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**Effect of crop rotation on canola seedling blight and soil
pathogen population dynamics**

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SUMMARY

Flexibility in rotation planning allows canola and field pea producers the ability to adapt to changing management practices and market opportunities. Current recommendations are to follow a one in four year rotation for canola or field pea on a particular field. The objective of this study was to determine the consequences of more intensive rotations of these crops on seedling emergence and other plant growth parameters, as affected by soil pathogens and soil population dynamics. Higher seedling emergence in the pasteurized soil and in the fungicide-treated seed than non-pasteurized soil and non-treated seed indicated that seedling emergence was affected by the soil pathogens. Seedling emergence and other growth parameters, including root vigour, shoot weight, root weight, seedling height and plant height were generally greater in the 4-year rotation and comparable among other rotations, although they were not always consistent. Soil pathogens associated with seedling blight and damping off of canola such as *Fusarium*, *Pythium* and *Rhizoctonia* were abundant in the soils collected from both Melfort and Scott. *Fusarium* had the highest soil populations, followed by *Pythium* and *Rhizoctonia*. This study indicated that a 4-year diverse crop rotation reduced the pathogen populations compared to intensive rotations. Soil pathogens were present in higher numbers in soils grown to hybrid canola compared to the conventional canola. Higher populations of *Fusarium* were present when pea was grown continuously.

INTRODUCTION

Canola cultivation practices have changed significantly in Canada during the last decade. Producers have adopted direct seeding and early seeding of hybrid and specialty oil canola varieties. Seedling blight and damping off of canola has a significant impact on crop establishment (Fig. 1). Losses have exceeded 10% at some locations. In 2005 and 2006, over 20% of fields surveyed in central Alberta suffered poor seedling establishment (DeMilliano, Orchard, pers. comm.).

The new hybrid and specialty oil varieties have different oil composition and lower plant vigour than conventional varieties, which may affect sensitivity to seedling blight. A significant input cost in growing hybrid/specialty canola is seed (up to \$45/acre plus user fees); therefore, control of seedling diseases has increased in importance to producers. Seedling blight of canola is caused primarily by *Rhizoctonia solani* (Fig. 2), and to a lesser extent by *Fusarium* sp. (Fig. 3) and *Pythium* sp. Cultural practices like crop rotation have been one of the primary tools for the management of crop diseases. Lower revenues from cereal crops and peas have resulted in pressure on growers to include canola more frequently in their rotations. Availability of herbicide and disease resistance in canola varieties has contributed to this trend. Research results from this project demonstrate that canola yields in the shorter crop rotation were reduced due to the combined effect of aggravated weed and disease pressure in the semi-arid Canadian prairie. Seedling blight is usually managed by application of fungicidal seed treatments, but producers have occasionally observed inconsistent protection against this disease.

Crop sequence research in western Canada has indicated that more diverse rotations tended to have less pest problems and lower production risk than rotations that were heavily cereal or broadleaf-based (Johnston et al. 2005). The recommendation to grow canola or field pea only once every four years is based primarily on the need to manage disease and weed pests. Growers frequently question whether improved weed control technology and cultivars with improved disease resistance can overcome these limitations. A 10% yield loss due to short rotation may more than compensate for a 20% price spread between canola and cereals in the short term. However, greater long-term yield losses may justify longer rotation periods. To address this question, a study was undertaken to compare a recommended 1 in 4 year crop rotation of canola and field pea with more intensive production of these crops in rotation with wheat and flax. The impact of fungicides was also examined within these rotations. To demonstrate the improvements made in canola technology since the original recommendation to grow canola only once in a four year rotation, a cultivar representing the latest technology (herbicide tolerant, blackleg resistant, hybrid) was compared with a cultivar that was commonly grown when this recommendation was made (conventional, blackleg susceptible, open-pollinated). Furthermore, the impact of these improvements on canola seedling blight severity and soil pathogen population dynamics was studied.

The implications of intensive production of canola and field pea were determined, while considering the cultivar and pesticide improvements that have been made since the one in four year rotation was recommended. The frequency of canola and field pea in the rotation impacted disease and weed control. Revised crop rotation recommendations for

canola and field pea, based on yield losses due to weeds have been evaluated, and the results of these studies have been reported earlier. The current report examines the impact of crop rotations on soil pathogen populations (*Fusarium*, *Pythium* and *Rhizoctonia*), and on the growth parameters of canola grown on soil collected from the rotation experiments conducted at Melfort and Scott, SK, under greenhouse conditions.

MATERIALS & METHODS

Soil sample collection

Soil samples were collected from field plots with various crop rotation histories, at Scott and Melfort, SK, after the cropping season in 2006 in collaboration with R. Kutcher, D. McLaren and B. Irvine. In 2007, soil samples were collected only from Scott, SK, as the Melfort site experiment was abandoned. Field experiments were designed as four-replicate split-plots with seven rotations as treatments (Table 1), with all phases of each rotation present every year. Fungicides were applied to sub-plots. Rotations with canola had cultivar as an additional factor. A total of 21 composite soil samples were collected from the seven crop rotations.

Assessment of soil fungal population

Growth medium

Three selective growth media, MPVM agar (selective for *Pythium*), PCNB medium (selective for *Fusarium*) and Ko and Hora (1971) medium (selective for *Rhizoctonia*) were used to isolate soil fungi.

Soil preparation

Plant debris was removed from the field soil samples. Sub-samples of about 20 g were taken from these samples and air-dried for 3-4 wk at room temperature. The soil samples were finely ground in a mortar with a pestle. Ten grams of soil from each sample were used to evaluate soil pathogen populations.

Soil dilution

The 10 g soil samples were added to 100 mL of a sterile 0.1% water agar medium to obtain a 10X dilution. The samples were diluted further to 50X and 250X using water agar medium.

Inoculation of plates

Using a 1000- μ L pipette, 0.5 mL samples from the 10X, 50X and 250X dilutions were plated onto the respective selective media for *Rhizoctonia*, *Pythium* and *Fusarium* using 4 or 5 Petri dishes for each sample. The Petri dishes were incubated at room temperature on a laboratory bench. Colony counts were performed after 3-4 days for *Pythium*, and after 5-7 days for *Fusarium* and *Rhizoctonia*.

Greenhouse bioassay study

Preparation of soil samples and seeding of canola

The remainder of the soil from each field sample collected in 2006 was divided into two equal portions. One half was pasteurized and the other portion was not pasteurized. The 2007 soil samples were not pasteurized. Each soil sample was potted into a medium size (3-inch) beer cup with holes at the bottom. Eight replicate cups were used for each soil

treatment in the first year trial, while four replicate cups were used for the 2007 samples. In 2006, the canola seed was not treated with fungicide. Since the soil collected in 2007 was not pasteurized, the seeds used were either treated with Apron Maxx (metalxyl-M + fludioxonyl) or left untreated. Ten canola seeds were seeded into each cup. The 2006 soil samples were arranged in a split-split-plot design with locations as the main plot, the soil treatment as sub-plots, and the soil samples in the sub-sub-plots. For the 2007 soil samples, the main plots were the seed treatments and sub-plots were the soil samples. This layout was also used for the Alberta and Saskatchewan soil samples.

Data collection

Seedling emergence was noted two weeks after seeding. After three weeks, the plants were harvested, the roots washed under running water to remove the soil and scored for development of root rot symptoms. Root vigour, dry root weight and shoot weight were also recorded.

Data analysis

Data from the fungal population study and greenhouse bioassay were subjected to analysis of variance. Data from Melfort and Scott from soil collected in 2006 were combined to identify if there were any significant differences between locations, pasteurized and non-pasteurized soil (2006). Similarly, data from treated and non-treated seeds were combined for soil samples collected in 2007. Since there were significant effects of the above-mentioned factors, although not all, on the response variables we ran

separate analysis of variance of the data. In addition, linear contrasts were made to compare different crop rotation effects on the same response variables.

RESULTS

Greenhouse bioassay study

Combined analysis of variance of the 2006 data indicated a significant site effect ($P \leq 0.01$) (Melfort vs. Scott) and soil pasteurization effect ($P \leq 0.01$). Seedling emergence was higher on Melfort soil compared to Scott soil, and in pasteurized than in non-pasteurized soil. The site effect was also highly significant ($P \leq 0.01$) on root vigour. Root vigour was greater in Melfort soil than in Scott soil. No significant site effect and pasteurization effect was found on dry shoot weight, although dry weight was higher for Melfort soil and lower in case of pasteurized soil.

Combined analysis of 2007 data indicated a significant seed treatment effect ($P \leq .01$) on seedling emergence, seedling height, and dry shoot and root weight. Seedling emergence, seedling height, dry shoot and root weight all were higher when the seeds were treated with Apron Maxx compared to non-treated seed.

Analysis of variance of the combined fungal population data for the soil from 2006 indicated there was no significant effect of location in the case of *Fusarium*, while the location effects were significant ($P \leq 0.01$) for both *Pythium* and *Rhizoctonia*. The *Pythium* population was higher in Melfort soil than in Scott soil; and the *Rhizoctonia* population was higher at Scott than at Melfort.

Linear contrasts of various rotations and treatments were conducted, including: four crops in a rotation CFWF/CWPW (canola-wheat- flusk-wheat/ canola-wheat-pea-wheat) vs. C (canola continuous); four crops vs. P (pea continuous); four crops vs. alternating C+P (canola + pea); four crops vs. three crops or less C+P+W (canola-wheat) +PW (pea-wheat)+ PCW (pea-canola-wheat); three crops PCW vs. CW +PW; hybrid vs. Westar; and fungicide vs. no-fungicide application.

Effect on seedling emergence

Results indicated that seedling emergence was not significantly different among most of the above-mentioned comparisons. Significant differences were observed when the 3-year rotation was compared with the 2-year rotation in sterilized soil and continuous crop in non-sterilized soil from Melfort (2006). Similar results were obtained for seedling emergence in Scott soil (2006); no significant differences were found among the comparisons in the sterilized soil. Significant differences were found when 4-year crop rotations were compared with rotations of 3 years or less and continuous pea; 3-year crop with continuous canola or pea; and continuous canola vs. continuous pea in non-sterilized soil. However, seedling emergence was higher in the 3- year rotation or continuous cropping. In 2007 (Scott soil), seedling emergence was not significantly different among the contrasts except 4-year rotation vs. 3 or less year rotation, where 3 or less year rotation had higher seedling emergence when Apron-treated seed was used. In the non-treated seed, the 4-year crop rotation had significantly higher seedling emergence compared to 1, 2, and 3-year crop rotations. In this trial, seedling emergence was significantly higher for Westar canola than for hybrid canola.

Effect on root vigour

Root vigour in 4-year rotations was higher compared to continuous cropping, 2-year, or 3-year rotations in both the sterilized and non-sterilized soil collected from Melfort in 2006 (Table 2). The 3-year rotation and continuous canola had higher vigour compared to continuous pea in sterilized soil, and the reverse was the case in non-sterilized soil.

However, not all of the comparisons were significantly different. In the soil collected at Scott in 2006, root vigour was lower in the 4-year rotation compared to others in both sterilized and non-sterilized soil, and all comparisons except two were statistically significant (Table 3). Root vigour in hybrid canola was higher in the Melfort soil and lower in the Scott soil in both sterilized and non-sterilized soil, although the difference was not statistically significant.

In 2007, root vigour was always higher in 4-year rotation relative to the other rotations, although the differences were also not statistically significant (Table 4). Significantly higher root vigour was found in the case of 1 and 2-year rotation in the treated seed, and 2-year rotation in the non-treated seed compared to 3-year rotation.

Effect on shoot weight

Among the contrasts, dry shoot weight was significantly higher in the case of the 4-year rotation compared to continuous, and in the 3-year rotation compared to alternating and continuous canola and/or pea when grown in soil collected at Melfort in 2006 that had been sterilized (Table 2). However, in non-sterilized soil from the same site, dry shoot weight was lower in the case of the 4-year rotation, compared to the 1 or 3-year rotations.

In both sterilized and non-sterilized soil, shoot weight was higher in soil from continuous canola than from continuous pea.

In the Scott soil, shoot dry weight was higher in the 3-year rotation compared to the 1 or 2-year rotations in both sterilized and non-sterilized soil.

No significant differences were observed in the case of shoot weight in the 2007 trial with fungicide treated seed, when the 4-year rotation was compared with less than 3-year rotations (Table 4). However, shoot weight was significantly higher in the soil collected from the plots that were treated with fungicide, 2-year crops and continuous crops, respectively, compared to plots without fungicide treatment and those with 3-year rotations. In the plots seeded with non-treated seed, the 4-year rotation produced significantly greater shoot weights compared to crop rotations of three years or less, and significantly higher shoot weights were found in the 1 or 2-year rotations compared to the 3-year rotation.

Effect on root weight

For the samples collected in 2007, significantly higher root weight was observed in the 4-year rotation compared to rotations of 3 years or less, and in the 3 years or less rotations compared to continuous crop or 2-year rotations for both treated and non-treated seed.

Effect on seedling height

For the samples collected in 2007, seedling height was significantly greater where fungicide seed treatments had been used compared to soils without fungicide treatment,

and under continuous cropping compared to 3-year rotations in the case of treated seed (Table 4). In the case of non-treated seed, seedling height was greater in 4-year crop rotations compared to crop rotations of 3 years or less, in the continuous crop compared to 3-year rotations and in samples where Westar canola had been seeded, compared to hybrid canola.

Effect of soil fungal populations

The greatest number of colony-forming units (CFU/10g soil) isolated from soil collected in 2006 represented *Fusarium* sp., followed by *Pythium* sp. and *Rhizoctonia* sp. (Table 5). In Scott, more fungal isolates were obtained from the soil collected in 2006 than from the soil collected in 2007 (Tables 5 and 6). In general, the number of CFU of each fungus was less in the 4-year rotations compared to the other rotations, although not all of the contrasts were statistically significant, at either Melfort or Scott. It was interesting to note that the number of CFU for all of the fungi was greater in the soil where hybrid canola was grown than in the soil where Westar was grown at both sites, and in both years for Scott. In most cases, these comparisons were statistically significant. Furthermore, the number of CFU of *Fusarium* was consistently higher in the soil where pea was grown continuously compared to the plots where canola was grown continuously. In soils where canola was grown continuously, consistently greater numbers of *Pythium* sp. were isolated, compared to soils where pea was continuously grown.

DISCUSSION AND CONCLUSIONS

In this study, seedling emergence and other growth parameters were higher in pasteurized soil and when the seeds were treated with fungicide, suggesting that pasteurization and seed treatment reduced the effect of soil pathogens. The most abundant of these pathogens were *Fusarium*, *Pythium*, and *Rhizoctonia*. Diverse crop rotations are known to reduce the quantity of pathogen propagules in the soil, and continuous planting of any crop increases diseases and pests specific to that crop, causing a reduction in the yield. Our greenhouse study also indicated seedling emergence and the growth parameters were greater in the diverse 4 year crop rotation. Similarly, the soil pathogen populations were reduced in the soil following four diverse crop rotations as compared to more intensive rotations. Selective increase of crop-specific pathogen populations was also noted in this study. For example, the population of *Fusarium* sp. was consistently higher in soil where pea had been grown continuously, and likewise the population of *Pythium* sp. was higher under continuous cropping of canola. Therefore, diversity in crop rotation can reduce disease pressure caused by soilborne pathogens. Crop rotation alone may not be effective for disease management, and an integrated approach including varietal resistance, other cultural practices and fungicides needs to be utilized for economic disease management.

Table 1. Rotation numbers and descriptions at Melfort and Scott, SK.

Rotation Number	Rotation Description
1	Continuous canola
2	Continuous field pea
3	Canola-wheat
4	Pea-wheat
5	Pea-canola-wheat
6	Canola-wheat-pea-wheat
7	Canola-wheat-flax-wheat

Table 2. Effect of crop rotation on canola growth parameters on Melfort soil (2006) in the greenhouse

Contrast	Emergence		Root vigour		Shoot wt. (g)	
Sterilized soil						
Four crop vs canola continuous single crop	8.92	8.89	2.30	1.88	1.11	1.11
Four crop vs continuous crop	8.92	8.94	2.30	2.13	1.11	1.12
Four crop vs pea continuous crop	8.92	8.75	2.30	1.43	1.11	1.05
Four crop vs less than 3 or less crops	8.92	8.83	2.30	1.96	1.11	1.09
Canola hybrid vs Westar	8.77	9.37	2.13	1.75	1.12	1.08
Fungicide vs no-fungicide	9.38	8.78	2.04	2.09	1.01	1.12
PCW rotation vs 2 year crop	9.21	8.47	2.13	1.99	1.22	0.97
PCW vs continuous	9.21	8.89	2.13	1.88	1.22	1.11
C-continuous vs P-continuous	8.94	8.75	2.13	1.43	1.12	1.05
Non-sterilized soil						
Four crop vs canola continuous single crop	8.11	7.79	2.23	1.93	1.01	1.10
Four crop vs continuous crop	8.11	7.68	2.23	1.59	1.01	1.10
Four crop vs pea continuous crop	8.11	8.00	2.23	2.18	1.01	1.01
Four crop vs less than 3 or less crops	8.11	8.20	2.23	1.95	1.01	1.16
Canola hybrid vs Westar	8.14	8.67	2.01	1.92	1.13	1.07
Fungicide vs no-fungicide	8.13	8.17	2.00	2.07	1.12	1.10
PCW rotation vs 2 year crop	8.79	8.38	1.58	2.25	1.36	1.10
PCW vs continuous	8.79	7.79	1.58	1.93	1.36	1.10
C-continuous vs P-continuous	7.68	8.00	1.59	2.18	1.1	1.01

Numbers in boldface are significantly different at the P= 0.05 level by linear contrast.

Table 3. Effect of crop rotation on canola growth parameters on Scott soil (2006) in the greenhouse

Contrast	Emergence		Root vigor		Shoot wt. (g)	
Sterilized soil						
Four crop vs canola continuous single crop	8.77	9.89	1.64	2.06	1.18	1.14
Four crop vs continuous crop	8.77	8.87	1.64	2.00	1.18	1.67
Four crop vs pea continuous crop	8.77	8.93	1.64	2.19	1.18	1.18
Four crop vs less than 3 or less crops	8.77	8.89	1.64	1.90	1.18	1.16
Canola hybrid vs Westar	8.70	8.63	1.76	1.84	1.14	1.20
Fungicide vs no-fungicide	8.56	8.89	1.98	1.73	1.13	1.18
PCW rotation vs 2 year crop	8.56	8.91	2.13	1.69	1.23	1.11
PCW vs continuous	8.56	8.89	2.13	2.06	1.23	1.17
C-continuous vs P-continuous	8.87	8.93	2.00	2.19	1.67	1.18
Non-sterilized soil						
Four crop vs canola continuous single crop	6.33	6.50	1.68	1.87	0.95	0.95
Four crop vs continuous crop	6.33	7.50	1.68	2.16	0.95	0.96
Four crop vs pea continuous crop	6.33	4.50	1.68	1.31	0.95	0.94
Four crop vs less than 3 or less crops	6.33	7.18	1.68	1.77	0.95	0.99
Canola hybrid vs Westar	7.09	6.66	1.71	2.09	0.95	0.98
Fungicide vs no-fungicide	6.93	6.79	1.66	1.75	0.91	0.98
PCW rotation vs 2 year crop	7.88	7.88	1.81	1.69	1.01	0.97
PCW vs continuous	7.88	6.50	1.81	1.87	1.01	0.95
C-continuous vs P-continuous	7.50	4.50	2.16	1.31	0.96	0.94

Numbers in boldface are significantly different at the P= 0.05 level by linear contrast

Table 4. Effect of crop rotation on canola grown on Scott soil (2007) under greenhouse conditions

Contrast			Shoot wt.		Root wt.		Root vigour		Plant ht.	
Apron-Maxx treated seed	Emergence		(g)	(g)					(cm)	
Four crop vs canola continuous single crop	7.17	6.43	0.68	0.69	0.31	0.27	4.00	3.67	11.19	11.47
Four crop vs continuous crop	7.17	6.58	0.68	0.67	0.31	0.26	4.00	3.56	11.19	11.01
Four crop vs pea continuous crop	7.17	6.88	0.68	0.73	0.31	0.30	4.00	3.67	11.19	12.35
Four crop vs less than 3 or less crops	7.17	6.27	0.68	0.62	0.31	0.24	4.00	3.68	11.19	9.87
Canola hybrid vs Westar	6.50	7.50	0.62	0.70	0.24	0.28	3.81	3.81	10.97	10.33
Fungicide vs no-fungicide	7.35	6.44	0.74	0.61	0.29	0.27	3.90	3.79	11.68	10.65
PCW rotation vs 2 year crop	5.25	6.31	0.30	0.66	0.14	0.30	3.20	4.00	9.55	10.03
PCW vs continuous	5.25	6.58	0.30	0.69	0.14	0.27	3.12	3.67	9.55	11.47
C-continuous vs P-continuous	6.43	6.88	0.67	0.73	0.26	0.30	3.56	3.87	11.01	12.35
Non-treated seed										
Four crop vs canola continuous single crop	4.80	6.43	0.53	0.41	0.26	0.02	4.00	3.67	9.81	9.27
Four crop vs continuous crop	4.80	6.58	0.53	0.40	0.26	0.17	4.00	3.56	9.81	9.35
Four crop vs pea continuous crop	4.80	6.88	0.53	0.44	0.26	0.24	4.00	3.87	9.81	9.1
Four crop vs less than 3 or less crops	4.80	6.27	0.53	0.35	0.26	0.18	4.00	3.67	9.81	8.57
Canola hybrid vs Westar	4.61	7.50	0.44	0.47	0.21	0.23	3.78	3.81	9.19	9.99
Fungicide vs no-fungicide	6.00	6.44	0.49	0.43	0.23	0.21	3.90	3.80	9.57	9.22
PCW rotation vs 2 year crop	5.63	6.31	0.21	0.41	0.12	0.20	3.13	4.00	8.06	8.82
PCW vs continuous	5.63	6.58	0.21	0.41	0.12	0.17	3.13	3.67	8.06	9.27
C-continuous vs P-continuous	6.43	6.88	0.40	0.44	0.17	0.24	3.56	3.87	9.35	9.1

Numbers in boldface are significantly different at the P= 0.05 level by linear contrast

Table 5. Effect of crop rotation on soil mycoflora (2006)

Contrast	<i>Fusarium sp.</i>		<i>Pythium sp.</i>		<i>Rhizoctonia sp.</i>	
Melfort soil						
Four crop vs canola continuous single crop	4338	5117	1555	1327	225	174
Four crop vs continuous crop	4338	4700	1555	1525	225	146
Four crop vs pea continuous crop	4338	5950	1555	930	225	230
Four crop vs less than 3 or less crops	4338	5584	1555	1517	225	175
Canola hybrid vs Westar	5440	5200	1576	1440	201	187
Fungicide vs no-fungicide	4967	5133	1800	1486	164	199
PCW rotation vs 2 year crop	5733	6175	2133	1340	172	180
PCW vs continuous	5733	4700	2133	1525	172	146
C-continuous vs P-continuous	5117	5950	1327	930	174	230
Scott soil						
Four crop vs canola continuous single crop	5022	6600	680	1017	321	371
Four crop vs continuous crop	5022	5125	680	945	321	402
Four crop vs pea continuous crop	5022	9550	680	1160	321	310
Four crop vs less than 3 or less crops	5022	5508	680	682	321	416
Canola hybrid vs Westar	5354	4225	625	875	378	354
Fungicide vs no-fungicide	6540	4913	1084	555	335	388
PCW rotation vs 2 year crop	3750	4750	220	410	404	489
PCW vs continuous	3750	5125	220	945	404	402
C-continuous vs P-continuous	6600	9550	1017	1160	371	310

Numbers in boldface are significantly different at the P= 0.05 level by linear contrast

Table 6. Effect of crop rotation on soil mycoflora in soil collected at Scott (2007)

Contrast	<i>Fusarium sp.</i>		<i>Pythium sp.</i>		<i>Rhizoctonia sp.</i>	
Four crop vs canola continuous single crop	1833	2417	433	425	184	171
Four crop vs continuous crop	1833	1718	433	456	184	169
Four crop vs pea continuous crop	1833	3813	433	363	184	175
Four crop vs less than 3 or less crops	1833	2573	433	406	184	185
Canola hybrid vs Westar	2288	1406	440	381	194	139
Fungicide vs no-fungicide	1875	2375	430	414	167	190
PCW rotation vs 2 year crop	1750	3218	425	369	234	183
PCW vs continuous	1750	1718	425	456	234	169
C-continuous vs P-continuous	2417	3813	425	363	171	175

Numbers in boldface are significantly different at P= 0.05 level by linear contrast

Fig. 1. Roots collected in a field survey showing root rot symptoms



Fig. 2. Roots infected by *Rhizoctonia* sp. in a greenhouse study showing symptoms of root rot



Fig 3. Roots infected by *Fusarium* sp. in a greenhouse study showing symptoms of root rot



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