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# Canadian Plant Disease Survey

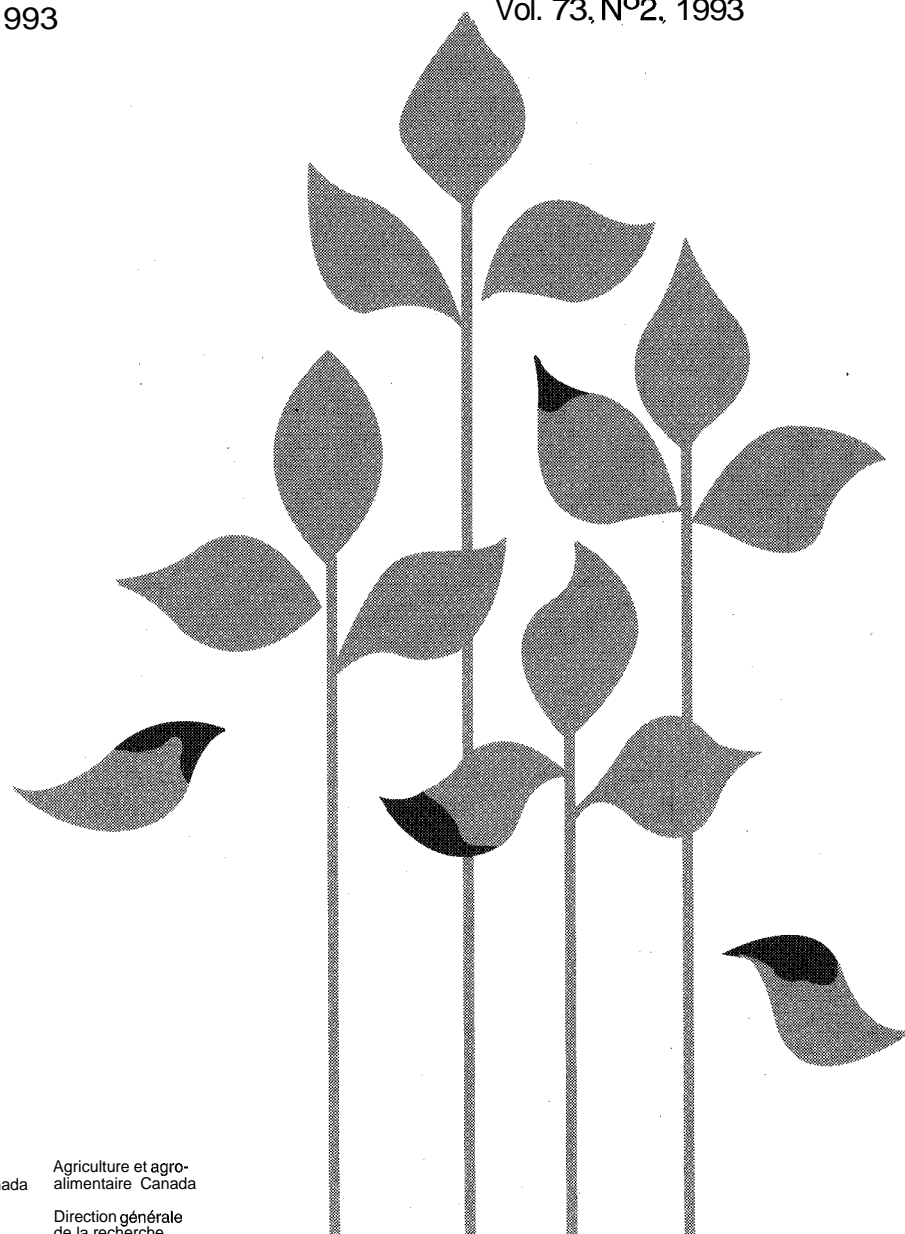
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# Inventaire des maladies des plantes au Canada

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# Canadian Plant Disease Survey

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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 periodique d'information sur la frequence des maladies des plantes au Canada, leur gravité, et les pertes qu'elles occasionnent. La redaction accepte d'autres communications originales notamment sur la mise au point de nouvelles methodes d'enquête et de lutte ainsi que sur l'évaluation des nouveaux produits. De temps a autre, il inclut des revues et des syntheses de rapports d'intérêt immediat pour les phytopathologistes.

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# Distribution of virulent blackleg on standing rapeseed/canola crops in Saskatchewan, 1982-1991

G.A. Petrie<sup>1</sup>

Surveys of standing crops of rapeseed/canola were conducted during July and August between 1982 and 1991. The virulent strain of *Leptosphaeria maculans* (blackleg) consistently was less prevalent and its average incidence was lower in the northern growing areas of Saskatchewan crop districts 8a, 9a, and 9b, than in areas farther south. Disease severity on infected plants also was often less in the north. Crops of both *Brassica rapa* and *B. napus* had less blackleg in northern areas. Blackleg incidences in *B. rapa* crops were consistently lower than those in crops of *B. napus*.

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Entre 1982 et 1991, pendant les mois de juillet et d'août, des études ont été effectuées sur des cultures sur pied de colza/canola. La souche virulente de *Leptosphaeria maculans* (jambe noire) était moins répandue et son incidence moyenne a été moindre dans les régions nordiques de croissance des districts agricoles 8a, 9a, et 9b de la Saskatchewan que dans les régions plus éloignées au sud. Au nord la maladie était souvent moins virulente sur des plants infectés. Les cultures de *Brassica rapa* et de *B. napus* ont été moins touchées par la jambe noire dans les régions nordiques. Les incidences de la jambe noire dans les cultures de *B. rapa* ont été moins prononcées que celles des cultures de *B. napus*.

## Introduction

After 1961, putative strains of *Leptosphaeria maculans* (Desm.) Ces. & De Not. (blackleg) weakly virulent on adult rape (*Brassica napus* L.) and turnip rape (*B. rapa* L.) plants were found intermittently on Cruciferae in crop district(s) (C.D.) 6 and 8b in central Saskatchewan (9,15). A virulent strain of *L. maculans* (LM-VIR) initially was found on stubble of the 1975 rapeseed crop in three widely separated Saskatchewan fields near Melfort (C.D. 8a), Humboldt (C.D. 8b), and Rosthern (C.D. 6b) (7,10). By 1980, infestations of LM-VIR had been found near Cutknife (C.D. 9b) and near the Manitoba border in crop district 5a (14). Spread of the pathogen was predominantly northwest and southeast from the original centers of infestation. In 1982, crops with high incidences of basal stem canker were widespread in crop districts 6 and 8b for the first time, with yield losses ranging up to 50% (11). Subsequently, blackleg prevalence, incidence and severity continued to increase along a northwest-southeast line from the southern part of crop district 9b to crop district 5. The disease spread into eastern Alberta Census Divisions 7 and 10 from crop district 9b (5,6), and into western Manitoba from crop district 5 (16).

The spread of LM-VIR into northern rapeseed/canola growing areas was not as rapid as its east-west movement. Since 1984, LM-VIR has been more widespread in crop district 8b than farther north and east in crop districts 8a and 9a (2,3,4,13). There are no published data which permit a comparison of levels of LM-VIR in southern and northern parts of crop district 9b, but LM-VIR only recently has been found in the Meadow Lake area in the extreme northern part of crop district 9b (R. Gugel, pers. comm.). The pathogen also has been slow to become established in north-central growing areas north and east of Prince Albert (C.D. 9a). The purpose of this paper is to further examine the distribution of LM-VIR in three paired areas: (1) southern and central parts of crop district 9b; (2) crop districts 6 and 9a; and (3) southern and northern parts of crop district 8.

## Materials and methods

The methods used in surveys conducted between 1982 and 1985 have been described (11,12,13). In 1991, 100 Saskatchewan canola fields were surveyed for blackleg between July 22 and August 31. Areas surveyed were similar to those done in 1971 (8, Fig. 2) except that crop districts 5b and 7b were not surveyed in 1991. All but a few crops were standing when sampled. Fifteen fields were sampled in crop district 6a, 20 in 6b, 11 in 9a, 23 in 9b, and 31 in crop district 8. Crop district 9b was subdivided into central (C) and southern (S) sectors, with about equal numbers of fields in each. In crop district 9b(C), the survey route was along highways 26 and 3, from north of North Battleford to

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the Alberta border. In crop district 9b(S), the route was along highway 16 to Maidstone, south on highway 21 to highway 40, and east on no. 40 to North Battleford. Crop district 8 was divided into northern and southern sectors and these were further subdivided into eastern (E) and western (W) sectors. The route travelled in crop district 8(NW) was along highways 25 and 3 from St. Louis to Melfort. In crop district 8(NE) the route was along grid 681 and highways 335, 35, 55, 23, and 3, linking Star City, Nipawin, Carrot River and Tisdale. In crop district 8(SW) the route was along grid 756 from Prud'homme to Annaheim, and in crop district 8(SE) it was along highways 35 and 349 from Sylvania to Naicam. Crop district 9a was surveyed between Prince Albert and Choiceland. Surveys in crop district 6 centred around Saskatoon. A three year comparison (1989-91) of blackleg on stubble of harvested crops around Prince Albert and Saskatoon will be the subject of another paper.

Two individuals going in different directions each walked an inverted V about 100 meters into and back out of a field, each randomly collecting 25 plants along the way. Plants were removed to the laboratory and stored at 1°C for 2 to 3 days until disease ratings were complete. Basal stem canker severity was assessed as "clean" (0), "slight" (1 = up to 33% of stem circumference girdled), "moderate" (2 = 34 - 66% of stem circumference girdled), or "severe" (3 = 67 - 100% girdling), and the presence of upper stem and leaf lesions recorded. Basal stem ratings (0 - 3) were converted to a percentage scale or disease severity index (8).

## Results and discussion

Virulent blackleg was very prevalent in Saskatchewan in 1991, occurring in 96% of the fields surveyed; basal stem cankers occurred in 93% of these fields. Reports were received early in the summer of severe stem canker developing in fields around Saskatoon. In August, many fields west of the city (C.D. 6b) had stem canker incidences of 100%. Crop district 9a had noticeably lower blackleg levels than crop district 6b (Table 1). The mean incidence of basal stem canker for all fields examined in crop districts 9a and 6b was 26% and 75.1%, respectively. Average severity ratings for infected plants (all fields) also were much lower in crop district 9a (Table 1). Although much of crop district 6 was surveyed 10 days later than district 9a, this probably did not have a large effect on the prevalence and incidence data. However, it may have influenced severity ratings somewhat.

In each of the four years crop district 9b was surveyed, LM-VIR was consistently more prevalent, and its overall incidence higher, in 9b(S) than in 9b(C), although the differences often were not large. The incidence of basal stem canker was higher in 9b(S) in three of the four years (Tables 1 and 2); this was also true of severity ratings. Only the 1991 severity data have been reported.

In 1991, the incidence of blackleg was considerably lower in fields in the northern part of crop district 8 than in those in the southern portion (Table 1). When the northern and southern parts were further subdivided, disease levels declined in the following order: crop districts 8(SW), 8(SE), 8(NW), and 8(NE) (Table 3). Very low blackleg incidences were found in crop district 8(NE), one of the areas where LM-VIR was found from 1975-1977 (7, 10). The average incidence of basal stem canker in seven *B. napus* fields there was only 2.9%. A comparison of data for southern and northern portions of crop district 8 for the years 1982 to 1985 also revealed consistently higher blackleg levels in the south, with one exception. In 1983, upper stem and leaf infections were more numerous in northern fields, but the reverse was true of basal stem cankers (Table 4).

In crop district 8(S), plant samples were collected from 11 fields of *B. napus* and from five fields of *B. rapa* in 1991; in crop district 8(N), 10 fields of *B. napus* and five of *B. rapa* were sampled. Each Brassica species had higher levels of basal stem infection in the southern part of crop district 8 (Table 5). The *B. napus* fields had a higher mean incidence of basal stem canker, in both the north and south, than the *B. rapa* fields. In crop district 9b, the frequency of basal stem infection on *B. napus* was slightly higher in 9b(C) than farther south, but only two fields were sampled in 9b(C). Stem canker incidence on *B. rapa* was higher in the south than in 9b(C) (Table 5).

An important consideration is whether blackleg incidence and severity in the north "catch up" to incidence and severity farther south by autumn. Reports of surveys conducted after swathing (4), and the author's post-harvest survey data indicate that the same general relationships observed in July are still apparent in October. In 1986, more than 90% of canola producers were using a combination seed treatment for seedling blight, blackleg, and flea beetle control (1). This likely slowed the spread of LM-VIR into more remote areas such as the Meadow Lake region of Saskatchewan and the Peace River region of Alberta. However, before seed treatment for blackleg became commonplace, LM-VIR may have entered northern areas repeatedly in infected seed. Higher rainfall levels and lower evaporation rates in the north probably adversely influenced blackleg levels by causing depletion of stubble-borne ascospore inoculum. The more favorable moisture regime in the north also permits more frequent field cultivation and more thorough burial of infected stubble.

## Acknowledgements

The assistance of Maurice Bahrey and Gordon Goplen is gratefully acknowledged.

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Table 1. Results of 1991 surveys of standing canola crops for virulent blackleg in three parts of Saskatchewan.

Crop district	No. fields	Prevalence%		Incidence%		Av. severity rating %	
		any infection	basal cankers	any infection	basal cankers	all plants	infected plants
9a	11	100.0	90.9	36.2	26.0	11.1	35.5
6a	15	100.0	100.0	75.5	65.7	52.1	62.3
6b	20	100.0	100.0	85.5	82.2	66.7	75.2
6	35	100.0	100.0	81.3	75.1	60.4	69.7
9b(C) <sup>1</sup>	11	81.8	81.8	24.2	16.2	5.9	26.5
9b(S)	12	100.0	91.7	30.2	20.8	9.6	32.3
9b	23	91.3	87.0	27.3	18.6	7.8	29.5
8 (N) <sup>2</sup>	16	93.8	87.5	16.9	11.8	4.7	23.5
8 (S)	15	100.0	93.3	45.0	32.0	15.7	41.8
8	31	96.8	90.3	30.5	22.6	10.0	32.4

<sup>1</sup> Subdivisions of crop district 9b: (C) = central part, (S) = southern part (see text).<sup>2</sup> Subdivisions of crop district 8: (N) = northern part, (S) = southern part (see text).

Table 2. Results of surveys of Saskatchewan rapeseed/canola crops for virulent blackleg, central and southern parts of crop district 9b, July, 1983-1985.

Year and sector <sup>1</sup>	No. of fields	% fields with plants infected		Mean % plants per field infected		
		on any part	at stem base	on any part	at stem base	at stem base
				all fields	infested fields	infested fields
1983 (C)	12	58	8	3	5	4
1983 (S)	12	92	50	27	30	11
1984 (C)	13	39	15	10	25	10
1984 (S)	15	60	20	16	27	27
1985 (C)	12	25	25	4	16	13
1985 (S)	18	50	39	7	14	11

<sup>1</sup> See text for descriptions of routes travelled.

Table 3. A comparison of the prevalence, incidence, and severity of virulent blackleg in four sectors of Saskatchewan crop district 8, July, 1991.

Sector <sup>1</sup>	No. of fields	% fields with plants infected		Mean % of plants per field infected		Basal stem canker severity (0 - 100)	
		on any part	at stem base	on any part	at stem base	all fields all plants	all fields infested plants
8(NE)	9	89	78	6	4	1	18
8(NW)	7	100	100	32	27	9	31
8(SE)	7	100	86	40	31	13	32
8(SW)	8	100	100	50	33	18	51

<sup>1</sup> See text for descriptions of routes travelled.

Table 4. Results of surveys of Saskatchewan rapeseed/canola crops for virulent blackleg, northern and southern parts of crop district 8, July, 1982-1985.

Year and sector <sup>1</sup>	No. of fields	% fields with plants infected		Mean % of plants per field infected		
		on any part	at stem base	on any part	infected fields	at stem base
				all fields	infected fields	infested fields
1982 (N)	11	82	0	12	15	0
1982 (S)	15	93	87	19	21	3
1983 (N)	16	56	0	15	27	0
1983 (S)	15	67	20	9	14	7
1984 (N)	17	59	6	7	12	16
1984 (S)	9	78	44	16	21	23
1985 (N)	19	58	21	5	8	7
1985 (S)	12	92	67	14	15	10

<sup>1</sup> See text for sector description.

Table 5. Incidence of basal stem infections of blackleg in standing crops of *Brassica rapa* and *B. napus* in northern and southern parts of crop district 8 and in central and southern parts of crop district 9b in 1991.

C.D. and sector <sup>1</sup>	Species <sup>2</sup>	No. fields	Mean incidence of basal stem canker (%)
8 (N)	<i>B. rapa</i>	5	12.1
8 (N)	<i>B. napus</i>	11	14.4
8 (S)	<i>B. rapa</i>	5	24.7
8 (S)	<i>B. napus</i>	10	35.7
9b (C)	<i>B. rapa</i>	9	13.9
9b (C)	<i>B. napus</i>	2	26.7
9b (S)	<i>B. rapa</i>	6	17.3
9b (S)	<i>B. napus</i>	6	24.3

<sup>1</sup> See text for descriptions of sectors.

<sup>2</sup> *B. rapa* = *B. campestris*.





# Post-harvest surveys of blackleg on stubble of rapeseed/canola crops in Saskatchewan, 1981-1991

G.A. Petrie<sup>1</sup>

In an area northeast of Prince Albert, Saskatchewan, the average incidence of the virulent strain of *Leptosphaeria maculans* (blackleg) on stubble in post-harvest surveys of rapeseed/canola crops increased from 14.5% in 1989 to 52.6% in 1991. Over the same three year period its average incidence west of Saskatoon increased from 59.4% to 92.6%. In both areas there was also a substantial increase in the frequency of fields with over 50% blackleg incidence. In the relatively remote areas west of Prince Albert and near Meadow Lake, the average incidence of virulent blackleg in 1990 was 25 and 21%, respectively. Since virulent blackleg was initially found in Saskatchewan in 1975, its prevalence and incidence have increased at a slower rate in northern growing areas than in those farther south.

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Dans une région au nord-est de Prince Albert, en Saskatchewan, l'incidence moyenne de la souche virulente de *Leptosphaeria maculans* (jambe noire) sur le chaume, lors des relevés en poste-récolte des cultures de colza/canola, a augmenté de 14,5 % en 1989 à 52,6 % en 1991. Durant cette même période de trois ans, son incidence moyenne à l'ouest de Saskatoon a augmenté de 59,4 % à 92,6 %. Dans les deux régions, on a remarqué que beaucoup plus de champs présentaient une incidence de la jambe noire à un taux de plus de 50 %. Dans les régions éloignées à l'ouest de Prince Albert et près de Meadow Lake, l'incidence moyenne de la jambe noire virulente en 1990 était de 25 et 21 %. Depuis que la jambe noire virulente a été trouvée pour la première fois en Saskatchewan en 1975, sa prédominance et son incidence ont augmenté beaucoup plus lentement dans les régions de croissance au nord que dans celles plus éloignées au sud.

## Introduction

The weakly virulent Puget Sound strain (LM-PS) of *Leptosphaeria maculans* (Desm.) Ces. & De Not. [anamorph = *Phoma lingam* (Tode:Fr.) Desm.] (14) was first found in Saskatchewan in 1957 in seed samples of rapeseed [*Brassica napus* L. and *B. rapa* L. (= *B. campestris* L.)] (16). Its occurrence on adult plants in commercial fields was confirmed four years later (17). Apart from one field north of North Battleford in crop district (C.D.) 9b (18), LM-PS was restricted to the southern half of crop district 8 from 1963 to 1967. However, during these five years its prevalence increased from less than 25% to 83% of the fields examined (10, 11, 12). By 1967, LM-PS had spread northeast to Melfort and by 1969, farther northeast to Aylsham (C.D. 8a) and into the far northwest near Meadow Lake (C.D. 9b). It was also detected on seed from south-central Manitoba and central Alberta (13).

A virulent strain of *L. maculans* (LM-VIR) was discovered in central Saskatchewan on stubble of the 1975 rapeseed crop (4, 6). Its subsequent spread in western Canada was reminiscent of that of LM-PS in the years following its discovery.

By 1982, LM-VIR had become a serious problem in crop district 6 and 8b (7). From 1984 to 1990, it was more prevalent in crop districts 6 and 8b than farther north and east in crop districts 8a and 9a (1, 3, 8). LM-VIR was first observed in the Meadow Lake area (C.D. 9b) in 1985 (R. Gugel, pers. comm.), although it had been present farther south in the North Battleford area in 1979 (9). This paper compares the results of post-harvest blackleg surveys conducted in Saskatchewan between 1981 and 1991 around Prince Albert (C.D. 9a) and Saskatoon (C.D. 6 & 8b), and includes the results of a 1990 survey of the Meadow Lake area (C.D. 9b).

## Material and methods

Crops of rapeseed/canola were sampled after harvest by pulling a stubble plant every five paces while walking through a field until at least 50 were obtained. Blackleg incidence was recorded, isolations made on V8 agar (5) and the proportions of LM-VIR and LM-PS present determined (4).

The following five areas, also illustrated in Figure 1, were surveyed in two or more years between 1989 and 1991: (1) Saskatoon west (STOON-W.): that part of crop district 6b consisting of the rural municipalities (R.M.) 345, 403, 404, and the western part of R.M. 344; (2) Saskatoon east (STOON-E): the area where crop district 6a, 6b and 8b converge, consisting of R.M. 343, 372, 373, and the eastern

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part of R.M. 344; (3) Prince Albert northeast (P.A.-NE): that part of crop district 9a consisting of R.M. 488, 490, 491, and 520; (4) Prince Albert south (P.A.-S): that part of crop district 9a between the North and South Saskatchewan Rivers (R.M. 461); and (5) Prince Albert west (P.A.-W): that part of crop district 9a consisting of R.M. 493, 494, and 496. The part of the Meadow Lake area surveyed in 1990 was in R.M. 588 in crop district 9b (Fig. 1).

## Results and discussion

Results of a 1981 survey from Wakaw (C.D. 8b) north to Meath Park (C.D. 9a) are in Table 1. Severe infections of LM-VIR occurred that year around Domremy and Wakaw. Virulent blackleg was seen at trace levels farther north near St. Louis and Birch Hills, but was not detected near Prince Albert and Meath Park. Table 2 contains the results of a 1985 survey that serves as an early Comparison of the extent of blackleg infestation in the area around Prince Albert with that west of Saskatoon. The incidences of infection were similar until the only heavily infected field near Shellbrook in R.M. 493 (Fig. 1), was removed from the data for crop district 9a. Then blackleg incidence in crop district 9a was much less than in crop district 6b. Two of the 12 remaining fields in crop district 9a were lightly infected.

Results of the 1989, 1990 and 1991 surveys are in Table 3. In Prince Albert north east, the mean incidence of LM-VIR increased from 14.5% in 1989 to 32.5% in 1990, and to 52.6% in 1991. The corresponding values for Saskatoon west were 59.4%, 60.5%, and 92.6%, and for Saskatoon east, 55.7%, 36.9%, and 94.7%. Prince Albert south had relatively high incidences of LM-VIR of 60.6% in 1990 and 73.3% in 1991. The drop in LM-VIR in Saskatoon east to 36.9% in 1990 was accompanied by an increase in LM-PS (Table 3). Prince Albert west was surveyed extensively only in 1990. The mean incidence of LM-VIR in Prince Albert west that year was only 25%, but the distribution of infestation in the area was suggestive of a spread north and west from the Shellbrook area, which had a severe localized infestation in 1985. Three fields near Debden, the northernmost locality surveyed in R.M. 494, had 0.0 – 3.0% LM-VIR and 14.6 – 51.1% LM-PS. Fields around Shellbrook were heavily infested by LM-VIR, but infections in the three fields farthest west (R.M. 496) ranged from 5.9 – 8.3% LM-VIR. The incidence of LM-PS in these three fields ranged from 5.0 – 66.3%.

Between 1989 and 1991, a steady increase in incidence levels of both strains of *L. maculans* occurred in Prince Albert northeast (Table 4). By 1991, all of the fields had 76 – 100% of the plants infected by either LM-VIR or LM-PS. In 1989, 6.0% of the fields in Prince Albert northeast had an LM-VIR incidence of over 50%; by 1991, this had increased to 47.7% of the fields. There also was a dramatic increase

in the incidence of LM-VIR in fields near Saskatoon between 1990 and 1991. In the 1990 survey, 65.2% of the fields near Meadow Lake had 25% or less LM-VIR (Table 4), and 56.5% had 25% or less of any blackleg strain. Upon isolation, an average 65.5% of the isolates were of the virulent type. The mean incidence of LM-VIR in surveyed crops in the Meadow Lake area was only 21.0% (Table 3).

There is persuasive evidence that LM-VIR and LM-PS are different species rather than variants of a single species (15). The two appear to be in direct competition. During the last ten years, in areas in which LM-VIR has become prevalent, the incidence of LM-PS has remained at a low level or declined. However, LM-PS still predominates in many northern fields and may suddenly increase in areas where levels of LM-VIR decline sharply, as in Saskatoon east in 1990. Conditions required by the two strains for sexual reproduction appear to differ (2), which may explain the preponderance of one or the other in different locations.

Survey data collected in Saskatchewan since 1981 clearly indicate a much slower increase in prevalence and incidence of virulent blackleg in a south to north direction than in an east-west direction. This is an apparent anomaly, as the moisture regime around Saskatoon would appear to be less favorable for blackleg development than that northeast of Prince Albert. Ascospores are the most important inoculum source for *L. maculans*. Ascocarps develop on stubble from the previous year's crop and, in the Saskatoon area, continue to produce spores for several years. Dry surface soil conditions, such as those that have prevailed around Saskatoon for a number of years, tend to conserve blackleg inoculum. Frequent showers and prolonged periods of high moisture under the crop canopy are an immediate stimulus to ascospore production and release, but the ultimate result is an accelerated decline in long-term sporulation potential (Petrie, unpublished). There is evidence that old canola stubble residue does not persist as long in northern areas as it does farther south. Moisture conditions farther north permit growers to cultivate summerfallow fields more frequently, accelerating the breakdown of infected canola tap-roots.

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Table 1. Results of 1981 blackleg survey of rapeseed/canola stubble crops in Saskatchewan crop districts 9a and 8b.

Subdivision of area surveyed & localities	Proportions of blackleg strains isolated (%)		Incidence of blackleg strains on stems in surveyed fields (%)	
	LM-VIR <sup>1</sup>	LM-PS <sup>1</sup>	LM-VIR	LM-PS
North (C.D. 9a)				
E. Meath Park	0.0	100.0	0.0	50.0
S. Meath Park	0.0	100.0	0.0	46.0
N.E. Prince Albert	0.0	100.0	0.0	60.0
S. Prince Albert	0.0	100.0	0.0	2.0
N. St. Louis	0.5	99.5	0.2	38.0
Averages (North)	0.1	99.9	0.04	39.2
Central (C.D. 8b)				
N. Birch Hills	33.3	66.7	4.0	8.0
S.E. Birch Hills	0.5	99.5	0.1	20.0
N. Hoey	0.5	99.5	0.1	10.0
Averages (Central)	11.4	88.6	1.4	12.7
South (C.D. 8b)				
Crystal Springs	90.3	9.7	87.3	9.4
Domremy	78.6	21.4	76.0	20.7
Wakaw	63.9	36.1	46.0	26.0
Averages (South)	77.6	22.4	69.8	18.7

<sup>1</sup> LM-VIR = virulent strain, LM-PS = weakly virulent Puget Sound strain.

Table 2. Results of 1985 blackleg survey in rapeseed/canola stubble crops in Saskatchewan crop district 9a near Prince Albert and crop district 6b near Saskatoon.

Sector and crop district	No. of fields	% fields with plants infected		Mean % plants/field infected		
		on any part	at stem base	on any part		at stem base
				all fields	infested fields	infested fields
North (9a)	13	23	23	7	29	19
	121	17	17	2	10	6
South (6b)	26	62	58	15	24	17

<sup>1</sup> Heavily infested Shellbrook field atypical of the district has been omitted (see text).

Table 3. Results of blackleg surveys in canola stubble crops near Saskatoon, Prince Albert and Meadow Lake, Saskatchewan, 1989-1991.

Year Surveyed areas' No. fields ( )	Proportions of blackleg strains isolated (%)		Mean incidence of blackleg on stems in surveyed fields (%)	
	LM-VIR <sup>2</sup>	LM-PS <sup>2</sup>	all strains	LM-VIR <sup>2</sup>
<b>1989</b>				
STOON-W. (11)	82.1	17.7	72.3	59.4
STOON-E. (9)	84.5	15.1	65.9	55.7
P.A.-W. (3)	60.6	39.5	69.3	42.0
P.A.-NE. (13)	34.1	65.6	42.5	14.5
<b>1990</b>				
STOON-W. (9)	88.0	12.0	68.7	60.5
STOON-E. (13)	64.2	35.6	57.5	36.9
P.A.-W. (19)	58.2	42.4	42.9	25.0
P.A.-NE. (7)	44.1	55.6	73.6	32.5
P.A.-S. (13)	90.2	9.4	67.0	60.6
MEADOW L. (23)	65.5	34.2	32.0	21.0
<b>1991</b>				
STOON-W. (22)	93.6	6.4	98.9	92.6
STOON-E. (18)	95.8	7.6	98.8	94.7
P.A.-NE. (15)	54.1	45.9	97.3	52.6
P.A.-S. (10)	73.8	26.2	99.3	73.3

<sup>1</sup> See text for description of survey areas.

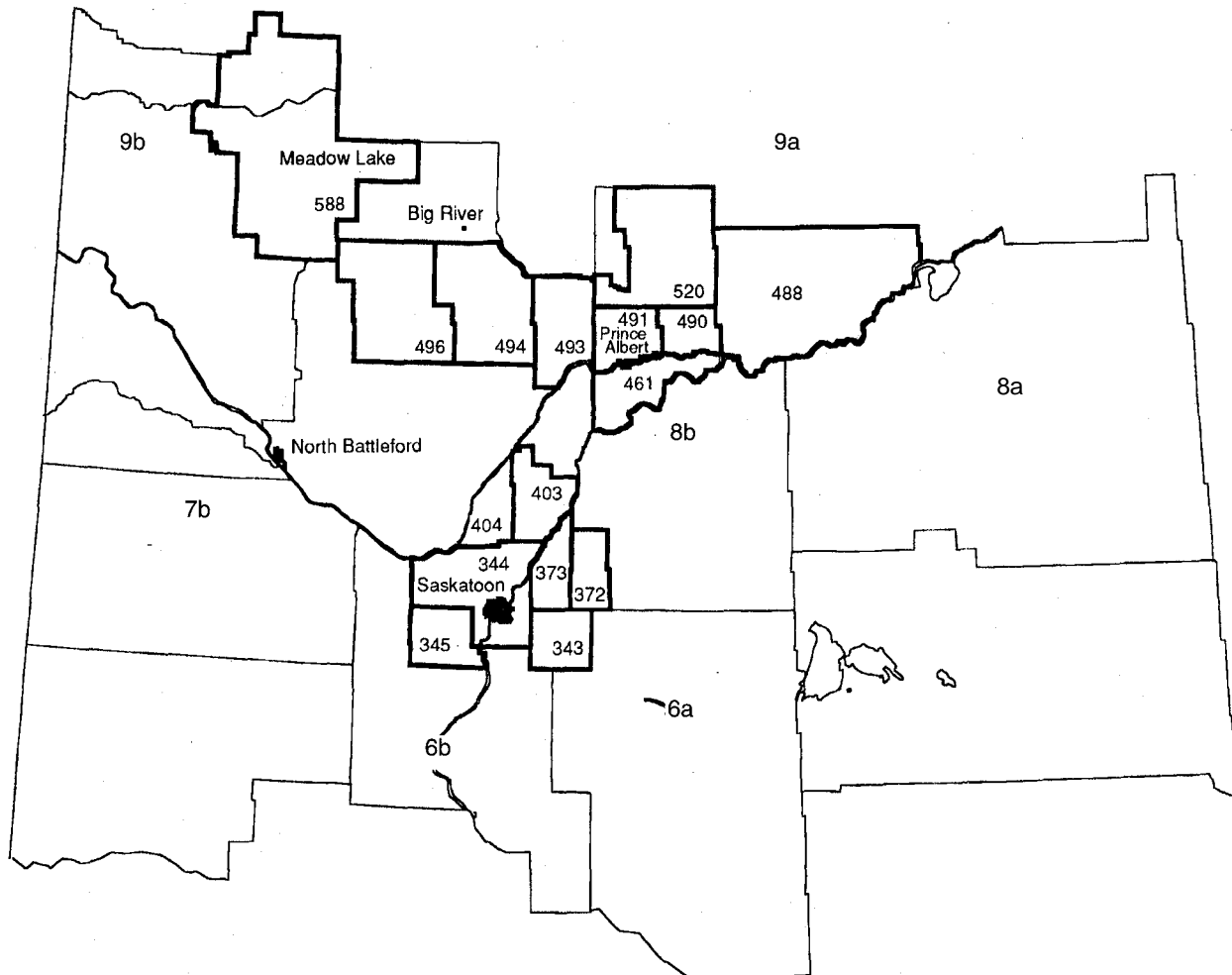
<sup>2</sup> LM-VIR = virulent strain, LM-PS = weakly virulent Puget Sound strain of *Leptosphaeria maculans*.

Table 4. Results of blackleg surveys in canola stubble crops near Saskatoon, Prince Albert and Meadow Lake, Saskatchewan, 1989-1991.

Year, strains, areas <sup>1</sup> , and No. fields ( )	Percent of fields in five incidence categories				
	0-10%	11-25%	26-50%	51-75%	76-100%
<u>1989, all strains</u>					
P.A.-NE & W (16)	12.5	18.8	25.0	31.3	12.5
STOON-W & E (20)	0.0	5.0	15.0	25.0	55.0
<u>1989, virulent strain</u>					
P.A.-NE & W (16)	50.0	31.3	12.5	0.0	6.3
STOON-W & E (20)	0.0	15.0	30.0	20.0	35.0
<u>1990, all strains</u>					
P.A.-W (19)	5.3	21.1	42.1	15.8	15.8
P.A.-NE (7)	0.0	0.0	28.6	14.3	57.1
STOON-W & E (22)	0.0	4.6	31.8	27.3	36.4
MEADOW L (23)	43.5	13.0	17.4	17.4	8.7
<u>1990, virulent strain</u>					
P.A.-W (19)	42.1	10.5	36.8	0.0	10.5
P.A.-NE (7)	57.1	0.0	14.3	0.0	28.6
STOON-W & E (22)	0.0	27.3	36.4	18.2	18.2
MEADOW L (23)	47.8	17.4	8.7	21.7	4.4
<u>1991, all strains</u>					
P.A.-NE (15)	0.0	0.0	0.0	0.0	100.0
STOON-W & E (40)	0.0	0.0	0.0	0.0	100.0
<u>1991, virulent strain</u>					
P.A.-NE (15)	0.0	20.0	33.3	13.4	33.3
STOON-W & E (40)	0.0	0.0	0.0	7.5	92.5

<sup>1</sup> See text for description of survey areas.

Figure 1. Parts of Saskatchewan where post-harvest surveys for blackleg were conducted in rapeseed/canola crops from 1989 to 1991. Larger units numbered 6a to 9b are crop districts and smaller areas in bolder outline numbered 343 to 588 are those rural municipalities surveyed.



# Survey for seed-borne diseases on weed species from screening samples obtained from seed cleaning plants across Canada in 1987/88

K. Mortensen and M.M. Molloy<sup>1</sup>

In search for potential biological control agents for weeds, requests for samples of Screenings from seed cleaning were sent out to seed cleaning plants across Canada in order to analyze for seed-borne diseases of weeds. Seven samples of screenings were received: two from Alberta, and one each from British Columbia, Saskatchewan, Manitoba, Ontario, and Prince Edward Island. A large percentage of the seeds (varying from 10 to 80%) developed fungal growth, of which very few affected germinated seedlings. Pathogenic fungi were isolated from diseased seedlings of wild oats: *Drechslera avenacea*, cow cockle: *Alternaria alternata*, stinkweed: *Alternaria raphani*, green foxtail: *Bipolaris sorokiniana*, wild buckwheat: *Botrytis* sp., from western Canada, and from a grass sp.: *B. sorokiniana*, and red clover: *Colletotrichum trifolii*, from eastern Canada. These results show that surveys for weed diseases can be conducted from samples of screenings submitted by cooperators. It is a quick and a relatively inexpensive method for weed disease surveying. However, as not all weed diseases are seed-borne, it cannot substitute surveys during the growing season.

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Des installations de nettoyage de semences, situées un peu partout au Canada, ont reçu des demandes pour faire analyser des échantillons de tamisage. Les demandeurs voulaient faire une analyse des pathogènes transmis par les graines afin de trouver des agents biologiques de lutte contre les mauvaises herbes. Sept échantillons de tamisage ont été reçus : deux de l'Alberta, et un pour chaque province dont la Colombie-Britannique, la Saskatchewan, le Manitoba, l'Ontario, et l'Île-du-Prince-Édouard. La croissance de champignons a été décelée sur un grand pourcentage des graines (de 10 à 80 %), mais très peu de champignons ont affecté les plantules émergées. Des champignons pathogènes ont été isolés à partir de plantules malades de la folle avoine : *Drechslera avenacea*, de la saponaire des vaches : *Alternaria alternata*, du tabouret des champs : *Alternaria raphani*, de la setaire verte : *Bipolaris sorokiniana*, de la renouée liseron : *Botrytis* sp., provenant tous de l'ouest du Canada et à partir d'une graminée : *B. sorokiniana*, et du trèfle rouge : *Colletotrichum trifolii*, provenant de l'est du Canada. Ces résultats montrent que des relevés pour les maladies de mauvaises herbes peuvent être menés à partir d'échantillons obtenus par des collaborateurs. C'est une méthode rapide et peu coûteuse d'inventorier les maladies des mauvaises herbes. Quoi qu'il en soit, puisque toutes les maladies de mauvaises herbes ne sont pas transmises par les graines, cette méthode ne peut remplacer des relevés menés pendant la saison de végétation.

## Introduction

Biological control of weeds with plant pathogens has received much attention in recent years (3,4,21,23) because of the pressures to decrease our dependence on synthetic herbicides. At present two bioherbicides are registered in the United States, *Colletotrichum gloeosporioides* (Penz.) Sacc. f. sp. *aeschynomene* 'Collego' for control of northern jointvetch [*Aeschynomene virginica* (L.) B.S.P.] and *Phytophthora palmivora* (Butler) Butler 'De Vine' for control of strangler vine (*Morrenia odorata* Lindl.) (20). In 1992, *C. gloeosporioides* f. sp. *malvae* was the first bioherbicide

registered in Canada under the tradename 'BioMal' for the control of round-leaved mallow (*Malva pusilla* Sm.) (11). The fungus in 'BioMal' was discovered as a seedling blight originating from infected round-leaved mallow seed (15).

Explorations for new bioherbicide agents are an integral part of the program on biological control of weeds with plant pathogens at the Agriculture & Agri-Food Canada, Research Station in Regina. Surveying for diseases on weeds can be done during the growing season, but this is very time consuming and expensive. As many plant diseases are seed-borne, analyzing weed seeds for disease causing organisms, might be an effective method of identifying organisms that parasitize weeds. In addition to being quicker and less expensive, representative samples of weed seeds from across Canada would provide a broader sampling base than one derived from surveys.

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The present work describes the results from an examination of seed samples from seed cleaning plants across Canada, the isolation of disease causing organisms from seedlings originating from weed seeds in the screening samples, and subsequently testing such organisms for their potential as bio-herbicide agents on the weeds from which they originated.

## Materials and methods

Requests for screening samples were sent out to representative seed cleaning plants selected from those listed in Inspection Memorandum 1-2-57,86-08-07, Agriculture and Agri-Food Canada, Food Production and Inspection Branch, Seed Section. The requests were made in the fall of 1987 to three seed cleaning plants in British Columbia, three in Alberta, four in Saskatchewan, five in Manitoba, four in Ontario, four in Quebec, and one in each of New Brunswick, Prince Edward Island, and Nova Scotia. Screening samples from seed cleaning procedures were received from seven cooperating seed plants located at: Dawson Creek, British Columbia (received 23 Feb. 1988); Barrhead, Alberta (received 3 Mar. 1988); Camrose, Alberta (received 27 Jan. 1988); Wiseton, Saskatchewan (received 29 Dec. 1987); Ste. Rose du Lac, Manitoba (received 16 Mar. 1988); Belleville, Ontario (received 21 Dec. 1987); and Montague, Prince Edward Island (received 16 May 1988).

The screening samples were sorted into different sizes of seeds using screens with grid sizes, of 2.34 mm, 2.73 mm, 3.12 mm (6/64", 7/64", 8/64", respectively) and a pan sample. From each of the screens, 100 seeds, if present, of the most common species were selected for tests.

Seeds were placed on moist filter paper (MFP) (Whatman No. 3, 9 mm diam) in a petri dish and incubated for 4 to 7 days with a 12 h light period provided by fluorescent light ( $28 \mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$ ) at  $24 \pm 0.5^\circ\text{C}$  and a 12 h dark period at  $21 \pm 0.5^\circ\text{C}$ . Seeds that germinated after four days were planted in autoclaved soil:peat moss:vermiculite (3:2:1) in 15 cm pots, covered gently with a thin layer of the same soil mixture and placed on greenhouse benches at  $23 \pm 4^\circ\text{C}$  with ambient lighting extended to a 16 h photoperiod with fluorescent and incandescent light ( $280 \mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$ ). After seven days of incubation, the filter paper with the germinated or non-germinated seeds was placed in pots with soil as above and placed on the greenhouse benches. Pan samples from the screening samples were planted in soil in 34 cm x 48 cm steel flats and placed on the greenhouse benches. Plants were kept for one month and monitored daily for disease development. Seedlings that developed disease symptoms were surface sterilized (0.6% sodium hypochlorite for 1 min), placed on potato dextrose agar (PDA) and on MFP and incubated as described above. The fungal pathogens were isolated and increased on PDA. Spore suspensions of pure isolates were used to inoculate

seedlings of the plant species from which they originated. Inoculation was done by spraying the spore suspensions until runoff using an air brush [Paasche Airbrush (Canada) Ltd., Type H-5]. The inoculated plants were placed in a dew chamber (Percival, Model E-54) at  $18 \pm 0.5^\circ\text{C}$  for 24 h in the dark, then returned to the greenhouse benches, and misted with water daily to maintain high humidity. If disease symptoms were not observed four weeks after inoculation, the fungal cultures were regarded as saprophytes and discarded. All material used in these tests was autoclaved before being discarded to prevent escape of weed seeds or diseases.

## Results and discussion

The screening samples received contained representative crop and weed seeds from an area near the seed cleaning plant. Five of the screening samples, Dawson Creek, British Columbia; Barrhead, Alberta; Camrose, Alberta; Ste. Rose du Lac, Manitoba; and Montague, Prince Edward Island; contained wheat seed; the Wiseton, Saskatchewan, screening sample had lentil seed; and the Belleville, Ontario, sample had rye grass (*Lolium* sp.) seed. The weed species varied among the screening samples. Table 1 shows the weeds identified in the western Canadian samples from selected seed samples and Table 2 shows weeds from the two eastern Canadian selected seed samples. The most common weeds from western Canada were wild oats (*Avena fatua* L.), wild buckwheat (*Polygonum convolvulus* L.), lamb's-quarters (*Chenopodium album* L.), green foxtail (*Setaria viridis* (L.) Beauv.), and stinkweed (*Thlaspi arvense* L.). Mustard spp. (not identified to species) were observed in pan samples planted in flats under greenhouse conditions from all western Canadian locations. Green smartweed (*Polygonum scabrum* Moench.), Russian thistle (*Salsola pestifer* Nels.), hemp nettle (*Galeopsis tetrahit* L.), lady's-thumb (*Polygonum persicaria* L.), cow cockle (*Saponaria vaccaria* L.), white cockle (*Lychnis alba* Mill.), and Russian pigweed, (*Axyris amaranthoides* L.) were observed from one or two locations in seeds samples, as well as in the pan samples. Chickweed (*Stellaria media* (L.) Vill.) was only observed in the pan sample from Barrhead (Table 1). From the Ontario screening sample, ragweed (*Ambrosia* spp.), a grass (*Lolium* sp. ?), and red clover (*Trifolium pratense* L.) occurred, red clover and grasses (not identified) also occurred in the pan sample. From the Prince Edward Island screening sample, grass (not identified), wild radish (*Raphanus raphanistrum* L.), wild buckwheat, barnyard grass (*Echinochloa crusgalli* (L.) Beauv.) and *Convolvulus* sp. (not identified to species) were most common, and corn spurry (*Spergula arvensis* L.), lamb's-quarter and *Polygonum* sp. (not identified to species) grew from the pan samples (Table 2).



Fungal growth developed on the coats of a large percentage of the seeds from most samples placed on MFP (Tables 1 and 2). Fungi growing on the seed coats and causing disease symptoms on the seedlings, were identified and used in pathogenicity tests. *Alternaria* spp. were most prominent on the seed coats. However, spores of *Bipolaris sorokiniana* (Sacc. in Sorok.) Shoem. occurred on some seeds from all of the weed species in the Prince Edward Island screening sample (Table 3). Since *B. sorokiniana* is a known pathogen on graminaceous species across Canada (5,12) and it occurred so frequently on seeds of non-graminaceous species (Table 3), one isolate from each weed species was inoculated on plants from four of the Prince Edward Island weed species in order to see if they were host specific. Pathogenicity did not vary significantly among the five isolates. They were most pathogenic on the unidentified grass sp., less so on barnyard grass, and only slight or no symptoms were observed on the wild buckwheat and wild radish (Table 3). *B. sorokiniana* was isolated from brown spots on cotyledons and first leaves of two grass seedlings from the Prince Edward Island location (Table 7), but the seedlings outgrew the disease. Symptoms were not observed on any of the other seedlings from weed species where *B. sorokiniana* spores occurred. Although the *B. sorokiniana* isolates originated from dicotyledonous weed seeds, they were not significantly different from isolates originating from graminaceous species.

Germination of weed seeds from the western Canadian samples was good with averages of 65% or higher for green foxtail, lamb's-quarter, stinkweed, cow cockle, and green smartweed; fair with averages of about 45% or slightly above for wild oats, wild buckwheat, lady's-thumb, and Russian pigweed; and poor with less than 25% germination for white cockle, Russian thistle, and hemp nettle (Table 4). Germination from the eastern Canadian screening samples were low, less than 16% for barnyard grass, grass sp. (not identified), wild radish, wild buckwheat, and ragweed; somewhat better (36%) for *Polygonum* sp. (not identified to species); while the red clover sample from Ontario had 71% germination (Table 5).

Disease symptoms developed on about 13% of wild oats seedlings from the five western samples (Table 4), which is 7% of all seeds tested. The symptoms on wild oats seedlings from these locations were caused by *Drechslera avenacea* (Curtis ex Cooke) Shoem. (Table 6). This is in agreement with other data showing that *D. avenacea* was isolated from 7.5% of wild oats seeds from 59 locations in the prairie provinces (unpublished data). Avenacea leaf blotch or leaf stripe (*D. avenacea*) occurs commonly on cultivated and wild oats in all provinces (5,12, 19). The seedling-blight stage has rarely been observed on cultivated oats under natural conditions (6). The present results showed that wild oat seedlings originating from infected

seeds can become severely infected under greenhouse conditions, but often only a streak was observed on the coleoptile and the first leaves and the plants remained alive. Under natural conditions these lesions would be sufficient to allow for secondary spread by spores and cause the common avenacea blotch often observed on upper leaves later in the season.

Seedling blight developed on 30% of the cow cockle seedlings from the Saskatchewan sample (Table 4) which is 21% of total seeds tested. All of the observed symptoms on cow cockle seedlings were caused by an *Alternaria* sp. Earlier observations indicated that seedling blight caused by *Alternaria alternata* (Fr.) Keissl. affected up to 65% of seedlings from a Regina seed lot of cow cockle (unpublished data). *A. sappanaria* (Pk.) Neerg. has been reported from cow cockle from Manitoba (2), from several states in the United States (7), and from western Europe (17). Another species, *A. dianthi* Stevens & Hall, was reported on cow cockle from Montana (13). Perhaps, several *Alternaria* species may attack cow cockle.

Disease symptoms developed on 7.9% of stinkweed seedlings (Table 4) which represents about 6% of all seeds tested. A pathogenic *Alternaria* sp. was isolated from five of these diseased seedlings, which were all from the Saskatchewan screening sample (100 seeds) (Table 6). This *Alternaria* sp. was submitted to National Identification Service, Ottawa, and identified as *Alternaria raphani* Groves & Skolko (Daom NO. 21 1978). *A. raphani* has previously been observed on stinkweed (18).

Disease symptoms developed on 1.7% of green foxtail seedlings (Table 4). The causal agent *B. sorokiniana* was isolated from two seedlings in the Manitoba sample (Table 6). Both isolates were rated as weakly pathogenic on green foxtail. Although inoculated tissues showed leaf spots, the plants outgrew the disease. In another study, *B. sorokiniana* was isolated from green foxtail at Regina, and a spore suspension was inoculated back onto green foxtail. Under optimum conditions these plants developed leaf spots but outgrew the disease. Under field conditions very little or no effect was observed when inoculated with this isolate of *B. sorokiniana* (unpublished data). *B. sorokiniana* has previously been isolated from crowns of green foxtail from Saskatchewan but to a much lesser extent than from cereal crops (10). These results indicate that *B. sorokiniana* has little potential as a biological control agent for green foxtail.

Disease symptoms were observed on wild buckwheat seedlings from three of the western locations, and *Botrytis* sp. was isolated from two seedlings originating near Barrhead, Alberta (Table 4). Both of these isolates caused slight leaf spotting when wild buckwheat was inoculated with these isolates. The symptoms were not regarded as suffi-

ciently pathogenic to warrant further study. *Rhizoctonia* sp. was isolated from diseased seedlings from two locations (Camrose, Alberta and Ste. Rose du Lac, Manitoba), but neither isolate was pathogenic.

The cause of the other seedling symptoms (Table 4) was not diagnosed. Species of *Alternaria*, *Pythium*, *Fusarium*, and *Phoma* (not identified to species) were isolated from the diseased seedlings, but in pathogenicity tests the injury was negligible. Perhaps a combination of stress in the greenhouse, including the activity of fungus gnats (Mycetophilidae) and these saprophytic fungi resulted in death of small seedlings.

From the two eastern Canadian locations, only two grass seedlings from the Prince Edward Island location showed disease symptoms which were attributed to *B. sorokiniana* and one seedling of red clover from the Ontario location showed symptoms (Table 5). A *Colletotrichum* sp. isolated from the red clover seedling (Table 7) caused typical anthracnose symptoms when red clover seedlings were inoculated with this isolate. This fungus was submitted to National Identification Service, Ottawa, and was identified as *Colletotrichum trifolii* Bain. Anthracnose of red clover caused by *C. trifolii* is a severe disease of red clover in the southern and mid-Atlantic United States, hence the name southern anthracnose (1,8,14). It has been recorded as far north as southern Canada, but is of little importance in the northern clover areas (9,24). Because *Colletotrichum* spp. have shown good potential as mycoherbicide agents (11,15,20,22) further studies were conducted for comparison with other *Colletotrichum* spp., and to determine its potential for biological control of black medick (*Medicago lupulina* L.), a serious weed in Canada (16).

The seven screening samples we received for this study during 1987-88 are not sufficient to give any accurate information on occurrence of seed-borne diseases of weeds. However, the present results show that surveys for diseases of weeds can be conducted from screening samples submitted by cooperators. This is a relatively inexpensive way of conducting weed disease surveys as it does not involve travel and accommodation expenses. A large geographical area can be covered and it can be done outside the growing season, at less busy periods of the year. This survey method works well for diseases that are seed-borne. However, it will not detect all diseases and, therefore, cannot substitute disease surveys during the growing season.

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Table 1. Weeds seeds identified in screening samples from western Canadian provinces and occurrence of fungi on seed coat when plated out on moist filter paper.

Weed species	Locations				
	Dawson Creek BC	Barrhead AB	Camrose AB	Wiseton SK	Ste. Rose du Lac MB
Wild oats	100*/46**	175116	43/81	75/77	71/31
Wild buckwheat	300/10	300147	300166	225170	200139
Lamb's-quarter	200111	(+)	100118	100181	
Green foxtail	(+)		100/19	(+)	200113
Stinkweed	10019	(+)	25/32	10116	
Mustard spp.	(+)	(+)	(+)	(+)	(+)
Green smartweed		100110		(+)	10018
Russian thistle		(+)		100184	
Hemp nettle		100162	50184		
Lady's-thumb			110128		
White cockle		100147			
Cow cockle				10014	
Chickweed		(+)			
Russian pigweed			201110		

\* indicates number of seeds selected from screening sample, plated on moist filter paper (MFP) then planted in pots,

\*\* indicates percent of seeds with fungi (not identified) on seed coats, and

(+) indicates that seeds were not selected, but that seedlings developed from pan sample, plated on MFP or in flats under greenhouse condition.

Table 2. Weeds seeds identified in screening samples from eastern Canada and occurrence of fungi on seed coat when plated out on moist filter paper.

Weed species	Locations	
	Belleville ON	Montague P.E.I.
Ragweed (not identif.)	225*/100**	
Grass (Lolium sp.)	3001100	
Grass (not identified)	(+)	100155
Red clover	250199	
Wild radish		100193
Wild buckwheat		300186
Corn spurry ?		(+)
Barnyard grass		100176
lambs-quarter		(+)
<i>Polygonum</i> sp.		(+)
<i>Convolvulus</i> sp.		3001100

\* indicates number of seeds selected from screening sample, plated on moist filter paper (MFP) then planted in pots,

\*\* indicates percent of seeds with fungi on seed coat (not identified) and

(+) indicates that seeds were not selected but that seedlings developed from pan sample, plated on MFP or in flats under greenhouse condition.

Table 3. Effect of *Bipolaris sorokiniana* isolates, originating from seed coats of five weeds, on plants from four of these weeds. Seed from screening sample received from Montague, Prince Edward Island.

Weed species	% seeds with fungus	Effect <sup>1</sup> of <i>B. sorokiniana</i> isolated from					Mean rating
		WB	G.sp.	WR	BG	C.sp.	
Wild buckwheat (WB)	3.0	1	0	2	2	1	1.6
Grass sp. (G.sp.)	10.0	2	3	4	4		3.3
Wild radish (WR)	14.0	0	0	0	1		0.3
Barnyard grass (BG)	15.0	1	1	1	2	2	1.4
<i>Convolvulus</i> sp. (C.sp.)	19.0						

<sup>1</sup> Disease rating using a scale from 0 - 9: 0 = no symptoms; 9 = plants dead

Table 4. Germination of weed seeds and percentages of seedlings with disease symptoms from western Canadian screening samples.

Weed species	Locations				
	Dawson Creek BC	Barrhead AB	Camrose AB	Wiseton SK	Ste. Rose du Lac MB
Wild oats					
Germination (%) <sup>1</sup>	24.0	86.3	44.2	24.0	43.7
Seedl.w.sympt.(%) <sup>2</sup>	8.3	13.9	15.8	5.5	19.4
Wild buckwheat					
Germination (%)	66.3	28.0	51.6	23.6	43.5
Seedl.w.sympt.(%)	0	2.3	0.6	0	11.5
Lamb's-quarter					
Germination (%)	73.5		67.0	81.0	
Seedl.w.sympt.(%)	0.7		1.5	2.5	
Green foxtail					
Germination (%)			87.0		76.0
Seedl.w.sympt.(%)			1.1		2.0
Stinkweed					
Germination (%)	64.0		76.0	60.4	
Seedl.w.sympt.(%)	3.1		5.3	18.0	
Green smartweed					
Germination (%)		66.0			63.0
Seedl.w.sympt.(%)		0			7.9
Russian thistle					
Germination (%)				10.0	
Seedl.w.sympt.(%)				0	
Hemp nettle					
Germination (%)		7.0	8.0		
Seedl.w.sympt.(%)		0	0		
Lady's-thumb					
Germination (%)			59.1		
Seedl.w.sympt.(%)			2.1		
White cockle					
Germination (%)		26.0			
Seedl.w.sympt.(%)		0			
Cow cockle					
Germination (%)				71.0	
Seedl.w.sympt.(%)				29.6	
Russian pigweed					
Germination (%)			51.7		
Seedl.w.sympt.(%)			0		

<sup>1</sup> (%) germinated of total number of seeds selected from screening sample (Table 1), plated on moist filter paper (MFP) then planted in pots.

<sup>2</sup> (%) of seedlings with disease symptoms, from which isolations were done.

Table 5. Germination of weed seeds and percentages of seedlings with disease symptoms from eastern Canadian screening samples.

Weed species	Locations		Weed species	Locations	
	Belleville ON	Montague P.E.I.		Belleville ON	Montague P.E.I.
Ragweed (not identif.)			Wild buckwheat		
Germination (%) <sup>1</sup>	2.6		Germination (%)		8.3
Seedl.w. sympt.(%) <sup>2</sup>	0		Seedl.w. sympt.(%)		0
Grass (not identif.)			Barnyard grass		
Germination (%)	17.3	12.0	Germination (%)		16.0
Seedl.w. sympt.(%)	0	16.7	Seedl.w. sympt.(%)		0
Red clover			Polygonum sp.?		
Germination (%)	71.2		Germination (%)		36.0
Seedl.w. sympt.(%)	0.6		Seedl.w. sympt.(%)		0
Wild radish			Convolvulus sp.?		
Germination (%)		9.0	Germination (%)		0
Seedl.w. sympt.(%)		0			

<sup>1</sup> (a) germinated of total number of seeds selected from screening sample, plated on moist filter paper (MFP) and planted in pots.

<sup>2</sup> (%) seedlings with disease symptoms, from which isolations were done.

Table 6. Fungi isolated from diseased seedlings from screening samples from western Canada.

Fungi isolates	Host species	Locations				
		Dawson Creek BC	Barrhead AB	Camros AB	Wiseton SK	Ste. du Lac MB
<i>Drechslera avenacea</i>	Wild oats	2(3.1%) <sup>1</sup>	21(12%)	3(6.9%)	1(1.3%)	6(8.4%)
<i>Alternaria raphani</i>	Stinkweed				5(5.0%)	
<i>Alternaria alternata</i>	Cow cockle				18(18%)	
<i>Bipolaris sorokiniana</i>	Green foxtail					2(1.0%)
<i>Botrytis sp.</i>	Wild Buckwheat		2(0.7%)			
<i>Rhizoctonia sp.</i>	Wild Buckwheat			1(0.3%)		1(0.5%)

<sup>1</sup> Number of seeds from which fungus has been isolated from selected seed sample (% of seeds)

Table 7. Fungi isolated from diseased seedlings from screening samples from eastern Canada.

Fungi isolated	Host species	Locations	
		Belleville ON	Montague P.E.I.
<i>Colletotrichum trifolii</i>	Red clover	1(0.4%) <sup>1</sup>	
<i>Bipolaris sorokiniana</i>	Grass sp.		2(2.0%)

<sup>1</sup> Number of seeds from which fungus has been isolated from selected seed sample (% of seeds).

# The Prevalence of tomato spotted wilt virus in weeds and crops in southwestern British Columbia

I. Bitterlich<sup>1</sup> and L.S. MacDonald<sup>2</sup>

A survey was conducted to determine the prevalence of tomato spotted wilt virus (TSWV) in southwestern British Columbia. Over 2600 samples from 38 commercial operations were collected and tested by enzyme-linked immunosorbent assay (ELISA). Twenty-five of the 38 sites had plants infected with TSWV. The incidence of the lettuce and impatiens strains of TSWV was equal outside the greenhouses, but the impatiens strain was more prevalent inside the greenhouses. TSWV was detected in four perennial weeds (*Trifolium* spp., *Cirsium arvense*, *Rumex acetosella*, and *Oxalis* sp.), one biennial weed (*C. vulgare*), three winter annuals (*Sfelleria media*, *Senecio vulgaris*, *Capsella bursa-pastoris*) and five annual weeds (*Cardamine oligosperma*, *Medicago lupulina*, *Galium* sp., *Geranium molle* and *Sonchus oleraceus*). Nine of the infected weed species grew outside the greenhouses. The only viruliferous thrips species collected during the survey was the western flower thrips (*Frankliniella occidentalis*) and it was the only thrips species collected inside greenhouses. TSWV is widespread in southwestern British Columbia and appears to be established in weeds outside the greenhouse operations.

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Une étude a été menée afin de déterminer l'importance du virus de la maladie bronzée de la tomate (TSWV) dans le sud-ouest de la Colombie-Britannique. Plus de 2600 échantillons provenant de 38 entreprises commerciales ont été recueillis et évalués à l'aide du test immuno-enzymatique ELISA. Vingt-cinq des 38 sites présentaient des plants infectés par le TSWV. L'incidence des souches de TSWV a été égale sur la laitue et les impatientes à l'extérieur des serres, mais elle a été plus marquée sur les impatientes à l'intérieur des serres. Le TSWV a été découvert sur quatre mauvaises herbes vivaces (*Trifolium* spp., *Cirsium arvense*, *Rumex acetosella* et *Oxalis* sp.), sur une mauvaise herbe bisannuelle (*C. vulgare*), sur trois annuelles d'hiver (*Sfelleria media*, *Senecio vulgaris*, *Capsella bursa-pastoris*) et sur cinq mauvaises herbes annuelles (*Cardamine oligosperma*, *Medicago lupulina*, *Galium* sp., *Geranium molle* et *Sonchus oleraceus*). Neuf de ces espèces de mauvaises herbes infectées ont poussé à l'extérieur des serres. Mais la seule espèce de thrips virulente capturée à l'intérieur des serres, durant l'évaluation, a été le thrips des petits fruits (*Frankliniella occidentalis*). Le TSWV est largement répandu dans le sud-ouest de la Colombie-Britannique et il semble s'être établi dans les mauvaises herbes à l'extérieur des opérations serres.

## Introduction

Tomato spotted wilt virus (TSWV) was first detected in British Columbia about 20 years ago (1). It is vectored by thrips, particularly the western flower thrips (*Frankliniella occidentalis*) which is native to British Columbia. These thrips did not become pests on greenhouse crops until after 1983, (R.A. Costello, personal communication) although they had infested field crops earlier. The occurrence of thrips on greenhouse crops and the greater movement of plant material into British Columbia have contributed to the increasing incidence of TSWV. Between 1987 and May 1991, TSWV was identified at 35 sites and caused significant losses to greenhouse crops. Many of the over 200

plant species listed as hosts are grown commercially or occur naturally in southwestern British Columbia (2).

While both the vector and hosts are present in British Columbia, it is unclear which hosts are potential sources of TSWV. Weeds were identified as reservoirs of TSWV in Hawaii (3) and because of the relatively mild winters in southwestern British Columbia it seemed possible that weeds could harbour TSWV. There are two predominant strains of TSWV in North America; the lettuce strain (L-strain) which is more common on vegetables and the impatiens strain (I-strain) which occurs more often on ornamental crops. However, both strains can infect vegetables or ornamental plants.

The results of this study in southwestern British Columbia show the range and incidence of TSWV, its presence in native weeds and the species of thrips that transmit the virus.

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## Materials and methods

One or more samples of crop and weed species were collected at 38 greenhouse sites between May 1991 and January 1992. At each site, at least one sample of the main weed and crop species were collected. Two or more samples were taken when plant species were present in large numbers or were ones previously identified as hosts. Of the 38 greenhouses surveyed, twenty-one had a history of TSWV; seven had crops which previously tested negative for TSWV; and ten sites had not been surveyed before. Any plants with ring spots, stunting and necrosis were also collected. Most plant samples were collected from within 5 m of the greenhouse but some plants were collected up to 100 m from the greenhouse. Plants were tested for TSWV by double antibody sandwich enzyme-linked immunosorbent assay (DAS-ELISA) using a polyclonal antibody against the impatiens strain and a monoclonal antibody developed against the lettuce strain. The antibodies were developed and supplied by D.J. MacKenzie and P.J. Ellis, Agriculture and Agri-Food Canada, Research Station, Vancouver.

Thrips were collected from different plant species in the greenhouse and the surrounding area. The protocol used by Cho *et al.* (4) was used to test the thrips.

## Results

There were 176 different plant species among the 2600 samples collected from the 38 commercial greenhouse ranges. Plants were infected with TSWV at 25 of the 38 sites (Table 1). TSWV had been previously diagnosed by the Plant Diagnostic Lab on crops at 14 of the 25 sites which had plants that tested positive during the survey. The 11 locations which had plants test positive during the survey had not been tested for TSWV before 1991. The virus was detected in weeds growing outside the greenhouse at 13 of the 38 sites surveyed. Seven of these 13 had a history of TSWV on crops, while plants from the other six sites had not been tested before. TSWV was detected in plants from Vancouver Island, the Fraser Valley, and Kelowna.

From the weed samples collected from the 38 sites, 2337 were tested for TSWV. TSWV was detected in 13 weed species and infected weeds were found at 20 sites (Table 2). Chickweed (*Sfelleria media*) was the most commonly infected weed. Over 13% of the 567 chickweed samples were TSWV positive and these plants were collected from 14 different sites. Infected plants and site distribution included clover (*Trifolium spp.*) plants from four of 30 sites, bull thistle (*Cirsium vulgare*) plants from three of 11 sites and black medick (*Medicago lupulina*) plants from two of 12 sites. TSWV also was detected in annual sowthistle (*Sonchus oleraceus*), bitter cress (*Cardamine oligosperma*),

Canada thistle (*Cirsium arvense*), shepherd's purse (*Capsella bursa-pastoris*) and wood sorrel (*Oxalis* sp.) collected in the greenhouse. Seven of these weeds overwinter every year and three others survive most winters in the region. Infected plants of chickweed, clovers, bull thistle, cleavers (*Galium* sp.), bitter cress, common groundsel (*Senecio vulgaris*), dove's foot geranium (*Geranium molle*), and black medick were collected outside the greenhouse. These outdoor infected weeds were symptomless and could be a source of infection in the spring. All of the 115 dandelion (*Taraxacum officinale*) plants tested were negative for TSWV.

The results did not show any relationship between weed species and virus strain. However, when only one strain was present on a crop, the infected weed(s) at the same site had the same strain. The I-strain was detected in 26 weed samples, the L-strain in 11 weed samples and both strains in three weed samples. At the sites surveyed, growers had good weed control within the greenhouse, but few growers were concerned with weeds around the greenhouse. Growers often had piles of culled plants beside the greenhouse which could facilitate the spread of TSWV to nearby weeds.

TSWV was detected in 16 ornamental or vegetable crops collected in the greenhouse while two samples of infected impatiens (*Impatiens wallerana*) were from outside (Table 3). In the ornamental crops, 381 crop plants were tested for TSWV and the most commonly infected plants were the impatiens, including the New Guinea varieties, where 26% of the plants were infected. TSWV was detected on impatiens at seven of 18 greenhouses surveyed and in 18 of the 68 samples tested. Tomato (*Lycopersicon esculentum*) plants were infected at five of nine sites and peppers (*Capsicum annum*) at three of seven greenhouses. Infected tomatoes were found at three greenhouses at very low levels, in what appeared to be healthy crops. This could be because thrips do not seem to multiply quickly on tomatoes grown in a well managed greenhouse. Even though the virus was present, the vector was not numerous enough to spread it. Brachycome (*Brachycome iberidifolia*) plants were infected with TSWV at two sites.

The impatiens strain was the most prevalent strain detected in the floriculture crops (Table 3) with 19 plants infected with only the I-strain, eight with only TSWV-L, and two with both strains. The tomato strain was more prevalent in the vegetable crops, with 12 plants infected with the L-strain, two with the I-strain, and two with both. Approximately two-thirds of the infected crop species exhibited symptoms associated with TSWV. In pepper, the lettuce strain was detected at one site and the impatiens strain at two sites. All of the tomatoes were infected with the lettuce strain, but two also had the impatiens strain.



Mixed operations, where vegetables and ornamental crops were both grown, had both strains present more frequently than one crop operations (Table 4). In addition, the percentage of mixed operations with TSWV was 75% while ornamental operations, or greenhouse vegetable operations with TSWV were 62% and 60%, respectively (Table 4).

Where TSWV was detected, both weeds and crops were infected at 60% of the sites. Weeds only were infected at 20% of the sites and crop plants only at the remaining 20%.

Thrips were observed at 35 of the 38 locations during the collection of plant samples (Table 5). Ninety-four samples of thrips were collected at 23 locations for identification and to test for the presence of TSWV. The ELISA test used for plants is also effective on individual thrips and results showed that 13% of thrips collected were carrying TSWV. All of the viruliferous thrips were collected from inside the greenhouse and were identified as western flower thrips by R.A. Costello. Viruliferous thrips were collected from chrysanthemum (*Dendranthema grandiflora*), lobelia (*Lobelia erinus*), geranium (*Pelargonium x hortorum*), spathiphyllum (*Spathiphyllum* sp.), alstroemeria (*Alstroemeria* sp.), pepper, chickweed and spiny annual sowthistle (*Sonchus asper*). Few thrips were found outside of greenhouses.

## Discussion

TSWV is established in southwestern British Columbia. Since 1987, TSWV has been detected at 47 sites overall. Of 38 commercial sites surveyed in 1991, TSWV was detected at 25. TSWV had been diagnosed at 14 of these sites prior to 1991 and the virus was still present in these sites when this survey was conducted. This is in contrast to only seven sites with a history of TSWV where it was not detected. Mixed operations, where both vegetables and ornamentals were produced, had a higher incidence of TSWV. This may be due to more movement of plant material and greater difficulty in controlling thrips in production schedules where the greenhouse cannot be completely emptied between crops. An average of 62 weed and 10 crop plants were tested per site. TSWV was detected in at least one and up to seven plants at each of the 25 operations. TSWV is more common than previously realized.

It is probable that TSWV overwinters in certain weeds. Seven of the weed species found to be infected with TSWV have no difficulty overwintering in this region. TSWV was detected in weeds located within 15 m of the greenhouse at 10 sites. These weeds will remain infected until they die. It is impossible to rely on the appearance of symptoms in weeds to determine if they are infected with TSWV, since all infected weeds in the survey had no symptoms. This shows the importance of maintaining a weed free buffer zone around the greenhouse.

The most common thrips were the western flower thrips and all of the viruliferous thrips were of this species. Thrips feeding on infected overwintering weeds are probably the main source for spread of the virus into the greenhouse in the spring.

In the instances where TSWV was only detected in weeds, it is probable that TSWV also was present in crops which may have been shipped to market or, the virus was present but not detected in the tests. TSWV is difficult to detect because it is located in discrete pockets within the plant and is unstable outside of the plant. As a result, it may not always be detected even though it is present, so that a negative result is not entirely reliable. The ELISA tests used are very specific, so positive test results indicate the virus is present.

TSWV puts British Columbia's \$84 million floriculture, \$21 million greenhouse vegetable and \$56 million field vegetable (5) industry at risk. Good weed control, effective sanitation, use of healthy transplants, and access to insecticides effective against thrips are essential to avoid major losses from TSWV.

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Table 1. Site histories and survey results for 1991.

History of site since 1987	Number of positive sites	Number of negative sites
No previous infections	11	6
Previous infections	14	7
Total from 1991 survey	25	13

Table 2. Summary of weed species infected with TSWV, 1991.

Weed	Growth habit <sup>a</sup>	TSWV strain <sup>b</sup>	Positive site/ total sites	No. infected/ total # tested	Location <sup>c</sup>
Annual sowthistle ( <i>Sonchus oleraceus</i> )	A	L	1/5	1/6	I
Bitter cress ( <i>Cardamine oligosperma</i> )	A	I	2/14	41/28	I,O
Black medick ( <i>Medicago lupulina</i> )	A	I,L	2/12	3/50	O
Bull thistle ( <i>Cirsium vulgare</i> )	B	I,L	3/11	5/132	I,O
Canada thistle ( <i>Cirsium arvense</i> )	P	L	1/7	1/20	I
Chickweed ( <i>Stellaria media</i> )	ANVA	I,L,IL	14/34	39/567	I,O
Cleavers ( <i>Galium sp.</i> )	A	L	1/1	1/1	O
Clover ( <i>Trifolium spp.</i> )	P	I,L	4/30	61/262	I,O
Common groundsel ( <i>Senecio vulgaris</i> )	ANVA	I	1/9	51/160	O
Dove's foot geranium ( <i>Geranium molle</i> )	A	I	1/1	31/240	O
Sheep sorrel ( <i>Rumex acetosella</i> )	P	O	1/1	1/4	O
Shepherd's purse ( <i>Capsella bursa-pastoris</i> )	A/WA	L	1/6	1/6	I
Wood sorrel ( <i>Oxalis sp.</i> )	P	I	1/10	1/11	I

<sup>a</sup> A = annual, B = biennial, P = perennial and WA = winter annual.

<sup>b</sup> I = impatiens, L = lettuce, IL = both strains.

<sup>c</sup> Location where samples were collected. I = inside greenhouse, O = outside.

Table 3. Summary of crop species infected with TSWV, 1991.

Crop	TSWV strain <sup>a</sup>	Positive sites/ total sites tested	# infected/ total #tested	Symptoms <sup>b</sup>
Pepper ( <i>Capsicum annuum</i> )	I,L	3/7	5/27	+
Tomato ( <i>Lycopersicon esculentum</i> )	L,IL	5/9	11/35	+
Begonia ( <i>Begonia x hiemalis</i> )	I	1/10	1/17	+
Brachycome ( <i>Brachycome iberidifolia</i> )	I,L	2/4	2/6	
Chrysanthemum ( <i>Dendranthemagrandiflora</i> )	L	1/10	3/79	+
<i>Episcia dianthiflora</i>	I	1/1	1/1	+
Exacum ( <i>Exacum affine</i> )	I	1/2	1/2	+
Fig ( <i>ficuselastica</i> )	I	1/5	1/7	+
Fuchsia ( <i>Fuchsia sp.</i> )	I	1/6	1/16	
Gloxinia ( <i>Sinningia speciosa</i> )	I	1/3	1/4	+
Impatiens ( <i>Impatiens wallerana</i> )	I,L,IL	5/12	9/21	+
Impatiens, New Guinea var.	I,L	3/14	4/21	+
Lantana ( <i>Lantanasp.</i> )	I	1/1	2/2	
<i>Laurentia fluviatillis</i>	I	1/1	1/2	
Geranium ( <i>Pelargonium x hortorum</i> )	L	1/6	1/25	
Spathiphyllum ( <i>Spathiphyllumsp.</i> )	I	1/1	1/1	+

<sup>a</sup> I = impatiens, L = lettuce, IL = both strains

<sup>b</sup> + = indicates the plants had leaf spots, stunting, tip dieback or fruit mottling.  
- = indicates no symptoms.

Table 4. Occurrence of TSWV on crops and weeds at vegetable, ornamental and mixed operations.

TSWV strain	Ornamental greenhouse	Vegetable greenhouse	Mixed greenhouse	Total sites
I	7	1	2	10
L	4	1	1	6
I + L	2	1	6	9
No TSWV	8	2	3	13
Total sites	21	5	12	38

Table 5. Summary of TSWV detected in thrips collected from 23 sites.

Collection area	No. of sites	No. of positive sites	Total samples	% WFT <sup>a</sup>	No. TSWV	I strain	L strain
Inside	20	5	56	100	44	10	2
Outside	16	0	38	97	38	0	0
Total	23	5	94	99	82	10	2

<sup>a</sup> The percentage of thrips that were western flower thrips.

# Prevalence of some seedborne fungi on soft white winter wheat seed from Ontario, Canada

R.M. Clear and S.K. Patrick<sup>1</sup>

To determine the mycoflora of grain samples of white winter wheat (*Triticum aestivum*), 435 samples collected over three years were examined for the presence of fungi by plating surface disinfected seeds onto potato dextrose agar. At least 59 species representing 35 fungal genera were recovered from seed. *Alternaria alternata*, *Epicoccum nigrum*, and species of *Arthrimum*, *Aspergillus*, *Cladosporium*, *Drechslera* and *Nigrospora* infected more than 1% of the seeds every year. *Bipolaris sorokiniana*, *Drechslera tritici-repentis*, *Fusarium graminearum*, *F. poae*, and *Septoria nodorum* infected more than 1% of the seeds in one or two years. Yearly differences in the quantity and time of precipitation and the frequency of a number of fungi such as the pathogens *B. sorokiniana*, *D. tritici-repentis*, and *S. nodorum*, including a 100 fold increase in the frequency of *F. graminearum* between 1988 and 1989, were recorded. Whereas forty years ago *B. sorokiniana* was the most common pathogen recovered from Ontario wheat seed, *F. graminearum* was the most frequently detected pathogen in this study.

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Afin de déterminer la mycoflore des échantillons de graines du ble tendre (*Triticum aestivum*), 435 échantillons récoltés durant trois années ont été examinés pour vérifier la présence de champignons en appliquant des semences sur des surfaces désinfectées d'agar composé de dextrose de pomme de terre. Au moins 59 espèces appartenant à 35 genres de champignons ont été récupérées de ces semences. *Alternaria alternata*, *Epicoccum nigrum* et des espèces de *Arthrimum*, *Aspergillus*, *Cladosporium*, *Drechslera* et *Nigrospora* ont infecté plus de 1% des semences à chaque année. *Bipolaris sorokiniana*, *Drechslera tritici-repentis*, *Fusarium graminearum*, *F. poae* et *Septoria nodorum* ont infecté plus de 1% des semences en une ou deux années. Des différences annuelles ont été enregistrées pour les quantités et les temps de précipitations et pour la fréquence du nombre de champignons comme les pathogènes *B. sorokiniana*, *D. tritici-repentis* et *S. nodorum*, incluant une augmentation centuplée de la fréquence de *F. graminearum* durant les années 1988 et 1989. Il y a quarante ans, *B. sorokiniana* était le pathogène le plus communément retrouvé dans les semences de ble en Ontario, alors que *F. graminearum* a été le pathogène le plus fréquemment détecté lors de cette étude.

## Introduction

Fungi can be recovered from surface disinfected wheat seed (*Triticum aestivum* L.), even though their presence in the seed is usually not evident until the seeds have been placed in an environment conducive to prolific fungal growth. Extensive fungal development on grain may reduce the value because of seed discolouration, chemical changes, loss of dry matter, objectionable odours, and mycotoxin accumulation (Christensen and Kaufmann 1974). If infested grain is used as seed, the seedborne diseases can reduce yield and the grain will be a source of inoculum. Grain buyers sometimes set tolerance limits for specific organisms which, if exceeded, will result in either rejection of the shipment or demands for a price reduction.

A knowledge of the mycoflora and their frequency on particular types of grain provides regulatory agencies with a basis to assess the risk associated with undesirable organisms and their metabolites. Since the last surveys of the mycoflora of Ontario grown wheat seed in 1942 (Greaney and Machacek) and 1951 (Machacek *et al.*), the introduction of new varieties and cropping practices may have changed the frequency of various seedborne fungi. The purpose of this study was to record the fungi associated with soft white winter wheat seed grown in Ontario in recent years and to compare these results with those obtained 40 to 50 years ago.

## Materials and methods

Ninety-nine 1 kg weekly composite samples and vessel loading samples of soft white winter wheat from Ontario were collected from terminal elevators in 1988, 259 in 1989 and 77 in 1990. Samples were collected by inspectors of the Grain Inspection Division of the Canadian Grain Commission and sent to the Grain Research Laboratory where they were documented, mixed, subsampled, and then

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stored at -15°C for up to 4 months. For mycological examination 100 seeds from each subsample were surface disinfected for 1 minute in a solution of 0.3% sodium hypochlorite then air dried in a laminar flow hood. The dry seeds were plated onto potato dextrose agar (10 seeds per plate) and incubated for 5 days on a 12 hr, 28°C light and a 12 hr 22°C dark cycle. Illumination was by a 4:1 mixture of fluorescent and long-wave ultraviolet lamps at 48 cm distance above the petri plates. The percentage of seeds in a sample which were infected by an organism and the average per year were recorded. Moisture and temperature conditions during the growing seasons were obtained from 11 Environment Canada weather stations within the white winter wheat growing areas of Ontario. The daily values were combined to obtain monthly averages for these environmental factors.

## Results

During this 3 year study, 59 species representing 35 fungal genera were recovered from the grain samples (Table 1). Every year more than 1% of the seeds were infected by species from six genera, *Alternaria* Nees ex Fr. (represented solely by *A. alternata* (Fr.) Keissler), *Arthrinium* Kunze ex Fr. (primarily the *Arthrinium* state of *Apiospora montagnei* Sacc., but also including *Arthrinium phaeospermum* (Corda) M.B. Ellis), *Aspergillus* Mich. ex Fr. (primarily *A. glaucus* group species), *Cladosporium* Link ex Fr. (primarily *C. cladosporioides* (Fresen.) de vries), *Drechslera* Ito, *Epicoccum* Link ex Schlecht. (represented solely by *E. nigrum* Link), and *Nigrospora* Zimmermann (primarily *N. oryzae* (Berk. & Br.) Petch). More than 1% of the seeds in one or two of the test years were infected with *Bipolaris sorokiniana* (Sacc.) Shoemaker, *D. tritici-repentis* (Died.) Shoemaker, *Fusarium graminearum* Schwabe, *F. poae* (Peck) Wollenw., and *Septoria nodorum* (Berk.) Berk.

Maximum levels of seed infection found in the samples ranged from 98% for *A. alternata* to 1% for many other fungi recorded (Table 2). Only *A. alternata* was found in every sample tested.

A lower overall incidence of infection was recorded in 1988 than in 1989 or 1990, but not all fungi were found less often in 1988. Precipitation during the growing season prior to July, 1988 was considerably less than that during 1989 and 1990 (Table 3). The growing conditions of 1989 appeared to be especially suited to the commercially important pathogens *F. graminearum* and *B. sorokiniana*. Two other commercially important pathogens, *D. tritici-repentis* (causal agent of tan spot) and *S. nodorum* (causal agent of glume blotch), were observed most often during the moister growing seasons of 1989 and 1990. *Fusarium graminearum* had the greatest yearly fluctuation in infection levels, ranging from 0.08% in 1988 to 11.89% in 1989, over a 100 fold increase.

## Discussion

During this three year study important fungal pathogens were isolated from the soft white winter wheat produced in Ontario. The diseases on the grain were similar to those recorded for seed in 1951 by Machacek *et al.* With the exception of species of *Pullularia* Berk., seven of the eight most common fungal genera reported by Machacek *et al.* (1951) were among the most common ones in this study. In the present study *Arthrinium* spp. and *D. tritici-repentis* were frequently observed on seed. These two species were not common among the samples examined by Machacek *et al.* (1951). Species of *Arthrinium*, previously called *fapularia* Fr., were isolated at just above trace amounts by Machacek *et al.* (1951), and *D. tritici-repentis* was not mentioned. During 1988-1990, *Nigrospora oryzae* was the dominant *Nigrospora* species isolated from Ontario winter wheat seed. Although Machacek *et al.* (1951) identified *N. sphaerica* (Sacc.) Mason as the only *Nigrospora* species observed, their spore size measurement of 15µ suggests that it most likely was *N. oryzae*.

*Alternaria* species have been isolated from wheat seed in different regions of Canada (Greaney and Machacek 1942; Machacek *et al.* 1951), and their growth within wheat seed can cause the discolourations known as blackpoint and smudge. However, those species along with *Arthrinium* spp., *Cladosporium* spp., *Epicoccum nigrum*, *F. poae*, and *N. oryzae*, appear to have a minimal effect on the health of wheat seed (Malone and Muskett 1964; Zillinsky 1983).

*Aspergillus glaucus* group species, *B. sorokiniana*, *D. tritici-repentis*, *F. graminearum*, and *S. nodorum* are all reported to affect seed health and occasionally seed appearance (Martens *et al.* 1984; Thorpe 1958; Valder and Shaw 1953). Machacek *et al.* (1951) and Greaney and Machacek (1942) found *B. sorokiniana* to be the most common pathogen recovered from wheat seed, and Machacek *et al.* (1951) reported yearly averages of seed infection to range from <0.1 to 12.0% of Ontario wheat. The results in this study show that *F. graminearum* was the most common pathogen recovered from wheat seed. Although the monthly precipitation averages for both May and June of 1989 and 1990 (Table 3) were similar, the frequency of *F. graminearum* in 1989 was ten times that of 1990 (Table 1). It seems likely that the conditions at time of anthesis, which are critical for both the infection by *F. graminearum* (Sutton 1982) and the production of tombstone kernels (Atanasoff 1920), were more suitable for infection in 1989 than 1990. The observation that tombstone kernels were an important degrading factor in 1989 but not 1990 (Anonymous 1989, 1990) is consistent with these results herein. The abundance of *B. sorokiniana* in 1989 may also be due to epidemiological considerations similar to those which favoured *F. graminearum*, as both fungi were several times more common on the seed in 1989. Greaney and Machacek (1946) reported that the amount of

rain during the growing season was the most important factor influencing the epidemiology of *B. sorokiniana*. However, Jorgensen (1974) reported temperature after sowing and not the frequency of moisture influenced the incidence of *B. sorokiniana* on barley seed.

The frequency of *F. graminearum* is important since it is a causal agent of fusarium head blight as well as root and crown rot of cereals (Martens *et al.* 1984). It also lowers the value of the crop due to the production of the degrading factor known as tombstone kernels and the fungus also produces mycotoxins such as deoxynivalenol (Sutton 1982). Previously, *F. graminearum* was seldom isolated from Ontario wheat seed (Gordon 1952), and it was not among the four predominant species isolated from cereal seed (Greaney and Machacek 1942), even though almost all their *Fusarium* isolates were from eastern Canada. This observation of increased recovery of *F. graminearum* from seed compared with 40 years ago is supported by recent surveys of Ontario wheat seed for *Fusarium* species by Duthie *et al.* (1986) and Clear and Patrick (1990). They found *F. graminearum* to be the most or second most common *Fusarium* species infecting soft white winter wheat seed grown in Ontario. The changes in the observed frequency of this pathogen may result from the same influences which resulted in several epidemics of fusarium head blight caused by *F. graminearum* since 1980.

Similarities in infection levels between years, such as for *S. nodorum* and *D. tritici-repentis* in 1989 and 1990, may be due to comparable weather conditions. Wet periods at heading favour seed infection by *S. nodorum* (Shipton *et al.* 1971), and this, as well as the reported wet harvest conditions of 1989 and 1990 (Anonymous 1989, 1990), may have been factors in the frequency of *S. nodorum*, *D. tritici-repentis* and *E. nigrum*.

*Aspergillus glaucus* group species were the only storage fungi commonly isolated in this study while much less common were members of the *A. flavus* group species. These two group species were the ones most often isolated by Machacek *et al.* (1951), and reflect storage conditions prior to sampling. Fewer seeds infected by the *Aspergilli* were recorded by Machacek *et al.* (1951), possibly because the samples they tested were destined to be used as seed and therefore may have been handled more carefully than grain. It is interesting that the highest observed incidence of the *A. glaucus* group species was in 1988, the year with the driest growing conditions. The higher incidence of this group and the higher bacterial levels observed during 1988 may have resulted from less overgrowth by other fungi masking their presence. Recovery of *A. glaucus* would likely have been higher if a media with a more optimal potential for their isolation had been used to culture the seeds. However, the scarcity of less xerophilic *Aspergillus* species shows the

grain was still in good condition at time of sampling.

Although Machacek *et al.* (1951) used potato sucrose agar and both Greaney and Machacek (1942) and Machacek *et al.* (1951) used an ethyl alcohol-mercuric bichloride solution for surface disinfection, it seems quite likely that these earlier studies and the present one provide a good estimate of the pathogens prevalent in soft white winter wheat seed over the survey periods. Even with some differences in methodology it still appears that the procedures used then and now would yield valuable data on the frequencies of seedborne fungi in this crop.

This study presents the principle species infecting soft white winter wheat seed produced in Ontario and shows some of the yearly and sample variation in infection levels that can occur over several survey years. The most abundant pathogenic species on the wheat seed appears to be *F. graminearum*, causal agent of fusarium head blight as well as diseases of the roots and crown. This pathogen appeared to be uncommon in Ontario wheat forty years ago, when *B. sorokiniana* was the most frequently identified seedborne pathogen.

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Table 1. Average level of seed infection (%) by various microorganisms on surface disinfected white winter wheat seed produced in Ontario during 1988, 1989, and 1990.

Microorganisms	Year no. of seed samples	Percentage of Seeds Infected*		
		1988 99	1989 259	1990 77
<i>Acremoniella atra</i>		0.00	0.09	0.00
<i>Acremonium</i> spp.		0.00	0.01	0.00
<i>Alternaria alternata</i>		58.99	71.99	84.05
<i>Arthrinium</i> spp.		2.59	1.35	1.43
<i>Aspergillus candidus</i>		0.04	0.01	0.04
<i>A. clavatus</i>		0.09	0.01	0.01
<i>A. flavus</i>		0.45	0.30	0.45
<i>A. fumigatus</i>		0.01	0.00	0.00
<i>A. glaucus</i>		6.33	1.02	1.86
<i>A. nidulans</i>		0.03	tr	0.00
<i>A. niger</i>		0.05	0.02	0.03
<i>A. ochraceus</i>		0.01	tr	0.00
<i>A. terreus</i>		0.01	0.01	0.00
<i>A. wentii</i>		0.02	0.00	0.00
<i>Aspergillus</i> not ID		0.05	0.01	0.00
<i>Aureobasidium pullulans</i>		0.16	0.18	0.20
Bacteria		5.41	2.79	3.84
<i>Bipolaris bicolor</i>		0.01	0.08	0.06
<i>B. sorokiniana</i>		0.42	2.67	0.50
<i>Botrytis cinerea</i>		0.00	0.02	0.01
<i>Cephalosporium</i> spp.		0.05	tr	0.00
<i>Chaetomium</i> spp.		0.02	0.02	0.02
<i>Cladosporium</i> spp		1.08	3.38	2.41
<i>Coelomycetes</i>		0.15	0.23	0.24
<i>Curvularia</i> spp		0.15	0.05	0.09
<i>Drechslera biseptata</i>		0.23	0.24	0.38
<i>D. tritici-repentis</i>		0.74	1.70	1.43
<i>D. teres</i>		0.07	0.06	0.07
<i>Epicoccum nigrum</i>		4.26	8.49	8.99
<i>Fusarium acuminatum</i>		0.04	0.18	0.12
<i>F. avenaceum</i>		0.05	0.43	0.25
<i>F. crookwellense</i>		0.00	0.01	0.01



Microorganisms	Year no. of seed samples	Percentage of Seeds Infected*		
		1988 99	1989 259	1990 77
continued				
<i>F. culmorum</i>		0.00	0.01	0.04
<i>F. equiseti</i>		0.21	0.05	0.13
<i>F. graminearum</i>		0.08	11.89	1.83
<i>F. oxysporum</i>		0.04	0.03	0.00
<i>F. pallidoroseum</i>		0.00	0.01	0.00
<i>F. poae</i>		0.16	0.98	1.26
<i>F. proliferatum</i>		0.00	0.01	0.01
<i>F. sporotrichioides</i>		0.16	0.47	0.23
<i>F. subglutinans</i>		0.00	0.01	0.00
<i>Fusarium</i> not ID		0.01	0.07	0.01
<i>Gonatobotrys</i> spp.		0.02	0.07	0.05
<i>Microdochium bolleyi</i>		0.01	0.06	0.02
<i>M. nivale</i>		0.00	0.01	0.00
<i>Mucor</i> spp.		0.15	0.13	0.15
<i>Nigrospora oryzae</i>		2.11	1.62	1.07
<i>N. sphaerica</i>		0.02	0.04	0.02
<i>Penicillium</i> spp.		0.49	0.17	0.36
<i>Phaeoramularia</i>		0.00	0.01	0.01
<i>Phomopsis</i> spp.		0.00	0.11	0.12
<i>Pithomyces</i> spp.		0.01	0.01	0.00
<i>Pseudomicrodochium</i> spp.		0.03	0.07	0.00
<i>Rhizopus</i> spp.		0.34	0.11	0.07
<i>Scopulariopsis</i> spp.		0.04	0.00	0.00
<i>Septoria odorum</i>		0.26	5.75	5.47
<i>Sordaria fimicola</i>		0.00	0.02	0.00
<i>Stemphylium</i> spp.		0.35	0.21	0.17
<i>Syncephalastrum racemosum</i>		0.01	0.02	0.00
<i>Trichoderma</i> spp.		0.02	0.01	0.00
<i>Trichothecium roseum</i>		0.01	tr	0.00
<i>Ulocladium</i> spp.		0.02	0.00	0.01
<i>Verticillium</i> spp.		0.00	tr	0.00

tr = &lt;0.01%

\* Results based on 100 seeds per sample plated onto potato dextrose agar at room temperature.

Table 2. Maximum incidence (%) of seed infection by various microorganisms on surface disinfected white winter wheat seed from Ontario during 1988, 1989, and 1990.

Microorganisms	Year no. of seed samples	Percentage of Seeds Infected*		
		1988 99	1989 259	1990 77
<i>Acremoniella atra</i>		0	3	0
<i>Acremonium</i> spp.		0	1	0
<i>Alternaria alternata</i>		95	96	98
<i>Arthrrium</i> spp.		14	13	8
<i>Aspergillus candidus</i>		1	1	1
<i>A. clavatus</i>		2	2	1
<i>A. flavus</i>		8	4	7
<i>A. fumigatus</i>		1	0	0
<i>A. glaucus</i>		48	14	15
<i>A. nidulans</i>		2	tr	0
<i>A. niger</i>		2	1	1
<i>A. ochraceus</i>		1	1	0
<i>A. terreus</i>		1	1	0
<i>A. wentii</i>		2	0	0
<i>Aspergillus</i> not ID		1	1	0
<i>Aureobasidium pullulans</i>		3	2	2
Bacteria		56	17	18
<i>Bipolaris bicolor</i>		1	2	1
<i>B. sorokiniana</i>		3	16	2
<i>Botrytis cinerea</i>		0	1	1
<i>Cephalosporium</i> spp.		1	1	0
<i>Chaetomium</i> spp.		1	1	2
<i>Cladosporium</i> spp.		7	14	23
<i>Coelomycetes</i>		2	3	3
<i>Curvularia</i> spp.		2	1	1
<i>Drechslera biseptata</i>		2	2	2
<i>D. tritici-repentis</i>		6	13	7
<i>D. teres</i>		1	1	1
<i>Epicoccum nigrum</i>		17	27	25
<i>Fusarium acuminatum</i>		1	2	1
<i>F. avenaceum</i>		1	4	3
<i>F. crookwellense</i>		0	1	1
<i>F. culmorum</i>		0	1	1
<i>F. equiseti</i>		3	1	1
<i>F. graminearum</i>		3	85	6
<i>F. oxysporum</i>		1	1	0
<i>F. pallidoroseum</i>		0	1	0
<i>F. poae</i>		2	5	5
<i>F. proliferatum</i>		0	1	1
<i>F. sporotrichioides</i>		3	7	3
<i>F. subglutinans</i>		0	1	0
<i>Fusarium</i> not ID		1	3	1
<i>Gonatobotrys</i> spp.		1	4	1
<i>Microdochium bolleyi</i>		1	2	1
<i>M. nivale</i>		0	1	0
<i>Mucor</i> spp		3	2	2
<i>Nigrospora oryzae</i>		9	11	4
<i>N. sphaerica</i>		1	1	1

Microorganisms	Year no. of seed samples	Percentage of Seeds Infected*		
		1988 99	1989 259	1990 77
continued				
<i>Penicillium</i> spp.		3	3	3
<i>Phaeoramularia</i>		0	1	1
<i>Phomopsis</i> spp.		0	2	1
<i>Pithomyces</i> spp.		1	1	0
<i>Pseudomicrodochium</i> spp.		1	2	0
<i>Rhizopus</i> spp.		5	3	1
<i>Scopulariopsis</i> spp.		1	0	0
<i>Septorianodorum</i>		3	24	11
<i>Sordaria fimicola</i>		0	2	0
<i>Stemphylium</i> spp.		2	3	3
<i>Syncephalastrum racemosum</i>		1	2	0
<i>Trichoderma</i> spp.		1	1	0
<i>Trichothecium roseum</i>		1	1	0
<i>Ulocladium</i> spp.		2	0	1
<i>Verticillium</i> spp.		0	1	0

\* Results based on 100 seeds per sample plated onto potato dextrose agar at room temperature.

Table 3. Average of the daily temperature and monthly rainfall recorded at eleven weather stations within the white winter wheat growing areas of Ontario in 1988, 1989, and 1990.

	April		May		June		July		August	
	C	mm	C	mm	C	mm	C	mm	C	mm
1988	6.3	59.3	14.3	50.1	17.8	17.3	22.4	106.0	21.4	75.5
1989	4.9	58.7	12.6	104.9	18.2	94.8	21.1	48.0	19.5	61.8
1990	8.2	67.4	11.7	106.5	18.0	85.4	20.1	98.3	19.5	97.5
30yr avg*	6.4	78.6	12.5	67.1	17.9	78.7	20.3	72.8	19.6	84.1

\* 1950-1980



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## Instructions to authors

The Canadian Plant Disease Survey is published twice a year, presenting articles on the occurrence and severity of plant diseases in Canada. Topics of interest include development of methods of investigation and control, including the evaluation of new materials. Original information, review papers and compilations of practical value to plant pathologists are accepted.

Peer reviewed articles and brief notes are published in English or French. Address the manuscript and all correspondence to Ms. Rosalyn McNeil, Information and Planning Services, Research Branch, Agriculture and Agro-Food Canada, Ottawa, Ontario K1A 0C6. Signatures of authors and the director of the establishment where the work was carried out should be supplied.

Diskette submission requirements. Please use a 3.5-inch IBM-compatible diskette. The diskette will be returned with author proofs. Send two letter-quality double-spaced printouts of the manuscript and a diskette containing all typed text, tables, figure and photo captions. Save the file, containing a single-spaced version of the article, in Wordperfect, if possible. Alternatively, save the file in ASCII format, instead of in the program's normal format. Consult your software manual for instructions on saving documents as ASCII files (sometimes called DOS files or printer files). Please label your diskette accordingly and indicate the document's full file name, including its extension.

Manuscripts should be concise and consistent in style, spelling, and use of abbreviations. They should be printed double-spaced throughout. Number all pages, including those containing abstract, tables, and legends. For general format and style, refer to recent issues of the Survey and to the *CBE Style Manual* 5th ed., 1983. Whenever possible, give numerical data in metric units (SI). Alternatively, provide the metric equivalents. Use square brackets to enclose the scientific name of a pathogen, following the common name of a disease, to denote cause.

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

Abstracts of less than 200 words should accompany each article, and should be provided in both English and French, if possible.

Figures should be planned to fit, after reduction, into one column (maximum 84 x 241 mm) or two columns (maximum 175 x 241). Trim them or add crop marks to show only essential features. Mount figures grouped in a plate tightly together, with no space between them. Provide a duplicate set of unmounted photographs and line drawings. Identify figures by number, author's name, and abbreviated legend.

Tables should be numbered using arabic numerals. Provide a concise title. Do not use vertical rules. Identify footnotes by reference marks (\*†§#¶\*\*†), particularly when they refer to numbers.

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## Recommandations aux auteurs

L'Inventaire des maladies des plantes au Canada est publié deux fois par année et contient des articles sur l'incidence et la gravité des maladies des plantes au Canada. Les articles portent surtout sur la mise au point de nouvelles méthodes d'investigation et de lutte comportant l'évaluation de nouveaux matériaux. Nous acceptons aussi des données de première main, des comptes rendus critiques de publications et les compilations qui peuvent être utiles aux phytopathologistes.

Les comptes rendus critiques et les courts résumés sont publiés en anglais et en français. Adresser le manuscrit et toute la correspondance à mademoiselle Rosalyn McNeil, Services d'information et de planification, Direction générale de la recherche, Agriculture et agro-alimentaire Canada, Ottawa (Ontario) K1A 0C6. Vous devez aussi nous faire parvenir la signature des auteurs et du directeur de l'établissement où le travail a été effectué.

*Exigences pour la soumission* des disquettes. Veuillez, utiliser une disquette IBM-compatible 3.5 pouces. La disquette vous sera retournée avec les corrections de l'auteur. Envoyer deux copies du manuscrit qualité lettre tapées à double interligne et une disquette contenant tout le texte, les tableaux, les figures et les photos. Sauvegarder le fichier contenant une version de l'article à simple interligne en Wordperfect si possible. Sinon, sauvegarder le fichier en format ASCII au lieu du format normal du programme. Dans votre manuel, voir les instructions de sauvegarde de documents en fichier ASCII (parfois appelés fichiers DOS ou fichiers de l'imprimante). Veuillez étiqueter votre disquette en conséquence et indiquer le nom complet du fichier du document incluant son extension.

Les *Manuscrits* doivent être concis et faire preuve de cohérence dans le style, l'orthographe et l'emploi des abréviations. Ils doivent être dactylographiés à double interligne. Numéroter toutes les pages incluant celles du résumé, les tableaux et les légendes. Pour plus de renseignements sur le format des feuilles et le style, prière de consulter nos dernières publications de *L'Inventaire* et le *CBE Style Manual* 5<sup>ème</sup> ed., 1983. Dans la mesure du possible, soumettre les données numériques en unités métriques, (SI). Sinon, fournir l'équivalent métrique. Utiliser des crochets pour identifier le nom scientifique d'un pathogène après le nom commun de la maladie dont il est l'agent causal.

Les *titres* doivent être courts et révélateurs, ainsi que le résumé qui les accompagne et les mots clés les plus utiles pour le classement et l'extraction de l'information.

*Chaque* résumé de moins de 200 mots devrait accompagner chaque article et devrait être rédigé en anglais et en français si possible.

Les figures doivent pouvoir, après réduction, entrer dans une colonne (maximum 84 x 241 mm) ou deux colonnes (maximum 175 x 241). Découpez les figures ou indiquez par des lignes quelle est la portion essentielle de la figure. Monter les figures groupées sur une planche côte à côte sans espace entre elles. Fournir un double des photographies non montées et des graphiques. Les figures doivent être numérotées, porter le nom de l'auteur et une légende abrégée.

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