Mycosphaerella tassiana on Cruciferae in Western Canada

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A species of **Mycosphaerella** commonly found in Western Canada on overwintered stubble of rape (**Brassica napus**) and turnip rape (**B. campestris**) and stems of weed species of the Cruciferae, Compositae and Chenopodiaceae has been identified as **M. tassiana** var. **tassiana**. The fungus in question is distinct from that causing ring spot of **B. oleracea**, **M. brassicicola**, authentic collections of which have apparently not been found to date on rape in Western Canada. Ascospore discharge in **M. tassiana** usually began during the first week of June, reached a peak by the third week of June, and then rapidly declined.

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Une espece de **Mycosphaerella** generalement trouvee au printemps dans l'ouest du Canada sur les chaumes de colza (**Brassica napus**) et de navette (**B. campestris**) et sur des tiges d'espèces adventices de **Cruciferes. Composées** et **Chénopodiacées** a ete identifiée comme etant **M. tassiana** var. **tassiana**. Ce champignon est different de **M. brassicicola**, agent de la tache annulaire chez **B. oleracea** dont on n'a vraisemblablement pas encore trouve de veritable specimen sur colza dans l'ouest du Canada. La decharge des ascospores de **M. tassiana** commence generalement la premiere semaine de juin, atteint son sommet au cours de la troisième semaine pour diminuer ensuite rapidement.

In 1958, Vanterpool collected in east-central Saskatchewan a disease of turnip rape (Brassica campestris L.) which was characterized by rather striking ashen grey stem lesions (Vanterpool, '1960). The disease appeared to be ring spot, which on this continent was reportedly confined exclusively to moist coastal regions of Washington, Oregon and California (Nelson and Pound, 1959). Although spermogonia were found in abundance in lesioned parts of the stems of the Saskatchewan material, the ascosporic state, Mycosphaerella brassicicola (Fr. ex Duby) Lind. was the object of an extensive but fruitless search over many years in Western Canada. However, since 1970, a species of Mycosphaerella has been collected on a number of occasions on overwintered rape stems both in Saskatchewan and Alberta. A study of this species was undertaken to determine whether it was in fact M. brassicicola. The widespread occurrence of ascocarps of the ring spot pathogen in the rape-growing area of Western Canada could be of considerable epidemiological significance. The results of these investigations are described in this paper.

Methods

Single ascospores cultures were secured in the following manner. Short pieces of infected rape stems were taped to the undersides of petri dish lids which were then placed over petri dish bottoms containing 15-20 ml of

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2% water agar. Following ascospore discharge, isolated single spores of *Mycosphaerella* were aseptically removed to plates of V8 juice agar (V8A) containing, per liter, 200 ml V8 juice, 0.75 g calcium carbonate and 20 g agar. They were then compared with V8A cultures of various fungi, including *Cladosporium herbarum* Lk., *Pseudocercosporella capsellae* (Ell. & Ev.) Deighton, and *Mycosphaerella brassicicola.* A culture of the lastnamed species was obtained from Dr. P. H. Williams, University of Wisconsin, Madison. Ascocarps from field material were also crushed on microscope slides in drops of lactophenol-aniline blue and ascospores of *Mycosphaerella* measured at a magnification of 800X.

The ascospore discharge pattern of the species of *Mycosphaerella* was studied in ascospore liberation tunnels over a three-year period (1975-77) using a method similar to that of McGee (1977). Samples consisting of short seg'ments of stems were stored out-of-doors. Each sample following thorough wetting with tap water was kept in a discharge tunnel for 1.5 h and spores collected on vaselined slides. Spore trapping was carried out at 4-week intervals.

Conidial suspensions from pure single ascospore cultures of the **Mycosphaerella** were atomized onto the leaves, flowers and young pods of plants of **B. napus** cv. Midas growing in the greenhouse, and the plants covered with plastic bags for periods of up to several days. Following uncovering, inoculated plants were examined periodically for several weeks for signs of infection.

Results

Between 1970 and 1977, many single ascospore cultures of the *Mycosphaerella* sp. were obtained from overwintered stems of turnip rape (*Brassica campestris* L.) and rape (*B. napus* L.) from the Peace River and

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Species	Source	Ascospore measurements (μ)	
		Average (L x W)	Range (L x W)
M. tassiana	Saskatchewan collections*		
var . tassiana	No. 16, Brassica sp.	22.6 x 7.4	18.2 - 32.5 x 6.6 - 8.8
	No. 78, <i>Brassica</i> sp.	24.1 x 7.2	20.4 - 28.6 x 6.1 - 8.3
	No. 109, <i>Brassica</i> sp.	23.2 x 7.8	20.9 - 30.8 x 6.6 - 9.4
	No. 5, Sisymbrium loeselii	22.0 x 6.9	17.6 – 27.0 x 5.5 – 7.7
	No. 12, Sonchus sp.	20.0 x 6.7	13.2 – 24.4 x 4.4 – 7.7
	No. 13, Tragopogonpratensis	21.1 x 7.0	18.7 – 26.4 x 5.5 – 7.7
M. tassiana			
var. <i>tassiana</i>	Wehmeyer (1963)		16 – 29 x 4.5 – 8.0
M. brassicicola	Dring (1961)		18-20-23 x 3.5 - 4.0
	Osmun and Anderson (1915) Weimer (1926)	24.5 x 4.3	15 — 25 x 3.5 — 5.5

Table 1. A comparison of reported dimensions of ascospores of two *Mycosphaerella* species with those of Saskatchewan collections of *Mycosphaerella* from Cruciferae and Compositae

Approximately 50 spores mounted in lactophenol-cotton blue were measured for each collection.

Lacombe areas of Alberta and, on several occasions, from the vicinity of Saskatoon, Saskatchewan. The ascospores were typically bicellular, but a second septum was frequently observed in germinating spores. All the isolations yielded only cultures identical with *Cladosporium herbarum,* the imperfect state of *Mycosphaerella tassiana* (de Not.) Johans. (Barr, 1958). They did not resemble the isolates of *Pseudocercosporella capsellae* or *Mycosphaerella brassicicola.* Unlike the typical greenish *Cladosporium* cultures, those of these two species were black to grey in color, considerably raised in the center, extremely restricted in their linear growth, and contained, in the case of P. *capsellae,* both spermogonia and cylindric to obclavate-cylindric conidia.

The species of Mycosphaerella was subsequently collected on overwintered stems of the following: Arabis holboellii Hornem., Brassica campestris. B. napus L., Axyris amaranthoides L., Cirsium sp., Descurainia sophia (L.) Webb, Erysimum asperum DC., E. parviflorum Nutt., Sisymbrium loeselii L., Sonchus sp., Thlaspi arvense L., and Tragopogon pratensis L. Ascospore dimensions are compared in Table 1 with those reported in the literature for *M. brassicicola* (Dring, 1961; Osmun and Anderson, 1915; Weimer, 1926) and M. tassiana var. tassiana (Wehmeyer, 1963). Although spore lengths from the Saskatchewan collections did not differ greatly from those reported for *M. brassicicola*, spore widths were consistently greater than those of that species and approximated those of *M. tassiana* var. tassiana. In fact, the morphology of the unknown species resembled that of the latter very closely.

In spring, *Cladosporium herbarum* often appears on rape stubble as a black sooty mould on shiny black, often

lens-shaped lesions extending from the stem base to a few cm above it or higher. This is the portion of the stem which often remains wet for a prolonged period during and after snow melt. It is from such material that ascospores of *M. tassiana* have been subsequently obtained. Striking symptoms of this type were observed in rape fields near Irma and Edgerton, Alberta, in May, 1973, and near Waldheim, Saskatchewan, in the early spring of 1975.

Ascospore discharge in *M. tassiana* generally increased rapidly from virtually nil in early June to a maximum during the third week of that month, and quickly declined thereafter (Fig. 1). In a few instances the high level of discharge was maintained until late July or early August.

The inoculated Midas plants remained free of symptoms for the duration of the greenhouse experiments. *C. herbarum* has been implicated as a cause of "pod drop" of rape (Petrie, 1973). However, blackening of the peduncles at the base of the pods, the symptom characteristic of pod drop, was not observed.

Discussion

It would appear that no authentic ascocarps of *M. brassicicola* have been found in the rape-growing area of Western Canada since "ring spot" was first described here (Vanterpool, 1960), nor has any other perfect state been linked to the disease as it occurs in this area. In 1968, the discovery of a *"Cercospora"* conidial state of "ring spot" was found in Saskatchewan. These and other pieces of evidence cast doubt on there being any connection of the disease, renamed "white leaf spot and grey stem", with *M. brassicicola* (Petrie and Vanterpool, 1975). The correct name of the conidial state of white

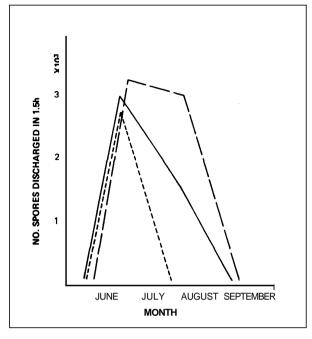


Figure 1. Ascospore discharge patterns for three representative collections of *Mycosphaerella tassiana* var. *tassiana* on rape stubble from Saskatchewan.

leaf spot and grey stem is *Pseudocercosporella capsel-*/*ae* (Ell. & Ev.) Deighton (Deighton, 1973). This subject will be examined in more detail in a later publication.

The species of Mycosphaerella commonly found on rape and various weed species in Western Canada would seem to be the ubiquitous M. tassiana var. tassiana. Evidence for this is the morphology of the perfect and imperfect states and the unequivocal demonstration of a connection between the two. The fungus in culture is totally unlike P. capsellae or M. brassicicola. The host range of the Mycosphaerella from rape also is much broader than that of *M. brassicicola* which is reportedly confined to varieties of Brassica oleracea L. (Dring, 1961; Weimer, 1926). M. tassiana var. tassiana was found on a number of dicotyledons in the present study; it also has recently been reported as part of the air spora over brome grass crops in the Saskatoon area (Shoemaker et al., 1974). Conners (1967) lists numerous hosts of this species.

The significance of **M.** tassiana on rape is open to speculation. Its imperfect state, **C.** herbarum, is common in seed samples of turnip rape and rape. It has been

associated with cotyledon yellowing of rape seedlings (unpublished data). In conjunction with *Alternaria alternata* (Fr.) Keissler it develops profusely on swathed rape under moist conditions and has been isolated from hypertrophies of the stem and inflorescence of turnip rape caused by *Albugo candida* (Pers. ex Lev.) Ktze. (*A. cruciferarum* S. F. Gray) (Petrie and Vanterpool, 1974). There is no evidence that *M. tassiana* is a pathogen of Cruciferae despite its association with pod drop of rape. Whether or not it in fact contributes in any way to the latter condition has not been satisfactorily demonstrated.

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