Root rot and wilt of mung bean in Ontario

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Root rot and wilt of mung bean caused severed losses in a seed increase field and a nursery in 1979 and 1980. Root rot was prevalent during the early growing season on clay soil. Wilt occurred during the flowering and late pod filling stages on clay and sandy soil. *Rhizoctonia solani, Thielaviopsis basicola, Fusarium oxysporum* and a *Fusarium* sp. isolated from diseased plants were evaluated for pathogenicity in the greenhouse. *R. solani* and *T. basicola* caused distinct lesions on roots and lower stems similar to those observed in the field. *F. oxysporum* and *Fusarium* sp. were non-pathogenic in greenhouse experiments.

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La pourriture des racines et la flétrissure du haricot mungo ont cause des pertes importantes dans un champ de multiplication de semence et une pépinière en 1979 et 1980. La pourriture des racines est répandue au debut de la saison de croissance en sol argileux tandis que la flétrissure apparait lors de la floraison et du remplissage des gousses sur les sols argileux et sableux. La pathogénicité de *Rhizoctonia solani, Thielaviopsis basicola, Fusarium oxysporum* et *Fusarium sp.* isolés a partir de plants malades, a été évaluée en serre. *R. solani* et *T. basicola* causent des lesions distinctes sur les racines et dans le bas des tiges, semblables a celles observées au champ. *F. oxysporum* et *Fusarium sp.* sont non-pathogenes dans des experiences en serre.

During August 1979, two fields of diseased mung bean [*Vigna radiata* (L.) Wilczek] were observed at King Grain Limited, Chatham, Ontario. One field consisted of clay loam and had been planted previously with soybean [*Glycine* max (L.) Merr.] and the other field consisted of sandy loam used as a breeding nursery. In the clay loam field, approximately 80% of the plants in a seed increase block of mung beans (cv. VC1089) were dead or had symptoms of wilt. Soybeans (cv. Premier) in adjacent border rows were healthy. In the sandy loam field, 90-100% of plants of certain lines were killed while others showed symptoms of wilt but no dead plants. White beans (*Phaseolus vulgaris* L.) and soybeans in border rows had no foliar symptoms of disease but roots had black or red lesions.

The root rot and wilt disease of mung bean had caused extensive plant loss and appeared to be a limiting factor in the commercial production of mung beans. This note reports the etiology of these diseases.

Materials and Methods

Wilted plants were collected in the field and transported to the laboratory in plastic bags. Leaf, stem and root segments were surface sterilized in a 1.25% sodium hypochlorite solution for 1-2 min and plated on potato dextrose agar (PDA). Clay soil collected from the vicinity of diseased plants was placed in 25 cm pots in a greenhouse at the Harrow Research Station. Three replicate pots were each planted with 25 seeds of one of the soybean cvs. Harcor or Evans or one of the mung bean cvs. VC1089, Kawa or M333. Plant stand and severity of root damage were determined 8 weeks from planting. The percentage or root surface that was discoloured or necrotic was assessed on a scale of 1-4, as follows: 1 = 0-10%. 2 = 11-25%, 3 = 26-50%. 4 = 51-100%. Sections of the lower stem and upper roots of 5 plants per replicate were surface sterilized in 1.25% sodium hypochlorite and plated on PDA. Five sections of root per plant were placed on carrot discs in a moist chamber to determine the incidence of *Thielaviopsis basicola*.

In 1980, isolations were made from field plots of VC1089 at three dates during the growing season. Ten plants from each of 3 replicates were selected randomly at 7, 9 and 12 weeks after planting. Three sections 5 mm in length from each stem were surface sterilized and plated on PDA.

The pathogenicity of isolates of Fusarium spp., Rhizoctonia *solani* and *T. basicola* were tested on soybean and mung bean in the greenhouse in 10 cm pots. Inoculum of *F. oxysporum* and an unidentified Fusarium sp. isolated from diseased mung bean roots was increased in sand-corn meal medium (SCM), 196 g sand 4 g corn meal 30 ml H_2O) for 4 weeks at 20-25°C. The inoculum and medium was mixed with steamed greenhouse potting mix at 0, 2.5, 5.0 and 10%(v/v). Ten seed of each cultivar were planted per pot. Treatments were replicated five times. Soybeans were harvested 8 weeks after planting and mung beans were harvested 15 weeks from planting.

Isolates of R *solani* from soybean and mungbean were cultured in CMS medium as described previously and mixed with greenhouse soil to obtain concentrations of 5 and 10%(v/v). Ten seeds were planted per pot and treatments were replicated five times. Observations were made 10 weeks from planting.

Inoculum of *T. basicola* from mung bean was prepared by washing the chlamydospores from culture plates of 20% V_8 juice agar and collecting them on a 22 μm mesh screen. Chlamydospores were mixed with soil to obtain inoculum densities of 1×10^2, 1×10^3 and 1×10^4 chlamydospore chains/g dry soil. Pots were planted with 10 seeds of VC1089, Kawa or M333. Treatments were replicated five times. Plants were harvested and examined for symptoms of root rot 8 weeks after planting.

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Table 1. Stand, disease rating and incidence of fungi isolated from soybean (cvs. Harcor and Evans) and mung bean (cvs. VC1089, M333 and Kawa) growing in field soil in the greenhouse.

Cultivar	Stand %	Disease rating*	Incidence of isolation (%)		
			F. oxysporum	R. *solani*	*T. basicola*
Harcor	69a**	1.9 ± 1.0	80	13	67
Evans	67a	1.9 ± 1.0	47	0	60
VC1089	41b	2.8 ± 1.0	67	7	33
M333	40b	2.6 ± 1.2	53	0	47
Kawa	24b	3.3 ± 0.6	40	20	60

*Root rot rating based on a scale of 1-4 where 1 = 0-10%. *2* = **11-25%, *3*** = ***26-50%*** and 4 = 51-100% of the root surface discoloured.
**Means followed by the same letter do not differ significantly according to Duncan's Multiple Range Test (P = *0.05).*
Note: Other fungi isolated included species of ***Alternaria, Chaetorniurn, Fusariurn, Phomopsis*** and unidentified fungi.

Results and Discussion

Field Observations

Three weeks after emergence, a high percentage of mung bean plants growing in clay soil showed symptoms of root rot. Leaves of infected plants wilted and stems became brown. Infected plants remained upright and red lesions in the cortex were evident on roots and stems near the soil line. Lower roots were completely rotted.

External discoloration or deep cankers were not as evident on plants observed later in the growing season. Diseased plants at a mid to late pod filling stage of growth showed symptoms of water stress. Lower petioles and leaves were collapsed. As the disease progressed, lower and upper leaves developed interveinal necrosis and eventually entire leaves became necrotic. Stems of affected plants appeared healthy although a salmon pink discolouration of the xylem was evident after stems were split longitudinally. The discoloured tissue extended **10-25** cm from the crown and lateral roots into the third or fourth internode and occasionally into the leaf petioles. A white or pink mycelium was frequently observed in the central cortex at the base of dead plants. Mycelium within xylem vessels was observed under the microscope.

Fungi isolated from diseased plants collected in the field consisted of ***F. oxysporum*** and a brown unidentified ***Fusarium*** s~., ***R. solani*** and species of ***Phomopsis, Alternaria*** and ***Chaetomium***. Chlamydospores of ***T. basicola*** in root lesions were observed under the microscope.

Greenhouse experiments

Organisms isolated from soybean and mung bean growing in field soil transported to the greenhouse were similar to those isolated from plants grown in the field (Table 1). Plant stands of mung bean as a percentage of stands in check pots containing steamed field soil were significantly lower than soybean stands. The root rot rating of mung bean was higher than soybean. Incidence of ***F. oxysporum,*** R. ***solani*** and ***T. basicola*** isolated from soybean and mung bean varied considerably and the incidence of isolation did not differ significantly (P = 0.05) among hosts. ***F. oxysporum*** was isolated frequently from mung bean plants growing in clay soil at 7, 9 and **12** weeks from planting (Table **2).** Other fungi including ***Fusarium*** sp, ***Phomopsis*** sp., ***Pythium*** sp. and ***R. solani*** were isolated significantly less frequency (P = 0.05).

Soil infestation with isolates of ***F. oxysporum*** and ***Fusarium*** sp. in greenhouse experiments did not result in infection or reduced stands of soybean and mung bean. Additional trials in which roots were dipped in a suspension of spores and transplanted into greenhouse soil failed to demonstrate the pathogenicity of the two species of ***Fusarium.*** Although the symptoms of mung bean wilt observed in the field and greenhouse resembled wilt induced by ***F. oxysporum.*** evidence of the involvement of this fungus was inconclusive.

Plant stands of mung beans were significantly less (P = 0.05) than soybeans after 10 weeks in soil infested with the mung bean isolate of R. *solani* (Table 3). Differences among cultivars

Table 2. Fungi isolated from mung bean (cv. VC1089) stems growing in field soil at 7, 9 and 12 weeks from planting.

Organism	Incidence in plant segments (%)		
	7	9	12
***Alternaria* sp.**	51 a*	50 a	32 bc
Fusarium oxysporum	50 a	49 a	68 a
***Fusarium* sp.**	12 b	33 b	34 b
Phomopsis sp.	8 bc	23 bc	17 cd
***Pythium* sp.**	O c	6 d	O d
Rhizoctonia solani	O c	2 d	6 d
Unidentified fungi	11 b	13 cd	1 d

* Means of 3 replicates, 10 plants/replicate, 3 segments/plant. Means followed by the same letter within a column do not differ significantly according to Duncan's Multiple Range Test (P = 0.05).

were less obvious but significantly different in pots containing the soybean isolate. Inoculum concentration did not significantly affect plant stands. Red cankers at or near the soil line were evident on most plants in the experiment and resembled those observed in the field 4 weeks after emergence. Plant losses occurred during emergence in pots in the greenhouse. Symptoms of wilt on older plants were rare. The vascular tissue of infected plants did not have the pink discoloration evident in wilted plants in the field during flowering and pod fill.

Plant stands of mung bean cultivars VC1089, Kawa and M333 were not affected by *T. basicola* infested soil (Table 4). Root rot rating increased significantly ($P = 0.05$) with increasing inoculum concentration. Significant differences in ratings among cultivars occurred at an inoculum concentration of 1×10^2 chlamydospore chains/g of dry soil but not at other concentrations. *T.* basicola caused brown to black lesions on tap and lateral roots but did not cause discolouration of the vascular tissue in the stem region. Wilt was not observed.

The pathogenicity of R solani and *T.* basicola to mung bean was demonstrated and it is probable that these fungi, especially *R.* solanicontribute to early season plant losses. The soybean cultivar Evans was more susceptible than Harcor when inoculated with the soybean isolate of *R.* solani but the mung bean isolate appeared less pathogenic on both soybean cultivars.

Table 3. Plant stand of soybean (cvs. Harcor and Evans) and mung bean (cvs. VC1089, Kawa and M333) after 10 weeks in soil infested with isolates of *Rhizoctonia solani* from soybean and mung bean.

	Isolate and inoculum concentration (v/v)			
	Soybean isolate		Mung bean isolate	
Cultivar	5%	10%	5%	10%
Harcor	81 a*	86 a	88 a	98 a
Evans	42 bc	44 b	84 a	80 a
VC1089	34 c	11 c	30 b	23 b
Kawa	47 bc	18 bc	41 b	41 b
M333	65 ab	20 bc	10 c	33 b

* Plant stand presented as a percentage of control. Means followed by the same letter within a column do not differ significantly according to Duncan's Multiple Range Test ($P = 0.05$).

The cause of late season wilt of mung bean was not determined but the disease symptoms and presence of *F. oxysporum* within infected tissue suggest this organism was responsible but additional research is required to confirm these observations.

Table 4. Effect of inoculum concentration of *Thielaviopsis basicola* on stand and root rot of 3 mung bean cultivars 8 weeks after planting.

	Inoculum concentration (spores/g soil)							
	0		1×10^2		1×10^3		1×10^4	
Cultivars	S*	R**	S	R	S	R	S	R
VC1089	8.6	(0)	8.4	(3.4)ab***	7.2	(4.1)	8.2	(4.9)
Kawa	7.5	(0)	8.0	(3.9)a	8.0	(3.7)	7.5	(4.6)
M333	8	(0)	8.8	(2.8) b	8.4	(4.2)	8.4	(4.8)

* Plant stand means of 5 replicate pots, 10 seed/pot.

** Root-rot rating (0-6) as follows: 0 = no lesion, 1 = lesion present but not coalescing to girdle the tap root, 2 = a girdling lesion 1-5 mrn long, 3 = a girdling lesion 6-20 mm long, 4 = a girdling lesion 21-40 mm long, 5 = a girdling lesion 41-60 mm long, 6 = a girdling lesion 60 mm long.

*** Means followed by the same letter within a column do not differ significantly according to Duncan's Multiple Range Test ($P = 0.05$).