

Studies on
the Pathogenicity of Species of Fusarium

A Thesis

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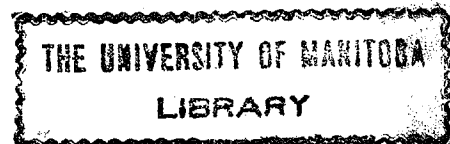
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INTRODUCTION

The most widespread and important root disease of small grain crops in Western Canada is Helminthosporium - Fusarium Root Rot, or Common Root Rot. It is present every year throughout the grain-growing area of the three prairie provinces, and affects all cereal crops. This root disease may be caused by several different soil-inhabiting fungi although Helminthosporium sativum P.K. and B., Fusarium culmorum (W.G. Sm.) Sacc., and other species of Fusarium, are most commonly associated with it. These fungi live in the soil and attack the roots, crowns, and lower portions of the stem of cereal plants. Certain of these fungi are, however, also capable of attacking the leaves, heads and seeds of different grain crops, causing diseases which are commonly referred to as "leaf spots", "head blights" and "seed discolorations". The greatest loss, however, from these fungi usually occurs through damage to the underground parts of the plants.

Common root rot attacks both seedlings and mature plants. As a seedling blight it may be very destructive, killing off a large number of seedlings. Affected plants that survive the seedling stage usually lack vigour and fail to tiller properly so that stands of affected crops are usually thin and weedy. In addition to causing serious

direct losses in yield and grain quality, the disease is also responsible for very important indirect losses.

The nature of the direct loss from common root rot is twofold. First, there is a loss in yield due to reduction in the number and size of kernels in the heads of grain; and second, a loss in grain quality due to the production of small shrunken kernels of inferior quality. It is important to point out that these inferior kernels, when sown, produce weak seedlings that are likely to be easily and seriously injured by root-rotting fungi inhabiting the soil.

The total actual loss due to common root rot is somewhat difficult to estimate. Judged by the results obtained in experimental plots at Winnipeg, Craigie (5) has estimated that the annual loss in wheat, oats, and barley is not less than five per cent. On a basis of five per cent loss in yield the annual cash loss to Manitoba alone for these three crops amounts to \$2,470,000. According to Broadfoot (2) and Simmonds (31), similar important annual losses are caused by common root rot in Saskatchewan and Alberta. The investigations of Henry (19), Dosdall (8), Christensen (6) and McKinney (23), have demonstrated the great economic importance of root diseases in cereal crop production.

A large number of pure-culture isolations from the roots of cereal plants collected in various parts of the

three prairie provinces of Canada by Henry (19), Simmonds (31), Broadfoot (2), Greaney and Bailey (15) and Sanford and Broadfoot (29), showed that fungi belonging to the genus Fusarium were very widely and consistently associated with root rots in Western Canada. These investigators carried out extensive pathogenicity tests to determine whether species of Fusarium found associated with the roots of cereal plants were saprophytic, weakly parasitic, or capable of causing severe injury under certain environmental conditions. Sanford and Broadfoot (29) studied the relative pathogenicity of numerous isolates of Fusarium culmorum obtained from wheat roots and found that the majority of them were only very weakly pathogenic. Samuel and Greaney (27) and Greaney and Johnston (16) studied the progressive invasion of the roots and crowns of wheat plants by fungi and found that Fusarium culmorum, and other species of Fusarium, were commonly associated with wheat roots and generally present in the soil, although in a number of cases they exerted no appreciable parasitic affect on the wheat plant.

Gordon (12) has made a taxonomic study of the species of Fusarium found to be associated with diseased roots and stem bases of wheat, oats, barley and rye in Manitoba. A study of approximately 4,100 isolations of these fungi indicated that twelve species and nine varieties representing seven sections of the genus Fusarium were present.

Studies on the microflora of the soil of permanent grain plots made at Winnipeg in 1936 and 1937 indicated that fungi of the genus Fusarium constituted an appreciable percentage of the total soil fungus flora of grain plots. In 1936, 13,850 isolations of fungi were made from one series of plots and Fusarium spp. accounted for 1,498 of this number, or approximately 10.8 per cent. Species of Fusarium were found to constitute 18.2 per cent of the total fungus flora isolated from the same series of grain plots in 1937. According to Gordon (12) the various isolates obtained from the soil in 1936 and 1937 comprised many species, varieties and forms of Fusarium belonging to eight sections of the genus.

Owing to the fact that Fusarium culmorum and other species of Fusarium are so commonly associated with the roots of cereal plants and so generally present in the soil, it was considered essential that some attempt be made to determine their pathogenicity. It was therefore decided to carry out a comprehensive series of greenhouse and field tests to determine the relative parasitic behaviour of the predominating species, varieties and forms of Fusarium isolated from diseased parts of cereal plants and from the soil.

The antagonism of microorganisms in culture media, in the host plant, and in the soil has been observed for many years, but it was not until more recently that the full significance of this factor in the study of root

diseases came to be appreciated by plant pathologists and soil microbiologists. The importance of the reaction of one organism upon another, not only in culture, but also in the soil itself is now well established. A review of the literature on the subject has been made by Fawcett (9), Machacek (22), Greaney and Machacek (17), Wiendling (33), Sanford and Broadfoot (30). The relation of biological antagonism to infection by a cereal root-infecting fungi has been studied by Simmonds (31), Broadfoot (3), Bisby, James and Timonin (1), and Greaney and Machacek (17). Bisby, James, and Timonin showed that under greenhouse conditions Trichoderma lignorum suppressed the virulence of Fusarium culmorum and Helminthosporium sativum. Greaney and Machacek demonstrated that in pot cultures the pathogenicity of Helminthosporium sativum was suppressed by the antagonistic action of Cephalothecium roseum.

The importance of soil temperature in relation to cereal root-rotting fungi has been investigated by McKinney (24), Dickson (7), Sanford (28), Hynes (20), and others. The results of these studies indicate that soil temperature is an important factor in the development of seedling root rot. In some cases, infection was accelerated with an increase in soil temperature, in others depressed, depending on the host and fungus studied.

A number of investigators including Dosdall (8),

Henry (19), McKinney (24), Mitra (25), Dickson (7), and Hynes (20), have studied the influence of soil moisture upon infection of wheat seedlings by parasitic fungi. The results show that both soil moisture and temperature are variable in their effect upon the pathogenicity of cereal root-rotting fungi. Further investigations are necessary to determine the actual effect of soil moisture and temperature on the occurrence of severe outbreaks of root rot of wheat under field conditions in Western Canada.

II. OBJECTS OF INVESTIGATION

The principal objects of the present investigation were: (1) to determine the relative pathogenicity to wheat of five species of Fusarium, and a number of cultures of Fusarium culmorum originally isolated either from diseased parts of cereal plants and grasses or from grain soils; (2) to study the influence of soil moisture and temperature on the pathogenicity of certain species of Fusarium; and (3) to determine the effect of some common soil-inhabiting fungi on the pathogenicity of Fusarium culmorum and Fusarium redolens Wr.

III. PATHOGENICITY STUDIES

Greenhouse Experiments

The results of previous greenhouse tests of pathogenicity of cereal root-infecting fungi by Greaney and Bailey

(15), Greaney and Machacek (17), and others, have emphasized the importance, if reliable and reproducible results are to be expected, of controlling factors influencing the soil environment. In the present investigation, therefore, every effort was made to standardize the conditions under which the various greenhouse pot tests were made.

Material and Methods

Greenhouse pot tests were made to determine the relative pathogenicity of twenty-four cultures of Fusarium culmorum isolated from the diseased parts of cereal and grass plants. A list of these isolates, together with information concerning the date and source of their isolation is given in Table 1.

Inoculum of each isolate of F. culmorum was prepared by growing the fungus on a sterilized sand and cornmeal mixture for about 15 days. This medium gave excellent growth of all isolates and species of Fusarium. When completely overgrown with the sporulating mycelium of the fungus, the sand-cornmeal inoculum was mixed with autoclaved soil, one part of inoculum to 30 parts of soil by weight, and placed in 6-inch pots. All soil for a given experiment was prepared at the same time. In order to allow the various fungi to become well established in the soil to which they were added, three days were allowed

Table 1. Number and source of isolates of Fusarium culmorum

Isolate No.	Year isolated	Host	Location
38	1932	Durum wheat	Rhodes, Man.
1263	1935	Reward "	Bowsman, Man.
1040	"	Common "	Brandon, "
937	"	Durum "	Pilot Mound, "
856	"	Reward "	Whitewater, "
854	"	Barley	Emerson, "
1119	"	Oats	Melita, "
1280	"	Marquis "	Griswold, "
1300	"	" "	Pipestone, "
1193	"	Reward "	Foxwarren, "
1202	"	Marquis "	Shoal Lake, "
1158	"	" "	Binscarth, "
1111	"	Common "	Clearwater, "
1122	"	Durum "	Deloraine, "
1197	"	Ceres "	Virden, "
39	1932	Durum "	Rhodes, "
19 A	1930	Common "	Saskatoon, Sask.
19 B	"	" "	" "
19 C	"	Oats	" "
285	1936	Common "	" "
529	1933	Brome grass	Winnipeg, Man.
6	1936	Marquis wheat	Pope, "
15	"	Common "	New Norway, Alta.
10 E	"	" "	Rothamsted, England.

to elapse between the time of soil preparation and date of sowing of the grain.

The seed used was Marquis wheat, selected by hand for uniformity and size. It was surface sterilized by rinsing in 95 per cent ethyl alcohol, immersing for three minutes in 0.1 per cent mercuric chloride solution, rinsing in alcohol, and then washing in sterile water. Before sowing, the surface-sterilized seed was inoculated, where required, by dipping it into a suspension of spores and mycelial fragments of the species of Fusarium to be tested. The control seed was dipped in sterile water. Immediately after treatment, the seed was sown in the prepared soil.

In each fungus series there were 4 pots, twenty-five seeds being sown in each pot. The complete test with 24 organisms and 24 control pots of uninfested soil sown with uninoculated seed consisted of 120 pots. The test was replicated four times. During the course of each test uniform conditions of light, moisture and temperature prevailed. The moisture content of the soil was maintained at a uniform level by adding sterile water at two-day intervals to bring the pots up to their original weight. To minimize place effect, the pots of each test were completely randomized on a large bench in the centre of the greenhouse.

Germination was usually complete 12 days after sowing at which time the number of emerged plants per pot were counted. At the end of the experimental period (28 days), non-emerged plants as well as the young seedlings were

lifted from the pots, washed free of soil, examined individually, and the extent of injury due to pre-emergence blight, seedling blight and root-rot was recorded. The classes and numerical ratings used to record the intensity of disease infection on individual seedlings and the method of computing the disease rating that was used to express the extent of the disease on the plants in each series of pots are given in Table 2.

After the disease data had been secured, the green weight of the plants of each inoculated and uninoculated series was recorded. In all experiments herein reported, plant emergence, disease, and yield data were analyzed according to the procedure described by Fisher (10) as the analysis of variance. To estimate the odds of significance, however, the direct ratio of the variances (the F value of Snedecor (32)) was used. The methods used in all greenhouse experiments with young plants herein reported were essentially similar to these described above.

In addition to the seedling test of pathogenicity with various isolates of Fusarium culmorum, the parasitic behaviour of five different species of Fusarium to adult Marquis wheat plants was studied. The species of Fusarium tested were strain No. 38 of F. culmorum (W.G. Sm.) Sacc., F. redolens Wr., F. avenaceum Fr. (Sacc.), F. oxysporum Schl. v. aurantiacum (Lk.) Wr., and F. equiseti (Cda.) Sacc. The methods used in these tests were briefly as

Table 2. Classes and numerical ratings used to record the degree of infection caused by Fusarium culmorum on wheat.

Class	Degree of infection on individual plants	Numerical rating
1	No infection	0
2	Small, scattered necrotic lesions on sheath, subcrown internode, or roots.	1
3	Distinct dark lesions on basal parts, particularly on subcrown internode and roots.	2
4	Large necrotic lesions on crown, subcrown internode, and roots; with loss of plant vigour.	3
5	Severe rotting of basal parts; plant chlorotic, often stunted; some culms dead.	4
6	Plant destroyed after germination, but before emergence. Dead plant.	5

$$\text{Disease rating} = \frac{\text{Sum of numerical ratings of individual plants} \times 5}{\text{Number of plants} \times 100}$$

follows. Surface sterilized Marquis wheat was sown in sterile quartz sand. When the healthy seedlings were six days old they were transplanted from the sand to 6-inch pots of sterilized soil containing inoculum of the fungus to be tested. At the time of transplanting, the entire root system of each seedling was inoculated by dipping it into a water suspension of spores and mycelium fragments of the respective fungi. Five seedlings were placed in each pot and grown to maturity. After the plants had been transferred, they were inoculated at regular intervals during the growing period (at 10, 20, 50, and 70 days) by means of water suspensions of spores which were added to the soil of each pot.

A complete experiment consisted of five pots (25 plants) of each species of Fusarium and five uninoculated control pots. Soil moisture content was maintained at a constant level in all pots throughout the experiment. The complete experiment was carried out at two different times. At maturity, the intensity of disease on individual plants was recorded according to the methods described by Greaney, Machacek and Johnston (18).

Experimental Results

The data of the tests with isolates of F. culmorum were examined by the analysis of variance method (Table 3). The results of the analyses in Table 3 show that the twenty-four isolates of F. culmorum tested differ greatly in their

Table 3. Complete analyses of variance for percentage of plants emerged, disease rating, and green weight of young Marquis wheat plants.

Variance due to	Degrees of freedom	Sum of squares	Mean square	F	5% point
Plant Emergence					
Replicates	3	4895.16	1631.72	10.43	1.60
Isolates	29	15691.85	541.09		
Error	87	4512.59	51.86		
Total	119	25099.60			
Disease Rating					
Replicates	3	4292.65	1430.88	15.79	1.60
Isolates	29	20816.39	717.80		
Error	87	3954.81	45.45		
Total	119	29063.85			
Green Weight					
Replicates	3	126840.66	42280.22	7.19	1.60
Isolates	29	16537.68	570.26		
Error	87	6900.72	79.31		
Total	119	150279.06			

pathogenic behaviour. In every instance, for plant emergence, disease rating and yield, the variance for organisms greatly exceeded the error variance. The complete results of the experiment, with the standard errors associated with the factors studied are summarized in Table 4.

It is evident from Table 4 that a wide variation exists in the virulence of the 24 strains of F. culmorum studied. On the basis of the standard error a significant necessary difference in disease rating between the organisms is 10.1. Using this as a basis for classification it was found that the various isolates of F. culmorum could be divided into three classes; namely, strong pathogenic, moderately pathogenic and weakly pathogenic. The great majority of those tested were only very weakly or weakly pathogenic to Marquis wheat. These results confirm the findings of Sanford and Broadfoot (29) who tested the comparative pathogenicity of some 219 cultures of F. culmorum isolated from the roots and crowns of wheat plants collected from widely separated fields in Alberta.

In the final analysis of the experiment, disease ratings and total green weight of the plants of the individual pot series were correlated. The significance of the correlation coefficient obtained was determined by the method described by Goulden (13). A highly significant negative coefficient of -0.9259 was obtained. This result substantiates the work of Livingstone (21) who studied the

Table 4. The relative pathogenicity of several isolates of Fusarium culmorum to Marquis wheat seedlings.

<u>Fusarium culmorum</u> isolate No.	Percentage of plants emerged	Degree of infection	Green weight of plants in grammes
38	47.0	64.0	42.6
1263	57.2	35.8	63.4
1040	59.2	25.8	62.4
937	59.8	35.9	60.9
856	60.2	20.6	67.6
854	61.2	24.6	66.0
10 E	61.8	27.8	68.5
1119	63.0	25.1	67.3
1280	63.0	24.4	70.0
1300	63.0	32.2	68.5
6	64.0	23.8	69.0
19 B	64.5	16.8	73.3
1193	65.0	21.1	66.7
1202	66.0	28.3	61.0
1158	66.2	22.4	71.6
1111	67.2	19.5	71.2
15	67.5	22.9	74.6
19 C	68.5	34.2	62.5
529	70.0	6.0	80.3
1122	72.2	16.0	76.7
39	74.0	11.8	78.3
19 A	75.2	16.4	78.0
285	77.0	10.0	79.0
1197	78.5	17.6	84.0
Control A	88.7	2.8	91.2
Control B	90.6	3.0	92.1
Standard error	\pm 3.60	\pm 3.37	\pm 4.45

relation between growth of roots and tops in wheat, and found that poorly developed wheat tops were associated with poor root development.

The results of the mature plant study with five species of Fusarium are given in Table 5. To economize space, the complete analysis of variance tables for disease ratings, weight of plants and grain are not given. However,

Table 5. The relative pathogenicity of five different species of Fusarium to mature plants of Marquis wheat in greenhouse pot tests.

Organism	Degree of root-rot infection (Disease Rating)	Weight of root and tops of plants (gm.)	Yield of Grain (gm.)
<u>Fusarium</u> <u>redolens</u>	15.6	125.6	19.2
" <u>oxysporum</u>	18.8	119.8	18.7
" <u>equiseti</u>	26.8	124.4	16.7
" <u>avenaceum</u>	32.4	113.2	15.0
" <u>culmorum</u> No. 38 <u>1/</u>	59.0	88.4	11.1
Control (no organism)	13.6	126.1	23.3
Standard error	<u>±</u> 3.73	<u>±</u> 4.22	<u>±</u> 2.05

1/ Plants first inoculated when 10 days old.

the results of these analyses established that the differences in pathogenicity observed between organisms in the experiment were very great. The complete results of the experiment in Table 5 show that of the species of Fusarium tested F. culmorum

(Isolate No. 38) was exceedingly pathogenic to wheat. Fusarium equiseti and F. avenaceum were weakly pathogenic, while F. oxysporum and F. redolens were not pathogenic to Marquis wheat. The comparative pathogenicity of these five fungi to young wheat plants is discussed in a later section of this paper. From Table 5 it is again evident that the intensity of root rot infection caused by species of Fusarium is negatively associated with yield in wheat.

Field Experiments

Field tests to determine the relative pathogenicity of five species of Fusarium; namely, F. culmorum, F. redolens, F. oxysporum, F. avenaceum, F. equiseti and of nine isolates of F. culmorum to adult wheat plants were made in 1938.

Methods and Materials

Seed of Renown wheat inoculated by the spore-suspension method was planted in artificially-infested soil (600 c.c. of oat-hull inoculum per rod-row where required). One part of oat-hull inoculum was used to nine parts of autoclaved soil, by volume. This soil inoculum was incubated for 15 days and then applied at seed level at the rate of 600 c.c. per rod-row.

The complete experiment consisted of four replicates of 16 plots each. Each plot contained two rod-rows, one in which 100 seeds were spaced in the row for estimating

the amount of disease and in the second row a weighed amount of seed was sown for furnishing the yield data. Data on emergence were taken 24 days after seeding and notes on the incidence of root rot and yield were recorded later. Details concerning the technique of infecting the soil of field plots with cereal root rot fungi, and of recording the amount of infection on mature plants used in the present investigations have already been given by Greaney, Machacek and Johnston (18).

Experimental Results

The analyses in Table 6 establish significant differences between organisms, with respect to plant emergence, disease rating and yield. The complete results of the field experiment showing the relative pathogenicity of different species and forms of Fusarium as indicated by percentage of plants emerged, degree of root rot infection and yield, are summarized in Table 7.

From Table 7, it is evident that all of the isolates of F. culmorum tested were under the conditions of the test in 1938, more or less pathogenic to Renown wheat. Again, increases in the amount of root-rot infection brought about corresponding decreases in yield. Of the other four species of Fusarium tested F. equiseti and F. avenaceum were, when compared with the controls, only slightly pathogenic to Renown wheat, while F. redolens and F. oxysporum gave results that did not differ significantly from the uninoculated controls. It is interesting to point out that

Table 6. Complete analyses of variance for percentage of plants emerged, disease rating and yield. (Winnipeg Field Experiment, 1938)

Variance due to	Degrees of freedom	Sum of squares	Mean square	F	5% point
Replicates Organisms Error	Plant Emergence				
	3	1605.56	535.19	25.68	1.91
	15	14748.94	983.26		
	45	1722.44	38.28		
Total	63	18076.94			
Replicates Organisms Error	Disease Rating				
	3	447.73	149.24	2.16	1.91
	15	824.46	54.96		
	45	1144.93	25.44		
Total	63	2417.12			
Replicates Organisms Error	Yield				
	3	439.33	146.44	5.32	1.91
	15	1633.68	108.91		
	45	920.45	20.45		
Total	63	2993.46			

Table 7. Relative pathogenicity of five species of Fusarium and of several isolates of Fusarium culmorum to Renown wheat grown under field conditions at Winnipeg, Man., in 1938.

Fungus	Isolate No.	Percentage of Plants emerged	Degree of Infection (Disease Rating)	Yield per acre bus.
<u>F. culmorum</u>	937	37	43.4	28.4
<u>F. "</u>	19A	28	43.2	27.4
<u>F. "</u>	529	30	41.4	24.2
<u>F. "</u>	1263	20	41.1	22.4
<u>F. "</u>	38	39	41.0	25.0
<u>F. "</u>	285	34	40.3	26.3
<u>F. "</u>	10E	35	39.2	26.5
<u>F. equiseti</u>		62	39.0	32.7
<u>F. avenaceum</u>		52	38.6	35.0
<u>F. culmorum</u>	19B	53	38.0	26.3
<u>F. "</u>	15	29	36.9	24.3
<u>F. redolens</u>		64	36.9	34.6
<u>F. oxysporum</u>		61	36.7	36.3
<u>F. culmorum</u>	19C	48	34.6	34.1
Control - A		66	31.0	35.9
Control - B		67	30.9	38.1
Standard error		\pm 3.09	\pm 2.52	\pm 2.26

the field results were in accordance with the results obtained in greenhouse pot tests with adult plants (Table 5).

IV. INFLUENCE OF ENVIRONMENTAL FACTORS ON PATHOGENICITY

Soil Moisture

Series of experiments were carried out in the greenhouse to determine the effect of soil moisture on the pathogenicity of F. culmorum, F. equiseti, F. avenaceum, F. redolens and F. oxysporium to Marquis wheat. The soil inoculation methods used were those employed in the pathogenicity tests with isolates of F. culmorum previously described. At the commencement of an experiment the soil in one half of the pots of each fungus was adjusted to 60 per cent of its moisture holding capacity (high moisture series), the soil in the remainder of the pots was adjusted to a moisture holding capacity of 40 per cent. (low moisture series). The moisture content of the soil was maintained by weighing the pots at two-day intervals and adding sufficient water to restore them to original weight. The two moisture series were tested at each of the following temperatures, 10°, 15°, 20°, and 25°C.

At the end of an experimental period of thirty days non-emerged plants, as well as the young seedlings, were lifted from the pots, and the extent of damage due to pre-

Table 8. Complete analyses of variance for plant emergence, disease rating and green weight of plants.
(Soil Moisture Studies, Experiment I)

Variance due to	Degrees of freedom	Sum of squares	Mean square	F	5% point
Plant Emergence					
Replicates	1	410.04	410.04	-	-
Soil moisture	1	36.00	36.00	1.76	4.15
Soil temperature	3	30.56	10.18	-	-
Organisms	3	1412.56	470.85	22.99	2.90
Soil M.x Soil T.	3	20.88	6.96	-	-
Soil M.x Org.	3	20.38	6.79	-	-
Soil T.x Org.	9	187.32	20.81	1.02	2.19
Soil M.x Soil T.x O.	9	39.74	4.42	-	-
Error	31	634.96	20.48	-	-
Total	63	2792.44			
Disease Rating					
Replicates	1	0.52	0.52	-	-
Soil moisture	1	0.02	0.02	-	-
Soil temperature	3	580.55	193.52	9.85	2.90
Organisms	3	73840.70	24613.56	1252.59	2.90
Soil M.x Soil T.	3	188.31	62.77	3.19	2.90
Soil M.x Org.	3	38.92	12.97	0.66	2.90
Soil T.x Org.	9	335.89	37.32	1.90	2.19
Soil M.x Soil T.x O.	9	114.63	12.74	0.65	2.19
Error	31	609.16	19.65	-	-
Total	63	75708.70			
Green Weight					
Replicates	1	2632.97	2632.97	-	-
Soil moisture	1	108.42	108.42	-	-
Soil temperature	3	6797.86	2265.95	10.88	2.90
Organisms	3	23104.64	7701.54	36.99	2.90
Soil M.x Soil T.	3	474.99	158.33	-	-
Soil M.x Org.	3	15.90	5.30	-	-
Soil T.x Org.	9	1869.77	207.75	0.99	2.19
Soil M.x Soil T.x O.	9	74.40	8.27	-	-
Error	31	6454.35	208.20	-	-
Total	63	41533.30			

emergence blight and seedling blight was recorded in the usual way.

The results of the analysis of variance tests for percentage of plants emerged, amount of disease and green weight of plants for the organisms, F. culmorum, F. equiseti and F. avenaceum appear in Table 8, while those for the fungi F. culmorum, F. oxysporum and F. redolens are given in Table 9.

It is clear from Tables 8 and 9 that the differences observed between soil moisture contents in these experiments were not significant. In no instance did the F. value exceed the 5 per cent point. There was, however, a very significant difference between organisms in these tests. Regarding the effect of temperature on the development of the species of Fusarium tested, it is evident from Tables 8 and 9 that temperature markedly influenced the degree of pathogenicity exhibited by various species of Fusarium on Marquis wheat seedlings. The complete results of the moisture experiments are summarized in Tables 10 and 11.

The evidence presented in Tables 10 and 11 show that soil moisture did not significantly increase or decrease the virulence of any of the species of Fusarium studied. In many instances, however, there was a tendency for low soil moisture content to increase the pathogenicity of these fungi, but the differences observed were not significant statistically.

Table 9. Complete analyses of variance for plant emergence, disease rating and green weight of plants.

(Soil Moisture Studies, Experiment II)

Variance due to	Degrees of Freedom	Sum of squares	Mean square	F	5% point
Plant Emergence					
Replicates	1	1190.25	1190.25		
Soil moisture	1	16.00	16.00	-	-
Soil temperature	3	124.12	41.37		
Organisms	3	4828.87	1609.62	36.56	2.90
Soil M. x Soil T.	3	78.87	26.29		
Soil M. x Org.	3	60.37	20.12		
Soil T. x Org.	9	107.26	11.91		
Soil M. x Soil T. x O.	9	55.26	6.14		
Error	31	1364.75	44.02		
Total	63	7825.75			
Disease Rating					
Replicates	1	3.66	3.66		
Soil moisture	1	66.62	66.62	1.09	4.15
Soil temperature	3	996.57	332.19	5.45	2.90
Organisms	3	68658.47	22886.15	375.67	2.90
Soil M. x Soil T.	3	68.15	22.71		
Soil M. x Org.	3	463.14	154.38	2.53	2.90
Soil T. x Org.	9	446.56	49.61		
Soil M. x Soil T. x O.	9	229.04	25.44		
Error	31	1888.74	60.92		
Total	63	72820.95			
Green Weight					
Replicates	1	2782.56	2782.56		
Soil moisture	1	2.80	2.80	-	-
Soil temperature	3	754.39	251.46	6.60	2.90
Organisms	3	7171.56	2390.52	62.80	2.90
Soil M. x Soil T.	3	141.01	47.00	1.23	2.90
Soil M. x Org.	3	30.88	10.29		
Soil T. x Org.	9	342.88	38.09	1.00	2.19
Soil M. x Soil T. x O.	9	128.52	14.28		
Error	31	1179.92	38.06		
Total	63	12534.52			

Table 10. The influence of soil moisture on the pathogenicity of Fusarium culmorum, Fusarium equiseti and Fusarium avenaceum to Marquis Wheat.
(Experiment I)

Organism	Temperature °C.	Percentage of plants emerged		Degree of infection (disease rating)		Green weight of plants (grammes)	
		High moisture	Low moisture	High moisture	Low moisture	High moisture	Low moisture
<u>Fusarium culmorum</u>	10	36	41	80.8	66.2	2.7	9.5
	15	37	42	87.4	87.5	5.0	9.3
	20	38	39	86.6	89.4	10.2	9.1
	25	36	38	88.1	90.2	8.8	7.4
<u>Fusarium equiseti</u>	10	45	47	2.7	2.2	25.5	32.4
	15	46	43	5.0	4.2	49.0	54.2
	20	48	49	4.8	8.1	62.7	65.1
	25	48	52	2.8	8.5	66.2	57.8
<u>Fusarium avenaceum</u>	10	43	45	7.4	2.9	25.2	34.0
	15	51	51	8.6	7.0	50.6	60.0
	20	49	46	9.1	10.0	59.0	66.6
	25	48	50	11.1	17.4	64.2	55.4
Control (no organ- ism)	10	52	53	3.2	2.2	27.2	33.8
	15	49	51	4.0	3.6	46.6	53.6
	20	49	50	3.6	5.1	63.5	67.0
	25	50	54	6.6	8.2	63.1	56.1
Mean of organisms		45	47	25.7	25.8	39.3	42.0

Table 11. The influence of soil moisture on the pathogenicity of Fusarium culmorum, Fusarium redolens, and Fusarium oxysporum to Marquis wheat.
(Experiment II)

Organism	Temperature °C.	Percentage of plants emerged		Degree of infection (disease rating)		Green weight of plants (grammes)	
		High moisture	Low moisture	High moisture	Low moisture	High moisture	Low moisture
<u>Fusarium culmorum</u>	10	40	32	75.2	86.4	5.1	4.1
	15	31	28	79.8	88.8	7.0	2.3
	20	30	31	88.1	92.4	3.0	1.8
	25	36	30	82.6	86.0	4.2	3.1
<u>Fusarium redolens</u>	10	57	55	1.6	1.2	22.6	22.5
	15	55	53	15.4	5.2	33.8	35.2
	20	52	52	15.8	6.7	26.4	36.3
	25	51	54	21.8	13.2	28.3	21.9
<u>Fusarium oxysporum</u>	10	54	52	1.5	1.4	20.4	21.1
	15	51	50	10.4	4.4	31.1	35.2
	20	51	53	18.4	11.6	29.8	35.0
	25	46	49	19.5	12.5	27.8	22.0
Control (no organ- ism)	10	55	52	1.8	0.8	22.9	23.3
	15	55	51	7.8	6.2	31.9	33.5
	20	52	52	16.3	7.3	29.5	36.0
	25	49	54	11.5	10.6	27.2	24.2
Mean of organisms		48	47	29.2	27.2	21.9	22.3

Soil Temperature

Another series of experiments were made to determine the direct effect of temperature on the pathogenicity of the five species of Fusarium mentioned above. Four pots of each fungus containing thirty seeds of Marquis wheat each were placed in each of four thermostatically controlled temperature tanks maintained at 10°, 15°, 20°, and 25° C., respectively. A uniform moisture content of 50 per cent of the total moisture-holding capacity of the soil was maintained in all pots. Each temperature experiment was carried out in duplicate, and the methods used for recording and analyzing the experimental data were essentially similar to those employed in previous greenhouse tests.

The data of two complete temperature experiments were examined by the analysis of variance method (Tables 12 and 13). The results of the analyses in Tables 12 and 13 show that the effects of temperature on the pathogenicity of species of Fusarium to wheat are very great. Although temperature did not markedly influence plant emergence, it had a very significant effect on the incidence of disease and on green weight of the plants. In no instance in the experiments was the interaction of temperature and organisms significant. From Tables 12 and 13 it is evident that highly significant differences exist between the degree of pathogenicity exhibited by various organisms tested. The complete results of the experiments, with the standard errors associated with the various factors studied, are summarized in Tables 14 and 15.

Table 12. Complete analyses of variance for plant emergence, disease rating and green weight of plants. (Soil temperature Studies, Experiment I)

Variance due to	Degrees of freedom	Sum of squares	Mean square	F	5% point
Emergence					
Replicates	3	455.06	151.69		
Temperatures	3	30.56	10.18	-	-
Organisms	3	1412.56	470.85	29.95	2.81
Organisms X Temp.	9	187.32	20.81	1.32	2.09
Error	45	706.94	15.71		
Total	63	2792.44			
Disease Rating					
Replicates	3	28.11	9.37		
Temperatures	3	580.56	193.52	9.43	2.81
Organisms	3	73840.70	24613.57	1199.49	2.81
Organisms X Temp.	9	335.88	37.32	1.82	2.09
Error	45	923.45	20.52		
Total	63	75708.70			
Green Weight					
Replicates	3	2810.07	936.69		
Temperatures	3	6797.86	2265.95	14.67	2.81
Organisms	3	23104.64	7701.55	49.86	2.81
Organisms x Temp.	9	1869.77	207.75	1.34	2.09
Error	45	6950.96	154.46		
Total	63	41533.30			

Table 13. Complete analyses of variance for plant emergence, disease rating and green weight of plants.

(Soil Temperature Studies, Experiment II)

Variance due to	Degrees of Freedom	Sum of Squares	Mean Square	F	5% point
Emergence					
Replicates	3	1215.25	405.08		
Temperatures	3	124.12	41.37	1.20	2.81
Organisms	3	4828.87	1609.62	46.72	2.81
Organisms X T.	9	107.26	11.91	-	-
Error	45	1550.25	34.45		
Total	63	7825.75			
Disease Rating					
Replicates	3	403.81	134.60		
Temperatures	3	996.58	332.19	7.61	2.81
Organisms	3	68658.47	22886.16	524.55	2.81
Organisms x T.	9	446.55	49.62	1.14	2.09
Error	45	1963.54	43.63		
Total	63	72468.95			
Green Weight					
Replicates	3	2790.66	930.22		
Temperatures	3	754.39	251.46	7.67	2.81
Organisms	3	7171.56	2390.52	72.93	2.81
Organisms X T.	9	342.89	38.10	1.16	2.09
Error	45	1475.02	32.78		
Total	63	12534.52			

From Tables 14 and 15 it is evident that temperature had a marked effect on the virulence of species of Fusarium. With all organisms, the greatest amount of injury to Marquis wheat seedlings occurred at the higher temperatures, that is at 20° C. and 25° C. It may be that the effect of temperature on the host is more important than the effect on the fungi. Dickson (7), for instance, obtained similar results to these obtained in the present study in his work on the seedling blight of wheat caused by Gibberella saubinetii (Mont.) Sacc., but the reverse with the seedling blight of corn caused by the same organism. His experiments lead to the conclusion that the plants are blighted most severely at temperatures which serve to predispose the respective plants to disease, that is, relatively high temperatures for wheat and low for corn. The present moisture and temperature studies, the results of which are given in Tables 10, 11, 14, and 15, indicate the importance of temperature on the occurrence of outbreaks of seedling blight and root rot of wheat caused by species of Fusarium. It is possible that soil moisture may also be an important factor in the occurrence of destructive epidemics of these root-infecting fungi. Further experiments are necessary to determine the significance of of this factor.

V. INFLUENCE OF ASSOCIATION OF OTHER SOIL FUNGI ON PATHOGENICITY

Pot culture tests were made to study the pathogenicity of Fusarium culmorum and Fusarium redolens alone, and in

Table 14. The influence of temperature on the pathogenicity of Fusarium culmorum, Fusarium equiseti and Fusarium avenaceum to Marquis wheat.
(Experiment I)

Temperature °C	<u>Fusarium culmorum</u>	<u>Fusarium equiseti</u>	<u>Fusarium avenaceum</u>	Control (no organism)	Mean
Percentage of plants emerged					
10	39	46	44	52	45
15	40	45	51	50	46
20	39	49	48	50	46
25	37	50	49	52	47
Degree of infection (Disease Rating)					
10	72.5	2.4	5.1	2.8	20.7
15	87.4	4.6	7.8	4.1	26.0
20	88.0	6.5	9.5	4.8	27.2
25	89.2	5.7	14.3	7.4	29.2
Standard error	± 2.267	± 2.267	± 2.267	± 2.267	± 1.132
Green weight of plants (gm)					
10	6.1	29.0	29.6	30.5	23.8
15	7.1	51.6	55.4	50.1	41.1
20	9.7	63.9	62.8	65.2	50.4
25	8.1	62.0	59.8	59.6	47.4
Standard error	± 6.214	± 6.214	± 6.214	± 6.214	± 3.107

Standard error of organisms - Plant emergence = ± 1.979

Disease rating = ± 2.267

Green weight = ± 6.214

Table 15. The influence of temperature on the pathogenicity of Fusarium culmorum, Fusarium redolens, and Fusarium oxysporum to Marquis wheat.
(Experiment II)

Temperature °C	<u>Fusarium culmorum</u>	<u>Fusarium redolens</u>	<u>Fusarium oxysporum</u>	Control (no organism)	Mean
Percentage of plants emerged					
10	36	56	53	54	50
15	30	54	50	54	47
20	31	52	52	53	47
25	33	53	48	52	46
Degree of infection (Disease Rating)					
10	78.9	1.4	1.5	1.3	20.8
15	88.4	9.3	7.4	6.9	28.0
20	83.8	11.2	14.1	11.8	30.2
25	84.3	16.4	16.0	12.1	32.2
Standard error	± 3.30	± 3.30	± 3.30	± 3.30	± 1.65
Green weight of plants (gm)					
10	4.6	22.6	20.7	23.1	17.8
15	2.6	34.5	33.2	32.7	25.8
20	4.4	31.3	32.4	32.8	25.2
25	3.6	25.1	24.9	25.7	19.8
Standard error	± 2.86	± 2.86	± 2.86	± 2.86	± 1.43

Standard error of organisms - Plant emergence = ± 2.93

Disease rating = ± 3.30

Green weight = ± 2.86

association with certain common soil-inhabiting fungi. The methods of soil infestation, planting of seed, harvesting, and of recording and analyzing experimental data used have already been described in a previous section of this paper. In the present experiment, however, wherever association effects were studied, the total quantity of inoculum incorporated into the soil was double the amount used where a fungus was tested singly. The soil fungi used in the investigation were isolated in 1936 and 1937 from the soil of permanent grain plots at Winnipeg, Man.

The particular pathogen investigated was a strain of Fusarium culmorum (No. 38) which was originally isolated in 1932 from diseased roots of durum wheat. The pathogenicity of this fungus on wheat was established by previous greenhouse pot tests (Table 4). The strain of Fusarium redolens used was isolated in 1935 from wheat roots.

In the first experiment (Experiment I) Fusarium culmorum (No. 38), Pyrenopeziza confluens (Pers.) Tul. and Trichoderma lignorum (Tode) Harz were studied singly; while Fusarium culmorum / P. confluens, Fusarium culmorum / T. lignorum and P. confluens / T. lignorum were studied in combination.

Fungi tested singly in the second experiment (Experiment II) were, namely, Fusarium culmorum, Fusarium redolens

Table 16. Complete analyses of variance for plant emergence, disease rating, and green weight of plants.
(Association Studies, Experiment I)

Variance due to	Degrees of Freedom	Sum of squares	Mean Square	F	5% point
Plant Emergence					
Replicates	3	1137.57	379.19	44.15	2.34
Temperatures	1	4.57	4.57		
Organisms	6	10958.25	1826.37		
Org. X Temp.	6	140.18	23.36		
Error	39	1613.43	41.37		
Total	55	13854.00			
Disease Rating					
Replicates	3	224.71	74.90	520.85	2.34
Temperatures	1	185.42	185.42		
Organisms	6	89566.85	14927.80		
Org. X Temp.	6	266.79	44.46		
Error	39	1117.96	28.66		
Total	55	91361.73			
Green Weight					
Replicates	3	1101.63	367.21	171.49	4.08
Temperatures	1	12156.96	12156.96		
Organisms	6	33083.03	5513.84		
Org. X Temp.	6	8892.18	1482.03		
Error	39	2764.78	70.89		
Total	55	57998.58			

Table 17. Complete analyses of variance for plant emergence, disease rating, and green weight of plants.
(Association Studies, Experiment II)

Variance due to	Degrees of freedom	Sum of squares	Mean squares	F	5% point
Plant Emergence					
Replicates	3	1070.89	356.96		
Temperatures	1	102.78	102.78	1.41	4.03
Organisms	8	11839.70	1479.96	20.29	2.13
Org. X Temp.	8	306.53	38.32	-	-
Error	51	3720.10	72.93		
Total	71	17040.00			
Disease Rating					
Replicates	3	884.48	294.83		
Temperatures	1	1855.42	1855.42	42.89	4.03
Organisms	8	84198.06	10524.75	243.35	2.13
Org. X Temp.	8	2747.13	343.39	7.93	2.13
Error	51	2205.89	43.25		
Total	71	91890.98			
Green Weight					
Replicates	3	1099.47	366.49		
Temperatures	1	3537.81	3537.81	87.31	4.03
Organisms	8	7299.66	912.46	22.52	2.13
Org. X Temp.	8	2082.09	260.26	6.42	2.13
Error	51	2066.57	40.52		
Total	71	16085.60			

Aspergillus flavipes and Penicillium intricatum; while those studied in combination were F. culmorum / A. flavipes, F. culmorum / P. intricatum, F. redolens / A. flavipes and F. redolens / P. intricatum. An adequate number of pots of uninfested sterile soil planted with surface-sterilized seeds of Marquis wheat served as controls in each experiment. The various experiments were adequately replicated.

The complete analysis of variance for percentage of plants emerged, disease rating and green weight of plants for Experiments I and II are given in Tables 16 and 17. An examination of the analyses in these tables shows that a high degree of significance can be attached to the differences observed between the pathogenicity of the various organisms and combinations of organisms studied in each test. Significant temperature differences were also obtained in each experiment for disease rating and green weight of plants. The summarized results of the experiments are given in Tables 18 and 19.

The results in Tables 18 and 19 show that the strain of F. culmorum used in these tests was very strongly pathogenic to wheat seedlings, while F. redolens was decidedly non-pathogenic. The virulence of F. redolens was not influenced in the presence of any of the other soil fungi. The important point in these experiments was the fact that the presence of the fungi Pyronema confluens and Trichoderma lignorum suppressed, to some degree

Table 18. The effect of Pyronema confluens and Trichoderma lignorum on the pathogenicity of Fusarium culmorum to wheat seedlings.
(Experiment I, Average of four trials)

Organisms added to sterile soil	Percentage of plants emerged		Degree of infection (Disease Rating)		Green weight of plants (grammes)	
	10°C	20°C	10°C	20°C	10°C	20°C
<u>F. culmorum</u>	17	16	86.1	91.3	1.6	4.4
<u>P. confluens</u>	48	50	3.4	2.9	28.5	73.6
<u>T. lignorum</u>	48	48	2.8	3.3	25.6	81.1
<u>F. culmorum</u> / <u>P. confluens</u>	28	21	73.6	84.1	3.2	6.0
<u>F. culmorum</u> / <u>T. lignorum</u>	25	23	77.7	87.0	3.3	2.3
<u>P. confluens</u> / <u>T. lignorum</u>	48	48	2.4	2.4	27.7	66.5
Control (no organism)	49	48	1.9	2.2	26.2	88.5
S.E. of organisms	± 3.21	± 3.21	± 2.67	± 2.67	± 4.21	± 4.21
Temperature mean	38	37	35.4	39.0	16.6	46.1
S.E. of mean of temp.	Insignificant ± 1.01				± 1.59	

Table 19. The effect of Aspergillus flavipes and Penicillium intricatum on the pathogenicity of Fusarium culmorum and Fusarium redolens to wheat seedlings.
(Experiment II, Average of 4 trials)

Organisms added to sterile soil	Percentage of plants emerged		Degree of infection (Disease Rating)		Green weight of plants (grammes)	
	10°C	20°C	10°C	20°C	10°C	20°C
<u>F. culmorum</u>	28	19	59.1	88.3	4.7	2.8
<u>F. redolens</u>	51	49	1.2	2.6	15.5	34.6
<u>A. flavipes</u>	51	52	1.6	2.5	14.4	36.0
<u>P. intricatum</u>	46	50	1.0	3.3	13.6	37.1
<u>F. culmorum</u> / <u>A. flavipes</u>	24	20	61.2	86.0	3.7	3.5
<u>F. culmorum</u> / <u>P. intricatum</u>	28	19	61.0	89.6	4.2	3.0
<u>F. redolens</u> / <u>A. flavipes</u>	50	50	0.6	1.9	13.2	36.5
<u>F. redolens</u> / <u>P. intricatum</u>	52	49	0.7	2.3	14.0	36.2
Control (no organism)	51	50	0.9	1.9	14.4	34.2
S.E. of organisms	± 4.26	± 4.26	± 3.28	± 3.28	± 3.18	± 3.18
Temperature mean	42	40	20.8	30.9	10.8	24.8
S.E. of mean of temperature	Insignificant		± 1.09		± 1.06	

at least, the virulence of F. culmorum, while Penicillium intricatum and Aspergillus flavipes did not.

The results of the experiments again established the fact that the pathogenicity of Fusarium spp. is appreciably influenced by temperature. In both experiments F. culmorum, whether alone or in combination with other fungi, proved to be more virulent at 20° C. than at 10° C.

The present experiments emphasize the importance of the reaction of one living micro-organism upon another in the soil, and confirm the experiments of Henry (19) Sanford and Broadfoot (30), Garrett (11), Bisby, James and Timonin (1), and others, who found that infection of wheat seedlings by root-rotting fungi was suppressed by the antagonistic effect of certain other soil fungi and bacteria. Further studies are still needed to determine the exact nature of the antagonism involved.

VI. DISCUSSION

A root disease of small grain crops, usually referred to as common root rot, is very destructive in the three prairie provinces of Canada. Fusarium culmorum has been consistently isolated from the diseased plants, while a number of other species of Fusarium have been consistently isolated from grain soils. These organisms are widely distributed throughout the grain-growing regions of Manitoba, Saskatchewan, and Alberta. For many years, plant disease

surveys and field observations and experiments have indicated that the incidence of the root disease of cereals caused by species of Fusarium, particularly by F. culmorum, varies greatly from year to year. The cause of this variation has been generally attributed to such factors as the occurrence of physiologic forms of the pathogens, to differences in soil and climatic conditions, and to the association effects of other soil-inhabiting micro-organisms. The present investigation was undertaken to obtain more definite information concerning the relative pathogenicity of several isolates of Fusarium, and to study various environmental factors influencing their pathogenicity to wheat.

Under the conditions of the experiments, the pathogenicity of the species of Fusarium tested was fairly consistent. The fungi that attacked wheat seedlings severely were also markedly pathogenic to adult wheat plants, and those exhibiting weak or non-pathogenic capabilities on seedlings were similarly weakly or non-pathogenic to adult plants. This result does not confirm the results of Sanford and Broadfoot (29) who found in their study of a large number of isolates of Fusarium (culmorum type) that no correlation existed between the degree of pathogenicity exhibited by these fungi on wheat seedlings and the degree of their pathogenicity on adult wheat plants.

The results of the present investigation suggests that virulent strains of Fusarium do not commonly occur in nature. It would seem that most isolates of Fusarium obtained from plants and soil are very weakly, if at all, pathogenic to wheat plants. If there are only a few

virulent strains of Fusarium occurring in nature, then the important practical problem of breeding wheat varieties resistant to Fusarium root-rot is considerably simplified.

The present experiments support the view that the natural soil flora has a marked inhibitive effect on the development of cereal root-rotting fungi. The pathogenicity of a very virulent strain of F. culmorum was markedly suppressed by the antagonistic action of certain common soil-inhabiting fungi such as Trichoderma lignorum and Pyronema confluens. These results confirm the intensive studies of this subject made by Henry (19), Sanford and Broadfoot (30), Garrett (11), and others, and emphasize the importance of the factor of biological antagonism in the field control of cereal root-rotting fungi.

In the present pot culture tests the severity of root rot of wheat caused by species of Fusarium was appreciably influenced by environmental conditions. The effect of soil moisture and soil temperature upon infection by these fungi can apparently be attributed to the combined influence on the vigour of the fungus and on the resistance of the host. Higher soil temperatures accelerated the growth of the fungus and inhibited the vigour of the host plant. It was also indicated that infection with F. culmorum increased with decrease of soil moisture content. Throughout the investigation it was obvious that the onset and progress of the Fusarium disease are greatly enhanced by weakened plant growth.

Brown (4), reviewing Dickson's work with Gibberella suabinettii (Mont.) Sacc. (Fusarium graminearum Schwabe), states that a significant correlation exists between resistance and a type of metabolism which is governed by the prevailing temperature. He reports as follows: "A low soil temperature the starch of the wheat endosperm is hydrolyzed much more rapidly than is the protein, with the result that the seedling is rich in sugar but poor in nitrogen. The cell walls therefore thicken rapidly by the deposit of cellulose material upon the original pectic framework, and on that account became much less susceptible to fungal attack. On the other hand, at high temperatures both the starch and the protein are rapidly hydrolyzed, the seedling is definitely richer in soluble nitrogen, growth is much more rapid and the cell walls remain much longer in the primary pectic condition. Hence the greater susceptibility to fungal attack." The experiments herein reported support these views, and demonstrate that the pathogenicity of species of Fusarium attacking cereal plants is markedly influenced by soil temperature.

Contrary to the results of Sanford and Broadfoot (29) the present work has indicated that seedling blight is a very important phase of root rot of wheat caused by Fusarium culmorum. The results of the present study, however, substantiate the work of Robertson (26) who made a histological study of wheat plants and demonstrated that the cell wall thickens and lignification sets in with advancing age, reaching a maximum when the plants are about 40 days old. This morphological change naturally

inhibits the invasion of the roots and crown tissues of wheat plants by pathogenic fungi. It appears, therefore, that the seedling stage in wheat plants is a very critical one from the standpoint of root diseases; the primary roots being quite susceptible to injury and invasion by fungi.

In Manitoba, seedling blight of wheat, and of other cereals as well, is responsible for important losses in yield. Fortunately, the seriousness of these losses are fully recognized and effective methods of control are now well within the power of the grower.

VII. SUMMARY

Greenhouse and field studies were made to determine the pathogenicity of five species of Fusarium, namely, F. culmorum (W.G.Sm.) Sacc.; F. avenaceum (Fr. (Sacc.)); F. redolens Wr.; F. equiseti (Cda) Sacc. and F. oxysporum Schl. v. aurantiacum (Ik.) Wr., and of twenty-four isolates of Fusarium culmorum to Marquis wheat. The species of Fusarium studied were those most commonly isolated from the soil of permanent grain plots at Winnipeg, Man. during the two-year period, 1936 and 1937. On the other hand, F. culmorum has been consistently isolated from the roots and stem bases of diseased cereal plants. These species of Fusarium, particularly F. culmorum, are very widely distributed throughout the grain-growing regions of Manitoba, Saskatchewan and Alberta.

In addition to studies on the pathogenicity of various species of Fusarium to seedling and adult wheat plants, greenhouse experiments were made to determine the effect of certain environmental factors on the parasitic behaviour of these important soil-inhabiting and cereal root-infecting fungi.

Of the twenty-four isolates of Fusarium culmorum tested only one was distinctly pathogenic to wheat in both seedling and mature stages of plant growth. The other isolates of this fungus were either not at all pathogenic, or only very weakly pathogenic to wheat. No marked pathogenicity was exhibited by the cultures of F. redolens, F. equiseti, F. avenaceum and F. oxysporum, tested. From the results of these studies it would seem that distinctly pathogenic strains of fungi belonging to the genus Fusarium are not commonly isolated from diseased parts of cereal plants, or from the soil.

In the present investigation it was found that a direct relation exists between the degree of pathogenicity exhibited by species of Fusarium and temperature. High temperatures always favoured root rot development. Numerous tests with a very virulent strain of F. culmorum indicated that this fungus was markedly pathogenic at temperature of 10°, 15°, 20°, and 25° C.; the degree of infection being most severe at the higher temperatures. Of the other species of Fusarium tested, F. avenaceum was slightly path-

ogenic at 25° C., but it was not pathogenic at temperatures of 20°, 15°, or 10° C. Fusarium redolens, F. equiseti and F. oxysporum were not pathogenic at any of the temperatures employed.

Under the conditions of the experiments the pathogenicity of various species of Fusarium was not appreciably influenced by the moisture content of the soil. In no case were the differences observed between the high and low soil moisture contents used statistically significant. It was observed, however, that the amount of root rot infection caused by a virulent strain of F. culmorum on Marquis wheat was greatest when the soil moisture content was relatively low, and smallest when it was maintained at a fairly high level throughout the experimental period. In general, the greatest amount of infection with species of Fusarium occurred under conditions unfavourable for the growth of the host.

A comprehensive series of greenhouse pot tests demonstrated that the pathogenicity of a virulent strain of F. culmorum was markedly suppressed in the presence of such common soil-inhabiting fungi as Trichoderma lignorum and Pyronema confluens. The results demonstrated that certain common saprophytic soil-inhabiting organisms are capable of exerting a measure of natural biological control over Fusarium culmorum. The importance of certain environmental factors and of biological antagonism on the patho-

genicity of species of Fusarium to cereal plants is discussed.

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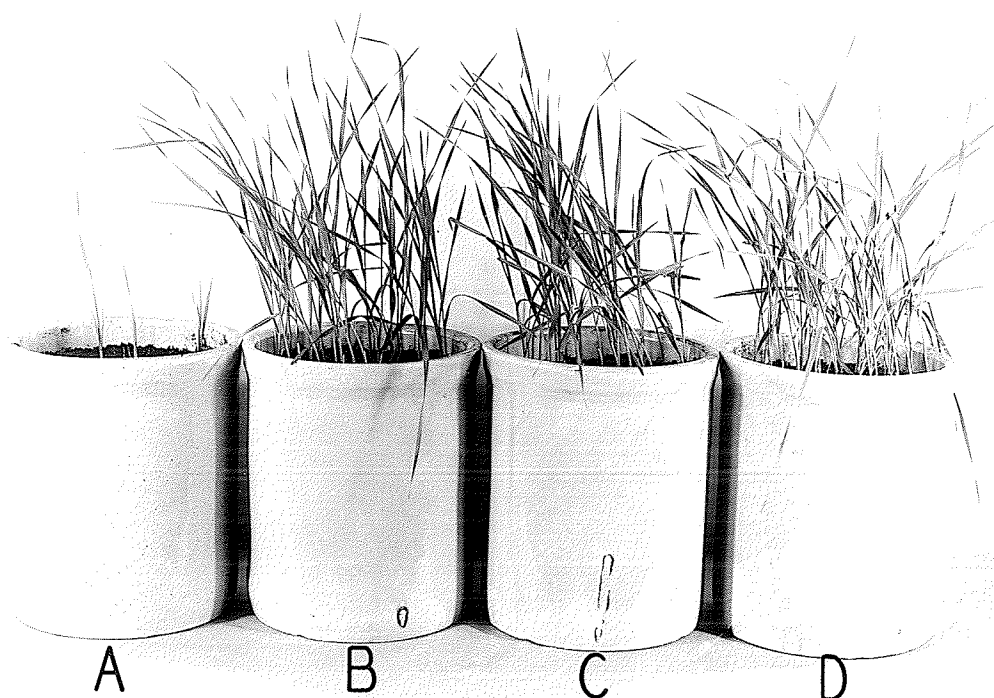
PLATES

Plate I



The relative pathogenicity of five isolates of Fusarium culmorum to Marquis wheat seedlings. Growth of plants at 22 days (25°C). Key to isolate number: A - 38, B - 1263, C - 1300, D - 937, E - 39, F - Control, no fungus, sterile soil.

Plate II



The relative pathogenicity of three species of Fusarium to Marquis wheat. Growth of plants at 22 days (25°C.). Arrangement of pot series: A - Fusarium culmorum (Isolate No. 38), B - Fusarium equiseti, C - Fusarium avenaceum, D - Control, seed and soil not inoculated.

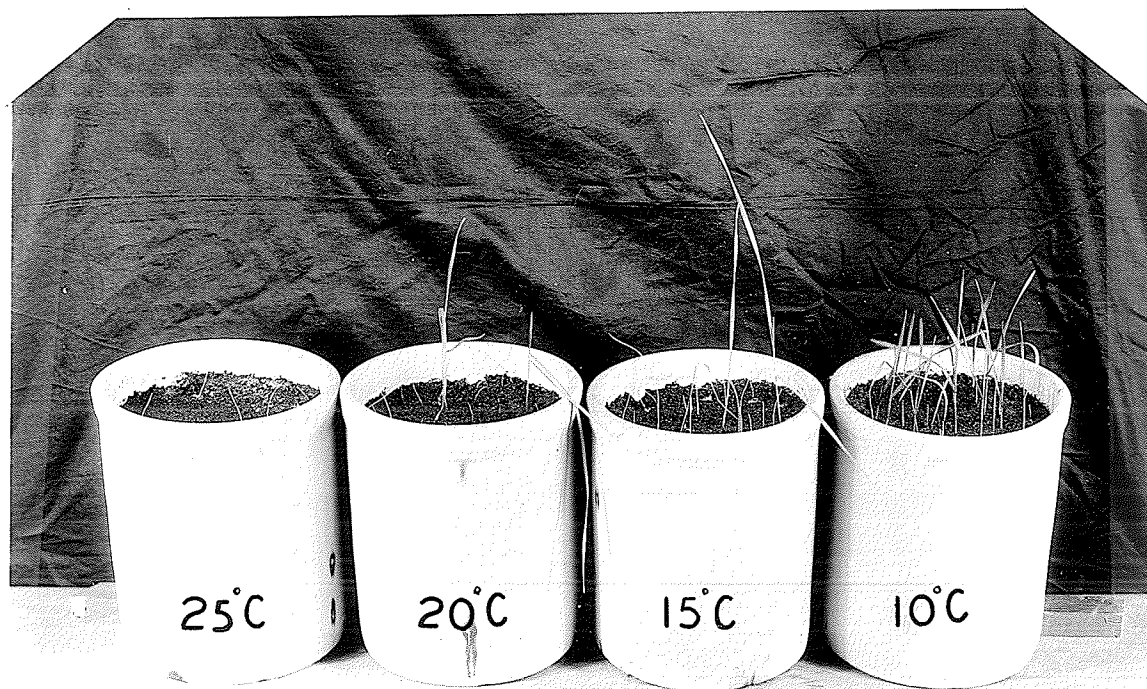
Plate III



The relative pathogenicity of three species of Fusarium to Marquis wheat. Growth of plants at 25 days (25°C.)

Arrangement of pot series: A - Fusarium culmorum,
B.-Fusarium oxysporum, C - Fusarium redolens, D - Control,
seed and soil not inoculated.

Plate IV



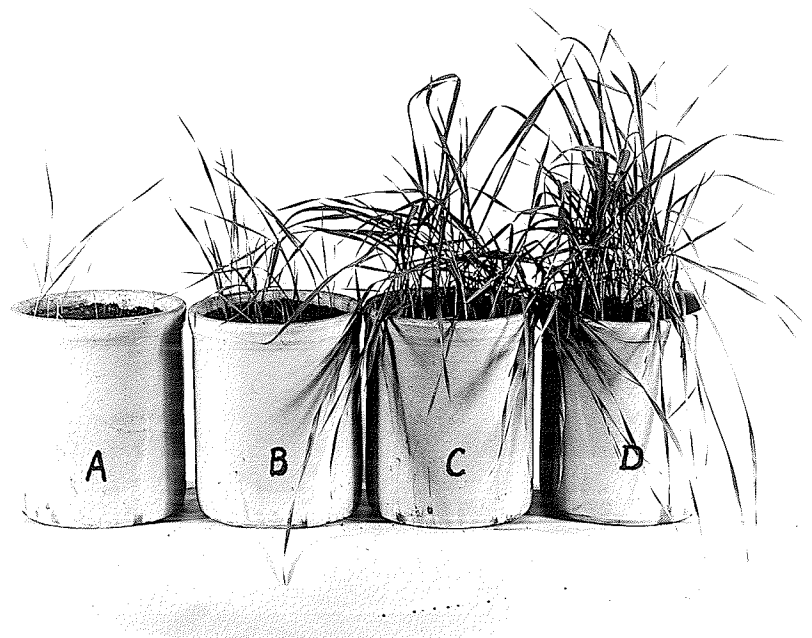
Effect of soil temperature on the pathogenicity of Fusarium culmorum (No. 38) to Marquis wheat. Growth of plants 20 days after sowing at four different temperatures.

Plate V



The pathogenicity of Fusarium culmorum (No. 38) as influenced by Trichoderma lignorum in greenhouse pot tests. Growth of Marquis wheat at 20 days (20°C.). Arrangement of soil series: A - Fusarium culmorum alone in sterile soil, B - Fusarium culmorum and Trichoderma lignorum in sterile soil, C - Trichoderma lignorum alone in sterile soil, D - Control, no fungus, sterile soil.

Plate VI



The pathogenicity of Fusarium culmorum (No. 38) as influenced by Pyronema confluens in greenhouse pot tests. Growth of Marquis wheat at 20 days (20°C.). Arrangement of soil series: A - Fusarium culmorum alone in sterile soil, B - Fusarium culmorum and Pyronema confluens in sterile soil, C - Pyronema confluens alone in sterile soil, D - control, no fungus, sterile soil.