VARIATION IN ISOLATES OF DIDYMELLA APPLANATA¹

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Introduction

During the years from 1949 to 1957, considerable variation was observed in the susceptibility of commercial raspberry varieties to spur blight, caused by <u>Didymella applanata</u> (Niessl) Sacc. Most of the varieties grown in Ontario and Quebec exhibited moderate to severe symptoms in years when there were prolonged humid conditions during May and early June. Isolations made from spur blight lesions on these varieties yielded a single cultural type of the organism which remained quite stable in culture. During this time, the variety Newburgh was grown beside the severely infected varieties without becoming infected. However, in 1958, Newburgh, growing beside the susceptible Rideau variety, became severely infected and developed typical spur blight lesions. This observation led to work on determining variation in pathogenicity.

Cultural differences among 3 isolates

D. applanata was isolated from commercial and wild raspberries growing—in Ontario and Quebec. Over 50 isolations from 22 localities yielded 3 fairly distinct cultural types. Differences in colony type and in microscopic characters were observed in culture. An isolate from wild raspberries, referred to as isolate #1 in this paper, produced a spreading depressed type of growth with abundant pycnidia produced in clumps (Fig. 1A). The medium (P.D.A.) became slightly discolored in 10 days. This isolate was fastgrowing and covered a petri plate in 28 days.

Isolate #2, from the commercial varieties, Trent, Rideau, Madawaska, Muskoka, Viking, September, Tweed, Latham, Carnival, Gatineau, and Ottawa yielded colonies that were more limited in lateral spread, produced more abundant aerial mycelium, and fewer pycnidia. This isolate also discolored the medium (Fig. 1B).

Isolate #3, from the variety Newburgh, produced a colony quite different from the other two. Very little aerial mycelium was produced. Pycnidla were produced in great quantity giving the colony a very dark appearance. The medium remained uncolored (Fig. 1C).

Isolates #1 and #2 produced pycnidia in 10 days. These were similar in color and wall consistency, being light-brown at first, and becoming black after 25 days. The walls were quite soft and did not become brittle until the cultures were 4 weeks old. Pycnidia produced by isolate #2 were considerably larger and more globose than those formed by isolate #1 (Table 1). Conldia produced by the 2 isolates fell in approximately the same size range, but the average size of those of isolate #2 was slightly larger.

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Fig. 1. A-Isolate #] B-Isolate # 2 C-Isolate # 3

Isolate #3 produced large numbers of pycnidia after 5 days. These were much more numerous than in cultures of the other isolates, and were considerably smaller (Table 1). They were dark in color and brittle in wall texture at this time. The conidia were considerably larger than those of the other 2 isolates and were exuded more freely onto the surface of the medium. The relative differences in measurements of pycnidia and conidia were also evident on the hosts (Table 2).

Table 1.	Size and number of pycnidia and size of conidia produced on P.D.A.
	3 isolates of D . a <u>pplanata</u> (28-day old cultures)

Number of		Size of pycnidia (p)		Size of conidia (p)	
T 1 /	pycnidia				
Isolate	per sq. cm.	range	average	range	average
#1	120	85-190x135-255	135x190	2.7-3.4x6.0	-6.7 2.8x6.4
#2	40	205-340x235-340	225x250	2.7-4.0x4.7	-6.7 3.3x6.6
#3	1400	75-185x 85-210	105x135	$2.0-4.0 \times 6.0$	-13.4 3.7x8.1

	Size of p	Size of pycnidia (u)		Size of conidia (u)	
Isolate	range	average	range	average	
#1	110-225x160-275	160x210	$2.7 - 3.8 \sim 6.0 - 7.2$	3.1x6.7	
#2	205-380x240-370	240x260	2.8-4.0x5.2-7.4	3.4x6.8	
#3	75-195x105-250	115x150	2.6-4.5~6.5-14.0	3.7x8.4	

Table 2. Size of pycnidia and conidia produced on raspberry canes after inoculation with 3 isolates of **D**. applanata.

Differences in life cycle

Several canes of the varieties Trent, Madawaska, Rideau and Newburgh, growing in pots, were inoculated with a conidial suspension of D, <u>applanata</u>. Isolates **#1** and **#2** produced severe lesions on the canes of Trent, Madawaska, and Rideau. Isolate **#3** produced a few lesions on all varieties. The plants were placed outdoors over winter and examined in the spring. The isolates were recovered from their respective hosts. Perithecia were found on the canes inoculated with isolates **#1** and **#2**. Only pycnidia were produced on the canes inoculated with isolate **#3**. It is probable that this isolate reproduces in nature by conidia only and does not employ the perfect stage in its life cycle. Perithecia were found in large quantities in the field on all varieties except Newburgh. Cultures from these were always of the type of isolate **#2**.

Pathogenicity

All 3 isolates were capable of causing infection on raspberry canes. Canes of the varieties Newburgh, Carnival, Madawaska, Trent, and Latham were infected by the 3 isolates when the bark was damaged and a conidial suspension introduced by means of a hypodermic needle. When the conidia were applied to uninjured canes and leaves with an atomizer, the results shown in Table 3 were observed. Isolate #3 caused severe leaf lesions and defoliation on the 5 varieties, whereas very few leaf lesions were produced after inoculation with isolates #1 and #2.

Inoculum used	Newburgh	Carnival	Madawaska	Trent	Latham
Isolate #1	0	S	S	S	s
Isolate #2	0	М	S	S	S
Isolate #3	S	S	S	S	S

Table 3. Results of inoculating 5 raspberry varieties with conidial suspensions of 3 isolates of **D**. <u>applanata</u>.

0 = no infection M = moderate infection S = severe infection

From Table 3, it can be concluded that isolates #1 and #2 were different from isolate #3 in ability to infect Newburgh. This difference in pathogenicity leads one to belive that isolate #3 constitutes a different race or possibly a new form since there are differences both in pathogenicity and morphology.

Discussion

Races of the fungus, <u>D</u>. <u>applanata</u> must be considered in breeding for spur blight resistance in raspberries. Since isolations made from wild raspberries in several localities in Ontario and Quebec yielded only one biotype, it is quite likely that, among the wild plants in this area, only one race occurs. Since no differences in pathogenicity were observed between this isolate and the common isolate from commercial varieties, isolates #1 and #2 described above must be considered to represent a single race. Newburgh, being resistant to this race, probably contains one or more genes for resistance to it, but not to the race described in this paper as isolate #3. It would seem, then, that additional races of the fungus may be found among isolates of <u>D</u>. <u>applanata</u> from other parts of the world where numerous varieties of red raspberry are grown. Work is being continued on this phase of the problem.

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