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Occurrence of fungi on leafy spurge in the prairie provinces from 1981 to 1983[1]

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Abstract:

Leafy spurge stands in the prairie provinces were surveyed for plant pathogens during the growing seasons of 1981, 1982 and 1983. In most sites surveyed, leafy spurge was found to be disease-free. The most frequent diseases observed were leaf spot and top dieback caused by *Alternaria* spp. and a leaf spot caused by *Septoria guepini*. Stem and root rot were observed on scattered plants from several sites. Several fungi were isolated of which *Fusarium* spp. were the most frequent. The potential of isolated fungi as biocontrol agents from leafy spurge is discussed.

Introduction

Leafy spurge (*Euphorbia esula virgata* complex) is an introduced herbaceous perennial weed that occurs throughout most of the northern half of the United States and across Canada. It is a serious weed in the prairie provinces and North Dakota, where the area infested with leafy spurge doubled during the 1973 to 1982 period (Best *et al*. 1979, Messersmith and Lym 1982). Chemical control of leafy spurge in pastureland on a large scale is not economical because retreatment is necessary every 3 to 5 years to get adequate control (Lym and Messersmith 1983). Biological control appears to be a satisfactory long-term solution. The purpose of this study was to survey and investigate the suitability of indigenous pathogens of leafy spurge as inundative biological control agents.

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Materials and methods

Surveys of leafy spurge were conducted in Saskatchewan from 1981 to 1983. The heavily infested areas at Jameson and Caronport were visited regularly during these growing seasons. Many of the leafy spurge infestations reported by Coupland *et al*. (1949-1955) in the early fifties in central and southeastern Saskatchewan were visited once (Harris, unpublished data) in 1981 and some of the sites were revisited in 1982 and 1983. A two-day survey of leafy spurge infested areas in Manitoba was conducted in each of 1982 and 1983. One leafy spurge site in Alberta was visited in 1981 and 1982. A few leafy spurge sites in interior British Columbia were surveyed in 1981 (Table 1).

Table 1. *Alternaria* leaf spot and top dieback observed on leafy spurge from the prairies during the three-year period 1981 to 1983.

Fungi isolated	Pathogen.* test (lab.) I	II	Field symptoms	Location and date
Alternaria alternata (Fr.) Keissl.	+		Irregular necrotic leaf spots	Weyburn, Sask. 7-7-83.
Alternaria sp.	(+)		Necrotic spots on stems	Caronport, Sask. 15-7-81
Cladosporium herbarum (Pers.) Link ex Gray	(+)			
Alternaria tenuissima (Kunze ex Pars) Wiltsh.	(+)		Top dieback	Caronport, Sask. 6-8-81.
Cladosporium sp.	(+)			
Botrytis cinera Pers.		+	Top dieback	Cardston, Alta. 19-8-81.
Fusarium sp.	–			
Alternaria sp.	–			
Bacteria	–			
Cladosporium herbarum (Pers.) Link ex Gray	(+)		Stem spots, purplish black	Mortlack, Sask. 1-6-82.
Alternaria sp.	+	++	Leaves turning blackish mainly on upper leaf surfaces	Caronport, Sask. 1-6-82.
Alternaria sp. ? (not sporulating)	(+)	+	Stem lesions (black)	Sask. Beach, Sask. 17-6-82.
Alternaria sp.	(+)		Flower and top dieback	Caronport, Sask. 12-7-82.
Alternaria sp. ? (not sporulating)	(+)			
Cladosporium herbarum (Pers.) Link ex Gray	(+)			
Alternaria sp.	+		Leaf dieback and necrotic leaf spots on flower bracts	Zehner, Sask. 21-7-82
Cladosporium herbarum (Pers.) Link ex Gray	(+)		Irregular necrotic leaf spots	Rounthwaite, Man. 27-7-82.
Alternaria sp.	++	–		
Alternaria sp.	+		Top dieback and necrotic leaf spots	Rounthwaite, Man. 27-7-82.
Fusarium sporotrichioides Sherbakoff	(+)			
Epicoccum purpurascens Ehrenb.	–			
Alternaria sp.	++	–	Top dieback and necrotic leaf spots	Rounthwaite, Man. 27-7-82.
Alternaria sp.	–		Top dieback and necrotic leaf spots	Treesbank, Man. 27-7-82.

Fungi isolated	Pathogen.* test (lab.) I	II	Field symptoms	Location and date
Alternaria sp.	++		Top dieback and necrotic leaf spots	Gainsborough, Sask. 28-7-82.
Alternaria sp.	–			
Fusarium sp.	–			
Alternaria sp.	++	–	Irregular necrotic leaf spots	Carnduff, Sask. 28-7-82.
Alternaria sp.	++	–	Irregular necrotic leaf spots	Estevan, Sask. 28-7-82.
Alternaria sp.	–		Flower dieback	Moose Jaw, Sask. 9-8-82.
Alternaria sp.	–		Necrotic leaf spots	Jameson, Sask. 18-8-82.
Alternaria sp. (or *Ulocladium* sp.)	(+)		Flower dieback	Jameson, Sask. 18-8-82.
Alternaria sp.	+		Top dieback	Jameson, Sask. 8-6-83.
Alternaria sp.	+		Flower dieback	Jameson, Sask. 23-6-83.
Alternaria sp.			Top dieback and necrotic leaf spots	Morden, Man. 12-7-83.
Alternaria sp.			Flower dieback	Cypress River, Mon. 13-7-83
Alternaria sp.			Necrotic leaf spots	Stockton Ferry, Man. 13-7-83.
Alternaria sp.			Necrotic leaf spots	Treesbank Ferry, Man. 13-7-83.
Alternaria sp.	(+)		Flower dieback	Rounthwaite, Man, 13-7-83.
Alternaria sp.			Leaves turning black	Caronport, Sask. 14-7-83.
Alternaria sp.			Leaves browning, stem and leaf petioles still green	Maxim, Sask. 19-7-83.
Alternaria alternate (Fr.) Keissler	(+)		Lesion at base of flower branch extending half up the branch	Caronport, Sask. 27-7-83.

* I. Wound test on stems; - = no effect; (+) = some discoloration with no or very slight lesion development in wound; + = some lesion development; ++ = plant part above wound wilting.
II. Spore suspension sprayed on plants; - = no effect; + = a few small lesions developed; ++ = severe leaf lesions developed.

Leafy spurge plants with disease symptoms were brought to the laboratory and analyzed for causal organisms. Plant material with distinct lesions or symptoms was surface sterilized in 0.6% sodium hypochlorite for 10 minutes, rinsed in sterile water and plated out on potato dextrose agar (PDA). If bacteria were suspected, the diseased plant material was cut up in small sections, placed in sterile water for 15 to 20 minutes and loopfuls of that water were streaked out on nutrient agar. Pathogenicity of isolated organisms was tested by wounding a stem area slightly with a scalpel, placing mycelium and/or spores or bacteria in the wound, wrapping the treated area with wet cotton and the treated plants were kept in a mist chamber for the following 18 to 24 hours. The plants were then left on greenhouse benches (temperature: 18-24º C., daylength: 14 hours, with cool fluorescence and incandescent light) for up to one month for regular inspection. Control plants, wounded and wrapped with cotton, were included with each test. In some instances additional pathogenicity tests were made by spraying a spore suspension onto undamaged plants, to test if a pathogen could enter and infect through the unwounded epidermis. Plants treated in this manner were kept for at least 48 hours in the mist chamber then moved to greenhouse benches. If no lesion development was observed after one month, the isolated fungi or bacteria were regarded non-pathogenic. Fungi that caused discoloration and lesion development were reisolated, compared with original cultures and sent to the Biosystematic Research Institute (B.R.I.), Agriculture Canada, Ottawa, or to Commonwealth Mycological Institute (C.M.I.), Identification Services, Kew, England, for identification.

Results and discussion

In most areas surveyed the leafy spurge population was found to be disease-free. The most prevalent disease problem observed was the *Alternaria* leaf spot and top dieback complex. The severity of the disease ranged from a few insignificant leaf spots to severe top dieback, occurring on about 10% of the plants. *Alternaria* spp. were consistently isolated from plants with such symptoms, although *Cladosporium, Fusarium* and other species of fungi were also frequently isolated. The latter were either non-pathogenic or only caused discoloration with almost no lesion development, whereas, most of the *Alternaria* spp. isolated were pathogenic (Table 1). All *Alternaria* spp. (except a few non-sporulating ones) produced conidia in chains. Considerable variation in amount of sporulation, color and type of mycelium was observed in cultures on PDA. Some cultures produced abundant aerial mycelium, whereas others had darker more flat mycelium. The latter usually produced more spores. Two species, *A. alternata* and *A. tenuissima*, were identified, but several, not identified to species by B.R.I. (Table 1), were distinctly different from *A. alternate* and *A. tenuissima*. *Alternaria* spp. have previously been reported from *Euphorbia* spp. in both Canada (Conners 1967) and in the United States (U.S.D.A. 1960, Krupinsky and Lorenz 1983).

Septoria leaf spot was widespread in Saskatchewan and was also found in Alberta and Montana, but not in Manitoba (Table 2). The severity of the disease varied from a few distinct leaf spots to larger lesions that coalesced, resulting in wilt of entire leaves. However, the attack was generally light and did little harm to the plants. Specimens sent to B.R.I. were identified as *Septoria* sp. similar to *S. jatrophae* Heald and Wolfe. Specimens sent to C.M.I. were identified as *Septoria guepini* Oudem. (Table 2). This is the first record of *Septoria* leaf spot on *Euphorbia* spp. in North America. *S. bractearum* Mont., *S. euphorbia* (Lasch.) Desm., and *S. guepini* Oudem. were reported on *Euphorbia* spp. in early literature from Europe (Harris *et al*. in preparation).

Powdery mildew was only detected from sites in interior British Columbia and from Jameson, Saskatchewan. Several mildew species have been reported on *Euphorbia* spp. (Harris *et al*. in preparation). Cleistothecia were not observed from either location and identification is difficult without the sexual stage. Powdery mildew is a serious problem under greenhouse conditions, but apparently does not do well under natural conditions on the prairies.

Stem and root rot were observed from several sites, but only on scattered plants. Plants affected showed stress and sometimes wilting of the entire plant. Several fungi were isolated of which *Fusarium* spp. were most frequent. *F. sporotrichioides* was only found in Manitoba and was the most pathogenic *Fusarium* sp. isolated. The higher pathogenicity agrees well with the more severe field symptoms observed from these sited (Table 3). Only *F. solani* and *F. acuminatum* have been reported from leafy spurge in Canada (Gordon 1959). *Rhizoctonia solani*, isolated from plants in the field as well as in the greenhouse with symptoms of root and stem rot, has previously been reported from *Euphorbia* in both Canada and the United Stated (Conners 1967, U.S.D.A. 1960). *Phomopsis euphorbia*, isolated from distinct lesions on stems of leafy spurge from Caronport, Saskatchewan has not previously been reported from North America. A hyphomycaete

(Cypress River, Man. 13-7-83) did not sporulate and so could not be identified. A few *Alternaria*-like spores in chains were observed immediately after isolation from diseased plant material, but the culture was different in appearance from the other *Alternaria* spp. isolated (Table1). This hyphomycete occurred on the upper part of the wilting leafy spurge stem, whereas *F. sporotrichioides* was isolated form lower parts of the stem. In wound tests this hyphomycete appeared slightly more pathogenic than the *F. sporotrichioides*. Perhaps both fungi are involved in the cause of the disease, which occurred severely in a patch about 5-6 meters in diameter, in contrast to only scattered plants as usually observed for stem and root rot (Table 3). *Curvularia inaequalis* and *Gliocladium roseum*, which showed discoloration with very slight lesion development on the wound tests, are common fungi, as are the nonpathogenic fungi, *Epicoccum pupurascens*, *Acremonium* sp., *Trichoderma* sp. And others frequently isolated. None of the bacteria isolated were found to be pathogenic and consequently were not identified.

Other symptoms observed on leafy spurge, from which no disease causing organisms were detected, are shown in Table 4. In the early part of the growing season in 1981 aborting flowers and slightly wilting bracts were observed at the Jameson site. Several fungi and bacteria were isolated, but none was found to be pathogenic (Table 4). These symptoms could possibly be caused by frost or cold weather conditions in the early part of the season.

Purple to reddish leaf spots and in some cases entire reddish plants were observed frequently. No disease-causing organisms were isolated from them. The phenomenon was especially common on sandy or poor soil where plants were stressed by drought. At some sites scattered wilting plants were observed; they did not show distinct stem lesions, rather a damaged area with a spongy appearance occurred at the base of the stem or just below soil level. No disease causing organisms were detected. At one site wilting plants occurred in an anthill.

At one site (Dundum, Sask. 30-6-82, Table 3) gray whitish worts or scabby symptoms appeared on a few plants in a roadside pasture. These only occurred in the epidermis, did not extend into the plant tissue, and apparently had no effect on the plants. *Alternaria* sp. and an ascomycete (not identified) were isolated but none of them caused any symptoms when tested on damaged epidermis of leafy spurge plants. The symptoms resembled oedema, which can be observed on some houseplants, caused by environmental factors.

Table 2. *Septoria* leaf spot and powdery mildew observed on leafy spurge from the prairies during the three-year period 1981 to 1983.

Fungi detected	Pathogen* test (lab.)	Field symptoms	Location and date
Septoria guepini Oudem.	+	Distinct leaf spots with brown margins and brownish centers	Cardston, Alta. 22-6-81.
Septoria guepini Oudem.	+	Distinct leaf spots with brown margins and brownish centers	Bethune, Sask. 2-7-81.
Septoria guepini Oudem.	+	Distinct leaf spots with brown margins and brownish centers	Regina Beach, Sask. 2-7-81.
Septoria guepini Oudem.		Distinct leaf spots with brown margins and brownish centers	Culbertson, Mont. 7-7-81.
Septoria guepini Oudem.		Distinct leaf spots with brown margins and brownish centers	Caronport, Sask. 16-6-82.
Septoria guepini Oudem.		Distinct leaf spots with brown margins and brownish centers	Silton, Sask. 17-6-82.
Septoria guepini Oudem.		Distinct leaf spots with brown margins and brownish centers	Zehner, Sask. 21-7-82.
Septoria guepini Oudem.		Distinct leaf spots with brown margins and brownish centers	Jameson, Sask. 18-8-82.
Septoria guepini Oudem.		Distinct leaf spots with brown margins and brownish centers	Jameson, Sask. 23-6-83.
Septoria guepini Oudem.		Distinct leaf spots with brown margins and brownish centers	Caronport, Sask. 14-7-83.
Erysiphe sp.		Powdery mildew, in reddish brown leaf spots	Kamloops, B.C. 26-6-81.
Erysiphe polygoni ? DC. ex /st. Amans.		Powdery mildew, from green-house plants	Regina, Sask. 15-12-81.
Erysiphe sp.		Powdery mildew and necrotic leaf spots	Jameson, Sask. 19-7-82.

*Spore suspension sprayed on leafy spurge plants; + = leaf spots developed.

Table 3. Stem and root rot observed on leafy spurge from the prairies during the three-year period 1981 to 1983.

Fungi and bacteria isolated	Pathogen.* test (lab.)	Field symptoms	Location and date
Epicoccum purpurascens Ehrenb.	–	Plant wilting, lesion at base of stem	Jameson, Sask. 10-6-81.
Trichoderma sp.	–	Lesion at base of stem	Langham, Sask. 5-6-81.
Fusarium acuminatum Ell. and Everh.	(+)	Distinct lesion at base	Caronport, Sask. 22-6-81.
Bacteria (not identified)	–		
Fusarium acuminatum Ell. and Everh.	(+)	Distinct lesion starting at base of stem, extending up one side, other side green	Cardston, Alta. 22-6-81.
Epicoccum purpurascens Ehrenb.	–		
Bacteria (not identified)	–		
Fusarium tricinctum (Corda) Sacc,	(+)	Lesion at base of stem (somewhat constricted)	Caronport, Sask. 6-8-81,
Gliocladium roseum (Link) Bainer	(+)		
Bacteria (not identified)	–		
Curvularia inaequalis (Shear) Boedjin	(+)	Lesion at base of stem (somewhat constricted)	Caronport, Sask. 16-6-82.
Acremonium sp.			
Fusarium solani (Mart.) Sacc.	–		
Fusarium tricinctum (Corda) Sacc.	(+)	Plant wilting, lesion at base of stem	Zehner, Sask. 21-7-82.
Alternaria sp.	(+)		
Epicoccum purpurascens Ehrenb.	–		
Alternaria sp.	(+)	Lesion at base of stem (somewhat constricted)	Caronport, Sask. 28-7-82.
Epicoccum purpurascens Ehrenb.	–		
Fusarium sporotrichioides Sherbakoff	+	Entire stem wilting from about 10 cm above soil level and up	Cypress River, Man. 13-7-83.
Hyphomycete (grayish culture, not sporul.)	+		
Rhizoctonia solani Kuhn	++	Gray stem lesion at soil level, entire plant wilting	Spruce Wood, Man. 13-7-83.
Rhizoctonia solani Kuhn	+	Rootstock rot (greenhouse)	Regina, Sask. 21-3-83.
Fusarium sporotrichioides Sherbakoff	+	Lesion at base of stem (entire plant wilting)	Stockton Ferry, Man. 13-7-83.
Fusarium sporotrichioides Sherbakoff	+	Lesion at base of stem (entire plant wilting)	Treesbank Ferry, Man. 13-7-83.
Fusarium solani (Mart.) Sacc.	(+)	Distinct lesion (light brown at base of stem of small plant)	Caronport, Sask. 14-7-83.
Fusarium equiseti (Corda) Sacc.	(+)		
Fusarium oxysporum Schlect.	(+)		
Phomopsis euphornbiae (Sacc.) Trav.	+	Lesion at base of stem, extending up one side (core of stem had brownish discoloration)	Caronport, Sask. 14-7-83.

*Wound test on stem; – = no effect; some discoloration with no or very slight lesion development;
+ = some lesion development; ++ = plant part above wound wilted.

Table 4. Physiological disorders observed on leafy spurge from the prairies during the three-year period 1981 to 1983.

Fungi and bacteria isolated	Pathogen.* test (lab.)	Field symptoms and comments	Location and date
Fusarium equiseti (Corda) Sacc.	(+)	Aborting flowers, frost ?	Jameson, Sask. 30-5-81.
Epicoccum purpurascens Ehrenb.	(+)		
Bacteria (not identified)	–	Aborting flowers, frost ?	Jameson, Sask. 30-5-81.
Helminthosporium sp.	*(+)*	Aborting flowers, frost ?	Jameson, Sask. 10-6-81.
Cladosporium sp.	(+)		
Epicoccum purpurascens Ehrenb.	(+)		
Cladosporium sp.	–	Flowers and flower branches wilting, frost ?	Jameson, Sask. 10-6-81.
Alternaria sp.	–		
Rhizoipus sp.	–		
Epicoccum purpurascens Ehrenb.	–		
None		Purple spots on leaves, physiological stress ?	Jameson, Sask. 10-6-81.
None		Brownish discoloration of leaves, physiological stress ?	Caronport, Sask. 16-6-82.
None		Reddish brown leaf spots, physiological stress ?	Jameson, Sask. 18-8-82.
None		Purple spots on upper leaves, physiological stress ?	Jameson, Sask. 30-6-83.
Secondary fungi (not identified)	–	Gray whitish scabby appearances on stems, not affecting plant, cause not detected	Dundurn, Sask. 30-6-82.
Secondary fungi (not identified)	–	Plants wilting in a patch, epidermis somewhat damaged at base of stem, cause unknown	Silton, Sask. 17-6-82.
None		Plants wilting in ant hill, stem epidermis damaged at soil level, cause unknown	Sask. Beach, Sask. 17-6-82.
Secondary fungi	–	Wilting tops, stem near soil level swollen, epidermis at soil level damaged, cause unknown	Jameson, Sask. 23-6-83.

*Tested by placing bacteria or mycelium directly on flower parts and kept moist under plastic bag for 2-3 days;
(+) = resulted in slight necrotic development on flower petals and bracts; - = no effect.

Conclusion

Several fungi were isolated from disease symptoms on leafy spurge plants in the prairie provinces. Most of the fungi isolated from plants with stem and root rot symptoms listed in Table 3 were non-pathogenic or weakly pathogenic and probably did not individually cause the symptoms observed. Perhaps the rot was induced by a complex of several fungi together, with environmental conditions. *Septoria* leaf spot and powdery mildew (Table 2) appeared to do little damage to leafy spurge plants in nature. Furthermore, they are difficult to culture in large quantities (powdery mildew is an obligate parasite and *Septoria* sp. was difficult to culture on agar media). Thus these fungi would be of little value as inundative biological control agents.

There is no doubt that *Alternaria* spp. caused the leaf spot and top dieback (Table 1). Symptoms observed at some sites were severe, especially in 1983, where up to 10% of the plants were attacked. Some of the *Alternaria* spp. showed good pathogenicity when spore suspensions were sprayed on leafy spurge plants and kept in a mist chamber for 3-4 days (Table 1), and thus might have potential as inundative biocontrol agents. However, a spore suspension of a culture (Caronport, Sask. 1-6-82), sprayed on a field stand of leafy spurge, did not result in infection. Perhaps these *Alternaria* spp. require too much moisture to develop consistently on the prairies. This would explain why leaf spot and top dieback appeared to be more severe in 1983, when there was above average precipitation in early July, than in the two previous years.

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