L. Avena sativa L. Triticum sp., Ambrosia artemisiifolia

L., Prunella vulgaris L. Chenopodium album L, and Rumex sp., These plants were tested by him because they were either used in rotation with bean or were weeds commonly found in the bean fields.

Moore (18, pp. 2-4) has reported that the damage due to root rot is in direct proportion to the depth of planting. He observed that infection was much more severe on bean plants from seed planted two inches deep than on those planted at one-half or one inch. This observation was confirmed by Walters and Burke (31), who reported that plants from seeds planted one inch deep showed less disease than those planted three inches deep.

Studies by Burkholder (8) on the effect of hydrogen-ion concentration on susceptibility of beans to dry root rot indicated that susceptibility was not affected by the hydrogen-ion concentration of the soil. Ion concentration appeared also to have little effect on the development of the casual organism.

Burkholder observed that plants infected with the dry root rot organism showed a greater reduction in yield when grown in dry than in medium wet or wet soils. Although healthy plants grown in dry soil produced normally upon the addition of moisture prior to blooming, infected plants gave little response under the same conditions.

The effect of frequency and extent of irrigation on the severity of dry root rot was studied by Walter and Burke (31) who reported that less root rot occurred when beans were given light, frequent irrigation than when given heavy, infrequent irrigation.

In Wyoming, Vaughn (10, p. 3) obtained no significant differences in the extent of dry root rot between plants receiving light frequent irrigations and those receiving heavy, less frequent irrigations. The light frequent irrigations, however, favored the production of adventitious roots.

Soil temperature is an important factor on the distribution and severity of many diseases (12, pp. 33-39) caused by soil-borne organisms. The effect of soil temperature on severity of dry root rot of bean was first studied by Reddick (21) in 1917. Using isolates of the pathogen obtained from Burkholder, he conducted experiments at soil temperatures of 15°, 22°, and 34° C., and showed that a 34 per cent reduction in yield was sustained by inoculated plants grown in the 22° C soil and a 25 per cent reduction in the plants grown at a soil temperature of 34° C. Neither healthy nor inoculated plants grown in the 15° C. soil produced any fruit. Burkholder (6) studied the effect of soil temperature on severity of the disease at soil temperature of 18°, 26°, and 33° C. A 46.6 per cent reduction was sustained by plants grown at a soil temperature of 18° C. and 52.6 per cent reduction from inoculated plants grown at a soil temperature of 26° C. No data were reported for the plants grown in the 33° C. soil as the plants were killed when the soil tanks became overheated. He concluded that there was little or no difference in severity of the disease between the two soil temperatures.



Harter and Zaumeyer (13, p. 23) are of the opinion that long rotations are an effective means of controlling the disease. A two or three year rotation is of little value, while one of six to eight years might be beneficial. No mention was made of crops that might be included in a good rotation plan or of the effects of other crop residues on the severity of the disease. Burkholder (5, p. 1027) conducted a survey of the bean fields in New York to determine if the crops grown in rotation with beans affected the disease to any extent, but was unable to draw any conclusions. Wilson (10, p. 5), using a seven year rotation, found that root rot was more prevalent when beans followed carrots, parsnips or beets. Soybeans and tomatoes, used as green manure, inhibited the growth of the root rot organisms. He noticed that beans grown after three years of alfalfa produced abnormal amounts of adventitious roots. His observation that continuous planting of beans resulted in the most root rot but the best yields, is contrary to reports by other workers (5, 13).

The first report of attempts to control the disease by the use of chemicals was given by Burkholder (5, pp. 1028-1030), who treated the soil with either calcium hypochlorite at the rate of 200 pounds per acre or a 1:100 solution of formaldehyde at the rate of 400 gallons to the acre. All plants in the treated soil were as severely infected as those in the untreated plots. In later trials he applied Cyanamid at rates of 50, 75, and 100 pounds to the acre drilled into the rows at time of planting. Control of the disease

was not obtained with any of the rates used, but germination was reduced and growth of the plants severely retarded. In 1923, Burkholder and Crosby (9, p. 23) stated that eradication of the dry root rot pathogen from the soil with the aid of chemical treatments was not possible since amounts of the fungicides sufficient to control the fungus were also injurious to the bean plant.

Between 1923 and 1947 there were no reports in the literature of control measures using chemicals. In 1947 Leach and Snyder (15) tested the effectiveness of chemicals applied to the seed in the rows at planting time. They obtained substantial reductions in the incidence of bean root rot with Dithane D 114 applied at the rate of one gallon per acre and the dust formulation (Dithane A 10) at the rate of two pounds per acre.

In 1950 Burke and Starr (3) in Wyoming reported encouraging results with the use of Dithane Z 78, Arasan, New Improved Ceresan, and Spergon applied as seed treatments. In the same year, however, Walters and Burke (31) reported that seed treatments with Arasan, Ceresan, and Dithane Z 78 did not give satisfactory control of the disease. Walters (30) reported in 1954 that significant control was not obtained in field trials using Ceresan, Arasan, Dithane Z 78, Spergon, Dow 9B, and a mixture of Arasan and Lindane as seed treatments. Watson (33) has reported that treatment of infested field soil with CBP 55 and formaldehyde gave good control of root rot of peas and beans. The rates of application of these chemicals were not given.

The importance of adventitious roots in keeping the infected plants alive has been mentioned by several workers (5, p. 1005) 13, p. 20). Menzies (17) is of the opinion that yield losses due to the disease can be reduced by hilling and by heavy irrigation to encourage the formation of adventitious roots.

### Rhizoctonia Root Rot

Rhizoctonia solani Kuhn, the imperfect stage of Pellicularia filamentosa (Pat.) Rogers, has been reported (13, p. 36) to cause root rot of bean in many of the United States and in several foreign countries. Infection of bean seedlings by this fungus usually results in damping-off. When older plants are infected sunken reddish-brown lesions develop on the main root. The fungus frequently enters the pith of the hypocotyl where it causes a brick-red discoloration.

The soil temperature relations of Rhizoctonia root rot have been studied by Richards (23), who found that while infection occurred at temperatures ranging from 9° to 29.5° C. the optimum soil temperature for disease development on beans was about 18° C. Because of this lower optimum temperature for disease development, bean root rot caused by R. solani would likely be found under conditions cooler than those favorable for development of dry root rot. That temperature relations may differ with different isolates of the fungus has been shown by Kendrick (14) who reported that in root rot of lima beans, caused by the same fungus, the soil

temperature relations may vary considerably with the race of the fungus.

Various isolates of the fungus may differ not only in temperature relations but also in degree of pathogenicity. Person (19) demonstrated a high degree of variation in the pathogenicity of R. solani isolates from beans, ranging from weak pathogens to very destructive ones. This same variation was observed on beans inoculated with isolates obtained from other hosts.

Crop rotation as a means of control of Rhizoctonia root rot is unsuccessful because the pathogen is parasitic on a large number of crops and weeds. Extreme care should be taken to avoid planting beans on ground that produced a crop of badly infected potatoes. Bean pods, resting on the soil, may become infected by the fungus and as a result, infection or infestation of the seed occurs. Some degree of control can be obtained if disease-free seed is used.

#### Fusarium Root Rot

In 1926, Weimer and Harter (34) described a disease caused by Fusarium aduncisporum Weimer and Harter, which was identical in symptoms with dry root rot. They described this as a new species because of morphological differences between its conidia and those of Fusarium solari f. phaseoli. In the Snyder and Hansen (27) system of classification of the genus Fusarium, F. aduncisporum is considered a synonym of F. solani f. phaseoli.



### Black Root Rot

Black root rot of beans caused by Thielaviopsis basicola (Berk. & Br.) Ferr., was reported by Burkholder (5, p. 1006), who found it in association with dry root rot. The symptoms of this disease are purple discolorations of the tap root early in the season, followed by coal-black lesions which vary from small streaks to cankers which half encircle the tap root.

Burkholder noted that black root rot generally occurred during the early part of the growing season and occasionally would reduce germination. Harter and Zaumeyer (13, p. 50) stated that black root rot has been observed so seldom and has been responsible for so little injury that no control measures have been worked out.

### Pythium Root Rot

A root rot caused by Pythium debaryanum Hesse. has been reported by Harter and Zaumeyer (13, p. 40). If the plants are attacked when very young, damping-off occurs and when half-grown plants are infected they usually wilt and die. Infection of older plants results in a semi-soft, colorless to dark-brown rot of the stem at the soil surface. While Pythium root rot rarely warrants control measures, Sherbakoff (13, p. 41) recommended a soil drench of a one per cent copper sulfate solution.

### Texas Root Rot

Bean has been reported (13, p. 47) to be highly susceptible to

Texas root rot caused by Phymatotrichum omnivorum (Shear) Dugg.

The above-ground symptoms include stunting, poor growth and sudden wilting. Symptoms on the underground parts are dark, sunken lesions. This disease, at present is limited to the southwestern United States.

## METHODS AND MATERIALS

Methods and materials which were utilized repeatedly throughout the course of these investigations will be described in this section. Those which were used only for a particular phase of the investigation are presented with the description and results of that phase.

### Culture Media

Potato dextrose agar, "PDA", was used for isolation and culture of the fungi associated with bean root rot and was prepared according to the following formula:

Liquid from 200 grams of boiled potatoes

Dextrose, 20 grams

Agar, 17 grams

Distilled water to one liter volume

The media varied from pH 6.4 to 7.0 after autoclaving for sterilization.

Acid PDA which would favor the growth of fungi over bacteria was prepared by the same formula except that after sterilization sufficient lactic acid was added to lower the pH range to from 3.5 to 4.0.

### Isolation

Prior to isolation attempts, all diseased bean roots were surface

sterilized by immersion in commercial sodium hypochlorite diluted one to ten with distilled water. At the beginning of the investigation an immersion period of two to four minutes was used; however, this resulted in such a copious growth of many fungi that the period of immersion was extended to five or six minutes. After the diseased tissue was removed from the chlorox solution it was washed in sterile water for one minute. This is not absolutely necessary as the sterilant is quite labile.

Sections of tissue approximately 2 mm by 2 mm were taken from the margin between healthy and diseased tissue whenever possible. These sections were pushed into the surface of the agar so that the tissue was exposed only at the surface of the medium. Ten sections were removed from each diseased root. Five of these were placed on PDA and five on acid PDA.

The cultures were incubated for three to ten days at room temperature. As colonies of fungi appeared hyphal tips were transferred to PDA slants and all isolated fungi were maintained on PDA slants. The cultures were examined to ascertain whether contamination had occurred. If contamination had occurred pure cultures were obtained by single spore isolation whenever possible. In the case of bacterial contamination, bacterial-free cultures of fungi were obtained by the method described by Raper (20).

#### Greenhouse Culture Methods

Bean plants (Phaseolus vulgaris L.) used in the various studies



were grown in a mixture of sandy loam and pulverized peat moss. This soil mixture was sterilized in an autoclave at 15 lbs. pressure for four to eight hours. After sterilization the mixture was watered and stored in the greenhouse for one to two weeks before being used. A complete commercial fertilizer was added at the rate of five grams for each No. 10 can of soil.

Unless otherwise stated, the FM-1 variety of beans, obtained from the Ferry-Morse Seed Company, Mountain View, California, was used throughout these investigations. The seeds were planted in No. 10 cans by placing them on the surface of the soil and covering them with a  $3/4$  inch layer of sterilized sand.

#### Inoculation Techniques

Artificial inoculations were made by the addition of water suspensions of spores to the seed and soil, placement of agar discs on which the fungi were growing next to the seed, and application of an agar-spore-water homogenate to the soil and seeds.

A water suspension of spores was prepared by adding sterilized distilled water to ten day old fungi cultures growing on PDA slants and shaking vigorously so that most of the spores were separated from the mycelium. By the addition of sterilized water, the spore concentration of the water suspensions was adjusted to one hundred thousand spores per ml. This concentration was determined with the aid of a haemocytometer. Ten ml. of spore suspension were applied to the seeds and soil in each No. 10 can.

The agar discs were cut from agar containing ten-day old cultures of the fungi. One agar disc was placed adjacent to or under each bean seed at planting time.

The agar-spore-water mixture was made by homogenization of seven to ten day old cultures with distilled water for one minute in the Waring Blendor. Additional distilled water was added to adjust the concentration of spores to one hundred thousand spores per ml. Ten to twenty ml. of this homogenate was added to the seeds and soil in each No. 10 can.

While inoculations appeared to be equally successful with all three methods, the first two were rather tedious. The water suspension and agar disc methods were therefore used mainly for determination of pathogenicity of the various fungal isolates. The agar-spore-water mixture was used primarily for the fungicide tests where inoculum was needed for several hundred cans of soil. In the 1955 fungicidal field trials the seeds were soaked in the agar-spore-water homogenate for several hours prior to planting.

In rechecking the possible pathogenicity of those isolates which showed negative results with the above methods of inoculation, wounding was employed prior to application of the inoculum. After removing the soil from a portion of the hypocotyl a wound was made with a dissecting needle. Water suspensions of spores was applied to the wounded hypocotyl and soil was then replaced around the wounded area.

### Standard Fusarium solani f. phaseoli Culture

A culture of F. solani f. phaseoli (YA-2), isolated from an infected bean root from Yamhill County was used in the temperature, estimation of yield losses, host range and laboratory and greenhouse control studies.

### Identification of Fusarium sp.

Positive identification of species within this group has been most difficult since an extensive knowledge of the key characteristics of the various species is a prerequisite to accurate identification. Preliminary identification to sections within the genus was made with the aid of keys by Wollenweber et al (35). Snyder and Hansen (26, 27, 28), in pointing out the unreliability of such key characters as conidial size and shape, type of chlamydospores, stromatol color and relative frequency of micro and macroconidia, have revised the genus so that it contains only nine species. Each species of the Snyder and Hansen system is made up of one or more sections of the Wollenweber system of classification. While the system of classification as proposed by Snyder and Hansen has greatly facilitated identification of members of this genus, the author was hesitant about stating that an isolate definitely was a particular species. Isolates, representing four groups of Fusarium associated with bean root rot were sent to Dr. Snyder at the University of California for positive identification. He confirmed all species identifications.

The identification to form follows the proposed use of host relationships as a basis upon which to separate the pathogenic forms within morphological species or units as outlined by Snyder and Hansen. However, as pointed out by Reinking (22), cross infection of host plants can occur between different pathological forms of F. solani, but infection is more severe on the host for which the form was named. In this study two distinct groups of F. solani were isolated; one group composed of virulent pathogens and the other of weak or non-pathogenic isolates. In order to distinguish between these two groups, the former is identified as F. solani f. phaseoli and the latter as F. solani.

#### Disease Classification

A system for rating disease severity was established to evaluate the extent of root rot in field and greenhouse studies. The ratings, based on extent of surface discoloration of the underground portion of the hypocotyl and the upper tap root, is as follows:

- 0 No infection
- 1 Infection on less than 20 per cent of the surface area
- 2 Infection on more than 20 per cent but less than 40 per cent
- 3 Infection on more than 40 per cent but less than 60 per cent
- 4 Infection on more than 60 per cent but less than 80 per cent
- 5 Infection on more than 80 per cent

This classification is intended for use only with plants up to six weeks of age and is of little value of rating the severity of the



disease in older plants.

## RELATIVE IMPORTANCE OF FUNGI ASSOCIATED WITH BEAN ROOT ROT

During the course of these investigations many bean fields were visited to determine the seriousness of the disease. In the summer of 1953 a preliminary survey was made of four bean fields in Lane County. This survey was extended to include all of the Willamette Valley during the summers of 1954 and 1955. All together, seventy eight bean fields, ranging in size from four to over two hundred acres, have been observed.

Bean roots showing root rot symptoms were collected from all of these fields. To determine the fungi associated with bean root rot and their relative frequency, isolations were made from bean roots representing all seventy eight fields.

Fusarium was the dominant genus among the isolates (Table 1). Approximately 250 isolates of F. oxysporum and F. solani f. phaseoli were obtained. The number of isolates of F. roseum and F. solani was 57 and 29 respectively. While a complete description of the macro and microscopic characteristics of these isolates would be desirable as reference for other workers, there is so much variation in characteristics, particularly among the isolates of Fusarium roseum and F. oxysporum, that the descriptions would be of little value. A short description of the isolates of F. solani and F. solani f. phaseoli is given below.

Short Description of Fusarium Solani f. phaseoli

Spores borne mostly in pseudopionnotes. Mature cultures blue or pink in color at room temperature. If stored at 32° F. the cultures will be yellowish to white in color. While the predominating number of septations of macroconidia of ten day old cultures is three, certain isolates produce predominately four-septate macroconidia. Macroconidia slightly curved near the rounded apex. Microconidia occasionally are abundant on five day old cultures, but usually are rare on older cultures. Chlamydospores terminal and intercalary, occasionally in chains.

Fusarium solani differs from the description given above in that the mature cultures are white at room temperature, production of spores in pseudopionnotes is rare and macroconidia are predominantly tri-septate.

Fungi isolated most frequently, other than Fusarium sp. included, in decreasing order of frequency: Phoma sp., Rhizoctonia solani, Botrytis sp., Penicillium sp., Trichoderma sp., Pythium sp., Mucor sp., and Sclerotinia sclerotiorum (Lib.) DBy. With the exception of Phoma sp. and Rhizoctonia solani all of the isolates occurred infrequently in the various bean fields. Botrytis sp. was isolated only from bean roots obtained from Washington, Multnomah and Clackamas Counties. Sclerotinia sclerotiorum was isolated from fields having past histories of severe white mold infection.

Table 1. Frequency of isolation of fungi from bean plants infected with root rot in the Willamette Valley in 1953, 1954, and 1955

	1953	1954	1955	Total
<i>Fusarium</i> sp.	60	250	290	600
<i>F. solani</i> f. <i>phaseoli</i>	21	122	109	252
<i>F. roseum</i>	9	17	31	57
<i>F. oxysporum</i>	27	102	133	262
<i>F. solani</i>	3	9	17	29
<i>Rhizoctonia solani</i>	4	24	29	57
<i>Pythium</i> sp.	12	7	19	38
<i>Trichoderma</i> sp.	3	24	17	44
<i>Aspergillus</i> sp.	0	3	0	3
<i>Phoma</i> sp.	0	32	41	73
<i>Mucor</i> sp.	0	13	25	38
<i>Pencillium</i> sp.	0	29	17	46
<i>Botrytis</i> sp.	0	19	32	51
<i>Stemphylium</i> sp.	0	2	11	13
<i>Sclerotinia sclerotiorum</i>	8	9	21	38
<i>Cladosporium</i> sp.	0	3	0	3
<i>Gliocladium</i> sp.	0	2	0	2
<i>Alternaria</i> sp.	0	6	8	14
<i>Rhizopus</i> sp.	<u>0</u>	<u>3</u>	<u>12</u>	<u>15</u>
Total identified	87	426	522	1035
Unidentified fungi	12	17	8	37
Bacteria	0	23	39	62
Sterile or contaminated	31	52	31	114
No. of roots from which isolates were made	130	518	600	1248



### Pathogenicity of Isolates

Of all the isolates tested, only representatives of Fusarium sp., (Figures 1 and 2), Rhizoctonia solani, (Figure 2), Botrytis sp., Sclerotinia sclerotiorum and possibly Pythium sp. produced disease symptoms on the bean plants. Inoculation with the isolates obtained in 1953 were made on Germain's 21 variety of bean. All subsequent inoculations were made on the variety FM-1.

### Pathogenicity of *Fusarium solani* f. *phaseoli* isolates

Pathogenicity was demonstrated for 203 isolates of this fungus. The greenhouse symptoms produced on FM-1 beans inoculated with these isolates were nearly identical with those described for severe field infections. Occasionally artificial infection resulted in death of the young plants, a symptom not encountered in the field. The reddish brown discoloration of the pith described under field symptoms was not encountered on any of the inoculated beans.

Two isolates selected at random from each county were used in a test to see if differences in pathogenicity could be detected. Fifty seeds of FM-1 beans were inoculated with each isolate by the agar-spore-water homogenate method at planting time. Disease ratings of the plants were made 28 days after planting.

While there are some differences in the average disease rating and range of disease rating there is no noticeable difference in pathogenicity of the various isolates (Table 2). The small differences in disease rating probably are due to the inaccuracy of

Table 2. Relative pathogenicity on FM-1 beans of two randomly selected isolates of Fusarium solani f. phaseoli from each county in the Willamette Valley

Isolates	Range of disease rating	Mean disease rating*
Lane		
LA 5c	2-5	4.18
LD 9	3-5	4.32
Linn		
LiE 11	3-5	4.26
LiA 3b	4-5	4.64
Marion		
MaB 8	3-5	4.42
MaC 2	3-5	4.36
Clackamas		
CD 7	3-5	4.18
DA 5a	3-5	4.44
Multnomah		
ME 13	3-5	4.26
MD 1	3-5	4.42
Washington		
WA 3	4-5	4.58
WC 8	3-5	4.32
Yamhill		
YA 2a	3-5	4.28
YB 7	3-5	4.28
Polk		
PB 5a	3-5	4.16
PA 6	3-5	4.48
Benton		
BC 7	2-5	4.12
BA 8c	3-5	4.54

\* based on 50 plants

the arbitrary disease rating classification. If major differences in pathogenicity of the isolates had occurred they would have been detected by this disease classification system.

#### Pathogenicity of Fusarium solani isolates

Less than 30 per cent of the isolates in this group proved to be pathogenic (Table 3). Those that did produce infection might well be considered as weak pathogens since very little damage to the bean plant was noted. Wounding of the plants prior to inoculation had no effect on the extent of pathogenicity of the isolates except on one wounded plant which developed lesions. Attempts to re-isolate the organism from this plant were unsuccessful.

FM-1 bean plants inoculated with the pathogenic isolates developed small, reddish brown, pin point lesions on the hypocotyl and tap root. The lesions were indistinguishable from those found on beans inoculated with isolates of F. solani f. phaseoli except that the lesions did not spread or coalesce. In no instance were more than five lesions noted on any one plant, whereas the number of lesions produced by F. solani f. phaseoli frequently was a hundred or more. No attempt was made to determine the pathogenicity of these isolates on other crops so that identification to form could be given.

#### Pathogenicity of Fusarium Oxysporum Isolates

No attempt was made to determine the pathogenicity of these

Table 3. Results of inoculation attempts on FM-1 beans with selected isolates of Fusarium solani

Isolate	Wounded	Not wounded
LB 3	0/6*	0/5
L4C 11	0/6	0/6
MaA 7	5/5	3/5
CC 12	0/4	0/6
MC 3	0/6	0/6
WA 8	1/6 <sup>x</sup>	0/5
PB 10	0/6	0/6
BC 4	6/6	6/6

\* The number of plants inoculated is given as denominator while the numerator denotes the number of plants showing infection.

<sup>x</sup> Culture not reisolated.

isolates on other crops so that identification to form cannot be given. Since several pathological forms are probably represented in this group, a variation in the relative pathogenicity of the isolates was anticipated. Symptom expression on FM-1 beans inoculated with the F. oxysporum isolates ranged from reddish brown pin point lesions to lesions covering almost 25 per cent of the surface area of the tap root and below ground portion of the hypocotyl. In no instance was the infection so severe that destruction of the tap root occurred. Approximately 50 per cent of



all the isolates proved to be pathogenic (Table 4). Some isolates were pathogenic only on wounded plants.

#### Pathogenicity of *Fusarium roseum* isolates

As in the case of *F. oxysporum* isolates of this species were not identified to form.

Pathogenicity was demonstrated for less than 50 per cent of the *F. roseum* isolates (Table 5). Wounding of plants before inoculation resulted in an increase in the number of plants infected. Symptoms produced on FM-1 beans by the pathogenic isolates were reddish brown pin point lesions indistinguishable from those produced by isolates of *F. solani* and certain isolates of *F. oxysporum*. Very little spread of these lesions occurred as the bean plants matured.

#### Pathogenicity of *Rhizoctonia solani* isolates

While approximately 60 per cent of the *R. solani* isolates were shown to be pathogenic (Table 6), they proved to be weak pathogens. Symptoms produced by the pathogenic isolates were small sunken reddish brown lesions on the below ground portion of the hypocotyl. Very little spread of these lesions occurred. None of the symptoms produced by these isolates approached in extent those described in the literature. None of the isolates produced reddish brown discoloration of the pith.

Table 4. Results of inoculation of FM-1 beans with selected isolates of Fusarium oxysporum

Isolate	Wounded	Not wounded
BA 10	5/5*	5/5
BE 5	3/6	2/5
LiA 13	0/6	0/6
LiD 1	4/5	4/4
WE 7	0/5	0/6
WB 2	3/6	0/6
MF 14	3/4	6/6
MF 6	0/6	0/6
CD 8	5/6	4/6
GA 1	3/5	0/5
MaC 15	4/5	5/5
MaA 4	5/6	3/5
PD 9	1/6 <sup>x</sup>	0/5
PC 2	0/5	0/6
LB 16	0/5	0/5
LA 1	5/5	4/5
YA 9	0/6	0/6
YC 11	0/6	0/5

\* The number of plants inoculated is given as the denominator, while the numerator denotes the number of plants showing infection.

<sup>x</sup> F. solani f. phaseoli isolated.

Table 5. Results of inoculation of FM-1 beans with selected isolates of Fusarium roseum

Isolate	Wounded	Not wounded
LC 6	4/6*	2/5
LB 1	0/6	0/5
L4C 7a	5/5	3/4
MaB 12	6/6	5/5
CF 11	0/6	0/6
CE 9	2/4	0/6
MH 1a	0/6	0/5
Md 13	0/6	0/6
WG 8	5/5	6/6
YD 7c	0/6	0/6
YA 4	0/4	0/5
PB 6	0/6	0/6
BD 4d	3/4	3/5
BA 5	0/6	0/6

\* The number of plants inoculated is given as the denominator, while the numerator denotes the number of plants showing infection.

Table 6. Results of inoculation of FM-1 beans with selected isolates of Rhizoctonia solani

Isolate	Wounded	Not wounded
LC 9	6/6*	4/5
LD 1a	0/6	0/6
LiC 6	0/6	0/5
LiD 13	0/6	0/6
MaD 3b	0/6	0/6
MaA 9	4/6	3/6
CB 5	4/5	2/5
CA 12	3/6	0/5
MB 17	2/6	0/6
MA 1c	0/6	0/6
WA 9a	4/5	4/6
WD 3	0/6	0/6
YA 9	6/6	6/6
YB 5c	3/5	3/6
PA 8	0/6	0/6
PC 2b	0/5	0/6
BB 10	0/4	0/6
BC 4d	0/6	0/6

\* The number of plants inoculated is given in the denominator while the numerator denotes the number of plants showing infection.



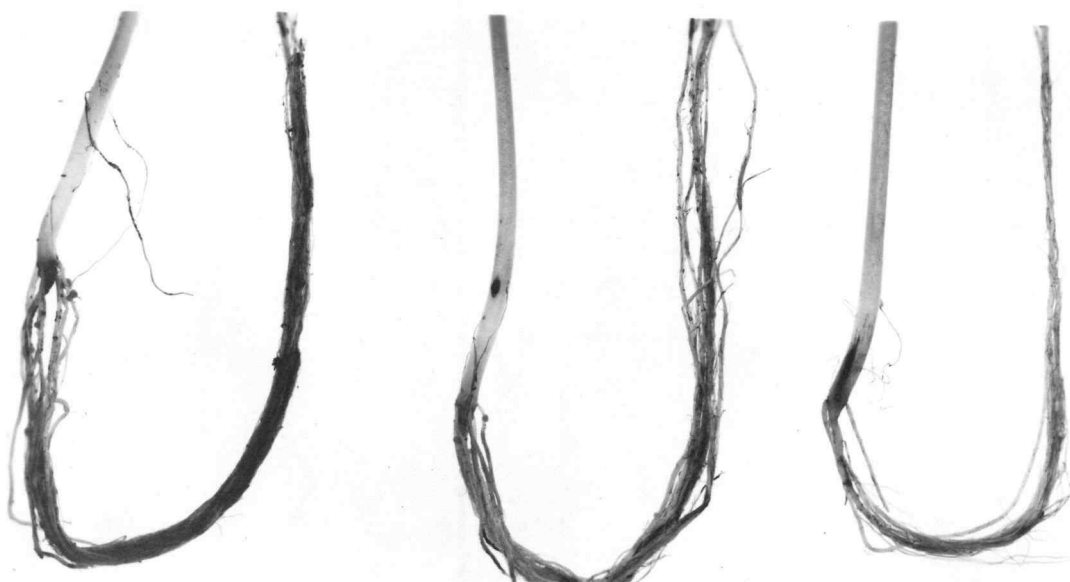


Figure 1. FM-1 bean plants inoculated with Fusarium roseum (left) Fusarium solani (center) and Fusarium oxysporum (right).

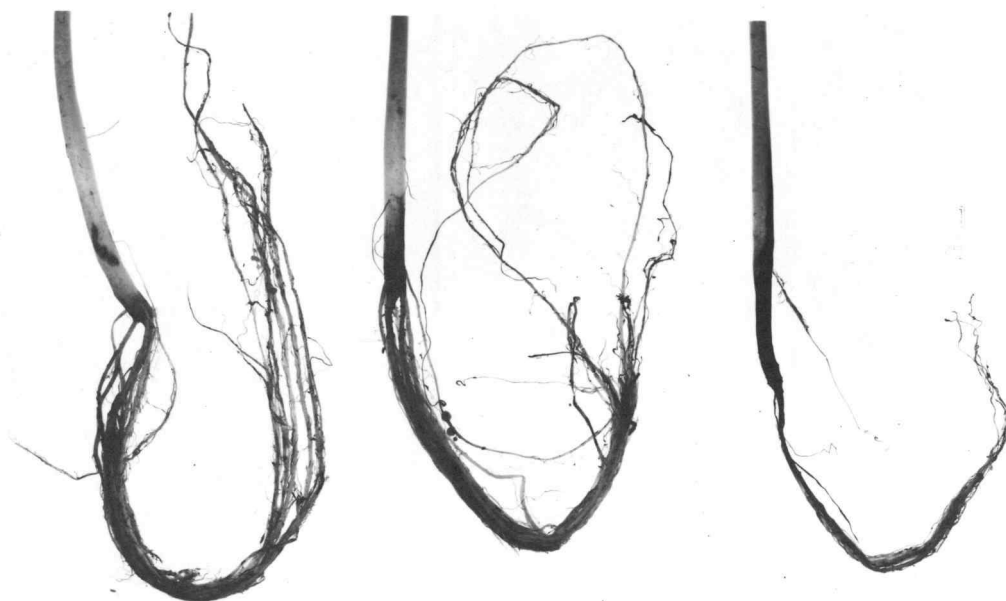


Figure 2. FM-1 bean plants inoculated with Rhizoctonia solani (left) and Fusarium solani f. phaseoli.

#### Pathogenicity of Botrytis sp. isolates

Most of the isolates of this genus were shown to be pathogenic. Infection brought about a wilting of the FM-1 bean seedlings. Several days after wilting was first noted the plants were dead and the fungus was fruiting abundantly on the above ground portion of the hypocotyl. Since no noticeable symptoms were observed on the below ground portions of the plants the author does not consider this fungus a root rot pathogen.

#### Pathogenicity of Sclerotinia sclerotiorum isolates

All of the isolates of S. sclerotiorum proved to be pathogenic. The symptoms produced by these isolates on FM-1 bean were small watery lesions at the soil line. Several days after the first appearance of these spots most of the plants were girdled by the disease. Discoloration and presence of the fungus on the tap root and hypocotyl was not noticeable until the plants were killed. Since typical root rot symptoms were not associated with infection by S. sclerotiorum, this fungus is not considered a root rot pathogen.

#### Pathogenicity of Pythium sp. isolates

Several FM-1 bean plants inoculated with isolates of Pythium sp. developed light brown watery lesions on the tap roots. While

these symptoms are similar to those described in the literature (12, p. 36), definite proof of pathogenicity was not determined as attempts to reisolate the organisms was unsuccessful. Later inoculation attempts with these isolates were unsuccessful.

#### Duplication of Reddish Brown Discoloration of the Pith

During the course of studies on the effects of combinations of pathogens on disease development, which will not be presented in this thesis, one observation was recorded which should be mentioned here. Two plants inoculated with a combination of F. solani f. phaseoli and F. oxysporum, developed a reddish discoloration of the pith as described under field symptoms of the disease. In the literature (12, p. 41) this type of reddish discoloration of the pith has been attributed to infection by Rhizoctonia solani.

#### Distribution of Root Rot Fungi by County

In order to obtain a better concept of the distribution of root rot organisms within the Willamette Valley, their distribution is summarized in Table 7. Isolates of Fusarium solani f. phaseoli and F. oxysporum were obtained from 77 of the 78 fields inspected. Rhizoctonia solani, F. roseum and F. solani were found in 35, 24, and 14 fields respectively.

The fungi obtained from diseased bean roots and shown to cause bean root rot included: Fusarium solani f. phaseol. F. oxysporum, F. roseum, F. solani and Rhizoctonia solani. Fusarium solani f.

phaseoli is considered as the dominant pathogen since most field symptoms have been reproduced on beans inoculated with pure cultures of this fungus. While the other fungi were capable of causing root rot, the disease caused by them was mild.

Table 7. Distribution of isolates of Fusarium solani, F. solani f. phaseoli F. roseum F. oxysporum and Rhizoctonia solani in the nine counties of the Willamette Valley

County	<u>F. solani</u>	<u>F. solani</u> f. <u>phaseoli</u>	<u>F. roseum</u>	<u>F. oxysporum</u>	<u>R. solani</u>
Benton	2/1*	9/5	8/3	27/5	4/4
Clackamas	3/1	17/6	14/6	21/6	5/2
Lane	6/2	42/11	5/2	37/11	9/7
Linn	2/1	29/9	0/0	29/8	3/2
Marion	1/1	37/15	8/5	43/15	9/5
Multnomah	9/3	46/10	11/4	38/10	11/6
Polk	1/1	26/7	4/1	32/8	5/2
Washington	5/4	27/7	0/0	16/7	5/3
Yamhill	0/0	21/7	7/3	17/7	5/4

\* The number of fields from which the fungi were obtained is given as the denominator. The numerator denotes the number of isolates.



## THE EFFECT OF SOIL TEMPERATURE ON THE SEVERITY OF DRY ROOT ROT

Although studies on the effect of soil temperatures on this disease have been reported by Reddick (21) and Burkholder (6), only two temperatures were studied in each case. This investigation was designed to determine the effect of six soil temperatures ( $16^{\circ}$ ,  $20^{\circ}$ ,  $24^{\circ}$ ,  $28^{\circ}$ ,  $32^{\circ}$ , and  $36^{\circ}$  C.) on severity of dry root rot.

Soil tanks were used to maintain the soil at the desired temperature. The  $16^{\circ}$  and  $20^{\circ}$  C. tanks were equipped with two by five foot refrigerator plates, and refrigeration was supplied by a one-quarter horse-power refrigeration unit. Temperature in each tank was controlled by individual thermostats and solenoid valves. Heating of the other four tanks was accomplished by spacing two sixty foot lead heating cables, of 400 watt capacity, on the bottom of each tank. The cables were activated by Fenwal thermostats. Circulation of the water within the tanks was accomplished by the use of one-thirtieth H.P. water pumps.

The soil containers used were water-proofed earthen crocks of three gallon capacity. A rubber drainage tube was inserted in the stopper in the bottom of the crock and connected to an exhaust pipe to carry away excess water. A watch glass was placed over the opening at the bottom of each crock so that sand and soil would not plug the outlet, and a four inch layer of sterilized sand was placed in the bottom of the crock to allow for adequate drainage. A six inch layer of sterilized potting soil was added next, and the

bean seeds were then planted on the surface of the soil and inoculated with culture YA 2 by the agar-disc method.

Burpee's Stringless bush bean was used in these tests in preference to the varieties grown in the Willamette Valley. Bush beans were easier to handle in the soil temperature tanks and the period of maturation of the fruits of this variety is short. Previous inoculation tests had shown that this variety was equally as susceptible to dry root rot as the varieties in commercial use. At each temperature, six crocks of this variety were inoculated and six served as controls.

With the exception of the 36° C. tank, water temperatures were maintained within  $\pm 2^{\circ}$  C. of the desired temperature and seldom varied more than  $\pm 1^{\circ}$  C. A week after the start of this investigation the 36° C. tank was found to be empty because of a broken drainage connection. This tank was refilled with hot water and adjusted to temperature within four hours.

The diurnal air temperature ranged from 16° C. to 26° C. during the course of this investigation. An attempt was made to maintain the daytime temperature between 20° C. and 25° C. Except for brief periods of intense illumination, this was accomplished by manual operation of the ventilators at various intervals during the day. Soil moisture was maintained by daily waterings, and excess water rapidly drained away.

The study was started December 4, 1954 and terminated on February 17, 1955. Recordings of soil and air temperature were

taken at least three times daily. The first seedlings to emerge were in the 32° and 36° C. tanks on December 6, 1954. All seeds had germinated by December 12, 1954.

During the course of these investigations there was considerable yellowing and dropping of the lower leaves of plants at all soil temperatures of both inoculated and check plants. This was thought to be due to the fact that the seeds were planted too soon after sterilization of the soil.

Records were taken of the severity of disease (Figures 3-8) and height of plants and yields (Table 8). There was considerable variation in the percentage of inoculated plants that produced fruit at the various temperatures. The plants grown in the highest (36° C.) and lowest (16° C.) soil temperatures showed the greatest percentage of yielding plants. The percentage of yielding plants was lowest (23.5 per cent) in the 28° C. tank. Fruit was produced by the uninoculated plants at all soil temperatures.

The greatest reduction in yield of inoculated plants occurred in the 28° C. tank, where an 86.5 per cent decrease was recorded. The least decreases occurred at the 16° C. soil temperature with a reduction of 39.3 per cent, closely followed by the 36° C. with a 41.0 per cent decrease.

The greatest reduction in height of the inoculated plants occurred in the 28° C. tank, closely followed by that of the plants in the 32° C. tank. Among the uninoculated plants the greatest mean height was obtained in the 28° C. tank and as the soil

temperature increased or decreased the mean height of the plants decreased.

Observation of the roots after harvest revealed that all inoculated plants, at all soil temperatures, showed severe root rot symptoms that completely covered the tap root and the below ground

Table 8. The effect of soil temperature on survival, height, and yield of Burpee's Stringless bush bean inoculated with Fusarium solani f. phaseoli

Temperature C°	Per cent of plants forming fruit		Mean height in cm		Mean decrease in yield
	C <sup>1</sup>	I <sup>2</sup>	C	I	
16	100	91.8	31.2	29.8	39.3
20	100	72.7	37.4	33.0	47.2
24	100	50.0	38.9	33.9	63.7
28	100	23.5	41.9	28.5	96.5
32	100	58.8	38.6	29.3	81.0
36	100	86.5	36.1	33.1	41.0

<sup>1</sup> Check

<sup>2</sup> Inoculated

portion of the hypocotyl. Most of the tap roots of the inoculated plants in the 28°, 32°, and 24° C. tanks were completely destroyed. Adventitious roots were produced abundantly by the plants grown at these soil temperatures. Less than five per cent of the tap roots of the inoculated plants in the 16°, 20°, and 36° C. were completely destroyed. The roots of five check plants showed symptoms of root



rot infection and the data obtained from these plants were eliminated.

Optimum disease development occurred at a soil temperature of 28° C. However, a greenhouse study of soil temperature effects on disease development is only of theoretical value unless a comparison is made with soil temperatures in the field. Field soil temperatures at the Vegetable Crops Experimental Area in Corvallis were recorded for the seasons of 1954 and 1955 (Table 9). During these two seasons the monthly averages of the mean soil temperatures at the two to three inch level ranged from 17.2° to 22.2° C. and the mean maximum soil temperatures from 20.0° to 27.8° C. The monthly mean minimum soil temperatures ranged from 12.2° to 18.3° C. The monthly average mean soil temperatures at the five to six inch level ranged from 16.1° to 22.2° C. and the mean maximum soil temperatures from 16.7° to 24.4° C. The monthly mean minimum soil temperatures ranged from 15.0° to 20.0° C.



Table 9. Mean soil temperature data, recorded in degrees centigrade for the 1954 and 1955 bean growing season at the Vegetable Crops Experimental Area, Corvallis, Oregon

	2-3 inch level			5-6 inch level		
	Min.	Max.	Mean	Min.	Max.	Mean
<u>1954</u>						
May 15-31	12.2	23.9	17.7	15.5	19.4	17.7
June 1-30	12.8	22.8	17.7	15.0	18.9	17.2
July 1-30	16.1	27.8	22.2	19.4	24.4	22.2
August 1-30	16.1	26.1	21.7	19.4	23.3	21.7
September 1-15	14.4	22.8	18.9	17.2	21.1	19.4
<u>1955</u>						
May 15-31	13.9	20.0	17.2	15.0	16.7	16.1
June 1-30	16.7	22.8	19.4	17.7	19.4	18.9
July 1-30	17.2	23.9	20.5	18.9	21.1	20.0
August 1-30	18.3	26.1	21.7	20.0	22.2	21.1
September 1-15	17.7	23.9	20.5	20.0	21.1	20.5

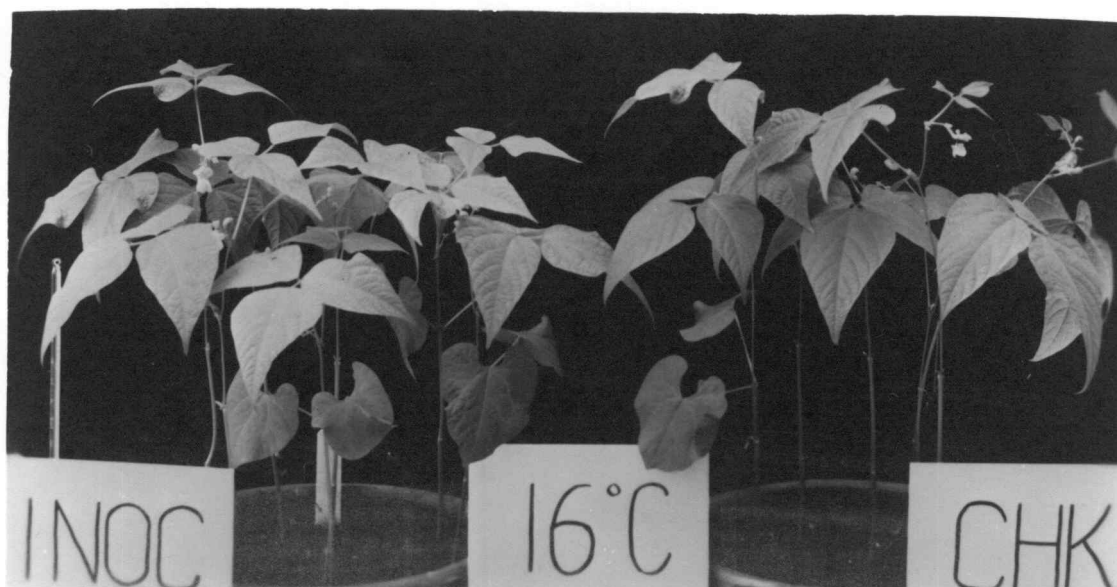


Figure 3. Comparison of diseased and healthy bean plants grown at a soil temperature of 16° C.

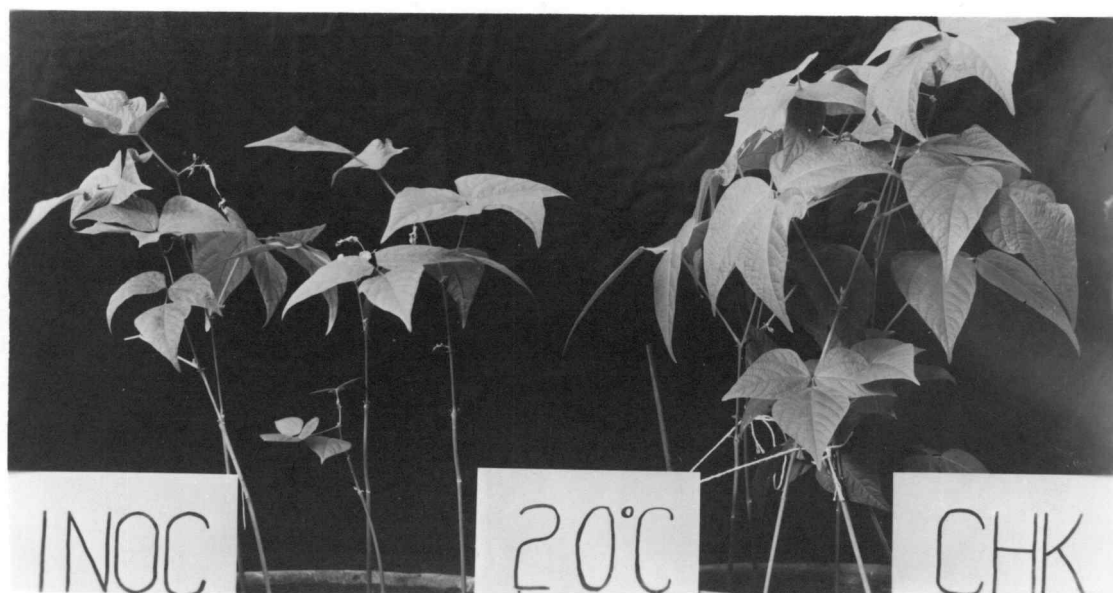


Figure 4. Comparison of diseased and healthy bean plants grown at a soil temperature of 20° C.



Figure 5. Comparison of healthy and diseased bean plants grown at a soil temperature of  $24^{\circ}\text{C}$ .



Figure 6. Comparison of healthy and diseased bean plants grown at a soil temperature of  $28^{\circ}\text{C}$ .

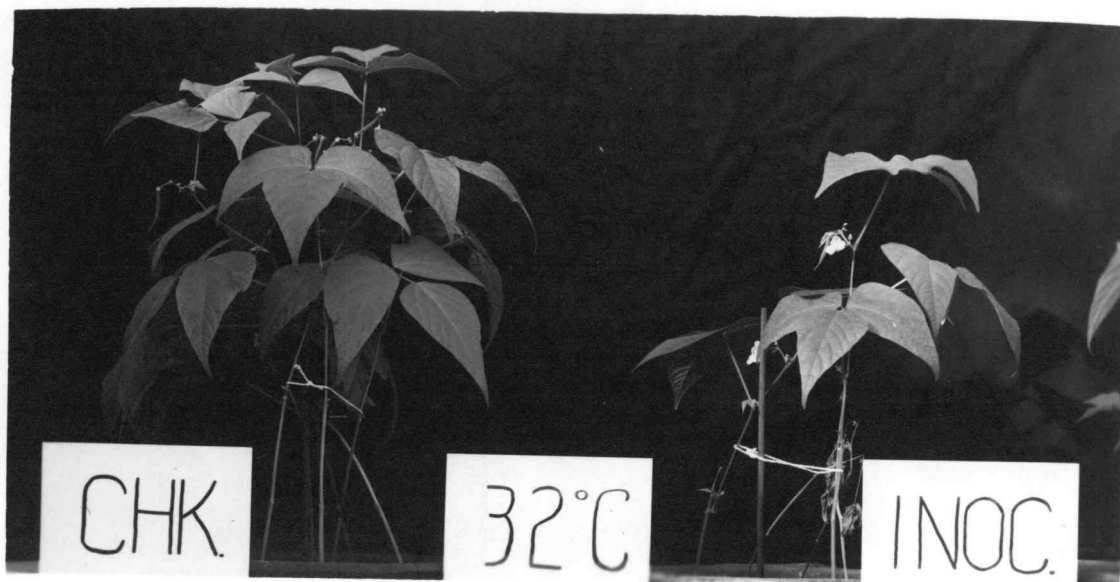


Figure 7. Comparison of healthy and diseased bean plants grown at a soil temperature of 32° C.



Figure 8. Comparison of diseased and healthy bean plants grown at a soil temperature of 36° C.

## ESTIMATION OF POTENTIAL FIELD YIELD LOSSES

An attempt was made to estimate disease losses in the field from data recorded during the field survey and yield data submitted by the growers at the end of the season. No correlation was found between disease ratings recorded early in the season and yields obtained. In some cases, the yields of fields with a low disease rating were several tons per acre less than from fields with higher disease ratings. A reliable comparison of disease severity and yields under field conditions could not be made without considering all other factors. Such factors as cultural practices, other diseases and variety of bean grown would directly influence the yield obtained. Since this information was not always available, estimates of losses were based on data obtained from greenhouse studies. While greenhouse data are not always applicable to field conditions, they may at least lend support to estimates of damage in the field.

Yield data of Burpee's Stringless bush beans, inoculated with Fusarium solani f. phaseoli and grown at different soil temperatures (Table 8) give an indication of losses which might be encountered as a result of infection under field conditions. However, these yield data were obtained from inoculated plants grown in sterilized soil and do not take into consideration all factors which might be encountered in field soils. Consideration of these data alone, might result in misleading estimates of disease damage.



The importance of microbiological factors in the soil must be considered in obtaining reliable estimates of yield losses due to disease. In sterilized soil the spread and development of the pathogenic organisms are unhampered by the presence of other organisms, while in unsterilized soil the microflora is quite complex and the various organisms are in competition for space and nutrients. Garrett (11), in his work with cereal root rot fungi, has demonstrated the importance of this complex microflora on disease severity. Burke (2), working with isolates of Fusarium solani f. phaseoli, reported a decrease in severity of infection on beans when inoculum was added to root rot-free soil as compared with plants in inoculated sterilized soil.

A study of yield losses was undertaken in the greenhouse, and consideration was given to the possible buffering action of soil microflora on the rapid development of disease. Soil samples obtained from the bean field at the Department of Botany and Plant Pathology farm were examined to determine the dominant fungi and bacteria. Isolation of these organisms was made by a modification of the method described by Warcup (32). Media prepared by adding five grams of yeast extract to a liter of PDA was adapted for isolating bacteria by adding crystal violet (25, p. 30) at the rate of 10 p.p.m. to discourage rapid growth of fungi. The pH of the media used for isolation of fungi was adjusted with lactic acid to pH 4 to eliminate growth of bacteria.

Fifteen ml of cooled media were added to five milligrams of soil in each petri dish and shaken to disperse the soil particles throughout the medium. Fifty petri plates, half with acid media, were incubated at room temperature and as soon as growth of micro-organisms was apparent transfers were made to PDA slants.

The fungi most commonly isolated from the soil included:

Aspergillus sp., Penicillium sp., Stemphylium sp., Alternaria sp., Chaetomium sp., Fusarium sp., Phoma sp., Graphium sp., Cephalosporium sp., Hormodendrum sp., Gliocladium sp., Cunninghamella sp., and Trichoderma sp. These fungi were identified to genus with the aid of Barnett's manual (1). With the exception of Fusarium sp., all these fungi were used in this study.

Because of the large array of media needed for identification of bacteria and because they were being used only as a research aid, no attempt was made to identify the bacterial cultures. The seven bacterial cultures most frequently encountered on the soil plates were maintained for greenhouse studies.

Three stock inocula were prepared by the agar-spore-water method and consisted of: 1. soil fungi, 2. bacteria, and 3. F. solani f. phaseoli. The inocula used in this study consisted of: 1. F. solani f. phaseoli, 2. F. solani f. phaseoli plus fungal and bacterial isolates, 3. F. solani f. phaseoli plus fungal isolates, 4. F. solani f. phaseoli plus bacterial isolates and 5. fungal and bacterial isolates. These inocula were prepared by mixing 200 ml of each of the desired stock inoculum.

Each inoculum was mixed with sufficient soil to fill ten No. 10 cans. After potting, the soil was watered and stored in the greenhouse for a month. The soil was checked periodically during this time to see that it did not become dry and water was added when needed.

The FM-1 bean seeds used in this study were planted as described in the section on general methods. Lath stakes, four feet in length, were used to support the bean vines. Roots of plants from five cans of each treatment were checked at the end of one month, at which time data was recorded on the severity of disease by use of the disease rating classification. Yield data were recorded from five cans of plants ninety five days after planting (Table 10).

The addition of fungi and bacteria to inocula containing the dry root rot pathogen resulted in a 22.8 per cent yield decrease as compared to a 38.4 per cent decrease when the pathogen was used alone. The addition of fungi and bacteria singly to the inoculum containing the pathogen resulted in a 24.1 and a 29.5 per cent yield decrease respectively.

Table 10. Comparison of disease ratings and yield decreases of FM-1 beans grown on soil infested with Fusarium solani f. phaseoli alone, and in combinations with bacterial and fungal isolates. Soil temperatures ranged from 18° to 26° C.

Inocula	Mean disease ratings	Mean per cent yield decreases
<i>F. solani</i> f. <i>phaseoli</i>	4.38	38.4
<i>F. solani</i> f. <i>phaseoli</i> plus bacterial and fungal isolates	2.85	22.8
<i>F. solani</i> f. <i>phaseoli</i> plus fungal isolates	3.15	24.1
<i>F. solani</i> f. <i>phaseoli</i> plus bacterial isolates	3.69	29.5
Check-Bacterial and fungal isolates	0.00	—

## HOST RANGE STUDIES

Severe root rot has been observed in bean fields where eight to twelve years have passed since the field was previously planted to beans. The maintenance of a high level of dry root rot inoculum for such a long period of time suggests the possibility that other plants may become infected and thus aid in maintaining the high level. To examine this possibility more closely a greenhouse study was undertaken in which attempts were made to infect horticultural crops used in rotation with beans and also weeds commonly found in and around bean fields.

The following crop plants were used in this study: barley (Hordeum vulgare L.), beets (Beta vulgaris L.), broccoli (Brassica oleracea var. botrytis L.) cabbage (Brassica oleracea var. capitata L.), carrots (Daucus carota L. var. sativa D.C.), corn (Zea mays L.), dill (Anethum graveolens L.), hops (Humulus lupulus L.), oats (Avena sativa L.), peas (Pisum sativum L.), rye (Secale cereale L.), and strawberry (Fragaria vesca L.). While peas are not grown in rotation with beans in the Willamette Valley they were included to determine if results obtained by Reinking (22) could be duplicated with the author's isolate of F. solani f. phaseoli.

Weeds used in the study included: Chenopodium album L., Polygonum hydropiper L., Festuca myuros L., Linaria vulgaris Hill, Senecio vulgaris L., Galium sp., Trifolium sp., Anthemis cotula L., Verbascum blattaria L., Stellaria media L., Cerastium viscosum



L., Geranium dissectum L., Brassica campestris L., Spergula arvensis L., Dactylis sp., Sonchus asper (L.) Hill, Equisetum sp., and Cyperus esculentus L.

Seeds of the horticultural crops, with the exception of hops and strawberries, were planted in vermiculite and the seedlings were allowed to grow for two weeks in that medium. Seedlings of hops were obtained from the Department of Farm Crops, while the strawberry plants were obtained by pegging down runner plants in the vermiculite. The seedlings of one set were wounded with a dissecting needle and transplanted into sterilized soil infested with a culture of Fusarium solani f. phaseoli, while the seedlings of another set of plants was planted directly in the infested soil without wounding.

Weed seedlings obtained from local bean fields were wounded in the same manner as the horticultural seedlings and planted in infested soil. A second set of weed seedlings was planted in the infested soil without wounding while a third set of seedlings was planted in sterilized soil for use in identification.

Roots of the plants used in this study were examined at two week intervals for a period of two months. Isolations were made from all roots showing discolorations. Of the horticultural crops inoculated, only peas showed any symptoms of root rot and these appeared on both the wounded and unwounded plants. The symptoms consisted of scattered brown pin point lesions on the main and secondary roots, similar in appearance to those described on

beans inoculated with pathogenic isolates of F. solani.

Root rot symptoms were found on the following weeds: Stellaria media, Equisetum sp., and Geranium dissectum. Symptoms ranged from small black lesions to black discoloration of a large portion of the main roots. Attempts to reisolate F. solani f. phaseoli were unsuccessful but isolates of Rhizoctonia solani and F. oxysporum were consistently obtained from the diseased tissue. Since seedlings were obtained from field soil, infection may have occurred before they were transplanted into the greenhouse, or the fungi may have been associated with the soil particles on the roots. Isolations made from the same weed species showing these symptoms also yielded R. solani and F. oxysporum.

## THE EFFECT OF CROP RESIDUES ON THE INCIDENCE OF DRY ROOT ROT

Since the development of severe root rot in fields where beans had not been planted for eight to twelve years could not be attributed to the presence of susceptible living hosts, a study of the influence of crop residues on the incidence of disease was carried out. It has been demonstrated with other diseases (12) 24) that organic residues in the soil may result in either a reduction or an increase in the incidence and severity of the disease.

In the Willamette Valley severe root rots on first year bean fields have been observed where the preceding crop was corn, hops, carrots, dill, oats, broccoli, or rye. Greenhouse studies of the effect of these crop residues were conducted. In addition, beets, barley, cabbage and strawberry were used since beans are frequently grown on land which had previously been planted to these crops. Peas, which are susceptible to infection by the dry root rot organisms were also included.

Naturally infested soil obtained from a Lane County bean field was thoroughly mixed with an equal quantity of steam sterilized soil before being used. Seedlings of hops and strawberries and seeds of the other crops were planted in flats containing the soil mixture. Two flats were prepared for each crop and the plants were allowed to grow for two months. The plants for each flat were then removed, chopped into small pieces and mixed with the soil in which they had grown. The flats were watered frequently for two months to

aid in the breakdown of plant tissues.

At the end of two months the soil from one flat of each crop was thoroughly mixed and divided into three aliquots which were mixed with sterilized soil in the following proportions: one part infested soil to four, nine and fourteen parts sterilized soil respectively. These dilutions were used to detect differences in the incidence of infection due to the residues. Five No. 10 cans were filled with each dilution of the soil from each of the crops. Eight seeds of the bean variety FM-1 were planted in each can of soil. After the plants had grown for a month they were removed from the soil and examined to determine the number of infected plants (Table 11).

The second flat for each crop was again planted with the same crop at the end of the two month watering period. After two months the plants were chopped up and mixed with the soil. Two months later soil aliquots were prepared in the manner described above and planted with FM-1 beans. Records were made of the number of plants showing infection one month after planting (Table 11).

Two flats of the soil mixture not planted to any crop served as controls. The flats were watered at the same time as the flats containing the crops. Aliquots of soil were mixed with sterilized soil in the proportions used for the other flats. The soil mixtures of one served as a control for the soil used after one planting, while the other was the control for soil used after two plantings. Any changes in the level of inoculum due to greenhouse

conditions should have been detected with the two check flats.

The highest percentages of infection of beans after one planting were obtained in soils where beans and peas had been grown. Using the per cent of infection as a criterion, the amount of inoculum in the soil increased after two consecutive plantings of beans and peas. No noticeable decrease or increase in the level of inoculum was apparent after any of the other crops.



Table 11. Effect of crop residues on incidence of dry root rot of bean in naturally infested soil in the greenhouse

Crop	soil dilution	Mean per cent infection					
		first planting			second planting*		
		1-5	1-10	1-15	1-5	1-10	1-15
Barley	100		77.5	67.5	97.5	70.0	60.0
Beans	100		95.0	77.5	100	92.5	87.5
Beet	97.5		85.0	62.5	97.5	70.0	65.0
Broccoli	100		80.0	60.0	95.0	57.5	55.0
Cabbage	95.0		82.5	60.0	90.0	72.5	52.5
Carrot	92.5		77.5	62.5	95.0	77.5	57.5
Corn	100		82.5	62.5	92.5	72.5	60.0
Dill	97.5		77.5	62.5	87.5	70.0	57.5
Hops	97.5		85.0	67.5	100	75.0	52.5
Oats	100		82.5	60.0	90.0	67.5	52.5
Pea	100		85.0	72.5	100	87.5	77.5
Rye	97.5		80.0	67.5	87.5	75.0	52.5
Strawberry	100		77.5	57.5	92.5	70.0	57.5
Check	100		82.5	65.0	95.0	77.5	57.5

\* two successive plantings

## CONTROL STUDIES

In undertaking studies on the control of dry root rot three control practices were considered, crop rotation, use of resistant varieties and fungicidal treatment of the seeds and soil. Of the three methods only the fungicidal treatments were studied in any detail, but brief mention of the reason for not undertaking work on the other methods is given below.

The value of crop rotation as a control measure for diseases depends on the length of time required to suppress the causal agent and is most effective in controlling diseases caused by fungi having a limited ability to survive as saprophytes. In the Willamette Valley, severe dry root rot has developed in fields which have been planted to crops other than beans for periods of eight to twelve years. Since no other hosts have been found in the area it is logical to assume that the dry root rot organism is able to compete successfully with the soil microflora as a saprophyte. Based on the observations and assumption given above, it is highly improbable that bean root rot in the Willamette Valley can be controlled by crop rotation. Furthermore, a thorough study of crop rotation as a means of control would require ten to twenty years for completion.

Studies of disease resistant varieties are currently in progress at Cornell University, Oregon State College, University of Idaho and elsewhere. However, it would probably be a number of years before

commercially acceptable varieties possessing a high degree of resistance to root rot will be available.

The use of fungicides as a means of controlling soil-borne diseases has increased in the past decade with the development of a variety of new chemicals. While control of dry root rot with fungicides has been reported in the literature (3, 15, 33), in most cases the observations are conflicting and at the present time no fungicides are being used in commercial fields for the control of dry root rot.

Tests to determine the value of fungicides as a means of controlling dry root rot in the Willamette Valley were conducted in the laboratory, greenhouse and field. A list of the materials tested is given in Table 12. Since several names have been given to these fungicides in the literature on dry root rot, the table lists the recognized common name, the chemical name and trade names.

#### Laboratory Testing of Fungicides

Fungicides were assayed in the laboratory to determine their possible value for control of Fusarium solani f. phaseoli. While the results of this testing are not always indicative of field performance, laboratory testing serves as a ready means of eliminating fungicides which are not effective against the disease organism.

Two criteria were used for the testing of the fungicides; inhibition of mycelial growth and of spore germination. Chemicals

Table 12. Fungicides used in the bean root rot control studies

Approved or common name	Chemical name	Trade names reported in the literature	Source
Chloranil	Tetrachloro-p-benzoquinone	Spergon	U. S. Rubber Co.
Diclonc	2,3-dichloro-1,4-naphthoquinone	Phygon	U. S. Rubber Co.
Maneb	Manganous ethylenebis(dithiocarbamate)		Rohm and Haas Co.
Nabam	Disodium ethylenebis(dithiocarbamate)	Dithane D 114	Rohm and Haas Co.
Zineb	Zinc ethylenebis(dithiocarbamate)	Dithane Z 78	Rohm and Haas Co.
Ferbam	Ferric dimethyldithiocarbamate		E.I. du Pont de Nemours and Co.
Thiram	Tetramethylthiuram disulfide	Arasan	E.I. du Pont de Nemours and Co.
Ziram	Zinc dimethyldithiocarbamate		E.I. du Pont de Nemours and Co.
Ceresan	Ethylmercury chloride		E.I. du Pont de Nemours and Co.
Ceresan M	N-(ethylmercuri)-p-toluenesulfonanilide		E.I. du Pont de Nemours and Co.



Table 12 con't. Fungicides used in the bean root rot control studies

Approved or common name	Chemical name	Source
New Improved Ceresan	Ethylmercury phosphate	E.I. du Pont de Nemours and Co.
Captan	N-(trichloromethylthio)-4-chlorohexene- 1,2-dicarboximide	California Spray Chemical Corp.
H.C.B.	Hexachlorobenzene	California Spray Chemical Corp.
P.C.N.B.	Pentachloronitrobenzene	Mathieson Chemical Corp.
Experimental Fungicides		
Bayer T.B. 4452	Quinoxime benzoyl hydrazone	Farbenfabriken Bayer A.G.
L.O. 737	Polyethylenethiuram tetrasulfide	Rohm and Haas Co.
L.O. 738	Polyethylenethiuram trisulfide	Rohm and Haas Co.
O.R. 56	Methyl endomethylene analog of Captan	California Spray Chemical Corp.
O.R. 101	Tetrahydro analog of Captan	California Spray Chemical Corp.
O.R. 127	Hexahydro analog of Captan	California Spray Chemical Corp.



tested by these methods included Diclone, Nabam, Thiram and Zineb, which have been reported (3, 15) to give control of bean root rot fungi. Other chemicals tested were Captan, HCB, PCNB, Maneb, Ferbam, Ziram, Bayer T. B. 4452, L.O. 737, L.O. 738, O.R. 56, O.R. 101 and O.R. 127.

Media containing one per cent dextrose, two per cent agar and fungicides at concentrations of .1, 1, 10, 100, and 1000 p.p.m. were prepared. Each series consisted of five petri dishes for each concentration of the fungicide to be tested for inhibition of spore germination or of mycelial growth. Because of the large number of petri plates needed, and the time required to check the results, the fungicides were assayed in groups of four over a period of several months. At the end of the testing four fungicides selected at random were reassayed to determine if the previous results could be duplicated. In all cases the results were similar to those obtained in the first assay.

Spores were obtained from seven day old cultures grown on PDA slants. Seven day old cultures were used in preference to younger cultures because the spores produced were more uniform with regard to percentage of germination obtained. The tests to determine the degree of inhibition of spore germination were conducted by placing drops of a water suspension of spores of F. solani f. phaseoli at five locations on the surface of the media of each petri dish. After twelve hours of incubation at 20° C., twenty spores were examined at each of the five locations, so that for each

concentration of fungicide, five replications of 100 spores each were checked (Table 13). Spores were considered to have germinated when the length of the germ tube was not less than half the diameter at the widest part of the spore. The spores in the check plates had germinated and in many cases had produced branching mycelia.

To determine the degree of inhibition of mycelial growth, 7 mm disc were cut from ten day old cultures of F. solani f. phaseoli on PDA and placed on the fungicide media. One disc was placed in each petri dish and incubated at 20° C. for one week. Measurements were taken of the increase in diameter of the colony (Table 13) and where variations occurred the largest diameter was recorded.

All of the fungicides at the 100 p.p.m. rate, with the exception of HCB, PCNB, and Bayer T.B. 4452, completely inhibited germination. Complete inhibition of spore germination was obtained at 10 p.p.m. with Diclone, Maneb, Nabam, L.O. 738, O.R. 101, and O.R. 127. Diclone and O.R. 127 were the only fungicides which completely inhibited germination at a concentration of 1 p.p.m.

All of the fungicides prevented mycelial growth at concentrations of 100 p.p.m. except Diclone, HCB, PCNB, Ziram, Bayer T.B. 4452, and L.O. 738. None of the fungicides completely inhibited mycelial growth at concentrations of less than 100 p.p.m., although Maneb greatly reduced the growth of the mycelia at concentrations of 10, 1, and .1 p.p.m.

Table 13. A comparison of the effects of sixteen chemicals, used at five rates, on spore germination and mycelial growth of Fusarium solani f. phaseoli grown on 1% dextrose agar at 20° C.

Treatment	p.p.m. 1000	Percent germination <sup>1</sup>				Diameter increase of mycelium in mm <sup>2</sup>				
		100	10	1	.1	1000	100	10	1	.1
Captan	0	0	3	97	100	0	0	16	29	35
Diclone	0	0	0	0	89	5	8	17	32	49
HCB	95	100	100	98	100	13	32	47	55	60
PCNB	95	100	99	100	100	23	36	36	43	57
Maneb	0	0	0	12	98	0	0	5	17	21
Nabam	0	0	0	62	95	0	5	14	43	51
Ferbam	0	0	53	72	99	0	0	19	38	50
Thiram	0	0	11	78	97	0	0	6	24	41
Zineb	0	0	94	100	100	0	0	17	32	47
Ziram	0	0	93	99	97	0	5	19	31	49
Bayer T.B. 4452	98	98	98	98	99	28	29	31	39	43
L.O. 737	0	0	5	79	92	0	0	13	34	57



Table 13 con't. A comparison of the effects of sixteen chemicals, used at five rates, on spore germination and mycelial growth of Fusarium solani f. phaseoli grown on 1% dextrose agar at 20° C.

Treatment	p.p.m. 1000	Percent germination <sup>1</sup>				Diameter increase of mycelium in mm <sup>2</sup>					
		100	10	1		.1	1000	100	10	1	.1
L.O. 738	0	0	0	67		98	0	12	35	32	48
O.R. 56	0	0	11	99		98	0	0	19	27	48
O.R. 101	0	0	0	8		98	0	0	23	36	51
O.R. 127	0	0	0	0		95	0	0	16	33	45
Check			99						64		

<sup>1</sup> Recorded after 12 hours

<sup>2</sup> Recorded after 1 week

### Greenhouse Seed Treatment Trials

The preliminary trials conducted during the summer of 1954, 100 gram lots of Germain's 21 bean seed were placed in paper bags with Thiram, Ceresan M and Maneb, respectively, and shaken until the seed were completely coated with the chemical. Seed of each treatment were planted in ten No. 10 cans of steam sterilized soil. A 10 mm disc of agar containing Fusarium solani f. phaseoli was placed adjacent to each seed. Untreated seed were inoculated in the same manner.

Three weeks after planting, the seedlings from five cans of each treatment were removed from the soil and severity of the disease recorded (Table 14). The plants in the other five cans were examined six weeks after planting and rated for disease severity.

Table 14. The effect of three fungicidal seed treatments, applied as dusts, on the severity of dry root rot of Germain's 21 beans grown in the greenhouse (1954)

Treatment	Mean disease ratings	
	3 weeks	6 weeks
Thiram	2.95	4.05
Ceresan M	3.30	3.85
Maneb	2.85	3.90
Check	3.85	4.10

All three treatments resulted in disease ratings lower than those of the check at the end of three weeks, but after six weeks



dry root rot infection among the treated plants was almost as severe as that among the checks.

Since the fungicides, applied as dusts, failed to give adequate control of the disease, seeds were pelleted with the same fungicides by immersing in a dilute solution of methocel, a cellulose acetate sticker, before application of the fungicide dusts. The method of infesting the soil for the second trial was changed since placement of all the inoculum adjacent to the seeds might not have given a true evaluation of the effectiveness of the fungicides. One liter of an agar-spore-water homogenate of the pathogen was added to and thoroughly mixed with sufficient soil for forty No. 10 cans. The mean disease ratings for the three and six week readings are given in Table 15.

Table 15. The effect of three fungicidal seed treatments, applied by pelleting, on severity of dry root rot of Germain's 21 bean grown in the greenhouse (1954)

Treatment	Mean disease ratings	
	3 weeks	6 weeks
Thiram	3.35	3.95
Geresan M	3.80	4.40
Maneb	2.95	4.05
Check	4.05	4.35

Application of the fungicides by pelleting did not increase the effectiveness of the treatments.

Further tests using seed of the bean variety FM-1 pelleted with Chloranil, Diclone, Ceresan, New Improved Ceresan and Zineb, were conducted during the spring of 1955. None of the treatments (Table 16) gave adequate control of the disease.

Table 16. The effect of five fungicidal seed treatments, applied by pelleting, on severity of dry root rot of FM-1 beans grown in the greenhouse (1955)

Treatment	Mean disease ratings	
	3 weeks	6 weeks
Chloranil	3.20	3.90
Diclone	3.45	4.10
Ceresan	3.15	4.05
New Improved Ceresan	3.50	4.25
Zineb	3.25	3.95
Check	3.65	4.20

#### Greenhouse Fungicidal Soil Treatments

The effects of fungicides incorporated into the soil on the severity of dry root rot were studied in the greenhouse during 1954 and 1955. The fungicides were tested at several rates to determine (1) the amount needed to obtain adequate control and (2) the phytotoxicity of the compounds on beans.

Fungicides tested included, Captan, Ferbam, Maneb, Nabam, Thiram, Zineb, Ziram, Formaldehyde, Diclone, Chloranil, L.O. 737, L.O. 738, PCNB, O.R. 56, O.R. 101 and O.R. 127. All except Nabam

and Formaldehyde were applied as one per cent dusts prepared with Attaclay as the diluent. Nabam and Formaldehyde were applied as ten per cent solutions. Each rate of each fungicide was mixed with sufficient infested soil to fill ten No. 10 cans. The soil was infested with F. solani f. phaseoli by mixing it with an agar-spore-water homogenate at the rate of 20 ml per No. 10 can. Ten seeds of the bean variety FM-1 were planted in each can the day after application of the chemicals.

In the preliminary tests Captan, L.O. 737, L.O. 738 and PCNB were applied at rates of 10 and 25 pounds per acre. In later tests Ferbam, Thiram, Ziram, Diclone, Zineb, Maneb and Chloranil were used at the same rates and Nabam and Formaldehyde at 2, 5, 8 and 10 gallons per acre. Further tests were conducted in 1955 using the fungicides at rates of 50 and 75 pounds per acre.

Disease ratings (Table 17) for the plants from five of the cans were recorded after three weeks and for the remaining five cans, six weeks after planting. Since the fungicides were tested at different times, the disease ratings of the check plants varied considerably. In Table 17, the disease ratings of the appropriate check plants are given with the ratings of each dosage of the fungicides.

Of all the fungicides tested, Maneb at all rates gave the best control of the disease, as is evident from the six week disease ratings. However, since this fungicide is very phytotoxic it is not a satisfactory chemical for controlling dry root rot.

Phytotoxicity symptoms produced by Maneb and some of the other fungicides in this greenhouse study included delay of germination, stunting of seedlings, and chlorosis and necrosis of leaf tissue.

### Field Control Trials

Field trials employing fungicides incorporated into the soil were conducted in 1954 and 1955 to determine the possibility of control of bean root rot by chemical treatment of the soil. Chemicals selected for the trials in 1954 had previously been shown to be effective against other soil fungi, or were experimental compounds that were reported to have shown promise in limited tests. Fungicides used in the 1955 trials included Captan and HCB, the most promising chemicals of the previous year's trials, plus Zineb (3), which was reported to give good control of the disease, and Maneb, which had given the best control in greenhouse tests.

A bean field near Coburg, Lane County, in which the disease was serious the previous year, was used for the 1954 trials. A randomized block experiment of six treatments, replicated six times, was set up so that the individual plots measured 5 by 33 feet.

The actual amounts of fungicides applied were determined by calculating the fraction of an acre being treated and multiplying by the desired pounds per acre. The fungicide dusts, containing 20 per cent active ingredient, were applied to the surface of the soil in a band two feet wide down the middle of each plot and were



Table 17. The effects of sixteen fungicides applied to the soil at four rates on dry root rot disease ratings of FM-1 beans

Treatment	Rate lb/acre	Mean disease ratings			
		3 weeks	Check	6 weeks	Check
Captan	10	2.95	3.97	4.54	4.48
	25	3.10	3.97	4.32	4.48
	50	2.63	4.15	3.98	4.30
	75	2.74	4.15	4.08	4.30
PCNB	10	3.84	3.97	4.65	4.48
	25	3.76	3.97	4.32	4.48
	50	3.45*	4.15	4.20*	4.30
	75	3.89*	4.15	4.23*	4.30
L.O. 737	10	3.45	3.97	4.13	4.48
	25	3.47*	3.97	4.28*	4.48
	50	3.64*	4.15	4.32*	4.30
	75	3.34*	4.15	4.15*	4.30
L.O. 738	10	3.78	3.97	3.94	4.48
	25	3.44*	3.97	4.20*	4.48
	50	3.84*	4.15	4.34*	4.30
	75	3.63*	4.15	4.18*	4.30
Chloranil	10	3.94	3.85	4.24	4.16
	25	3.67	3.85	4.05	4.16
	50	3.45	3.55	3.96	4.08
	75	3.34	3.55	3.87	4.08
Diclone	10	3.65	3.85	4.13	4.16
	25	3.93	3.85	4.05	4.16
	50	3.21	3.55	4.15	4.08
	75	3.17	3.55	3.84	4.08
Ferbam	10	3.79	3.85	4.32	4.16
	25	3.27	3.85	4.07	4.16
	50	3.05	3.55	3.84	4.08
	75	3.30	3.55	4.05	4.08
Thiram	10	3.62	3.85	4.24	4.16
	25	3.37	3.85	4.05	4.16
	50	2.95	3.55	4.15	4.08
	75	3.34	3.55	3.98	4.08

\* phytotoxic



Table 17 continued. The effects of sixteen fungicides applied to the soil at four rates on dry root rot disease ratings of FM-1 beans

Treatment	Rate lb/acre	Mean disease ratings			
		3 weeks	Check	6 weeks	Check
Ziram	10	3.62	3.55	3.98	4.12
	25	3.22	3.55	3.81	4.12
	50	3.85*	3.97	4.12*	4.08
	75	3.76*	3.97	3.89*	4.08
Zineb	10	2.95	3.55	3.87	4.12
	25	3.09	3.55	3.96	4.12
	50	2.84*	3.97	3.72*	4.08
	75	3.01*	3.97	4.12*	4.08
Maneb	10	2.93*	3.55	2.85*	4.12
	25	2.24*	3.55	2.30*	4.12
	50	2.16*	3.97	2.24*	4.08
	75	2.33*	3.97	2.15*	4.08
Nabam	2 gal/acre	3.05	3.55	4.11	4.12
	5	3.23	3.55	3.95	4.12
	8	3.74	3.97	4.22	4.08
	10 gal/acre	3.47	3.97	4.09	4.08
Formaldehyde	2 gal/acre	3.15	3.55	3.88	4.12
	5	3.22	3.55	4.04	4.12
	8	2.84	3.97	3.78	4.08
	10	3.08	3.97	3.92	4.08
O.R. 56	10	3.88	4.13	4.46	4.38
	25	3.62	4.13	4.22	4.38
	50	3.68	4.13	4.32	4.38
	75	3.38*	4.13	4.17*	4.38
O.R. 101	10	3.76	4.13	4.25	4.38
	25	3.45	4.13	4.18	4.38
	50	2.72	4.13	4.05	4.38
	75	2.34*	4.13	3.96*	4.38
O.R. 127	10	3.83	4.13	4.42	4.38
	25	3.66	4.13	4.15	4.38
	50	2.68	4.13	4.17	4.38
	75	2.26*	4.13	4.31*	4.38

\* phytotoxic

incorporated into the top six inches of soil with a rotovator.

The plots were planted to FM-1 beans within a day after application of the fungicides. Staking, fertilization, cultivation and irrigation of the plots were carried on by the cooperating grower, who gave the experimental plots the same care as he did the surrounding beans grown for processing. One month after planting the disease ratings of fifteen bean plants selected at random from each replicate in the experimental were recorded (Table 18). Yield data were recorded at each of seven pickings.

The low yields obtained from the plots treated with PCNB occurred because the fungicide at a rate of 30 pounds per acre was phytotoxic to the bean plants. Phytotoxic symptoms of PCNB on bean plants included reduced germination and stunting of the plants throughout the growing season. The data on these plots were not included for the statistical analysis. The data obtained from the other treatments were analyzed statistically and the analysis of variance did not show any significant differences due to treatments.

In 1955 the field experiments were conducted at the Department of Botany and Plant Pathology farm in Linn County and consisted of five treatments, replicated five times, in a randomized block. Each plot measured 5 by 30 feet. Four of the treatments consisted of fungicides incorporated into the soil. The fifth treatment, hilling of the bean plants, has been reported in the literature (17) to be a possible means of reducing yield losses due to the disease.

Application of fungicides and recording of data on disease

Table 18. Effect of five fungicides, incorporated into the soil, on root rot disease ratings and yields of FM-1 beans grown in Lane County (1954)

Treatment	Rate of application in pounds per acre	Mean disease ratings*	Mean yields in tons per acre
L.O. 737	15	3.68	6.95
L.O. 738	15	3.73	6.96
PCNB	30	3.95	3.72
Captan	30	2.97	7.27
HCB	30	3.54	7.10
Check	—	3.85	6.20

\* Based on 15 plants per replicate, recorded one month after planting.

severity and yields (Table 19) were conducted in the same manner as for the 1954 field trials. The hilling of the bean plants was done three weeks after planting and consisted of a three inch layer applied in a ten inch band to both sides of the rows of bean plants.

Statistical analysis of the data for the 1955 field trials indicated that none of the treatments was effective in controlling the disease.

Table 19. Effect of fungicides incorporated into the soil, and of hilling, on root rot disease ratings and yields of FM-1 beans grown in Linn County (1955)

Treatment	Rate of application in pounds per acre	Mean disease ratings*	Mean yields in tons per acre
Captan	50	3.08	7.82
Zineb	15	3.35	7.73
Maneb	15	2.15	8.42
HCB	50	3.52	7.80
Hilling	—	3.76	8.03
Check	—	3.64	7.85

\* Based on 15 plants per replicate, recorded one month after planting.



VERTICAL DISTRIBUTION OF *FUSARIUM SOLANI* F. *PHASEOLI* IN THE SOIL

Failure of soil fungicides to control bean root rot prompted a study to determine the vertical distribution of the pathogen in the soil. If the fungus is abundant in the soil below 8 to 10 inches, then fungicides applied to the top six inches would have a slight chance of reaching the fungus or preventing the disease.

In 1955 a series of soil samples were obtained from a bean field in Lane County, known to be heavily infested with *F. solani* f. *phaseoli*. A trench three feet deep, two feet wide and four feet in length was made in the soil. Samples were taken at intervals of four inches to a depth of two feet. Care was taken to prevent contamination of the lower soil samples by the upper soil layers. The uppermost soil sample was obtained from one end of the trench; each succeeding sample was taken at six inch intervals along the length of the trench. The shovel and trowel used for collecting the soil samples were washed periodically in a 25 per cent solution of commercial chlorox.

In the greenhouse each soil sample was divided into two lots. One lot of soil was mixed with sterilized soil at the rate of one part field soil to three parts sterilized soil. Five No. 10 cans were filled with the mixture and five with the undiluted field soil.

Each can was planted with ten seed of the bean variety FM-1. At the end of one month records were made of the number of plants showing infection (Table 20). Random samples of the infected beans were selected for isolation to ascertain that the pathogen in the



lower soil levels was Fusarium solani f. phaseoli.

Table 20. Vertical distribution of Fusarium solani f. phaseoli in the soil of a Lane County bean field as indicated by infection of FM-1 bean plants

Depth of soil sample in inches	Number of plants			
	Field sample		Diluted sample	
	Healthy	Infected	Healthy	Infected
0-4	0	47	0	48
4-8	7	39	10	37
8-12	29	18	40	9
12-16	33	15	46	3
16-20	46	3	46	1
20-24	47	1	49	0

Fusarium solani f. phaseoli was found at depths down to two feet, but was more abundant in the upper eight inches. The greatest concentration of the fungus was in the upper four inches. Fusarium solani f. phaseoli was isolated from all the samples of infected beans.

## DISCUSSION AND CONCLUSIONS

Based on the frequency of isolation and occurrence in bean fields Fusarium solani f. phaseoli is the most important pathogen causing bean root rot in the Willamette Valley. Most symptoms observed in the field can be reproduced on beans inoculated with pure cultures of the pathogen. While isolates of F. solani, F. oxysporum, F. roseum and Rhizoctonia solani also are capable of causing root rot, the damage caused by these pathogens is small, and in some cases infection by these fungi occurred only after wounding.

Experiments failed to establish the presence of more than one physiologic form of the pathogen among the isolates identified as F. solani and considered by the author to belong to the form phaseoli. However, if the group of isolates identified as F. solani be considered as belonging to the same pathogenic form, the conclusion would have to be made that at least two strains of the pathogen are present. Because of the differences in cultural characteristics and the fact that the isolates were weak pathogens or non-pathogenic these isolates are not considered as belonging to the same form. It is more likely that they may be other pathogenic forms capable of causing mild infections on beans as has been demonstrated by Reinking (22).

Severity of the disease increased with increasing soil temperatures from 16° C. up to 28° C. and decreased with higher soil temperatures. A comparison of these data with field temperature data for the growing season of 1954 and 1955 revealed that the

average field soil temperatures were below that required for optimum development of the disease. Therefore, to evaluate the seriousness of the disease in the Willamette Valley, emphasis should be placed on the data for soil temperatures of 16°, 20°, and 24° C. as these soil temperatures are more typical of field conditions. While infection can occur over a wide range of temperatures it is concluded that the severity of the disease depends upon seasonal variation of soil temperatures.

This conclusion contradicts that given by Burkholder (6) who presented data to show that high (26° C.) and low (18° C.) soil temperatures had little effect on the severity of the disease. One possible explanation for the difference in conclusions might be the existence of physiologic forms, even though the existence of physiologic forms has not been reported in the literature. Kendrick (14) has suggested that differences in soil temperature relations may vary with different races of Rhizoctonia solani. Therefore, it is possible that different physiologic forms of Fusarium solani f. phaseoli may also react differently at the same soil temperature.

An estimate of field yield losses due to the disease, based on field data, is difficult because of the influence of factors such as cultural practices and other diseases. No correlation was found between the field disease ratings, recorded early in the season and yield obtained from these fields.

Yield losses obtained in greenhouse studies, in which the



pathogen was added to sterilized soil, are higher than would be encountered in the field, as no consideration was given to the influence of soil microflora. It was shown that the addition of non-pathogenic fungi and bacteria to the greenhouse soil would decrease yield losses by approximately 40 per cent.

If the data obtained from the controlled soil temperature studied, field soil temperatures, and the influence of soil microflora are taken into consideration, it is estimated that yield losses of from 15 to 25 per cent might be sustained in severely infested bean fields in the Willamette Valley. This estimate is very conservative when compared with that of Burkholder's (5, pp. 1022-1027) who reported yield losses of 50 per cent in field trials with artificially infested soil. However, in his trials, the lack of precipitation during the latter part of the growing season contributed to these greater yield losses. In the bean fields of the Willamette Valley, which are under irrigation, sufficient moisture can be maintained throughout the growing season.

Attempts to explain the high level of dry root rot inoculum in soils where crops other than beans have been grown for eight to twelve years, on the basis of other hosts were unsuccessful. Peas, the only other host found, are not normally grown in rotation with beans in the Willamette Valley.

Although no apparent effects of crop residues other than that of beans and peas on the level of inoculum was detected in greenhouse studies, no conclusions can be made concerning the effect of



such residues under field conditions. The incorporation of these crops in rapid sequence does not simulate the natural sequence of cropping since only a single crop would have been grown in one year. The significance of the physiological age and condition of residues under field conditions has not been considered. The effect that soil temperatures might have on colonization of the residues by various organisms also has not been considered. The greenhouse studies were carried out in hopes that some indication of the effect of crop residues could be detected. Since these studies were unsuccessful it is likely that field studies over a period of years will be necessary before any conclusions concerning the effects of crop residues can be made.

The possibility of controlling the disease by fungicidal seed treatment seems unlikely. Treatment of the seed with Maneb, which was shown to be effective in reducing the disease when incorporated into the soil, failed to give adequate control. Seed treatment might be a satisfactory means of control if infection occurred only during the seedling stage but Harter and Zaumeyer (13) have shown that plants in all stages of development appear to be susceptible. From the results of seed treatment studies, it is concluded that none of the fungicides tested were effective in controlling the disease.

While many of the fungicides were effective in inhibiting spore germination and mycelial growth when incorporated with dextrose agar, they were not effective in reducing the severity of the

disease when incorporated into the soil. The efficiency of the fungicides is apparently greatly reduced by the buffering action of the soil.

Maneb incorporated into the soil in greenhouse studies was effective in reducing the severity of disease, but when used in the field at a rate which was not phytotoxic to the bean plants, it did not significantly reduce yield losses. From the data obtained from greenhouse and field studies, it is concluded that none of the fungicides tested were effective in controlling the disease when applied at rates which were not phytotoxic to the bean plants.

Hilling of the beans to stimulate development of adventitious roots, as suggested by Menzies (17) did not significantly reduce yield losses.

Fusarium solani f. phaseoli was found at a depth of two feet in the soil. While the placement of fungicides in the top six inches of soil would be in the area of the highest concentration of the pathogen, the fungi in the lower levels could serve as a source of inoculum later in the season as the roots penetrate deeper into the soil.

## SUMMARY

1. Fusarium solani f. phaseoli was found to be the most important pathogen causing bean root rot. No differences in pathogenicity were detected among the isolates of this pathogen. F. solani, F. roseum F. oxysporum and Rhizoctonia solani were also found to be root rot pathogens but the infections caused by these pathogens was mild and in some cases occurred only after wounding.
2. The effects of soil temperatures on the severity of dry root rot were studied in the greenhouse at soil temperatures of 16°, 20°, 24°, 28°, 32° and 36° C. The greatest yield losses were sustained by plants grown in the 28° C. soil. The average field soil temperatures for the 1954 and 1955 growing seasons were below that required for optimum disease development.
3. Yield losses recorded in greenhouse studies in which the pathogen was added to the sterilized soil were reduced by approximately 40 per cent when other fungi and bacteria were added. Based on these data plus those obtained from the soil temperature studies and the field soil temperatures, yield losses that might be sustained in severely infested fields were estimated at 15 to 25 per cent.
4. Attempts to correlate the high level of dry root rot inoculum in soils where crops other than beans have been grown for eight to twelve years, with the presence of other hosts, were unsuccessful.

5. Studies of the effects of thirteen crop residues on the incidence of dry root rot were carried out in the greenhouse. The incidence of the disease was increased after two successive crops of beans or peas and none of the crop residues reduced the incidence of the disease.

6. In the laboratory some chemical inhibited the growth and spore germination of the pathogen. Chemicals tested in the greenhouse and field did not reduce the severity of dry root rot when used at rates which were not phytotoxic to the bean plants. Hilling of the beans to stimulate development of adventitious roots did not significantly reduce yield losses.

7. Fusarium solani f. phaseoli was found in the soil at depths as great as two feet.



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