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The Canadian Plant Disease Survey is a periodical of information and record on the occurrence and severity of plant diseases in Canada and on the assessment of losses from disease. Other original information such as the development of methods of investigation and control, including the evaluation of new materials, will also be accepted. Review papers and compilations of practical value to plant pathologists will be included from time to time.

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L'Inventaire des maladies des plantes au Canada est un périodique d'information sur la fréquence des maladies des plantes au Canada, leur gravité, et les pertes qu'elles occasionnent. La rédaction accepte d'autres communications originales notamment sur la mise au point de nouvelles méthodes d'enquête et de lutte ainsi que sur l'évaluation des nouveaux produits. De temps à autre, il inclut des revues et des synthèses de rapports d'intérêt immediat pour les phytopathologistes.

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## The 1979 blue mold epidemic of flue-cured tobacco in Ontario and disease occurence in subsequent years<sup>1</sup>

S.K. Gayed<sup>2</sup>

The incidence, spread, and severity as well as factors that contributed to the severity of the 1979 blue mold epidemic are documented. Losses in subsequent years were negligible. The origin of the 1979 epidemic in Ontario is discussed.

Can. Plant Dis. Surv. 65:2, 23-27, 1985.

L'incidence, l'étendu et la sévérité de même que les facteurs ayant contribués à la sévérité de l'épidémie de moisissure bleue de 1979, sont documentés dans cette article. Les pertes les années suivantes furent négligeables. L'origine de l'épidémie ontarienne de 1979 est discutée.

#### Introduction

Downy mildew of tobacco, commonly known as blue mold, and caused by *Peronospora tabacina* Adam, was first recorded in Ontario in 1938. The incidence and severity of the disease up to 1950 were well documented by Stover and Koch (12). From 1951 to 1966 blue mold which appeared sporadically in greenhouse seedbeds causing little economic loss (3), was considered a seedbed disease in both the U.S.A. and Canada (8). The disease was not recorded in Ontario from 1966 to 1978 (3). In the present publication, the incidence and severity of the disease as well as the factors that influenced the epiphytotic are documented. The source of the causal pathogen *P. tabacina* in Ontario is also discussed.

#### The Progress and Severity of the Epidemic

In 1979, blue mold caused by P. tabacina was first reported on 12 July in a field 3.2 km east of Tillsonburg, Oxford County, on tobacco plants started from container-grown seedlings (speedlings) imported from Florida. Yellow circular chlorotic spots, 12-20 mm in diameter, were observed on the adaxial surface of the lower leaves, with a corresponding greyish-blue downy growth of P. tabacina on the abaxial surface. On the same date and in the same field blue mold infection was also noticed on tobacco plants grown from seedlings produced by the grower in his own greenhouse. Plants from both sources were similar in size and color, but the blue mold lesions were fewer and smaller on the leaves of local plants. By 20 July, blue mold was reported on five more tobacco farms in the Haldimand-Norfolk Region, again on plants raised from Florida seedlings. The number of farms reporting the disease increased daily and by the end of July, blue mold was reported on hundreds of farms in the main tobacco area in Ontario (Table 1). Following the early appearance of blue mold symptoms on the lower leaves (Fig. 1), diseased spots coalesced forming large necrotic areas (Fig. 2). Gradually, leaf spots of P. tabacina appeared on the middle and even on the Systemic infection symptoms were totally absent at mid-July in tobacco plants including those from imported seedlings. Early systemic infection causes stunting of the plant as well as twisting and malformation of leaves (7). However, another form of systemic infection was noticed in mature plants. The plants were not stunted at this stage, but the pathogen was able to move within the vascular tissue of the leaf (Figs. 3 and 4) and reached the stem. Accordingly, tobacco plants in severe cases of systemic infection were weakened and toppled by the wind or by the weight of their foliage (Fig. 5). Necrosis of the vascular cylinder was evident when these plants were cut longitudinally (Fig. 6). In Australia and the U.S.A. this type of systemic infection is referred to as "stem infection" (7, 9). In August 1979, it was common to notice twisting and puckering of the tip leaves of tobacco plants although such leaves were visually free from leaf infection (Fig. 7). Similar symptoms were reported in France (12).

#### Factors that Favored the 1979 Epidemic

About 82% of the Ontario flue-cured tobacco crop in 1979 was grown within a 50 km radius of Delhi. Approximately 75% of the farms grew the cultivar Virginia 115; most of the remainder grew the cultivar Delhi 76. Although both cultivars are susceptible to blue mold, symptoms on the latter were more advanced and sporulation of *P. tabacina* was more active on the larger leaves of this cultivar. Such concentration of farms growing cultivars susceptible to the disease provided the pathogen with a vast area of susceptible host, a factor that contributes to epidemics (13).

In Ontario, tobacco plants were young in early July and it is known that young, growing plants are susceptible to blue mold (7). Moreover, the tobacco plants were tender and therefore more susceptible to the pathogen (14).

Downy mildew epidemics including those of *P. tabacina* are influenced by weather conditions (11, 12). In Ontario, at the end of June 1979, the weather was cloudy, cool, and humid (Fig. 8). Although dry weather prevailed during July, the tobacco leaf was wet for several hours daily, with dew. Dew has been proven to provide sufficient moisture for the germination of conidia of maize downy mildew (2). Moreover, most growers sprinkle irrigated their fields during July, which probably assured additional moisture for conidial germination. Periods of

tip leaves of many tobacco plants at the end of the season.

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cool nights were recorded between 18-22 July and 25-28 July (Fig.8). Thus weather conditions in July were favorable for blue mold infection and sporulation. The cool, cloudy weather in August was ideal for continued severity of the disease and random winds during the season contributed to the spread of the pathogen.

Sporulation of the pathogen was very active on tobacco leaves particularly the larger leaves of cv Delhi 76. It was common to notice sporulation of *P. tabacina* on both surfaces of the leaf. Since the pathogen can produce as many as one million conidia per square centimeter of sporulating leaf surface (8), spore load in the air in the main tobacco area was very high during the season. At the end of the season in midseptember, viable conidia were collected from lawns and foliage of trees; and during October floating conidia in the air gained entrance to a greenhouse and infected healthy tobacco seedlings.

In 1979, there were no fungicides registered in Canada to protect tobacco plants in the field from blue mold infection or to stop its spread, and tobacco companies made it clear that they would not accept any tobacco that had been treated with unregistered fungicides.

Table 1. The approximate accumulative number of tobacco farms that recorded blue mold in the main tobacco counties in Ontario at different periods during the 1979 epidemic.

Period between	Haldimand- Norfolk	Oxford	Brant	Elgin	Middlesex
10-20 July	5	1	0	0	0
21-29 July	15	10	0	3	0
30 July - 13 Aug.	682	180	154	85	16
14-22 Aug.	. 822	201	164	190	17
23-30 Aug	. 1048	210	184	241	61

Total: 1744 farms

#### Blue Mold in Ontario 1980-1984

In 1980, blue mold was active in certain tobacco areas in the U.S.A. causing considerable losses particularly in Georgia, Kentucky, North Carolina and Virginia (5). However, in 1980, there was no trace of blue mold in the main tobacco area in Ontario which had suffered badly from the 1979 epidemic. The disease was only reported on 5 August 1980 on one farm near Mount Brydges in Middlesex County and on a few farms 230 km to the east in Northumberland County (Fig. 10). The pattern of blue mold distribution in Ontario in 1979 (Fig. 9) was totally different from that in 1980 (Fig. 10). In those counties where the disease was recorded in 1980, tobacco growers did not adequately protect their crops with Ridomil sprays, whereas in the main tobacco area, growers were keen to protect their crops with the systemic fungicide. Fortunately crop losses were negligible in Ontario in 1980. In 1981, blue mold was reported in early August on one farm near Straffordville, Elgin County (6) and 3 farms in 1983 near Silver Hill, Haldimand-Norfolk Region, without measurable loss. In the 1982 and 1984 seasons, blue mold was not recorded in Ontario.

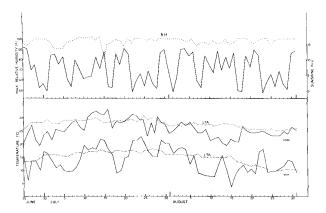
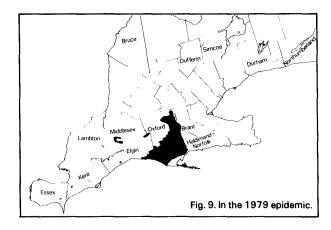
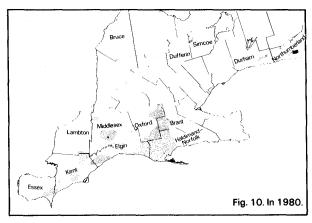


Figure 8. Maximum relative humidity, hours of sunshine, maximum and minimum temperature in the summer of 1979. (LTA - long term average).





Figures 9-10. Distribution of blue mold in the tobacco growing areas in Ontario. Dotted areas are the tobacco-growing areas, dark-shaded areas indicate the blue mold infected areas.

#### **Discussion**

For the past forty years, the source of *P. tabacina* infecting tobacco in Ontario has been a matter of speculation. It was believed that conidia from Kentucky and Ohio, carried by prevalent southwest wind, were the most logical source for infecting tobacco in Ontario (12). In the 1943 and 1944 seasons, it was reported that blue mold occurred in Ontario but not in Kentucky and Ohio (12). In both years, the nearest source for *P. tabacina* conidia was in Virginia, which the authors thought was too far away to be the source of spore showers that resulted in blue mold infection in Ontario. Although evidence was lacking, they attributed blue mold infection in both years to oospores of *P. tabacina* from previous crops which had overwintered in tobacco soil in Ontario (12).

Similarly in 1979, blue mold infection started in Ontario before Ohio and Kentucky (4). This probably lead tobacco specialists to believe that blue mold was introduced to Ontario on those seedlings imported from Florida and that the spread of the epidemic was mainly the result of these local infections (4).

The facts are against the theory that the disease was carried on the imported seedlings for the following reasons:

- The lack of early systemic infections which are common in the seedling stage. The systemic infections in the 1979 epidemic were only on mature plants.
- 2. The rapid spread and the devastating nature of the epidemic cannot be logically attributed to spread of local infections. This observation supports the view that spore showers from U.S.A. were the source of the epidemic in Canada (7). This assumption was clarified when the Department of Environment was contacted and Bhartendu (1) computed backward air trajectories and indeed he speculated that conidia from Virginia had arrived in the main tobacco area in Ontario on 29 and 30 June 1979. Similarly, air-borne conidia form Virginia might have been the source of blue mold in 1943 and 1944. There is no evidence to demonstrate tobacco infection with oospores of *P. tabacina*. Even after the 1979 epidemic, all trials to infect tobacco with oospores were unsuccessful (10).

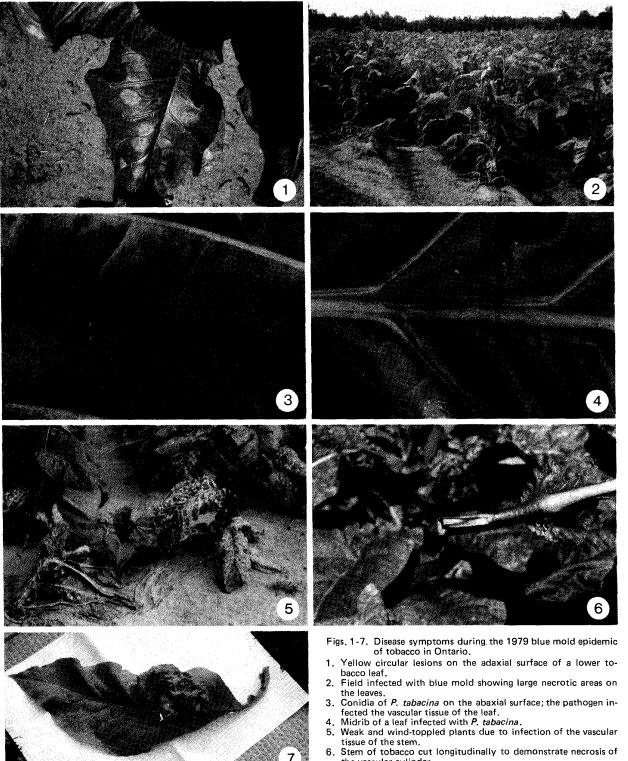
Still, a question remains unanswered: Why blue mold in 1979 in Ontario tended to show first on plants from imported tobacco seedlings as compared to those from local seedlings? According to the importers of the Florida seedlings, imported seedlings when received were much smaller than their own local seedlings and almost yellow in color as they had been raised in medium very poor in nitrogen. On July 12, on the farm where blue mold was first noticed in 1979, both plants from the grower's greenhouse and from imported seedlings were similar in size. This suggests that the imported plants were growing at a faster rate, probably as a result of having a better developed root system than those seedlings produced locally in greenhouse ground beds. There is evidence that rapidly growing plants develop more tender tissue which is susceptible to *P. tabacina*.

#### Acknowledgements

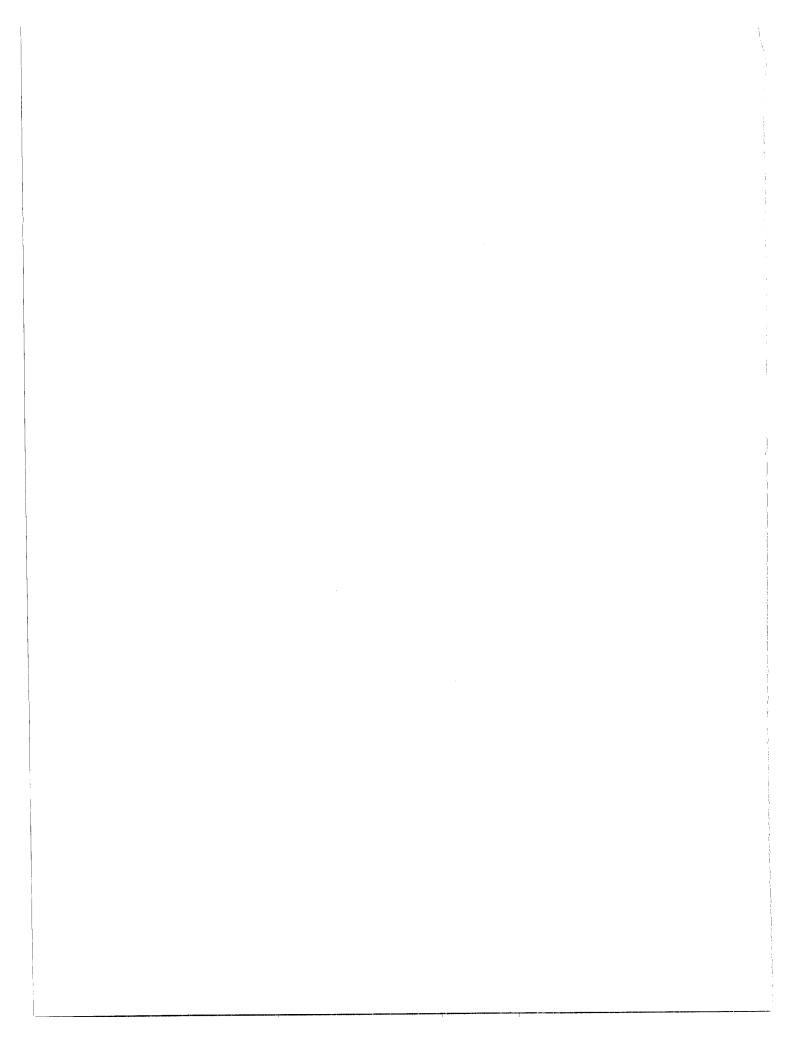
The author thanks D.A. Brown for drawing Figures 8, 9 and 10. Thanks are due to Dr. H.H. Cheng for providing data on the atmospheric RH during the 1979 season. The author is also grateful to Dr. P.W. Johnson, Director of Delhi Research Station for critically reading the manuscript.

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- the vascular cylinder.
  7. Puckered tobacco tip leaf.



## Effect of fungicides on Ramularia leaf and stalk spot of rhubarb in coastal British Columbia

D. J. Ormrod<sup>1</sup>, M. E. Sweeney<sup>2</sup> and L. S. MacDonald<sup>1</sup>

A fungus which caused economically damaging leaf and stalk spot of rhubarb in coastal British Columbia in 1983 and 1984 was identified as *Ramularia rhei* Allescher. Crop loss was reduced in 1984 through the application of fungicides applied four times between March 19 and May 3. The most cost effective fungicide was chlorothalonil followed by captan.

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Un champignon causant des taches sur les feuilles et les pétioles de la rhubarbe en 1983 et 1984 a été identifié comme *Ramularia rhei* Allescher. Les pertes ont été réduites en 1984 grâce à quatre applications de fongicides entre le 19 mars et le 3 mai. Le fongicide le plus économiquement rentable fut le chlorothalonil suivi du captane.

#### Introduction

Rhubarb (*Rheum Rhabarbarum* L.) is commonly grown in home gardens but is a minor commercial crop in coastal British Columbia. There are approximately 25 hectares in commercial field production with an annual farm value of \$100,000.

In the spring of 1983, leaf and stalk spot caused extensive damage rendering a three hectare crop unmarketable. Isolations were made and cultures submitted to the Biosystematics Research Institute of Agriculture Canada in Ottawa were identified as *Ramularia* sp. The disease occurred again in the spring of 1984 and a sample of infected leaves and stalks yielded *Ramularia rhei* Allescher and was deposited in the National Mycological Herbarium as accession DAOM 189221.

The fungus has been reported from Alberta, Saskatchewan, Manitoba, Quebec and Prince Edward Island but has not been reported previously from British Columbia (1).

Leaf infections of *Ramularia rhei* first appear as small red dots. These gradually enlarge to form more or less circular spots one cm or more in diameter. Large spots are white to tan coloured with purplish halos (Fig. 1). Stalk infections, which occur later, first appear as small spots which become elongated as the stalks grow (Fig. 2). The larger ones become tan coloured sunken lesions up to one cm in length. A white accumulation of conidia may be present in the centre of both leaf and stalk spots.

#### **Materials and Methods**

In June of 1983, an experimental plot was marked out in a severely diseased six-year-old planting of cv. Crimson at Deroche, B.C. Plots were 1 m  $\times$  8 m, replicated four times in a randomized complete block design. Sprays of Benlate 50 W (benomyl), DPX 3866 75 DF (benomyl), Bravo 500 F (chlo-

rothalonil), Captan 50 W (captan) and Rovral 50 W (iprodione) were applied June 7, 1983; and March 19, April 4, April 18 and May 3, 1984. Applications were made with a hand sprayer using a volume of approximately 600 litres of water and 100 ml of Super Spred surfactant/ha.

Each plot was rated for leaf infection on May 3, 1984 by assigning a value of one for slightly, two for moderately and three for severely infected leaves. Stalk infections were determined on May 15 when the entire experiment was harvested and stalks were sorted into three groups; those with no spots, those with 1-20 spots and those with more than 20. Stalks with more than 20 spots were considered to be unmarketable, (Fig. 3).

#### **Results and Discussion**

Spring rainfall was above average in 1983 and 1984 and leaf spot was severe in both years. Stalk infections were most numerous below heavily infected leaves suggesting that conidia washing down from the leaves were a source of inoculum for stalk infection.

Results of the fungicide trial are given in table 1. Bravo gave significantly lower numbers of leaf spots, unmarketable stalks (> 20 spots) and total infected stalks than any other treatment. Captan gave the second lowest unmarketable stalks although the two benomyl treatments were slightly lower than captan in leaf spot. Rovral was only slightly effective at the rate used. Economical disease reduction was achieved with Bravo and captan but the best treatment left 22% of the stalks with at least one infection. This may be due to the dense canopy which made thorough spray coverage extremely difficult.

Stalks with more than 20 spots are normally not acceptable either in the fresh or processed market and, in this trial, were rated culls. Heavily infected stalks were usually found under heavily infected leaves. This, combined with the difficulty of protecting stalks due to the dense canopy, suggests that fungicides should be applied in the early spring to protect the newly emerging and enlarging leaves.

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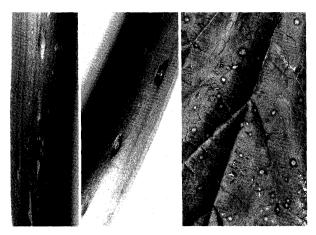
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Table 1. Cost effectiveness of fungicide applications for reduction of rhubarb leaf and stalk spot caused by Ramularia rhei.

Fungicio		Rate of Appn. (kg. a.i./ha)	Leaf Spot Rating*	% Stalks > 20 spots	% Stalks 1-20 spots	% Stalks Infected	Cullage (\$/ha)**	Cost of Ttmt. (\$/ha)***	Net Saving ( /ha) <sup>++</sup>
Bravo 50	00 F	1.75	20.7 a <sup>+</sup>	3.4 a	18.7 a	<b>22</b> .1 a	436	156	1238
Captan !	50 W	1.5	36.0 с	<b>5.9</b> b	24.1 a	30.0 b	761	101	968
Benomy 7	75 DF	0.9	32.0 b,c	8.2 c	28.0 a	36.2 c	1055	_+++	_
Benlate !	50 W	0.9	28.3 b	9.1 c,d	<b>22.1</b> a	31.2 b	1175	306	349
Rovral !	50 W	0.9	43.5 d	10.6 d	30.6 a	41.2 d	1357	363	110
untreated		-	71.5 e	14.2 e	29.7 a	43.9 d	1830	_	_

<sup>\*</sup>Visual rating total for each treatment where 3 = severely infected leaf; 2 = moderately infected leaf; 1 = slightly infected leaf (mean of 4 replications).

<sup>+++</sup> Cost not available.



Figures 1-3. Ramularia rhei on rhubarb.

1. Leaf spot. 2. Moderately infected stalk. 3. Heavily infected stalk (unmarketable).

Of the two most effective fungicides, captan would be preferred for grower use as it is already registered for use against *Botrytis* gray mould on rhubarb and could probably be approved for leaf spot control through the minor use program. It is also the lowest cost treatment. Bravo would be a suitable alternative but would have to be cleared for use, also probably through the minor use program. With this in mind, residue samples from the Bravo treated plots were collected and submitted for analysis.

#### Acknowledgement

We wish to thank Don Faulkner for the use of his crop, Cindy Holbrook for laboratory assistance and George P. White of the Biosyostematics Research Institute for identification of the fungus.

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<sup>\*\*</sup>Cullage calculated as value of crop lost due to >20 spots/stalk where crop is 45 tonnes/ha with a grower selling price of \$286/tonne.

<sup>\*\*\*</sup> Total cost of four applications including cost of fungicide at grower cost price plus \$40 allowance for 4 spray operations at \$10 each.

<sup>\*</sup>Means within columns followed by the same letter are not significantly different at the 5% level of Duncan's Multiple Range Test.

<sup>\*\*\*</sup>Compared to the untreated control.

cankers.

### Observations on the occurrence of European Canker in New Brunswick

E.N. Estabrooks<sup>1</sup>, K. Lynch<sup>2</sup> and G.W. Reed<sup>2</sup>

European canker (Nectria galligena Bres.) was found to be widespread throughout New Brunswick apple orchards following a survey of orchards in all regions. Severity of canker varied between sites. Although distinct differences in the amount of canker was not always consistent between cultivars, differences due to rootstocks showed more of a trend. Trees on more vigorous rootstocks such as B.A. and M106 usually show more

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Une étude menée au Nouveau-Brunswick a révélé que le chancre européen du pommier (*Nectria galligena* Bres.) est largement répandu dans les vergers de la province. La sévérité du problème varie selon les endroits. Bien que la présence de chancres ne semble pas toujours dépendre du cultivar, certains portegreffes pourraient accentuer le problème. Les porte-greffes vigoureux tels B.A. et M106 rendent habituellement leurs scions plus susceptibles au chancre.

#### Introduction

European canker (Nectria galligena Bres.), a disease common to most apple-growing areas of the world, was first reported in North America in Nova Scotia and New York in 1899 (1). Spores are known to be washed by rain or blown by wind into openings or wounds in the bark. Infections have been noted to occur in winter-damaged bark, pruning cuts, burrknots and through leaf-scars. After leaf drop in the fall, the open scars become sites for infection and these can be observed as infected buds and spurs in the following growing season. Observations in New Brunswick indicate that most cankers originate from this type of leaf scar infection and progress to perennial cankers which destroy tree productivity through girdling of trunks and scaffold limbs. The fruit rot stage of the disease has not been observed here. Although canker has been of some concern in New Brunswick since at least the 1960's, the increasing prevalence of disease in young orchards has resulted in great concern among commercial apple producers during the past five years.

Differences between cultivars in canker susceptibility have been reported in California orchards (2). In British Coliumbia the Summerland McIntosh and Harrold's Red Delicious were found to be susceptible to buildup of canker, whereas Spartan and Golden Delicious showed some tolerance (3). Interstock cultivar was reported to have little effect on tree mortality whereas scion cultivar was a highly significant factor. Moore in England reported differences in susceptibility of apple rootstocks to European canker as early as 1934 (4).

#### **Materials and Methods**

A preliminary assessment of European canker was conducted in three commercial apple orchards during the winter of

1981-82. In 1982 a follow-up survey was conducted in demonstration blocks in each of the five major apple-growing regions of the Province. Each block consisted of about 300 trees with four cultivars, and five rootstocks in various combinations. Seven trees of each cultivar/rootstock combination comprised one sample plot and each plot was replicated three times. All trees were approximately the same age having been planted between 1975 and 1978.

In addition, in 1983, two research orchard blocks at the Fredericton Research Station were surveyed for the presence of canker on the trunks of 12-year-old McIntosh on six different rootstocks. Cultivar/rootstock plots consisted of 6 trees replicated nine times in block 1 and 10 trees replicated nine times in block 2. All McIntosh (Mac) were of the Summerland strain except for one spur McIntosh (Dewar strain). Rootstocks surveyed were Beautiful Arcade (BA), Malling 106 (M106), Ottawa #5 (05), Malling 26 (M26) as well as interstem Ottawa #3 on Beautiful Arcade (03/BA) and Beautiful Arcade on Malling 26 (BA/M26).

Table 1. Cultivar/rootstock effect on prevalence of European Canker in three N.B. orchards.

Orchard	Cultivar/Rootstock	Age (Yrs)	No. of trees with Canker*
1	Spur Mac/M111	6	29
1	Cort./M7	6	19
1	Mac/Rob.5	18	45
1	Cort./Rob. 5	18	19
2	Mac/Rob. 5	20	43
2	Cort./Rob. 5	20	17
2	Mac/M7	6	20
3	Cort./M7	10	12
3	Spur Mac/M111	10	15

<sup>\*50</sup> trees of each cultivar/rootstock were rated for disease.

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Table 2. Rootstock effect on prevalence of European Canker in five regional apple plantings.

	Perc	Percent trees with canker						
Location	M9/M111	M26	M111	M106	M7	Ave.		
York County	64.9	67.3	71.7	57.4	87.9	67.8		
Carleton County	20.4	46.9	58.3	55.6	68.1	49.9		
Queens Count	y 31.6	32.3	45.0	37.2	72.4	43.7		
Charlotte County	13.6	18.3	17.5	46.7	39.7	27.2		
Westmorland County	8.5	25.9	34.3	25.4	45.3	27.8		
Ave.	27.8	38.1	45.4	44.5	60.7			

#### **Results and Discussion**

In the preliminary assessment of European Canker conducted in three commercial apple orchards during the winter of 1981-82, there was an apparent difference in the amount of canker between cultivars (Table 1). In orchard one, 90 per cent of the McIntosh on Robusta #5 trees (Mac/Rob. 5) were infected with canker compared to only 38 per cent of the Cortland on Robusta #5 (Cort./Rob. 5). In orchard 2 the ratio was similar with 86 per cent of the Mac/Rob. 5 trees infected compared to 34 per cent of the Cort./Rob. 5. In both of these orchards the trees had sustained considerable winter damage showing up as both southwest injury and limb crotch damage. These areas appeared to have become sites for canker infection. The ability of the Cortland cultivar to minimize winter damage compared to McIntosh may, in part, explain the differences in canker severity between the two cultivars.

From the 1982 survey of the five regional demonstration orchards it was apparent that the amount of canker varied between sites ranging from an average of 27.2% trees infected in south-western N.B. (Charlotte County) to 67.8% trees infected in York County (Table 2). No distinctive differences in the number of trees with canker could be detected between cultivars (Table 3). The number of trees having canker varied according to rootstock from an average of 27.8% for Malling 111 with interstem M9 (M9/M111) to 60.7% for trees on Malling 7 (M7) rootstock when considered over all five regional blocks.

In block 1 of the research orchard at the Fredericton Research Station containing the McIntosh cultivar on six rootstocks, distinct differences in the number of tree trunks with canker infections were noted. Mac/B.A. and Mac/M106 showed significantly more trees with cankers than any other combination present (Table 4). More than 20% of the trees on these rootstocks had cankers while less than 5% of the trees on M26 and BA/M26 rootstocks had cankers. No attempt was made to categorize the severity of canker in this block.

Table 3. Cultivar effect on prevalence of European Canker in five regional apple plantings.

	Percent trees with canker						
Location	Spur Mac	Cortland	Mac	Jerseymac			
York County	61.4	72.5	71.7	100.0			
Carleton County	35.8	57.5	62.8	37.9			
Queens County	30.2	28.9	49.2	50.0			
Charlotte County	37.5	17.9	40.0	61.1			
Westmorland County	23.0	26.1	42.0	38.6			
Ave.	37.6	40.6	53.1	57.5			

In block 2 containing the McIntosh cultivar on slightly different rootstocks, a similar trend was observed (Table 4). Mac and Spur Mac on BA rootstock and Mac/M106 had significantly more canker than Mac/M26, Mac/O3/BA and Mac/BA/M26. These and other observations indicate that European Canker has become a major threat to continued apple production in some N.B. orchards.

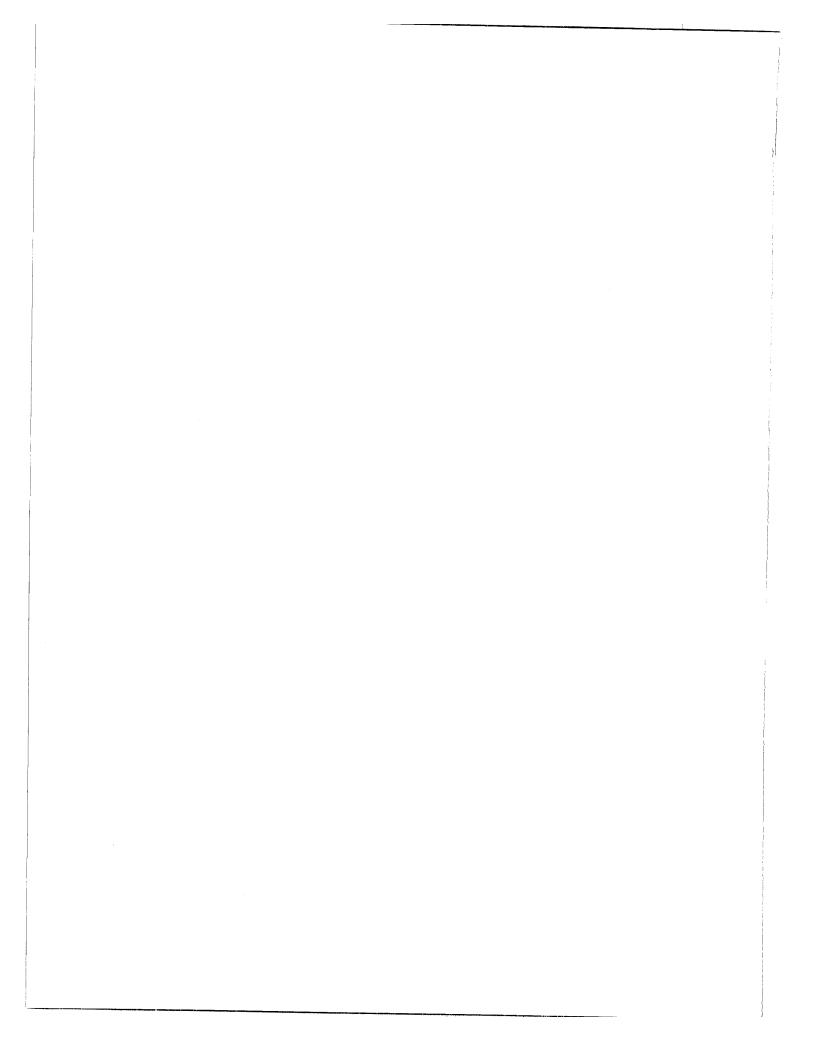
Despite the fact that European Canker has been known since the 1800's and research dates back to 1914, the main means of control is through good cultural practices, early detection and prompt removal of cankers. Good tree vigour also plays an important role in resisting and containment of the disease. Resistant varieties and rootstocks are not available to avoid the problem.

Table 4. Cultivar/rootstock effect on prevalence of European Canker, Fredericton Research Station, 1983.

	Bloc	k 1	Block	< 2
Cultivar/ rootstock	No. trees examined	% trees with cankers	No. trees examined	% trees with cankers
Mac/BA	53	22.6 a <sup>1</sup>	72	33.3 a <sup>1</sup>
Spur Mac/BA	50	14.8 ab	74	<b>28</b> .9 ab
Mac/M106	46	21.1 a	72	18.9 b
Mac/05	53	7.4 bc	_	
Mac/M26	51	3.7 bc	84	4.4 c
Mac/03/BA	_	_	80	1.1 c
Mac/BA/M26	39	1.9 с	82	0.2 c

<sup>&</sup>lt;sup>1</sup>Values followed by the same letter do not differ significantly at the 5% level (Duncan's Multiple Range Test).

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## Blackleg and other diseases of rapeseed in Saskatchewan, 1978 to 1981<sup>1</sup>

G.A. Petrie, K. Mortensen<sup>2</sup> and J. Dueck<sup>2</sup>

The prevalence and incidence of the virulent strain of blackleg (*Leptosphaeria maculans*) increased tenfold in standing crops of rapeseed (*Brassica napus* and *B. campestris*) in Saskatchewan between 1978 and 1981, but the overall yield loss from basal stem cankers was slight. Virulent blackleg was most prevalent in northeastern and central areas (crop districts (C.D.) 6b and 8), occurring in over 40% of the fields sampled over the 4-year period. It was least prevalent in the southeast (C.D. 1, 2 and 5), occurring in only 1.5% of the fields. Damping-off and seedling blight, caused primarily by *Rhizoctonia solani* were severe in 1979. Footrot (*R. solani* and *Fusarium roseum*) affected 81% of the fields that year with a mean incidence of 16%. White rust (staghead) declined in importance on *B. campestris* over the 4 years, but a different race of *Albugo candida* became more prevalent on *B. juncea*. Grey leaf spot or alternaria black spot (*A. brassicae* and *A. raphani*) was often widespread, but rarely severe. White leaf spot and grey stem (*Pseudocercosporella capsellae*), pod drop (cause unknown) and aster yellows were generally of minor importance. "Hybridzation nodules" on the roots of rape plants were prevalent in dry areas in 1979.

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La distribution et la fréquence d'une souche virulente du champignon de la jambe noire (*Leptosphaeria maculans*) ont augmenté par un facteur de dix dans les champs de colza (*Brassica napus* et *B. campestris*) de la Saskatchewan entre 1978 et 1981, mais la perte de rendement due au chancre de la tige est demeurée peu élevée. La jambe noire virulente était plus répandue dans le nord-est et le centre de la province (région agricole (R.A) 6b et 8), étant présent dans 40% des champs échantillonnés sur une période de 4 ans. Elle était moins répandue dans le sud-est de la province (R.A. 1, 2 et 5), n'étant présente que dans 1.5% des champs. La fonte des semis et de brûlure des plantules, causées principalement par *Rhizoctonia solani* ont été sévères en 1979. La pourriture des racines (*R. solani* et *Fusarium roseum*) affecta 81% des champs cette année-là, avec une fréquence moyenne de 16%. La rouille blanche diminua en importance sur *B. campestris* durant ces quatre ans, mais une race différente d'*Albugo candida* gagna de l'importance sur *B. juncea*. La tache grise ou la tache noire (*Alternaria brassicae* et *A. raphani*) furent souvent très répandues mais rarement sévères. La tache blanche foliaire et la tige grise (*Pseudocercosporella capsellae*), la tombée des siliques (cause inconnue) et la jaunisse étaient généralement de peu d'importance. La présence de "Nodules d'hybridization" sur les racines des plantes de colza était générale dans les régions sèches en 1979.

#### Introduction

Our primary objective in annual disease surveys of Saskatchewan rapeseed crops has been to monitor the spread of a virulent form of blackleg (*Leptosphaeria maculans* (Desm.) Ces. & de Not.), first found in the province on residue of the 1975 crop (5, 13). At that time it was detected at low levels in three widely separated fields, at Star City (crop district (C. D.) 8a), Humboldt (C. D. 8b) and Rosthern (C. D. 6b). In 1977, a severe localized outbreak of this strain occurred near Star City (13). Its incidence was 100% in a large portion of one field examined. In another field that had been seeded or had volunteered in stubble from the previous year, the yield loss from blackleg basal stem cankers was estimated conservatively at 20%. Abnormally high summer precipitation in northeastern Saskatchewan in 1977 contributed to the spread of the pathogen (1).

Other objectives of the annual surveys have been to monitor damping-off and footrot (*Rhizoctonia solani* Kühn and *Fusarium roseum* Lk. emend. Snyder and Hansen), several diseases of lesser importance, and damage from environmental or other factors.

#### Methods

Two series of surveys were conducted each year in fields having rapeseed that year, one series in standing crops and one in stubble. The first was started in late June or early July and was largely completed by early August. In this survey, plants were examined for early L. maculans infections on the leaves and stems, because such infections cause the maximum loss in yield from basal stem cankering (6). The stubble survey began late in August and was usually completed by the end of September. The extent of L. maculans infection on stubble is an indicator of inoculum levels (numbers of ascospores) in subsequent years. Routes were mapped out each year, and approximate locations of fields to be sampled marked equidistantly along them. In standing crops, five plants were pulled at each of five sites along an inverted "V" course. In stubble surveys, 10 stubble plants were pulled at each of five sites per field. Blackleg and footrot were rated independently using the 0-3 scale of severity established for footrot in earlier surveys (11). Other diseases were rated as in earlier surveys (8, 10).

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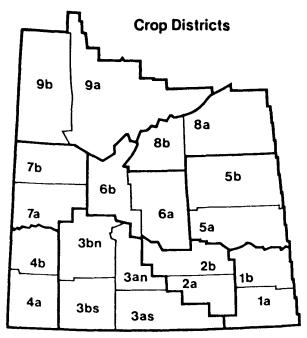


Figure 1. Saskatchewan crop districts and sub-districts.

The principal cultivars of Argentine rape (*Brassica napus* L.) and Polish or turnip rape (*B. campestris* L.) grown each year and the percentage of the total hectares of rapeseed seeded to each cultivar were available from the annual crop variety survey published by the Canadian Co-operative Wheat Producers Ltd. (2) (Table 1). As relatively few fields of *B. campestris* were encountered, the two species generally are not differentiated in the subsequent tables. The crop districts referred to throughout this report are illustrated in Fig. 1 and Fig. 2.

#### Results

#### 1978

The Star City-Nipawin area, location of the 1977 outbreak of blackleg, was visited in June and early July, 1978, prior to the principal surveys. On June 28, abundant leaf spots caused by the virulent strain were found in fields planted next to two of the 1977 fields. In one of these, stem girdling was noted on a small number of the young plants. The area was visited again on July 7. Additional infections were not apparent; spots were confined to the lower leaves and many of these were senescent. On July 14, scattered leaf spots were observed in fields throughout northeastern Saskatchewan. Although rainfall in the area was above normal in June, July precipitation was below normal (1). As part of the July-August 1978 survey, which covered much of the province, the northeast was visited again on July 26. The incidence of virulent blackleg was low, as it was in other areas where it had occurred previously. None was found in southeastern or east-central districts. In late August, 16 standing crops examined in C.D. 8a and 8b (the northeast) were affected, but the incidence of infection did not exceed 10%.

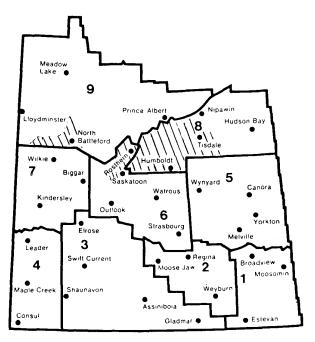


Figure 2. Major areas of infestation by the virulent strain of blackleg. Cross-hatched areas are those in which fields having more than 10% infection by the virulent strain were located. Data from all surveys (1978 to 1981) combined.

In August and September of 1978, 56% of the stubble fields examined were infected, with the Rosthern area having the highest levels. No blackleg occurred in the five mustard fields surveyed. Brown or oriental mustard (*Brassica juncea* (L.) Coss) and yellow mustard (*B. hirta* Moench = *Sinapis alba* L.) are resistant to the virulent strain (Petrie, unpublished). All the blackleg survey data are summarized in Tables 2, 3 and 4. As with other diseases, mean severity ratings for blackleg generally were very low throughout the 4-year period. Therefore, only data for prevalence and incidence appear in the tables. Figure 2 illustrates the major areas of infestation by the virulent strain between 1978 and 1981.

#### 1979

A preliminary survey was again conducted in the Star City area in 1979. In that year, May was abnormally cool and wet, and heavier than normal precipitation continued in June. Relatively little blackleg was seen on a July trip. Symptoms consisted of scattered leaf spots that likely had arisen from ascospore infection during the fourth week of June. The first general survey was started July 19 and continued through August. Although the virulent strain was prevalent in the Melfort-Star City and Rosthern areas, occurring in 60% of fields examined, its average incidence and severity were low in all areas. In July, the most heavily infected field (16%) occurred near Cutknife in crop district 9b. Levels of infection of this magnitude had not previously been found west of Saskatoon. The virulent strain also was found in three fields not examined in the general survey. These were near Laird (C.D 6b), Rosthern (C.D 6b) and Wakaw (C.D. 8b).

In the August-September 1979 survey of stubble fields, blackleg incidence was again generally low, although in areas in which it had become well established (Melfort, Humboldt, and Rosthern) it occurred in up to 88% of the fields. The virulent strain again was not detected in southeastern and east-central crop districts 1 and 5a, or in the Meadow Lake (northwest) or Asquith-Delisle areas, which were surveyed extensively.

#### 1980

Disease development was retarded by a prolonged dry spell. Little blackleg was found in standing crops, apart from those in the Melfort-Star City area. Although most of the fields there had 10-15% infected plants, one had over 80%. In the fall stubble survey, one specimen infected by the virulent strain was obtained from a field near Churchbridge, near the Manitoba border. This is the only instance in which this strain has been found in the east-central or southeastern part of the province prior to 1983. In a heavily infected field south of Laird, the virulent strain occurred on 50% of the plants. The most likely source of the early basal stem infections of these plants was ascospores from old rape residue in the field.

#### 1981

Precipitation in June and July was above normal and plants in a few fields had basal stem infections by late June. A high incidence of the virulent strain of blackleg was found in four fields in the Waldheim-Rosthern area and in one near Humboldt. In late June, 20% of the plants were infected in two fields west of Rosthern. In late July, the blackleg incidence in one of these fields had reached 100%, whereas in the other the incidence was much the same as it had been a month earlier, and plants cankered at the time of the initial survey were prostrate under the plant canopy. Discharge of ascospores in close proximity to the second field likely did not continue during July. In late July two other fields in the same general area were found with blackleg incidences close to 70%. Infection consisted of small lesions on the upper parts of the stems. A field near Fulda, in the Humboldt area, had 48% infection in early July. Ascosporebearing residue from the previous rape crop occurred in this field. As in the case of the Waldheim and Rosthern fields, leaf and upper stem infections predominated; only 8% basal stem infection was recorded.

In the August-October 1981 survey, the virulent strain occurred in all fields entered in C.D. 6b and 8. There was also an increase over the preceding two years in fields having over 10 and 20% incidence of infection (Table 3). In September basal cankering and premature ripening were noted in fields near Cudworth and Fulda in crop district 8b. All eight fields in the Asquith-Delisle area west of Saskatoon had the virulent strain, whereas two years earlier it could not be detected in this area. In a sample of foundation seed from near Melfort, infection by the virulent strain of blackleg exceeded 8%, an unusually high level of seed infection for this disease (14).

Damping-off, seedling blight, and footrot (*Rhizoctonia solani* Kühn, *Fusarium* spp., and *Pythium* spp.). The data for footrot from 1978 to 1981 are presented in Table 5. This disease complex is prevalent in at least some parts of Saskatchewan every year. Several heavily infected fields were encountered in a earlier survey (11). The disease was particularly noteworthy in 1979 (Table 5). Damping-off and seedling blight caused widespread damage in the spring, especially between North Battleford and the Alberta border. Cool wet conditions stimu-

Table 1. Rapeseed/canola varieties grown in Saskatchewan, 1978-1981.\*

Species	% of	% of total seeded hectares for years					
and variety	1978	1979	1980	1981			
B. campestris							
Candle	5.9	12.6	19.4	20.8			
Torch	20.6	18.3	10.7	7.2			
B. napus							
Altex	0.0	0.6	8.9	17.9			
Midas	24.3	11.9	6.7	4.0			
Regent	1.9	26.5	38.3	38.5			
Tower	44.8	27.2	14.5	8.2			
Other varieties	2.5	2.9	1.5	3.4			
Total hectares,							
Sask.x1,000	1,133.1	1,335.5	890.3	526.1			
Acres × 1,000	2,800.0	3,300.0	2,200.0	1,300.0			

<sup>\*</sup>Crop Varietal Survey, Canadian Co-operative Wheat Producers Ltd. Reprinted in (Rapeseed) Canola Digest. "Canola" refers to cultivars having low levels of erucic acid and glucosinolates; i.e., Candle, Altex, Tower and Regent.

lated the proliferation of seedling blight fungi while retarding seedling development. Later in the season, numerous enquiries were received dealing with further manifestations of the problem. For example, "wire-stem" symptoms were common in mid-June in a field near Lone Rock (C.D. 9b), and rotting and breakage of stems at soil level occurred in fields at North Battleford, Cutknife, and Maidstone (C.D. 9b) in July and August. Premature ripening often accompanied the basal stem rot. Rape plants with white mould on the roots were received from Meath Park (C.D. 9a), and yellow mustard rotting at the base and at the lower leaf nodes was collected north of Birch Hills (C.D. 8b). In these two instances, Fusarium roseum Lk. emend. Snyder and Hansen was isolated.

Sclerotinia stem rot (Sclerotinia sclerotiorum (Lib.) de Bary). The surveys described in this paper frequently could not properly assess sclerotinia stem rot. Surveys completed in July will either miss Sclerotinia entirely, or often greatly underestimate the amount of stem rot because the disease does not develop until crops are in the mid-flowering stage (7). Ascospore infection occurs during flowering and often this is not complete until the end of July or later. Stubble surveys also will underestimate the incidence of stem rot, as upper stem infections cannot be assessed.

Sclerotinia stem rot was found most consistently in the northeast, less regularly in northwestern and north-central areas, and infrequently in central areas and in the southeast. In areas where the disease was detected in standing crops in late July or in August, or in stubble crops, between 20 and 30% of the fields were usually affected. In 1978, 80% of stubble fields in the Melfort area had stem rot.

4-year

average

	Virulent strain		nt strain	All st	trains	% fields with over	
Year	No. fields	% fields infected	% plants infected	% fields infected	% plants infected	10% virulent blackleg	20% virulent blackleg
1981	41	53.7	4.3	68.3	5.0	14.6	2.4
1980	46	45.7	3.8	45.7	3.8	8.8	6.6
1979	85	20.0	0.6	31.8	1.2	2.4	2.4
1978	125	5.6	0.2	13.6	1.1	0.0	0.0

39.8

2.8

Table 2. Prevalence and incidence of blackleg in standing crops of *Brassica napus* and *B. campestris* in Saskatchewan, 1978-81.

White rust (staghead) (Albugo candida (Pers. ex Lév.) Ktze. Leaf rust was widespread in most fields of turnip rape (B. campestris) and brown mustard (B. juncea) (Table 6). Hypertrophies of the inflorescence (stagheads) generally occurred in 50% or more of the fields examined in a given year, but the mean incidence was relatively low. A distinct race of Albugo candida occurs on each of B. campestris and B. juncea (Petrie, unpublished). Brassica napus cultivars grown in Canada are immune to both races, and Canadian yellow mustard (Sinapis alba) cultivars are resistant to both. The B. juncea race has been found in plots at Saskatoon for at least ten years. Its severity has increased since 1977, both in commercial fields and in breeders' plots. For example, severe staghead infection occurred in a field near Blaine Lake in 1977; the estimated loss in yield (4) was 15%. In April 1978, three commercial seed samples of brown mustard were examined for oospores of A. candida using a washing and filtration technique (12). All three contained oospores and two had relatively high levels of 85 spores per gram of seed. In the 1978 survey two widely separated fields of B. juncea each had a leaf rust incidence of 100%. In one, 16% of the plants had terminal hypertrophies on one or more branches.

31.3

2.2

Grey leaf spot or alternaria black spot (Alternaria brassicae (Berk.) Sacc. and A. raphani Groves and Skolko). In 1978 both the prevalence and incidence of Alternaria infection were high in standing crops of all Brassica species, but disease severity was low. In July, infection was recorded in 70% of the fields; in late August it occurred in 94% of the fields, with a mean incidence of 69%. The situation in 1979 was similar. The disease was recorded in 76% of the fields, with a mean incidence of 49%. However, crops in the Humboldt area and in southeastern Saskatchewan had relatively little black spot. It was most severe in fields in the Meadow Lake area, which were examined relatively late in the growing season. It also was severe on pods of swathed plants in several fields in the Middle Lake and Melfort areas in mid-September, 1979. Black spot was less apparent in 1980 and 1981 surveys, being recorded in 10.9% and 23.7% of the fields, respectively. Incidence of infection was also low.

White leaf spot and grey stem (*Pseudocercosporella capsellae* E11. and Ev.) Deighton). In earlier survey reports this disease was called ringspot (*Mycosphaerella brassicicola* (Duby) Lind.) (10). However, in a recent paper a case was made for changing the names of both disease and pathogen (15). White leaf spot was of minor importance from 1978 to 1981, and little grey stem was found in standing crops. In stubble, however, the

grey stem phase of the disease was prevalent in 1979 and 1981. It occurred in 80% of the fields in 1979 (mean incidence: 36.4%) and 94.4% of the 1981 fields (mean incidence: 56.4%). The grey stem phase of the disease is thought to cause little loss in yield because it appears late in the growing season.

6.5

2.9

Pod drop (cause unknown). Pod drop (10) was noteworthy only in 1978. In July it occurred in 74% of the *B. napus* fields in northeastern areas (primarily crop districts 8a and 9a). In late August, 93.8% of 16 fields of *B. napus* in C.D. 8 were affected, with a mean incidence of 36.5%. Pod drop was also frequently observed in fields of cultivated mustards in 1978 but was rare in fields of *B. campestris*. These differences in occurrence on different species are consistent with observations made in earlier surveys (10).

Aster yellows mycoplasma. Aster yellows was of minor importance over the four-year period. In 1978 and 1979 trace amounts were observed in a few fields, mostly in central and east-central areas. In 1981, 71% of *B. napus* fields and 33% of *B. campestris* fields in the northwestern area (C.D. 9b) were lightly infected. The disease was also of little significance in recent Manitoba surveys (16) and earlier Saskatchewan surveys (8).

Root nodules up to several mm in diameter were common on rape plants in parts of the province in 1979. Most affected were central areas, particularly around Humboldt, where plants in 77% of the fields displayed these symptoms. In the southeast (C.D. 1a and 5a) 57% of the fields were affected. Not only were large numbers of fields involved; in some fields all the plants pulled had root nodules. The mean incidence of infection was 46% in the Humboldt area; in the southeast it was 45%. There were lesser amounts in the Meadow Lake, Rosthern and Vonda areas. In 1981, 30% of the fields in the Humboldt area were affected, with a mean incidence of 23%.

Hail damage was widespread in 1978 and in a few cases severe; 20% of the fields in the northwest were affected, 13% in the northeast, and 12% in central areas. Fungi often had entered the hail wounds and produced lesions around them. Stem lesioning of this kind caused by *Alternaria* spp. was pronounced in two fields of *B. campestris* in crop districts 1a and 7b. Hail damage was also thought to have enhanced *Alternaria* infection in two mustard fields (one yellow and one brown) in C.D. 1a. In 1980 the virulent strain of blackleg and hail injury

Table 3. Prevalence and incidence of blackleg in stubble fields of Brassica spp. in Saskatchewan, 1978-81

		Virulent strain		All s	All strains		% fields with over	
Year	No. fields	% fields infected	% plants infected	% fields infected	% plants infected	10% virulent blackleg	20% virulent blackleg	
1981	45	73.3	8.6	100.0	20.6	24.3	13.2	
1980	42	54.8	4.1	71.4	7.6	11.9	4.8	
1979	50	46.0	2.0	84.0	8.3	4.0	4.0	
1978	32	56.3	7.3	90.6	27.2	18.8	12.5	
4-year average	42	57.6	5.5	86.5	15.9	14.8	8.6	

were associated in a North Battleford field. In 1981 hail damage was reported in six fields. *Alternaria* was associated with the damage in fields near Aberdeen and Fulda (C.D. 8b). *Fusarium roseum* was isolated from black lesions around hail wounds on material from one field.

Herbicide injury was observed in each of the four years. Profuse development of the weakly virulent strain of blackleg on herbicide-damaged plants has been reported (9). In 1978 this strain was again associated with pronounced stem twisting and basal stem proliferations in a Brooksby field (C. D. 8b). In 1979 herbicide-induced basal stem galls and the virulent strain were associated in a field near Waldheim.

Table 4. Geographical distribution of the virulent strain of blackleg within Saskatchewan, 1978 to 1981.

	C	% of fields having the virulent strain (standing plus stubble crops)					
Area	Crop districts*	1978	1979	1980	1981	1978-81	
Northeast	8a	17.4	62.5	90.0	100.0	43.8	
Northwest	8b	4.4	11.1	60.0	30.0	19.1	
Central	8b, 6b	36.4	35.1	51.9	72.4	48.7	
Southeast	1,2,5	0.0	0.0	7.1	_	1.5	
Averages		15.9	29.6	50.0	65.1	35.4	

<sup>\*</sup>See Figure 1.

Heat canker on rape was reported from Dundurn (C.D. 6a) and Chelan (C.D. 8a) in mid-June. The condition occurred uniformly throughout the Dundurn field and resulted in many plants breaking off at the narrowly constricted point at soil level. Four fields were severely affected in the Chelan area. Vanterpool has described this symptom in flax, and a similar one caused by frost (17, 18).

#### **Discussion**

The virulent strain of blackleg is most damaging when it attacks the crown of the plant, severing the stem at its base. The younger the plant, the more susceptible it is to basal stem infection. Only mild basal cankering results when plants are infected after the six-leaf stage of growth, that is after the end

of June (6). Observations made in 1978 and 1981 left no doubt that infection can occur in June. The prevalence of early season infection largely depends upon when ascospore production by the pathogen commences. This differs on one- and two-year-old rape stubble residue (6). The subject will be discussed further in a subsequent paper. The survey results indicate that, fortunately, infection of young plants was not widespread during this four-year period. However, it is apparent that the prevalence, incidence and severity of blackleg increased between 1978 and 1981. In addition, the disease has spread into parts of Saskatchewan where it had not been found prior to 1978. In 1981 it occurred throughout the Asquith-Delisle area, although it had not been found there two years previously. In 1980 it was detected for the first time in east-central Saskatchewan. It still has not been found in parts of northwestern Saskatchewan including the Meadow Lake area and in the part of north-central Saskatchewan to the northeast of Prince Albert. Rimmer and Platford (16) did not find the virulent strain of blackleg in recent surveys in Manitoba. No reports of it in Alberta crops appeared prior to 1983.

Table 5. Footrot prevalence and incidence in standing and stubble crops of *Brassica* spp. in Saskatchewan, 1978-81.

	No. of	% fields	Mean %	% fields with over	
Year	fields	infected	incidence	10% footrot	20% footrot
1981	86	74.3	5.6	17.1	8.6
1980	88	71.7	4.5	8.8	6.6
1979	135	80.7	16.0	52.6	26.7
1978	157	41.4	6.7	14.7	8.3
Mean	s 117	67.0	8.2	23.3	12.6

Damping-off and seedling blight, caused primarily by *Rhizoctonia solani*, are often important, as in 1979, in preventing adequate stand establishment in rapeseed fields. Footrot, in which most of the same pathogens are involved, is less important, occurring as it does late in the growing season. An exception to this generalization is premature ripening in mid-season resulting from basal stem infections by *Rhizoctonia* and *Fusarium*. Prematurely ripened plants setting little seed may often be seen scattered throughout rapeseed fields.

Table 6. Prevalence and incidence of Albugo candida (white rust and staghead) in standing crops of turnip rape (Brassica campestris), brown mustard (B. juncea) and yellow mustard (B. hirta).

Species and year	No. of fields	Crop Districts* Represented				pertrophies leads)** % plants
B. campestris				N		
1978	14	1, 5, 7, 8, 9	57.1	31.4	50.0	3.6
1979	9	1, 2, 8, 9	100.0	82.2	55.6	6.1
1981	3	9	100.0	90.7	100.0	2.0
3-year average			85.7	68.1	68.5	3.9
B. juncea						
1978	2	1, 8	100.0	100.0	50.0	8.0
B. hirta						
1978	3	1,8	0.0	0.0	0.0	0.0

<sup>\*</sup>See Figure 1.

The percentage loss in yield of *B. campestris* due to staghead (*Albugo candida*) during the 1978-81 period was much less than the six and nine percent calculated for the years 1971 and 1972, respectively (8). In addition, there has been a reduction in the percentage of the total rapeseed acreage sown to *B. campestris* from 63% in 1970 to 28% in 1981 (2). The 147,000 hectares planted to this species in 1981 represent only 22.4% of the total sown a decade earlier. The almost complete replacement of the cultivar Torch, by Candle between 1978 and 1981 has had little effect on losses caused by white rust, as both are susceptible, but the new cultivar Tobin, licensed in 1981, is resistant. The race attacking *B. juncea* remains a potentially serious problem.

Alternaria black spot was frequently widespread but only occasionally damaging. In recent Manitoba surveys (I6) and an earlier series in Saskatchewan (10) the pattern of high prevalence and incidence and low severity was also found.

The nodules observed on the roots of rape plants appear to be the result of a physiological disorder and have been referred to as "hybridization nodules" in crosses of swedes and rape (3). Our observations led us to relate them to drought stress. In 1979 in the southeastern part of the province and in the Humboldt area, nodules were abundant on plants in many fields that suffered from drought and were uneven in height. In some fields nodulation was clearly more common on plants on more exposed, drier sites.

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<sup>\*\*</sup>Average loss in yield due to staghead for the three years estimated at < 2.0%.

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# Yield losses in Saskatchewan rapeseed/canola crops from basal stem cankers of blackleg (*Leptosphaeria maculans*) in 1982, with notes on other diseases<sup>1</sup>

G. A. Petrie

The virulent strain of blackleg (*Leptosphaeria maculans*) occurred in 67.9% of 53 fields of *Brassica napus* and *B. campestris* surveyed in Saskatchewan in July, 1982. An average of 11.0% of the plants per field had leaf or stem infections, whereas only 0.8% had basal stem cankers. Thirteen of the 53 fields plus four others were sampled again in August. In these I7 fields from 0.0 to 78.0% of the plants had severe basal stem cankers, and yield losses from 0.0 to 56.0%. Seven of the fields which were near Humboldt in central Saskatchewan had an average loss of 29.9%. The estimated average yield loss for the whole province was six percent. Footrot (*Rhizoctonia solani* and *Fusarium roseum*) occurred in 79.3% of the 53 fields sampled in July, and had a mean incidence of 8.5%. White rust (*Albugo candida*) occurred in 71.4% of seven fields of *B. campestris* but its mean incidence, 24.6%, was relatively low. Other diseases were of little consequence in the July survey. Footrot, alternaria black spot (*Alternaria brassicae* and *A. raphani*), sclerotinia stem rot (*Sclerotinia sclerotiorum*), and pod drop (cause unknown) were prevelant in the 17 fields sampled in August but caused minor damage. Aster yellows and grey stem (*Pseudocercosporella capsellae*) were observed only rarely.

Can. Plant Dis. Surv. 65:2, 43-46, 1985.

Une forme virulente du champignon de la jambe noire (Leptosphaeria maculans) était présente dans 67.9% de 53 champs de Brassica napus et B. campestris inventoriés en Saskatchewan au mois de juillet 1982. En moyenne, 11.0% des plants avaient des infections foliaires ou des tiges, tandis que seulement 0.8% des plants étaient infectés à la base de la tige. Treize des 53 champs plus quatre autres furent échantilonnés de nouveau en août. Dans ces 17 champs, de 0.0 à 78.0% des plants étaient sérieusement affectés par la jambe noire avec des pertes de rendement allant de 0.0 à 56.0%. Sept de ces champs situés près de Humbolt au centre de la Saskatchewan avaient une perte moyenne de 29.9%. La perte de rendement movenne pour toute la province a été estimée à 6%. On a retrouvé de la pourriture des racines (Rhizoctonia solani et Fusarium roseum) dans 79.3% des 53 champs échantillonnés en juillet et en moyenne chez 8.5% des plants. La rouille blanche (Albugo candida) était présente dans 71.4% des 7 champs de B. campestris mais son incidence moyenne était relativement basse, 24.6%. Les autres maladies inventoriées en juillet furent de peu de conséquence. La pourriture des racines, les taches grises et noires (Alternaria brassicae et A. raphan), la pourriture sclérotique (Sclérotinia sclerotiorum), et la tombée des siliques (cause inconnue) étaient présents dans les 17 champs échantillonnés au mois d'août, toutefois ils n'ont causé que des dommages mineurs. La jaunisse de l'aster et la tige grise (Pseudocercosporella capsellae) furent rarement observées.

#### Introduction

Surveys of rapeseed/canola<sup>2</sup> fields (*Brassica napus* L. and *B. campestris* L.) in Saskatchewan from 1975 to 1981 revealed an increasing trend in the prevalence and incidence of the virulent strain of *Leptosphaeria maculans* (Desm.) Ces. and de Not., the cause of blackleg or basal stem canker (10, 12). Although substantial yield reductions occurred in a few fields, losses of the magnitude of those reported in parts of Australia (3, 8) and England (5, 6) were not observed. A more serious outbreak of blackleg occurred in central Saskatchewan in 1982, however.

#### Methods

Fifty-three rapeseed/canola fields made up the principal

survey which was conducted between July 7 and 21. Survey and disease rating procedures were as previously described (12), with the exception of stem canker ratings, where McGee's method was used (7). In some fields sampled in July, basal stem canker appeared to have the potential to become severe. Therefore 13 fields that had more than 10% incidence of the disease in July were sampled again in August, along with four additional fields not previously sampled.

Severity of basal stem canker was assessed on 60 plants per field, with ten plants pulled at each of six sites. Plants with over 50% of the stem circumference cankered were considered severely infected when estimating yield loss (8). The 13 fields sampled in July and August were again sampled in September following harvest, and blackleg severity reassessed. Presence and severity of other diseases were routinely recorded only in the July and August surveys.

#### **Results and Discussion**

In the July survey (Table 1), as earlier years (12), fields in western (Cutknife) and north-central (Meath Park) areas had less blackleg than those in northeastern (Melfort) and central (Rosthern and Humboldt) areas. The overall prevalence and inci-

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<sup>2 &</sup>quot;Canola" refers to cultivars of rapeseed low in erucic acid and glucosinolates.

Table 1. Prevalence and incidence of blackleg and footrot, July 1982

		Viruler		Footrot			
	Mean	M	lean % plants	with lesions	Mean	Mean	
Areas surveyed and no. fields	% fields infected	any part	leaves	upper stems	stem bases	% fields infected	% plants infected
Rosthern (11)	72.7	13,8	12.4	1.5	0.7	81.8	5.7
Humboldt (10)	100.0	20.1	18.8	8.0	1.2	90.0	7.0
Cutknife (10)	50.0	1.0	0.9	0.1	0.2	70.0	7.7
Meath Park (11)	36.4	6,6	6.6	0.4	0.7	63.6	3,7
Melfort (11)	81.8	13.6	12.1	0.4	1.1	90.1	18.2
All areas (53)	67.9	11.0	10.2	0.6	0.8	79.3	8.5

Table 2. Blackleg development from July to September, 1982, in selected fields heavily infected by the virulent strain.

				Mean % plants with					
No. Area fields	Dates sampled	lesions on any part	leaf lesions	upper stem lesions	basal stem lesions	girdling basal stem lesions	virulent strain found on isolation		
Rosthern 6	7 July	22.7	21.3	2.0	0.7	0.0	100.0		
		<b>1</b> 1 Aug.	47.7		16.1	40.9	2.2	100.0	
		<b>20</b> Sept.*	83.2		83.2	75.0	37.9	91.0	
Humboldt	7	12 July	35.4	33.7	1.7	3.3	0.0	100.0	
		17 Aug.	80.7		41.5	76.9	15.7	100.0	
		22 Sept.*	94.8		94.8	78.9	55.9	84.2	

<sup>\*</sup>Stubble plants only.

dence of the virulent strain were considerably greater than in 1981 (12). Leaf infections were much more common than stem infections in the July survey (Table 2), as ascospores infected the leaves in June and July and the disease only subsequently spread to the lower stem to cause cankers near soil level.

By mid-August, basal stem cankers, which are the major cause of yield loss from blackleg, were more common than upper stem lesions. However in a field at Laird, north of Saskatoon, 96% of the plants had small upper stem infections at this time, and none had detectable basal cankers. Estimated yield losses in the sampled fields varied from 0 to 56% (Table 3). The mean loss for the Humboldt fields was 29.9%. On the basis of random surveys of stubble crops, it appeared that a yield loss figure for Saskatchewan as a whole would be close to six percent.

In the September survey a high incidence of basal stem lesions was observed in the 13 fields (Table 2). These lesions, which generally showed little vertical development in August, often had developed considerably upward from soil level on the stubble plants. The upper portions of the decapitated stalks had also been colonized by *L. maculans*. This represented the post-harvest flush of saprophytic growth also noted by

others (5). Consequently the August estimates of yield loss were considered to be more reliable than the September estimates. A very high proportion of the isolates from stubble represented the virulent strain (Table 2). This is not always so when samples taken at this time of year are plated out.

Infection of the young plant is a prerequisite for severe stem canker development (9). Cool, wet conditions in the spring of 1982 delayed seeding and retarded crop growth over much of the province. A severe snow and rainstorm occurred at the end of May, and frost damage necessitated reseeding of some fields. Canola development by mid-June lagged well behind that in 1981 (1). On the other hand the onset of ascospore production by the virulent strain of blackleg on stubble of the previous year occurred sooner in 1982 than in 1981, and many more spores were produced early in the growing season (Petrie, unpublished data). Precipitation remained above normal for most of the summer, with mean temperatures near or slightly below normal (2). This combination of factors, which favored infection of young plants, probably explains the heightened severity of blackleg in 1982.

The introduction of stem canker (blackleg) resistant cultivars of French origin has eliminated at least temporarily a threatening problem in England (5, 6). Resistant cultivars are currently

Table 3. Estimated loss in yield due to the virulent strain of blackleg in selected fields in central Saskatchewan, August, 1982.

	Me	Mean % of plants with							
Area and field no.	basal stem infections	severe basal* cankers	% loss in yield**						
Rosthern									
Ros-1	28.2	8.1	3.0						
Ros-2	6.7	0.0	0.0						
Ros-3	35.9	3.4	2.0						
Ros-4	56.7	8.3	3.0						
Ros-6	51.7	20.0	13.0						
Ros-9	65.9	22.6	16.0						
Means	40.9	10.4	6.2						
Humboldt									
Hum-1	33.3	10.0	8.0						
Hum-2	76.7	53.2	39.0						
Hum-3	60.0	5.0	3.0						
Hum-6	80.0	40.0	29.0						
Hum-7	100.0	50.9	47.0						
Hum-9	90.0	36.7	27.0						
Hum-10	98.3	78.0	56.0						
Means	76.9	39.1	29.9						
Prud'homme	,								
Prud-9	68.0	24.0	17.0						
Prud-8	12.0	0.0	0.0						
Prud-7	20.0	4.0	2.0						
Means	33.3	9.3	6.3						

being developed at Saskatoon and elsewhere in western Canada, but will not be ready for release to growers for at least a few years. In Canada, at present, producers must rely upon crop rotation, burial of infected stubble, and seed treatment for blackleg control (4, 11).

Other diseases. Footrot (*Rhizoctonia solani* Kühn and *Fusarium roseum* Lk. Snyder and Hansen) was more prevalent in July, 1982 (Table 1) than in July, 1981 (12), although the mean incidence was similar. Compared to the 1978-81 period (12) there was a relatively high number of fields (32.1%) having over 10% footrot incidence in 1982. The disease was common in the 17 selected fields in August but was not severe (Table 4).

Alternaria black spot (Alternaria brassicae (Berk.) Sacc. and A. raphani Groves and Skolko) was recorded in five of the 53 fields in July; its highest incidence was 32%. Despite being very prevalent in August it did not cause severe infections (Table 4). A similar situation prevailed in August with regard to pod drop (Table 4), the cause of which is unknown. In July, white rust (Albugo candida (Pers. ex Lév.) Ktze.) occurred in 71.4% of seven fields of Brassica campestris with a mean incidence of 24.6% and highest incidence of 68% in a field in the Cutknife area. Sclerotinia stem rot (Sclerotinia sclerotiorum (Lib.) de Bary) occurred in 47.1% of the 17 selected fields, with a mean incidence of 3.3%. Aster yellows and grey stem (Pseudocercosporella capsellae (E11. & Ev.) Deighton) were rarely observed (Table 4).

Table 4. Prevalence and incidence of secondary diseases in 17 fields selected for high incidence of blackleg, August 1982

	Foo	trot*	Alter	naria**	Pod	Drop**	Scler	otinia*	Aster	Grey
Area and no. fields	% fields	% plants	% fields	% plants	% fields	% plants	% fields	% plants	yellows % fields	stem % fields
Rosthern (6)	100.0	9.4	100.0	62.3	100.0	48.3	50.0	4.9	0.0	0.0
Humboldt (7)	100.0	3.3	100.0	97.4	100.0	80.5	57.1	1.0	0.0	14.3
Other areas (4)	50.0	2.0	50.0	8.0	0.0	0.0	25.0	5.0	25.0	0.0
All areas (17)	88.2	5.2	88.2	64.0	76.5	50.2	47.1	3.3	5.9	5.9

<sup>\*</sup>Mean severity ratings for 17 fields (0-3 scale) = 0.1

<sup>\*&</sup>gt;50% of the stem girdled.

<sup>\*\*</sup>Estimated from Fig.1(a), McGee and Emmett (1977).

<sup>\*\*</sup>Mean severity ratings for 17 fields (0-3 scale) = 0.5

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### Saskatchewan rapeseed/canola disease survey, 1983<sup>1</sup>

G.A. Petrie

In July, 1983, the virulent strain of blackleg (*Leptosphaeria maculans*) occurred in 61.3% of Saskatchewan rapeseed fields surveyed in crop district (C.D.) 8; 75.0% of those in C. D. 9, and 19.4% of those in C. D. 1 and 5. Its average incidence in the three areas was 12.1%, 15.0%, and 1.2%, respectively. Overall loss in yield due to blackleg was slight. Five of the 86 fields surveyed had small numbers of plants with severe basal stem cankers. Footrot (*Rhizoctonia solani* and *Fusarium roseum*) was less prevalent than in the period 1978 to 1981, whereas alternaria black spot (primarily *A. brassicae*) and late-season aster yellows were more prevalent. White rust (*Albugo candida*) was unimportant due to widespread use of resistant cultivars. A mid-May survey of 22 stubble fields from the 1982 crop in eastern Alberta failed to detect the virulent strain of blackleg.

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Au mois de juillet 1983, la lignée virulente du champignon de la jambe noire (*Leptosphaeria maculans*) était présente dans 61.3% des champs de colza inventoriés en Saskatchewan dans la région agricole (R.A.) 8; dans 75.0% de ceux inventoriés dans la R.A. 9 et dans 19.4% de ceux inventoriés dans les R.A. 1 et 5. Son incidence moyenne dans ces trois régions était respectivement de 12.1%, 15.0% et 1.2% et la perte de rendement totale causée par la jambe noire fut légère. Cinq des 86 champs inventoriés avaient un petit nombre de plants avec chancres à la base de la tige. La pourriture des racines (*Rhizoctonia solani* et *Fusarium roseum*) était moins répandue qu'en 1978 à 1981, tandis que la tache grise (principalement *Alternaria brassicae*) et la jaunisse des asters de fin de saison, étaient plus répandues. La rouille blanche (*Albugo candida*) était peu importante due à l'usage de cultivars résistants. Un inventaire, effectué à la mi-mai, de 22 champs de l'est de l'Alberta contenant des débris agricoles de la récolte 1982 n'a pas permis de détecter la lignée virulente de la jambe noire.

#### Introduction

This survey of rapeseed/canola<sup>2</sup> fields (*Brassica napus* L. and *B. campestris* L.) conducted during the 1983 growing season is one in a continuing series (6, 7, 8) to monitor the prevalence and severity of the virulent strain of blackleg (*Leptosphaeria maculans* (Desm.) Ces. and de Not.) and other rapeseed diseases. Yield losses from stem cankers of blackleg in central Saskatchewan in 1982 were much higher than in previous years. As the southeastern part of Saskatchewan was not visited in 1981 or 1982, considerable attention was devoted to this area, which was largely free of the disease prior to 1981. The virulent strain had not been reported in commercial fields in Alberta before 1983, and recent surveys in Manitoba also failed to detect it (9). As it is prevalent in Saskatchewan near the Alberta border, the 1983 survey was extended into eastern Alberta.

#### Methods

In the survey for the virulent strain of blackleg in Alberta, stubble plants from 22 fields from the 1982 rapeseed crop were collected in mid-May, 1983. The survey went west to Killam and Viking, north to Vermilion and Lloydminster and south to Wainwright and Hughendon. A random sample of 50 stubble plants was taken in each field, consisting of ten plants at each

of five sites. A sample of selected blackleg-infected plants was also collected. Isolations were made from all material suspected of having blackleg, using the technique described previously (6) and blackleg strains identified.

The principal disease survey in Saskatchewan was carried out between July 12 and 27. Thirty-one fields in the northeast (crop district 8) were sampled between July 12 and 14, 24 fields in the northwest (western half of C. D. 9) were sampled from July 19 to 20, and 31 in the southeast (C. D. 1 and 5) were sampled from July 25 to 29. Survey procedures and disease rating schemes have been previously described (3, 4, 5, 6, 8). One modification involved the sampling technique within fields. In the 1983 survey, one plant was collected every five paces along an inverted "V" course until 25 plants were obtained. Severity of blackleg basal stem cankers was assessed as in 1982 (7), with McGee's method (1, 2).

The fall survey of stubble fields was conducted in central Saskatchewan. Of particular interest were the incidence and severity of basal stem cankers. Two stubble plants were pulled every ten spaces for a total of 50 plants per field.

#### Results

Alberta blackleg survey. All Alberta isolates of *L. maculans* from the previous year's stubble were identical to Saskatchewan's most common weakly virulent strain. Its presence was confirmed in 91% of the fields. The mean blackleg incidence per field was 22%. In six fields over 40% of the plants had blackleg symptoms; in one, 65% infection was recorded. The infections were superficial, probably developing late in August and in September, 1982.

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<sup>2 &</sup>quot;Canola" refers to cultivars of rapeseed low in erucic acid and alucosinolates.

Table 1.	Prevalence and incidence of	the virulent strain of	f blackleg in standing crops of	f <i>Brassica</i> spp. in 1983
	·····			· · · · · · · · · · · · · · · · · · ·

	% of fie	lds having	g plants with	infections	% of fields	f fields with over % of plants per field having infe			infections	
Crop District	on leaves	on stems	at stem base	on any part	10% blackleg	20% blackleg	on leaves	on stems	at stem base	on any part
8	58.1	3.2	6.5	61.3	32.3	19.4	11.2	0.2	0.7	12.1
9*	66.7	16.7	29.2	75.0	37.5	20.8	12.9	2.4	2.9	15.0
1, 5	9.7	6.5	6.5	19.4	3.2	0.0	0.6	0.4	0.3	1.2
Means	44.8	8.8	14.1	51.9	23.3	12.8	8.2	1.0	1.3	9.4

<sup>\*</sup>Western half

Saskatchewan July blackleg surveys. Much less of the virulent strain was found in the southeast (C. D. 1, 5) than in the other two areas (Tables 2 and 3). Five of the six infected fields found in the southeast occurred in C. D. 5b, the sixth was found near Langenburg in C. D. 5a. No infected fields were found in C. D. 1. Crop districts are shown in an earlier paper (8).

The most severely affected fields encountered in C. D. 9 were near Marshall, Baldwinton, and Cutknife, and in C. D. 8, near Tisdale, Ridgedale, and Humboldt. All six had an incidence of virulent blackleg of over 40%. Symptoms consisted mostly of leaf spots. Only five of the 86 fields had plants with severe basal stem cankers (over 50% of the stem circumference cankered), and usually only one plant per 25-plant sample was this severely infected. In the most heavily diseased field, which was in the Cutknife-Baldwinton area of C. D. 9b, 23% of the plants had severe basal cankers. This would represent a 17% loss in yield (2).

In the fall stubble survey 36 fields were sampled, 31 of canola, three of yellow mustard (*B. hirta* Moench = *Sinapis alba* L.) and two of brown mustard (*B. juncea* (L.) Coss). The average incidence of blackleg (all strains) was 18% for the 31 canola fields and 10% for the five mustard fields. The virulent strain occurred in all the canola fields and in 60% of the mustard fields. A mean of 65% of the isolates from canola were virulent and 35% weakly virulent. Only 2.6% of the isolates from the five mustard fields were virulent and 97.5% weakly virulent. Both mustard species are highly resistant to the virulent strain (Petrie, unpublished). Stubble plants of canola with severe basal stem cankers were uncommon in this survey.

Other diseases in Saskatchewan surveys. Footrot (*Rhizoctonia solani* Kühn and *Fusarium roseum* Lk. emend. Snyder and Hansen) occurred in 50% of the 86 standing crops, with a mean incidence of 6.7%. These values are somewhat lower than the average for the four years 1978 to 1981 (Table 2). In 1983, the prevalence and incidence of footrot were highest in C. D. 8 and lowest in C. D. 1 and 5. Crop district 8 also had a large proportion of the fields with over 20% incidence of infection; C. D. 1 and 5 had none (Table 2).

The July survey was too early in the growing season to adequately assess sclerotinia stem rot (*Sclerotinia sclerotiorum* (Lib.) de Bary). However, 25% of the fields entered in C. D. 9 had stem rot, with a mean incidence of 1.8%. The disease was much less prevalent in the other two areas in July, occurring in approximately three percent of the fields.

Alternaria black spot (*A. brassicae* (Berk.) Sacc. and *A. raphani* Groves and Skolko) occurred in approximately 90% of the 86 standing crops, including all fields in C. D. 9 (Table 3). Leaf spot incidence was high in all areas but severity was low. Pod and stem spotting were scarcely observed in the two earlier surveys but were prevalent in fields in C. D. 1 and 5 which were surveyed later (Table 3). Other observations indicated that *Alternaria* pod and stem infections were common by the end of July throughout the province. However, stem and pod spot severity averaged less than 0.5 on a 0-3 rating scale (4). The most severely damaged fields were two of the high erucic acid *B. campestris* cultivar, R-500, located near Blucher and Lanigan in central Saskatchewan, which were examined in late August. Numerous pod spots and elongated, coalesced stem lesions were noted.

Pod drop (4) occurred in several fields but was generally of minor importance. Virtually no aster yellows was recorded on plants collected in July. By late August, however, infection was unusually plentiful in several fields, including some in the Shellbrook and Shell Lake areas of C. D. 9a, west of Prince Albert (L. Burgess, personal communication).

White rust (*Albugo candida* (Pers. ex Lév.) Ktze) was uncommon, leaf symptoms occurring in only 14% of the 86 fields sampled. In only two fields did leaf rust incidence exceed 50%, and stagheads were observed in only two fields. The highest incidence of staghead was 12% in a field south of Grenfell in C. D. 1b.

Hail damage on stems and pods was noted in seven fields in the southeast. In a field north of Ituna, 74.1% of the plants were slightly damaged. Hail injury was moderately severe on 53.6% of the plants in a field near Leslie. Both fields are in C. D. 5.

#### Discussion

There has been a striking increase in the prevalence and incidence of the virulent strain of *L. maculans* during the last six years (7, 8). Fortunately, with the exception of 1982 (7), severe basal stem cankering has occurred only sporadically during this period. Stem canker severity is directly related to earliness of infection, which in turn depends upon ascospores being released from infected canola residue prior to the end of June.

A large area of southeastern Saskatchewan has remained essentially free of the virulent strain of blackleg. In 1980 it was collected in one field in the Churchbridge-Langenburg area in

Table 2. Prevalence and incidence of footrot (Rhizoctonia solani and Fusarium roseum) in standing crops of Brassica spp. in 1983.

Crop	% fields	% plants	% fields	with over
district	infected	infected	10% footrot	20% footrot
8	54.8	10.7	32.3	22.6
9*	54.2	6.5	20.8	8.3
1, 5	41.9	2.8	6.5	0.0
Means	50.0	6.7	19.8	10.5
4-year means (1978-81)	67.0	8.2	23.3	12.6

<sup>\*</sup>Western half.

C. D. 5a, the only occurrence reported prior to 1983 from the southeast (8). The virulent strain likely is scarce here because the southeast is not a major canola-producing area. More intensive cropping of canola in central and northern agricultural areas of the province no doubt contributed greatly to its rapid spread in those areas. Discovery of several infected fields in C.

D. 5b indicates that the virulent strain of blackleg may be moving gradually into the southeast. Continued fungicidal treatment of seed sown in southeastern crop districts should help to delay spread of infection throughout these districts.

The attempt to find the virulent strain of blackleg in the part of Alberta closest to the heavily infected Cutknife-Maidstone area of Saskatchewan was unsuccessful. However, Saskatchewan's most common weakly virulent strain was prevalent in eastern Alberta. It had been found in several fields there as early as May, 1973 (Petrie, unpublished data). It may be safe to conclude that the virulent strain was rare in eastern Alberta, if it occurred there at all, in 1982.

Footrot was less prevalent in 1983 in Saskatchewan than in previous years, whereas alternaria black spot and late-season infections of aster yellows were more noticeable than they had been for several years. *Alternaria* pod and stem lesions developed rapidly from the latter part of July to harvest, so that they were detected only in the last of the three survey trips (Table 4).

White rust is declining in importance except on *B. juncea*, because of the widespread use of race 7-resistant cultivars, including the new *B. campestris* cultivar Tobin.

Table 3. Prevalence, incidence, and severity of alternaria black spot in standing crops of Brassica spp. in 1983

		Leaf spots		Pod :	spots*	Stem	spots
Crop Districts	% fields infected	% plants infected	Severity (0 - 3)	% fields infected	% plants infected	% fields infe <b>c</b> ted	% plants infected
8	80.7	51.8	0.4	0.0	0.0	0.0	0.0
9**	100.0	90.3	8.0	8.3	2.1	0.0	0.0
1, 5	96.6	73.5	0.7	71.0	35.4	41.9	15.0
All areas	89.5	70.5	0.6				

<sup>\*</sup>Severity of pod and stem spots < 0.5 on 0-3 scale.

#### **Acknowledgements**

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<sup>\*\*</sup>Western half.



## Differences in mosaic disease virus profiles between three potato cultivars

John G. McDonald<sup>1</sup>

As a result of an enzyme-linked immunosorbent assay (ELISA) survey in Prince Edward Island, major differences were found in the virus profiles of mosaic-diseased Shepody, Red Pontiac, and Green Mountain potato cultivars. Potato viruses Y, (PVY) and X, (PVX) were associated with mosaic disease in all three cultivars, but to differing degrees; PVY was present in 89% of the samples of Shepody showing severe mosaic but was present in only 59% and 32% respectively, of those samples of Red Pontiac and Green Mountain. Potato virus A (PVA), however, was only found in the Green Mountain cultivar.

Can. Plant Dis. Surv. 65:2, 51-52, 1985.

A la suite d'un inventaire de plusieurs cultivars de pommes de terre dans L'Île du Prince Edward en utilisant le test d'immunoadsorbant lié à un enzyme (ELISA), on a découvert des différences majeures dans les profils des virus associés à la mosaïque chez les cultivars Shepody, Red Pontiac et Green Mountain. Les virus Y (PVY) et X (PVX) de la pomme de terre sont associés avec la mosaïque chez les trois cultivars, mais à des degrés différents. On retrouve le PVY dans 89% des échantillons de Shepody montrant des symptômes sévères de mosaïque mais dans seulement 59% et 32% des échantillons de Red Pontiac et de Green Mountain. Toutefois, le virus A de la pomme de terre (PVA) n'est présent que chez le cultivar Green Mountain.

In a recent study (2) of the association of mosaic-inducing viruses with the potato (*Solanum tuberosum*) cultivar Russet Burbank in Prince Edward Island, it was found that potato virus X (PVX) was the virus most frequently associated with plants showing mild mosaic, while mixed infections of potato virus A (PVA) and PVX accounted for the majority of plants showing severe mosaic. As previously discussed (2), knowledge of which viruses are the cause of mosaic symptoms in a particular cultivar is essential for selecting appropriate control measures and in applying serological assays for seed potato certification.

Of the other commercial potato cultivars produced in Prince Edward Island, the three with the highest susceptibility to mosaic are Shepody, Red Pontiac and Green Mountain. As the prevalence of the different mosaic-inducing viruses in a particular cultivar and region will vary according to complex ecological factors including cultivar susceptibility (1), this study was undertaken to determine the principal causes of mosaic in each of these cultivars.

Leaf samples were collected from six fields of Shepody, five of Red Pontiac and five of Green Mountain. Both mosaic-affected and symptomless samples were taken from each field and the severity of the symptoms were categorized as before (2). The enzyme-linked immunosorbent assay (ELISA) methods used to identify the presence of potato virus Y (PVY), PVA and PVX in leaf samples were also the same as previously reported (2).

The results, shown in Table 1, indicate major differences in virus profiles between the three cultivars. While PVX, PVY and PVA were all associated with mosaic disease in Green Moun-

tain, only PVX and PVY were detected in Red Pontiac and Shepody. This would suggest a high degree of resistance to PVA in these latter two cultivars as it is known that they are not immune (unpublished data of the author).

PVY was found to be the major cause of mosaic in Shepody as it was associated with 89% of all the samples showing severe mosaic and with 71% of all the mild mosaic samples. For Red Pontiac, PVX appeared to be a more significant cause of mosaic as PVY was only detected in 59% of the samples showing severe mosaic. In the case of Green Mountain where both PVA and PVY were detected, PVA was detected in 42% of the samples showing severe symptoms while PVY was detected in 29%; PVX in single infection accounted for 34% of these samples. In plants showing mild symptoms, PVX in single infection accounted for 46% of the samples, while PVA and PVY were present in 30% and 26% of these samples, respectively.

The number of symptomless samples of Shepody, Red Pontiac and Green Mountain were 71, 12 and 56, respectively and levels of PVX were 63%, 50% and 88%. This level of occurrence of presumably symptomless strains of PVX is similar to the 69% level reported for Russet Burbank(2). PVY was also found in 2% of the Shepody and Green Mountain samples, probably reflecting recent current season spread.

In conclusion, this study clearly demonstrates the effect of cultivar on the prevalence of the different mosaic-inducing viruses. In some cultivars (e.g. Green Mountain and Russet Burbank), PVA plays a significant role as a cause of mosaic, while in others (e.g. Red Pontiac and Shepody) it does not.

The relative importance of either PVX or PVY as causes of mosaic was found to vary considerably. This study does, however, reinforce the importance of PVX as a cause of mosaic and suggests that efforts to control this often latent virus will be helpful in reducing the occurrence of mosaic disease.

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Table 1. Potato viruses X, A, and Y detected by ELISA in mosaic-diseased (severe and mild) samples of the Shepody, Red Pontiac and Green Mountain cultivars.

Minus Cambinations	Shepody		Red Po	ontiac	Green Mountain		
Virus Combinations	Severe Mosaic	Mild Mosaic	Severe Mosaic	Mild Mosaic	Severe Mosaic	Mild Mosaic	
PVX alone	13	32	22	16	16	51	
PVA alone	0	0	0	0	2	3	
PVY alone	85	25	17	2	2	0	
PVA + PVX	0	0	0	0	38	28	
PVY + PVX	204	54	15	4	17	27	
PVA + PVY	0	0	0	0	0	0	
PVA + PVY + PVX	0	0	0	0	7	2	
Total	302	111	54	22	82	111	

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# Incidence of a "Take-All Like Fungus" recovered from the crowns, stems and roots of winter wheat grown in Manitoba<sup>1</sup>

A.V. Sturz and C.C. Bernier

The occurence of winter wheat with take-all symptoms is reported for Manitoba in the 1983-84 growing season. Some of the different methods of measuring take-all in the field are discussed. The methodology used to characterize the main fungal components of the crown-root rot complex is outlined. Fungi isolated included a *Gaeumannomyces graminis* var. *tritici* "like" fungus, *Fusarium culmorum*, *F. avenaceum*, *F. equiseti*, and *Cochliobolus sativus*. The effects that different cropping systems had on the occurrence of take-all and the other principal fungi in the crown-root rot complex are briefly assessed.

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La présence de symptômes de piétin-échaudage sur le blé d'hiver au Manitoba est rapportée pour la saison 1983-84. Quelques unes des différentes méthodes d'évaluation sur champ du piétin-échaudage sont discutées. La méthodologie employée pour charactériser les composantes cryptogamiques principales du piétin est décrite. Les micro-organismes isolés comprennent un champignon semblable à *Gaeumannomyces graminis* var. *tritici, Fusarium culmorum, F. avenaceum, F. equiseti,* et *Cochliobolus sativus*. Les effets des différents régimes de récoltes sur la fréquence piétin-échaudage et des autres principales composantes du complexe sont évalués brièvement.

#### Introduction

In recent years acreages of winter wheat, Triticum aestivum, have increased in the prairie provinces of Canada with the introduction of the winter hardy variety Norstar and the development of improved management systems. With increased winter wheat production has come an increase in the incidence of take-all, a crown and root disease caused by the fungus Gaeumannomyces graminis (Sacc.) Von Arx & Olivier var. tritici Walker. In the 1983-84 growing season, brief surveys assessing the extent of take-all in wheat fields in South-Western Manitoba found levels of the disease ranging from slight to moderately severe (unpubl. Sturz and Bernier). An early maturing winter wheat crop accelerated by dry conditions in the latter part of the growing season masked the true extent of the disease and take-all levels may well have been higher. This paper describes 1) some of the different methods used to assess the incidence of take-all in the field, 2) briefly outlines the methodology used to characterize the main fungal components in the crown-root rot disease complex found at the experimental winter wheat plots, Minto, Manitoba, and 3) reports on the effects that different cropping systems had on the occurrence of take-all and the other principal fungi in the crown-root rot disease complex at that site.

#### **Materials and Methods**

The incidence of take-all in fields and experimental plots was assessed by counting the number of diseased plants found within randomly selected blocks 20.7 m<sup>2</sup>, comprising 14 rows of winter wheat. Plants were examined at approximately growth stage 10.54-11.1 Feekes. Estimations of disease inci-

Surveys were made in a number of experimental plots growing winter wheat following different cropping sequences. Disease

Table 1. Occurrence of winter wheat with take-all symptoms, at growth stage 11 (Feekes) following different previous crops, in the growing season 1983-84, at the experimental ground, Minto.

Winter Wheat Sown On	Mean No. Plants Diseased	Mean No. Foci	Mean Size Largest Foci (M <sup>2</sup> )	Mean Size Foci (M <sup>2</sup> )	Mean No. Plants Per Foci	%** Area T. A.
Winter Wheat	22.5*	10	0.4	0.02	1.8	3.7
Barley	80.5	70	0.4	0.007	1.1	3.5
Rape	20.5	13	1.1	0.007	1.7	6.0
Flax	15.5	13	0.04	0.007	1.2	0.5
Oats	8.0	8	0.005	0.005	1.0	0.3

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dence were based on whole plants not individual tillers. No attempt was made to assess disease severity in individual plants. Assessments were based on head-development criteria [(i) nil through (ii) rudimentary to (iii) white head being considered as presence of the disease and (iv) normal head development as absence] and presence or absence of the characteristic root-discoloration and/or stem-base blackening. Periodically, infected plants were removed so that symptomed tissue could be checked in the laboratory for the causal organism.

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<sup>\*</sup>Counts were taken from a randomly selected block of winter wheat 20.7  $\ensuremath{\text{M}}^2$ .

<sup>\*\*</sup>The size of a one plant take-all (T.A.) foci was estimated as  $0.005\,\text{M}^2.$ 

characteristics examined included the number of disease foci, the mean size of the largest and the average foci found, the number of diseased plants per foci, and the percentage area of wheat lost to take-all.

Infected tissue was surface sterilized with a 1-2% solution of sodium hypochlorite (Na O CI) for 3 min, rinsed with sterile distilled water to remove chlorine and plated onto potato dextrose agar (amended with antibiotics). To reduce surface tension Tween 20 (polyoxyethelene sorbitan monolaurate) was incorporated into both the sodium hypochlorite solution and the sterile distilled water at approximately 0.1 parts per thousand. Plates were incubated in the dark at 22°C for 7 days.

A seedling-baiting technique (modified after Hornby 1969) was used to isolate the take-all causal organism and the other component fungi of the root-crown disease complex. The root, crown and lower stem tissues of wheat plants displaying take-all symptoms, as well as the rhizosphere soil and plant debris of winter wheat plants grown in different crop rotations, were studied. Fusaria were identified to the species level after the method described by Sturz and Johnston (1983).

#### Results

Higher levels of take-all were recorded after rotations of wheat, barley and rape as compared to oats or flax (Table 1). It was found that the incidence of take-all based on the

Table 2. Percent recovery of fungal species from the crown, root and stem tissues of Norstar winter wheat seedlings inoculated at the primary root with wheat stem sections displaying take-all symptoms.

Fungal Spp. Isolated	Inoculated %	Control %
Microdochium bolleyi	2.5 <sup>++</sup>	0
"Ggt" +	95	0
Fusarium equiseti	15	0
Other fusaria <sup>+++</sup>	10	0
Cochliobolus sativus	5	0
Alternaria spp.	0	90
Trichoderma spp.	0	10
Other fungi	0	70
Plants with crown/stem rot symptoms	65	0

<sup>+&</sup>quot;Ggt" Gaeumannomyces graminis var. tritici 'like' fungus.

++Sample of 40 seedlings.

Table 3. Percent recovery of fungal spp. from the root, crown and stem tissues of Norstar winter wheat seedlings sown in soil and plant debris taken from the rhizospheres of winter wheat plants grown after different crop sequences.

Fungal Spp. Isolated	Wheat Stems with	Soil and Plant Debris from the Rhizosphere of Winter Wheat Grown in Plots after Different Crop Sequences**						
i ungar 3pp. isolateu	Take-all Symptoms* Only (%)	Winter Wheat (%)	Barley (%)	Oat (%)	Flax (%)	Rape (%)		
"Ggt" <sup>†</sup>	50***		_	_		17		
Microdochium bolleyi	17	83	42	42	75	33		
Fusarium oxysporum	33		_	_	_	_		
F. avenaceum	25	33	33	8	42	17		
F. culmorum	17	17	33	17	_			
Other fusaria ++	8	_	17	_	_	8		
Cochliobolus sativus	_	_	_	_	_	8		
Rhizoctonia cerealis	_	_	17		41			
Other fungi <sup>+++</sup>		58	41	33	25	17		
Plants with crown/stem rot symptom	ns 42	8	17	0	8	8		

<sup>&</sup>quot;"Ggt" Gaeumannomyces graminis var. tritici 'like' fungus.

<sup>+++</sup> F. avenaceum, F. acuminatum Ellis & Everhart, F. solani and unidentified fusaria.

<sup>++</sup> F. equiseti, F. arthrosporioides (?), F. sporotrichioides.

<sup>+++</sup> Alternaria spp., Trichoderma spp., Mucor spp., Rhizopus spp.

<sup>\*</sup>Debris ≥ 2.0 mm.

<sup>\*\*</sup>Rhizosphere soil and plant debris < 2.0 mm.

<sup>\*\*\*</sup>Sample of 12 seedlings.

parameter, number of diseased plants per plot did not correspond well with those estimates of incidence measured as the percent aea of wheat lost to take-all.

Recovery of the take-all organism (*Ggt*) proved to be extremely difficult. Isolations from the crown and stem sections of 20 wheat plants displaying take-all symptoms, yielded only *Fusarium equiseti* (Corda) Sacc. (55%), *F. solani* (Mart.) Sacc. (25%), *F. oxysporum* Schlect (30%) and small amounts of *Cochliobolus sativus* (Ito & Kurib.) (15%). The seedling-baiting technique (modified after Hornby, 1969) was more successful (Table 2). By this method a *Ggt* "like" fungus (95%) was recovered from 'inoculated' seedlings. *Microdochium bolleyi* (Sprague) de Hoog (2.5%) was also recovered along with isolates of *F. equiseti* (15%). Of 40 seedlings baited after this method, 65% developed typical take-all stem and root blackening symptoms.

Repeating the Hornby technique, using as an inoculum source the rhizosphere soil and plant debris of wheat plants grown following different crop sequences, higher levels of *M. bolleyi* were isolated [winter wheat following-winter wheat (83%), — barley (42%), — oats (42%) — rape (75%) and — flax (33%)] (Table 3). Species of fusaria were also recovered and included *F. culmorum* (W.G.Sm.) Sacc. (17-33%), *F. avenaceum* (Fr.) Sacc. (25-44%), *F.equiseti* (7%) and *F. sporotrichioides* Sherb. (7%). The *Ggt''*like' fungus and *C. sativus* were only recovered from winter wheat following rape (17% and 8% respectively). *Rhizoctonia cerealis* van der Hoeven was recovered in winter wheat following barley (17%) and winter wheat following flax (41%) rotations.

#### **Discussion**

In this study disparities between the different assessment methods were thought to be mainly attributable to seedling and early plant mortality, thereby lowering the accuracy of those methods involving counts of visibly diseased plants. However, diseased plant counts when related to mean number of foci and mean foci size were superior to percent area takeall assessments in estimating the spatial distribution of the disease. "Residue effects" resulting in differences in the maturing times of winter wheat following different crop sequences contributed to the problems of devising a uniformly accurate method for assessing this disease in the field. It is concluded that additional earlier measurements, where possible, would be advantageous in estimating the incidence of the disease.

Difficulties encountered in isolating the take-all causal organisms are ascribed primarily to the age and dried state of symptomed material at the time of sampling. Fusaria such as  $F.\ culmorum,\ F.\ avenaceum$  and  $F.\ equiseti$ , are all documented as severe pathogens of the crowns and roots of overwintering cereals. Differences in recovery levels of the Ggt "like" fungus from rhizosphere soils and associated plant debris as compared to wheat stem fragments may in part be due to the better survial of the Ggt "like" fungus in the larger wheat debris fractions. It cannot be inferred from these results that the Ggt "like" fungus is absent from the rhizosphere fractions of the soils tested, only that it is absent from soil particles and plant debris  $< 2.0\ \text{mm}$  in size.

#### **Acknowledgement**

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### **Eumartii wilt of potato in Alberta**

S.F. Hwang<sup>1</sup> and I. R. Evans<sup>2</sup>

This paper deals with *Fusarium* species isolated from diseased potato tubers held in storage or from wilted plants growing under field conditions. It reports the first documented occurrence of Eumartii wilt of potatoes in Alberta.

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Cet article traite des espèces de *Fusarium* isolées à partir de tubercules de pommes de terre malades entreposées ou à partir de plants flétris poussant au champ. On y note le premier cas documenté en Alberta de flétrissure de la pomme de terre causée par *F. solani* var. *eumartii*.

#### Introduction

Fusarium species cause a variety of potato diseases. About 20 species are pathogenic on potatoes. They can cause wilt, tuber rot, dry rot of tubers in storage, and seed-piece decay. According to Fusarium species, potato wilt can be classified as Eumartii wilt, Oxysporum wilt or Avenaceum wilt. Eumartii wilt is generally the most aggressive and results in the greatest damage to potatoes. Oxysporum wilt is usually milder than Eumartii wilt and the disease is typically a vascular wilt in contrast to the other Fusarium wilts described here, which are more nearly cortical rots. The mycelia of F. oxysporum are closely limited to the xylem vessels of the stem, while those of F. avenaceum occur abundantly in both the vascular and cortical tissues of the lower stem, while the mycelia of F. solani var. eumartii are most abundant in the stem cortex (McLean and Walker, 1941). Avenaceum wilt is comparable in severity to Oxysporum wilt.

The first symptoms of Eumartii wilt are light green areas between the veins of the top leaves. (Ohms et al., 1961). Later, the leaves develop reddish to purplish spots, giving infected plants a bronzed appearance (Fig. 1). If the epidermal tissue of the stolon is removed, a brown discoloration of the remaining tissue is evident, sometimes confined to the vascular elements. In severe cases, the entire stolon my be completely rotted (Fig. 4). Affected tubers show a vascular discolaration which originates at the point of stolon attachment and extends into the tuber. Eumartii wilt has been reported as widespread in the United States in Pennsylvania, New York, Ohio and Idaho (Haskell, 1916; Goss, 1924). The disease has been serious in Idaho causing severe losses in some growing areas, due to internal tuber damage (Ohms and Fenwick, 1961). This is not the case in Canada and as far as we are aware this is the first confirmed report of this disease in Alberta.

#### **Materials and Methods**

During 1984, isolations of *Fusarium* species were made from diseased tubers collected from storages and one commercial potato field in Alberta. Isolation of the causal fungus was

made as follows: different portions of the diseased plant were cut into small pieces approximately 1 cm<sup>2</sup> and surfacesterilized in 95% ethyl alcohol for one minute, followed by immersion in 1:10 commercial bleach solution (12% sodium hypochlorite) for one minute. They were then transferred aseptically, through three rinses of sterile distilled water onto Nash and Snyder's pentachloronitrobenzene (PCNB) medium (Nash and Snyder, 1962) in order to isolate Fusarium sp. After making sure that the cultures were pure by the method of single-sporing described by Snyder and Smith (1962), inoculations were made on healthy plants. Inoculum concentrations of several selected isolates identified as F. solani var. eumartii and F. avenaceum were washed from PDA tubes and spore numbers determined by hema-cytometer counts. The inoculum level was adjusted with sterile distilled water to approximately 2 imes 10 spores/mL. In order to avoid significant changes of inoculum viability, they were refrigerated at 4°C after preparation and used within a 4 hour period.

On July 2nd, 1984, seed pieces of two potato plants cultivars, Russet Burbank and Warba, grown for one month in the greenhouse in pots, were inoculated by injecting a suspension  $(2 \times 10^5 \text{ spores/mL})$  of spores into the stems just below soil level. Four plants were inoculated with each isolate. Comparable plants of each cultivar were injected in the same way, but no inoculum was introduced. Seed tuber pieces of Russet Burbank were also inoculated. Prior to inoculation, seed tubers were washed, dipped into 0.5% NaOCI and allowed to dry. The tubers were cut and dipped at once in the spore suspensions and planted immediately in steamed soil. Twenty seed pieces were used in each treatment. All the plants were kept in the greenhouse at 24°C. Data were recorded by measuring the length of stem discoloration one week after the stem moculation test and by measuring the height of potato plants one month after inoculation test and by measuring the height of potato plants one month after inoculation of the seed-pieces.

#### **Results and Discussion**

Fusaria were successfully isolated form vascular discolored stem tissue below or close to the soil line and from discolored vascular tuber tissue. From 80% of the isolations there developed a species of *Fusarium* having a pale olive-buff color on the PDA medium. It was readily obtained from the potato roots and less readily from stem pieces (Fig. 3). This species was almost identical to *F. eumartii* described by Carpenter in 1915 (1). The isolate had sparse aerial mycelium, light brown

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sporodochia, macroconidia nearly straight in the lower half, slightly curved in the upper half, 3 to 4 septate, with occasional microconidia.

In the stem inoculation tests, *F. solani* var. *eumartii* produced complete disorganization of the stem at the point of inoculation (Fig. 5). The average lengths of stem discoloration by several isolates of *F. solani* var. *eumartii* and *F. avenaceum* are presented in Table 1. Three weeks after inoculation, the plants were dead and the below ground portions of the stems were found to be nearly rotted through while check plants remained healthy.

A reduction of shoot height was found to occur when seedpieces were inoculated by *F. solani* var. *eumartii* and *F. avenaceum* prior to planting. The results in Table 2 indicate that

Table 1. The average length of stem discoloration of two varieties of potatoes one week after inoculation with strains of *Fusarium solani* var. *eumartii* and *F. avenaceum*.

		Stem Discoloration (mm)		
Species and Strain No.	Russet Burbank	Warba		
F. solani var. eumartii				
7	32	17		
15	22	19		
2	9	14		
F. avenaceum				
3	6	4		
6	6	4		
Check	0	0		

inoculation with *F. solani* var. *eumartii* could cause a 50% failure of seed-piece emergence. Even when the plants emerged, the average height of one-month-old inoculated plants were markedly lower (11.4 cm) compared to the control plant, (28.9 cm). *F. avenaceum* did not have an effect on seed-piece emergence; however, the height was reduced (22.4 cm).

Inoculation experiments show that F. solani var. eumartii is an extremely virulent parasite capable of causing seed piece

Table 2. The emergence percentage and height of potato shoots (Russet Burbank) one month after seed-pieces were inoculated with *F. solani* var. *eumartii* and *F. avenaceum*.

Species and Strain No.	Emergence %	Green Height cm				
F. solani var. eumartii						
7	50	17.1				
24	50	16.6				
2	70	11.4				
28	80	18.1				
F. avenaceum						
6	100	22.8				
21	100	22.4				
Check	100	28.9				

emergence failure, severe growth reduction, stem-end rot and internal discoloration of tubers. It is probable that there are other weak parasites, capable of causing similar host reactions when environmental conditions are favorable. Such weak pathogens might be *F. equiseti* and *F. trichothecioides* which were also isolated in this project from diseased plants and tubers.

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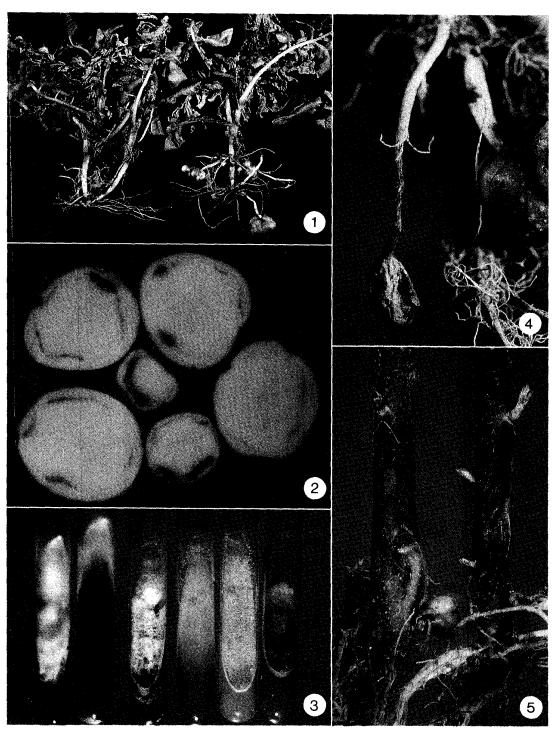


Figure 1. Symptoms of Eumartii wilt in Russet Burbank showing bronzed appearance.

Figure 2. Symptoms of Eumartii wilt in Norgold Russet potato tuber from storage — Note bacterial ring rot like symptoms. Figure 3. Four cultures on the right side illustrated for *F. solani* var. *eumartii* and the left two cultures represented

Figure 4. Symptoms of a severely infected stolon and small tubers in Russet Burbank.

Figure 5. Symptoms of Eumartii wilt showing the effect of inoculation with *F. solani* var, *eumartii* on Russet Burbank, Note the brown stem discoloration on the right, check on left.

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# Evaluation of polyacrylamide gel electrophoresis, bioassay and dot-blot methods for the survey of potato spindle tuber viroid

R.P. Singh<sup>1</sup> and C.F.Crowley<sup>2</sup>

Three methods (e.g. polyacrylamide gel electrophoresis (PAGE), bioassay using *Solanum berthaultii* and dot-blot) were compared for the detection of potato spindle tuber viroid (PSTV). Composite samples ranging in dilution from 1:3 to 1:500 (infected discs:healthy discs) were used. Extracted nucleic acid was used for all the tests. PAGE could not detect PSTV beyond 1:10 dilution whereas both bioassay and dot-blot detected PSTV reliably up to 1:300 dilution. Using bioassy and dot-blot, PSTV was surveyed in 103 tablestock fields of potato. Tablestock fields of potato cultivars Kennebec, Russet Burbank, Shepody and Superior planted with seed grade Elite II to Certified were surveyed. No PSTV was found in any of the 618 composite samples tested, which represented 51,500 leaves. Freedom of the seed potato crop from PSTV was thus confirmed by laboratory tests.

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On a comparé la capacité de détection du viroide de la filosité de la pomme de terre (PSTV) à l'aide de trois techniques, l'électrophorèse au gel de polyacrylamide (PAGE), l'essai biologique avec *Solanum berthaultii* et la tache-point, en utilisant des échantillons composés dont la dilution varie de 1:3 à 1:500 (disques infectés: disques sains). Tous les tests ont utilisé des extraits d'acide nucléique. PAGE n'a pas permis la détection du PSTV dilué au-delà de 1:10, tandis que l'essai biologique et la tache-point l'ont détecté sans erreur jusqu'à une dilution de 1:300. La présence du PSTV fut inventorié dans 103 champs de pommes de terre de table en utilisant les techniques de l'essai biologique et de la tache-point. Cet inventaire couvre des champs de pommes de terre de table des cultivars Kennebec, Russet Burbank, Shepody et Superior plantés avec de la semence de catégorie Elite II à Certifiée. Aucun PSTV ne fut détecté dans les 618 échantillons composés représentant 51,500 feuilles. Les tests en laboratoire ont ainsi permis de confirmer l'absence du PSTV des pommes de terre de semence.

#### Introduction

Although potato spindle tuber viroid (PSTV) was first observed on the North American continent in the early 1920s (1), it is not an economic problem now (6,8). However, in order to ensure that the potato seed crop is free of PSTV, it is necessary to monitor the crop during the growing season. Visual indexing has been used for a long time and has been successful in reducing the PSTV incidence in the seed crop (8). However, availability of additional methods of PSTV detection make it necessary to determine their applicability for monitoring of large acreages of potato fields for PSTV. Therefore, the method of polyacrylamide gel electrophoresis (PAGE) (2,4,5), bioassay using *Solanum berthaultii* (7), and nucleic acid hybridization (3) were compared for large-scale testing. The latter two methods were also used to survey the potato crop for PSTV in New Brunswick.

#### **Materials and Methods**

Viroids and Detection Procedures — In order to obtain fieldgrown potato plants with current-year PSTV symptoms, as an aid for visual indexing, the following was done. Virus and viroid-free tubers of several cultivars were obtained from the Potato Breeding Program of the Fredicton Resarch Station. Single-eyed tuber pieces were planted in peatmoss and when the plants were 10-12 cm in height, they were inoculated with PSTV containing nucleic acid extract (5,7). One day after the inoculation, plants were transplanted into the field (May 20, 1984). Plants were observed for symptoms every two weeks. As a comparison, viroid-free plants grown from the same tubers were planted in the adjoining plot.

For the comparison of test methods, nucleic acid was extracted as described previously (4,7). One PSTV-infected leaf disc (7 mm) was mixed with 2,4,9,49,99,249,299,399 and 499 leaf discs of viroid-free potato. For nucleic acid extraction, leaf to extracting buffer ratio of 1:3 (w/v) was always used. The extracted nucleic acid was dissolved in distilled water. Three  $\mu$ I of each nucleic acid sample was applied to the dotblot membrane (Agdia, Inc., Mishawake, Indiana, U.S.A.); 250  $\mu$ I of the nucleic acid was diluted equally with glycine phosphate buffer and used to inoculate *S. berthaultii* seedlings (7); and another 100  $\mu$ I of the nucleic acid extract was used for the PAGE test (4,5).

Survey of Potato Fields for Potato Spindle Tuber Viroid — One hundred and three potato fields, selected at random were used for the survey of PSTV. All the fields were planted for processing or tablestock purposes. Because processing or tablestock fields are the farthest removed potatoes from nucleus virus-free stocks, under the Seed Potato Certification Program, it was expected that if there is any PSTV present, it should be more easily found in tablestock fields than in seed fields, which are inspected thoroughly every year. Once fields were selected, the planting date, cultivar, source and grade of seed used for planting were recorded. The survey field was

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Fig 1. The field symptoms of potato spindle tuber viroid in cultivars, Kennebec, Russet Burbank, and Shepody from experimental plot. Upright growth, reduced upper leaves, and bushy appearance of plant is evident in all cultivars.

Table 1. Comparison of potato spindle tuber viroid detection in composite leaf samples of potato by various methods.

	_	Methods			
Dilution Ratio	Test	PAGE* 1 2 3	Bioassay** 1 2 3	Dot-blot <sup>+</sup> 1 2 3	
1:3		+++	+++	+++	
1:5	•	+ + +	+ + +	+++	
1:10		+ + +	+ + +	+ + +	
1:50		+	+ + +	+ + +	
1:100		NT	+++	+ + +	
1:250		NT	+ + +	+ + +	
1:300		NT	+ +	++?	
1:400		NT	+	+	
1:500		NT			

<sup>\*</sup>Polyacrylamide gel electrophoresis.

observed by two to four persons, each covering a large crosssection of the field and collecting leaflets at random. A total of 500 leaflets (in lots of 100) were collected. In case of suspicious looking plants, additional samples were collected. All leaflet samples were kept cold in a styrofoam box containing ice. On arrival at the laboratory, discs were cut (7 mm) and nucleic acid was extracted as described (7). For each field, one PSTV control was used. A PSTV control consisted of one PSTV-infected disc mixed with 99 viroid-free discs of same potato cultivar. Extracted nucleic acid was used for bioassy and dot-blot tests.

#### Results

Field Symptoms of Current Year Infection by PSTV — Because PSTV is very rare in potato-growing areas, the procedure of growing plants in the greenhouse and their transplanting into the field soon after inoculation was the nearest thing to a natural PSTV infection in the field. This procedure provided close to 100% infection in 29 potato cultivars inoculated in this way (10 plants in each cultivar). All inoculated plants developed moderate to severe symptoms within six weeks of transplanting. The symptoms of three cultivars used in the survey are shown in Fig.1. Availability of PSTV symptomatic cultivars, particularly the newly released Shepody (9), was of assistance in visual inspection of fields to be surveyed.

Comparison of Detection Procedures — Three experiments were done with field-grown leaf material using PAGE, bioassay and dot-blot tests as shown in Table.1. PSTV was detected from potato leaves by PAGE up to a dilution of 1:5 to 1:10 but not beyond. Both bioassay on *S. berthaultii* and dot-blot nucleic acid hybridization detected PSTV up to a dilution of 1:300. Although PSTV was detected at higher dilutions with both bioassay and dot-blot (Table 1) the detection was not consistent. Therefore, for survey purposes, a dilution of 1:100 was selected.

<sup>\*\*</sup>Bioassay on Solanum berthaultii plants.

<sup>&</sup>lt;sup>†</sup>Nucleic acid hybridization, using Agdia Inc. kit. NT = Not tested.

Table 2. Survey and detection of potato spindle tuber viroid in potato fields planted for tablestock use.

Potato	Seed Grade	No. of	Detection of PSTV		
Cultivar	Planted	Fields	Bioassay*	Dot-blot**	
Kennebec	EII	1	0/5+	0/5	
Kennebec	EIII	1	0/5	0/5	
Kennebec	F	4	0/20	0/20	
Kennebec	С	2	0/10	0/10	
PSTV controls <sup>+</sup>	+ _	8	8/8	8/8	
Russet Burbank	EIII	20	0/100	0/100	
Russet Burbank	F	27	0/135	0/135	
Russet Burbank	С	6	0/30	0/30	
<b>PSTV</b> controls	_	53	53/53	50/53	
Shepody	EIII	6	0/30	0/30	
Shepody	F	9	0/45	0/45	
Shepody	С	8	0/40	0/40	
<b>PSTV</b> controls	_	23	23/23	21/23	
Superior	ΕII	1	0/5	0/5	
Superior	EIII	3	0/15	0/15	
Superior	F	8	0/40	0/40	
Superior	С	7	0/35	0/35	
PSTV controls		19	18/19	19/19	

<sup>\*</sup>Bioassay on Solanum berthaultii plants.

Survey of PSTV in Potato Fields — A total of 103 potato fields were surveyed. The four major cultivars and their respective number of fields surveyed were: Russet Burbank 53, Shepody 23, Superior 19, and Kennebec 8. A total of 51,500 leaflets were collected and 618 nucleic acid extractions were made. The seed grades planted ranged form Elite II,2 fields; Elite III, 30 fields; Foundation, 48 fields; and Certified, 23 fields. No PSTV was found in any field either by bioassay or by dot-blot test (Table 2) irrespective of cultivar or seed grade used for planting.

#### Discussion

In an earlier study (8) we showed that visual inspection data indicated that PSTV was not present in seed potato fields since 1980. This study further extends that observation to the tablestock fields, as well as by the sensitive detection procedures of bioassay and dot-blot (Table 2). The probable reasons for the absence of PSTV in seed potato fields have been discussed before (8) and may apply to tablestock fields as well.

The procedures of dot-blot and bioassay both are more sensitive than PAGE. However, the bioassay requires large greenhouse space for testing, while the dot-blot as performed by a commercial agency (Agdia Inc.) is quite inexpensive. The problem of membrane dispatching and minimizing delays by mail could further improve the dot-blot's usefulness.

#### **Acknowledgements**

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<sup>\*\*</sup>Nucleic acid hybridization, using Agdia Inc. kits.

<sup>\*</sup>From each field 5 samples consisting of 100 leaflets each were used to extract nucleic acid.

<sup>+++</sup>For each field there was one PSTV control included, consisting of 1 PSTV infected leaflet combined with 99 healthy leaflets of the same cultivar.

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Titles should be concise and informative providing, with the Abstract, the key words most useful for indexing and information retrieval.

Abstracts of no more than 200 words, in both English and French, if possible, should accompany each article.

Figures should be planned to fit, after reduction, one column (maximum 84 × 241 mm) or two columns (maximum 175 × 241 mm), and should be trimmed or marked with crop marks to show only essential features. Figures grouped in a plate should be butt-mounted with no space between them. A duplicate set of unmounted photographs and line drawings is required. Figures should be identified by number, author's name, and abbreviated legend.

Tables should be numbered using arabic numerals and have a concise title; they should not contain vertical rules; footnotes should be identified by reference marks (\*  $\dagger$  § # ¶ \*\*  $\dagger$ †) particularly when referring to numbers.

Literature cited should be listed alphabetically in the form appearing in current issues; either the number system or the name-and-year system may be used. For the abbreviated form of titles of periodicals, refer to the most recent issue of Biosis List of Serials published by Biosciences Information Service of Biological Abstracts or to the NCPTWA Word Abbreviation List, American National Standards Institute.

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Les articles et les communiqués sont publiés en anglais ou en français. Les manuscrits (l'original et une copie) et toute la correspondance qui s'y rapporte doivent être envoyés à M. H.S. Krehm, Service des programmes de recherche, Direction de la recherches, ministère de l'Agriculture du Canada, Ottawa (Ontario) K1A OC6.

Les manuscrits doivent être concis et faire preuve de suite dans le style, l'orthographe et l'emploi des abréviations. Ils doivent être dactylographiés à double interligne, de préférence sur des feuilles à lignes numérotées. Toutes les pages doivent être numérotées y compris celles portant le résumé, les tableaux et les légendes. Pour plus de renseignements sur le format des feuilles et le style, prière de consulter nos dernières publications et le CBE Style Manual (3e ed. 1972) de l'American Institute of Biological Sciences, Washington (DC). Dans la mesure du possible, les données numériques doivent être exprimées en unités métriques, (SI) ou être suivies de leur équivalent métrique. L'emploi de crochets est autorisé pour l'identification du nom scientifique d'un microorganisme pathogène après le nom commun de la maladie dont il est l'agent causal.

Les titres doivent être courts et révélateurs en contenant, avec le résumé, les mots clés les plus utiles pour le classement et l'extraction de l'information.

Chaque article doit être accompagné d'un résumé d'au plus 200 mots en anglais et en français, si possible.

Les figures doivent pouvoir, après réduction, remplir une colonne (maximum 84 × 241 mm) ou deux colonnes (maximum 175 × 241 mm) et devraient être taillées ou montrer les parties essentielles à garder. Les figures groupées sur une même planche doivent être montées côte à côte, sans intervalle. L'article doit être accompagné d'un double des photographies non montées et des graphiques. Les figures doivent être numérotées, porter le nom de l'auteur et une légende abrégée.

Les tableaux doivent être numérotés en chiffres arabes et avoir un titre concis. Ils ne devraient pas avoir de lignes verticales. Les renvois doivent être identifiés par un signe typographique particulier (\* † § # ¶ \*\* ††) surtout lorsqu'il s'agit de nombres.

Les références bibliographiques devraient être citées par ordre alphabétique comme dans les livraisons courantes. On peut utiliser le système de numération ou le système nom-et-année. Pour l'abrégé du titre des périodiques, on suivra l'édition la plus récente de Biosis List of Serials publiée par les Biosciences Information Services de Biological Abstracts ou la NCPTWA Word Abbreviation List et l'American National Standards Institute, Standards Committee Z39.