Virulence Differences Between *Fusarium roseum* 'Acuminatum' and *F. roseum* 'Avenaceum' in Red Clover

J. C. Stutz and K. T. Leath

Graduate assistant and adjunct professor, respectively, Department of Plant Pathology, Pennsylvania State University, University Park 16802. Second author is also a research plant pathologist, USDA-ARS.
Current address of senior author: assistant professor, Division of Agriculture, Arizona State University, Tempe 85287.
Contribution 1387, Department of Plant Pathology, The Pennsylvania Agricultural Experiment Station. Authorized for publication as Journal Series Paper 6617, and Contribution 8306 from the U.S. Regional Pasture Research Laboratory, University Park, PA 16802.
Mention of a trademark, vendor, or proprietary product does not constitute a guarantee or warranty of the product by the U.S. Department of Agriculture and does not imply approval to the exclusion of other products that also may be suitable.
Accepted for publication 22 June 1983.

ABSTRACT

Stutz, J. C., and Leath, K. T. 1983. Virulence differences between *Fusarium roseum* 'Acuminatum' and *F. roseum* 'Avenaceum' in red clover. Phytopathology 73:1648-1651.

The virulence of isolates of *Fusarium roseum* 'Acuminatum' was compared to that of isolates of *F. roseum* 'Avenaceum' in roots of 5-wk-old red clover plants and also in red clover seedlings. Penetration and colonization of roots by isolates of the two pathogens were observed. In 5-wk-old plants, isolates of *F. roseum* 'Avenaceum' were more virulent than isolates of *F. roseum* 'Acuminatum' and caused more frequent and severe necrosis at all inoculation sites above the root tip. There were distinct differences in fungal development between the two pathogens in roots of 5-wk-old plants inoculated at least 2 cm from the root tip. *F. roseum* 'Avenaceum' colonized the cortex via distributive hyphae, and caused necrosis in this area. *F. roseum* 'Acuminatum' penetrated the epidermis in this area, but further colonization was limited to the formation of chlamydospores in the cortex. Neither distributive hyphae nor necrosis was observed. When 5-wk-old plants were inoculated at the root tip, both pathogens penetrated and colonized the root, formed distributive hyphae and caused necrosis. In inoculated seedlings, there were no differences in virulence between isolates of *F. roseum* 'Acuminatum' and 'Avenaceum' as measured by frequency and length of necrosis. Both pathogens formed distributive hyphae in the cortex of seedling roots.

Fusarium roseum (Lk.) emend. Snyd. and Hans. 'Acuminatum' and *F. roseum* (Lk.) emend. Snyd. and Hans. 'Avenaceum' are often among the *Fusarium* species isolated from root and crown tissue of red clover plants with cortical root rot (9,11,18,20). Determining from previous research whether differences in virulence exist between these two groups in red clover is difficult. Many experiments designed to screen isolates of *F. roseum* 'Acuminatum' and 'Avenaceum' for pathogenicity and virulence were performed by inoculating red clover seedlings (9,18,20). Unfortunately, red clover seedlings are often more susceptible to these fungi than are mature plants (4,6). Although investigations have shown differences between isolates of *F. roseum* in older red clover plants (4,6), the isolates tested were not identified to be a particular cultivar of *F. roseum*. Experiments that confirmed the pathogenicity of *F. roseum* 'Acuminatum' and 'Avenaceum' in older red clover plants were conducted with too few isolates to determine if real virulence differences exist (11).

The objectives of this research were to determine if and how, as a group, *F. roseum* 'Acuminatum' differed from *F. roseum* 'Avenaceum' in virulence to red clover roots. Differences in virulence between isolates were determined by measuring the length of necrosis in inoculated red clover roots and by testing the ability of isolates to cause necrosis at inoculation sites above the root tip. The nature of any differences in virulence was examined by observing fungal penetration and colonization of red clover roots.

MATERIALS AND METHODS

Isolates of *F. roseum* 'Acuminatum' and *F. roseum* 'Avenaceum,' originally isolated from diseased plant tissue, mostly red clover (Table 1), were maintained in the culture collections of the U.S. Regional Pasture Research Laboratory, University Park, PA, and the Fusarium Research Center at Pennsylvania State University. Identification of the isolates was verified by experienced personnel at the Fusarium Research Center. Isolate 959 had previously been identified as *F. roseum* 'Acuminatum' (11). Reexamination of this isolate revealed this identification to be in error, and the identity of isolate 959 has now been verified to be *F. roseum* 'Avenaceum.' Cultures, started from single spores, were stored in lyophilized colonized carnation leaves (5) and grown on V-8 juice agar (13). Isolates 927, 959, and 1055 had previously been shown to be pathogenic (11). All other isolates were screened for pathogenicity by making root tip inoculations as specified in the slant-board technique to eliminate nonpathogenic isolates (11). Inoculum was prepared by growing the fungi on autoclaved white 100% cotton threads, 1.5 cm long, laid on the surface of V-8 juice agar plates. After 6 days, the threads, covered with mycelium and occasional macroconidia, were lifted from the agar and used to

TABLE 1. Isolates of *Fusarium roseum* 'Acuminatum' and *Fusarium roseum* 'Avenaceum' used for red clover root inoculation

Cultivar and isolate	Original host	Original location
F. roseum 'Acuminatum'		
Isolate 927[a]	*Medicago sativa*	Pennsylvania
Isolate 1055[a]	*Coronilla globosum*	Pennsylvania
Isolate 5207[b]	*Trifolium pratense*	Wisconsin
Isolate 5653[b]	*Trifolium pratense*	Wisconsin
Isolate 5654[b]	*Trifolium pratense*	Wisconsin
Isolate 5655[b]	*Trifolium pratense*	Wisconsin
F. roseum 'Avenaceum'		
Isolate 959[a]	*Trifolium pratense*	West Virginia
Isolate 5181[b]	*Trifolium pratense*	Nova Scotia
Isolate 5215[b]	*Trifolium pratense*	Wisconsin
Isolate 5702[b]	*Trifolium pratense*	Vineland, Canada
Isolate 5703[b]	*Trifolium pratense*	Vineland, Canada

[a] Isolate identification number from the U.S. Regional Pasture Research Laboratory culture collection, University Park, PA.
[b] Isolate identification number from the Fusarium Research Center culture collection, The Pennsylvania State University.

The publication costs of this article were defrayed in part by page charge payment. This article must therefore be hereby marked "*advertisement*" in accordance with 18 U.S.C. § 1734 solely to indicate this fact.

inoculate plant roots. All cultures were maintained in the dark at 18 C. Both 4-day-old red clover (*Trifolium pratense* L. 'Arlington') seedlings and 5-wk-old plants were inoculated by laying an individual colonized thread over a plant radicle or root. Colonized threads were positioned at selected distances above the root tip, which are referred to as inoculation sites.

Seedlings were grown on 1.5% water agar from seeds that were treated for 30 sec in 95% ethyl alcohol, 10 min in 1% sodium hypochlorite, and then washed with sterile distilled water. At 4 days after emergence of the cotyledons, five seedlings were placed in sterile petri plates on No. 2 filter paper soaked with 4 ml of half-strength Hoagland's solution (7). At this stage, the mean radicle length was 2.6 ± 0.5 cm. The seedlings were inoculated midway between the radicle tip and the hypocotyl. Petri plates were incubated flat, and radicles were shaded from light by wrapping the lower half of the plates with aluminum foil. All seedlings were grown in a chamber with a 12-hr daily photoperiod, a light intensity from cool-white fluorescent lamps of 30 $\mu Ein \cdot m^{-2} \cdot sec^{-1}$, and a constant temperature of 22 C.

Seedlings were inoculated with isolates 927 and 5207 of *F. roseum* 'Acuminatum' and isolates 959 and 5215 of *F. roseum* 'Avenaceum.' Forty-five seedlings were inoculated with each isolate. Observations were made at 2, 4, and 6 days, and the length of necrosis was measured and the frequency of necrotic lesions determined.

Five-week-old plants were grown by using the slant-board technique (8), and all methods were as previously described (11) unless otherwise noted. For inoculations, the roots were spread fanlike on individual slant-boards, and 10 roots per plant were inoculated. The length and frequency of necrosis were determined 6 days after inoculation.

The effect of inoculation site on the virulence of *F. roseum* 'Acuminatum' and *F. roseum* 'Avenaceum' was compared in 5-wk-old plants inoculated at 0, 1, 2, 3, and 5 cm above the root tip with isolates 927 and 1055 of *F. roseum* 'Acuminatum' and isolates 959 and 5215 of *F. roseum* 'Avenaceum.' Each plant was inoculated with two isolates. Six replicates of 16 plants each were observed.

Five isolates of *F. roseum* 'Acuminatum' and five isolates of *F. roseum* 'Avenaceum' were compared for ability to cause necrosis at the 4-cm inoculation site. Ten roots of each plant were inoculated individually, at random, with all 10 isolates. Twenty-five plants were inoculated. After necrosis measurements, stained roots were examined for internal hyphae at ×400.

Root inoculation sites were characterized by examination at ×40, and measurements of the length of the root cap and meristematic zone, region of cell elongation, region of cell differentiation, and the area of secondary root formation were determined with 168 uninoculated roots from 20 plants used as controls.

Root tissue for microscopic observation was stained by boiling in 0.015% aniline blue lactophenol for 4 min (19, with decreased stain concentration). Seedling radicles for microscopic examination were inoculated with isolates 927 and 5207 of *F. roseum* 'Acuminatum' and isolates 959 and 5215 of *F. roseum* 'Avenaceum.' For each isolate, 10 roots were harvested at 6, 12, 24, 48, 72, and 96 hr after inoculation. Roots of 5-wk-old plants were inoculated at the root tip and at the 4-cm inoculation site with isolate 927 of *F. roseum* 'Acuminatum' and isolate 959 of *F. roseum* 'Avenaceum' and harvested at 3, 6, 12, 18, and 24 hr and every 24 hr for the first 5 days after inoculation. Twelve roots were observed for each isolate at each time period. Roots were observed at ×400 for fungal penetration of root epidermal cells and colonization of the root cortex.

Statistical significance was determined for all experiments by analysis of variance of a randomized complete block design at $P = 0.05$.

RESULTS

The test isolates of *F. roseum* 'Acuminatum' and *F. roseum* 'Avenaceum' consistently caused necrosis of 4-day-old seedlings (Table 2). By 6 days after inoculation, almost all of the seedlings had brown necrotic lesions that extended from the inoculation site toward the root tip and hypocotyl. Isolates did not differ in the frequency of necrosis caused at any of the three sampling periods.

All four isolates developed similarly in roots of red clover seedlings. By 12 hr after inoculation, all isolates had directly penetrated the epidermis of more than 50% of the seedling roots examined. No appressoria were formed, and penetration was intercellular and intracellular. Between 24 and 48 hr, hyphae of all isolates had begun to colonize the root cortex in all roots examined. Colonization was intercellular at first, and then intracellular. After extensive colonization of the root cortex, all isolates formed long parallel distributive hyphae (3), which grew longitudinally in the intercellular spaces of the root cortex, proceeding away from the inoculation site toward the hypocotyl and root tip. By 72 hr, distributive hyphae were present in more than 50% of the roots inoculated with isolates 959 and 5215 of *F. roseum* 'Avenaceum.' In comparison, isolates 927 and 5207 of *F. roseum* 'Acuminatum' required 120 hr before the majority of roots contained distributive hyphae.

In 5-wk-old plants, all isolates caused a more frequent and more extensive necrosis when inoculated at the root tip or at the 1-cm site than at sites further above the root tip (Table 3). When roots were inoculated above the 2-cm site with isolates 927 and 1055 of *F. roseum* 'Acuminatum' less than 10% of the inoculated roots had necrotic lesions at 6 days. In contrast, over 60% of the roots inoculated similarly with isolate 959 of *F. roseum* 'Avenaceum' and over 95% of the roots inoculated at the 2-cm site with isolate 5215 of *F. roseum* 'Avenaceum' had necrotic symptoms. At all inoculation sites, the two isolates of *F. roseum* 'Avenaceum' caused a significantly greater length of necrosis than the two isolates of *F. roseum* 'Acuminatum.' Only isolate 959 of *F. roseum* 'Avenaceum' caused a significantly higher frequency of necrosis at the 5-cm inoculation site in comparison to the 2- and 3-cm inoculation sites.

Roots of 5-wk-old uninoculated control plants had a well defined root cap and meristematic zone at the root tip. The region of cell elongation started at 0.08 ± 0.02 cm and extended to the region of cell differentiation, which started at 0.92 ± 0.24 cm from the root

TABLE 2. Frequency and length of necrosis on roots of 4-day-old red clover (cultivar Arlington) seedlings inoculated with isolates of *Fusarium roseum* 'Acuminatum' and 'Avenaceum'

	Day 2[x]		Day 4		Day 6	
Cultivar and isolate	Frequency[y] (%)	Length (mm)	Frequency (%)	Length (mm)	Frequency (%)	Length (mm)
F. roseum 'Acuminatum'						
Isolate 927	31.0 a[z]	2.0 b	66.7 a	5.9 c	97.6 a	15.3 b
Isolate 5207	42.6 a	1.9 b	81.0 a	7.6 bc	100.0 a	18.8 a
F. roseum 'Avenaceum'						
Isolate 959	50.0 a	3.8 a	85.7 a	11.1 a	100.0 a	18.6 a
Isolate 5215	40.5 a	3.1 ab	78.6 a	10.0 ab	100.0 a	20.4 a

[x] Days after inoculation when measurement occurred.
[y] Percentage of inoculated roots that had necrotic lesions.
[z] Different letters indicate that values are significantly different ($P = 0.05$) according to Duncan's multiple range test.

TABLE 3. Frequency and length of necrosis on roots of 5-wk-old red clover (cultivar Arlington) plants inoculated with isolates of *Fusarium roseum* 'Acuminatum' and 'Avenaceum' at different inoculated sites

Inoculation site (centimeters above the root tip)	*F. roseum* 'Acuminatum'				*F. roseum* 'Avenaceum'			
	Isolate 927		Isolate 1055		Isolate 959		Isolate 5215	
	Frequency[y] (%)	Length (mm)	Frequency (%)	Length (mm)	Frequency (%)	Length (mm)	Frequency (%)	Length (mm)
0	83.3 a[z]	4.1 a	64.8 a	2.8 a	91.7 a	14.1 a	100.0 a	19.3 a
1	50.0 b	2.9 b	37.0 b	2.3 a	72.9 abc	7.6 b	97.9 a	19.7 a
2	24.1 c	1.0 c	9.3 c	0.4 b	60.4 c	4.0 b	97.9 a	17.0 ab
3	7.4 d	0.3 c	5.5 c	0.1 b	60.4 c	4.7 b	97.9 a	14.6 b
5	5.6 d	0.3 c	3.7 c	0.1 b	79.1 ab	5.0 b	95.8 a	14.5 b

[y] Percentage of inoculated roots that had necrotic lesions 6 days after inoculation.
[z] Different letters indicate that values are significantly different ($P = 0.05$) according to Duncan's multiple range test for columns only.

TABLE 4. Time required by isolates of *Fusarium roseum* 'Acuminatum' and 'Avenaceum' to penetrate and colonize roots of 5-wk-old red clover (cultivar Arlington) plants when inoculated at the root tip or 4 cm above the root tip

Stage of host-pathogen interaction	*F. roseum* 'Acuminatum' Isolate 927 inoculation site[a]		*F. roseum* 'Avenaceum' Isolate 959 inoculation site	
	0 cm (hr)[b]	4 cm (hr)	0 cm (hr)	4 cm (hr)
Penetration of epidermis	8	24	8	18
Colonization of cortex	18	48	18	48
Distributive hyphae in cortex	48	Absent	24	96
Chlamydospores in cortex	Absent	120	Absent	Absent

[a] Distance of inoculation site from root tip.
[b] Hour of sampling when more than 50% of inoculated roots were observed to be at each stage based on inoculation of 108 roots per isolate per inoculation site.

TABLE 5. Frequency and length of necrosis on roots of 5-wk-old red clover (cultivar Arlington) plants 6 days after inoculation at 4 cm above the root tip with isolates of *Fusarium roseum* 'Acuminatum' and 'Avenaceum'

Cultivar and isolate	Necrosis	
	Frequency[y] (%)	Length (mm)
F. roseum 'Acuminatum'		
Isolate 927	0.0 d[z]	0.0 f
Isolate 5207	4.2 d	0.1 f
Isolate 5653	4.2 d	0.5 ef
Isolate 5654	0.0 d	0.0 f
Isolate 5655	0.0 d	0.0 f
F. roseum 'Avenaceum'		
Isolate 959	66.7 b	3.4 cd
Isolate 5181	41.7 c	2.5 de
Isolate 5215	95.8 a	11.2 a
Isolate 5702	87.5 a	5.4 bc
Isolate 5703	87.5 a	6.0 b

[y] Percentage of inoculated roots that had necrotic lesions.
[z] Different letters indicate that values are significantly different ($P = 0.05$) according to Duncan's multiple range test.

tip. Secondary roots started to emerge 6.41 ± 1.31 cm from the root tip.

The pattern of penetration and colonization of roots of 5-wk-old plants (Table 4) inoculated at the root tip with isolate 927 of *F. roseum* 'Acuminatum' and isolate 959 of *F. roseum* 'Avenaceum' was similar to penetration and colonization of 4-day-old seedlings. Penetration by both isolates occurred most often in the region of cell elongation or just adjacent to this area in the region of cell differentiation. Penetration, defined as the presence of fungal hyphae in the root epidermis, and colonization, defined as the presence of fungal hyphae in the root cortex, occurred much more rapidly for both isolates when plants were inoculated at the root tip in comparison to the 4-cm site. Isolate 959 of *F. roseum* 'Avenaceum' also developed distributive hyphae, intercellularly in the root cortex, more rapidly in roots inoculated at the root tip in comparison to the 4-cm site.

Roots inoculated with *F. roseum* 'Acuminatum' isolate 927 at the 4-cm inoculation site had hyphae in the root epidermis and cortex by 48 hr after inoculation (Table 4). Colonization of the cortex was not as extensive as when isolate 927 was inoculated at the root tip. Isolate 927 did not form distributive hyphae at the 4-cm site, but did at the root tip. The major difference between roots inoculated with isolate 927 of *F. roseum* 'Acuminatum' at the root tip and the 4-cm site, was the presence of chlamydospores in the cortex of roots inoculated at 4 cm.

The five pathogenic isolates of *F. roseum* 'Avenaceum' consistently caused a greater frequency and length of necrosis at the 4-cm inoculation site than did isolates of *F. roseum* 'Acuminatum' (Table 5). In fact, the five pathogenic isolates of *F. roseum* 'Acuminatum' caused little or no necrosis at the 4-cm inoculation site. Isolates were originally screened for pathogenicity by inoculation at the root tip. After staining, a microscopic examination of these roots inoculated at 4 cm showed that all isolates colonized the root cortex. All isolates of *F. roseum* 'Acuminatum' inoculated at 4 cm formed chlamydospores in the root cortex but did not form distributive hyphae. All isolates of *F. roseum* 'Avenaceum' formed intercellular distributive hyphae in the root cortex, even in those roots that did not have necrotic symptoms at 6 days after inoculation.

DISCUSSION

Although previous research showed that isolates of *F. roseum* were pathogenic in red clover roots (4,6,9,11,18,20), there are no reports that isolates of *F. roseum* 'Acuminatum' as a group differ in virulence in red clover roots from isolates of *F. roseum* 'Avenaceum.' This research presents evidence that there are distinct differences in virulence and pathogenic behavior between the two groups in roots of 5-wk-old red clover plants. Isolates of *F. roseum* 'Avenaceum' were consistently more virulent than were isolates of *F. roseum* 'Acuminatum,' causing more frequent and more severe necrosis at all inoculation sites in 5-wk-old plants. In addition, there were distinct differences in pathogenic behavior between the two *F. roseum* cultivars in roots inoculated at 2 cm or more from the root tip.

Isolates of *F. roseum* 'Acuminatum' caused little or no necrosis at these inoculation sites, and they formed chlamydospores in the root cortex. This type of asymptomatic colonization of the root

cortex may explain surveys in which *Fusarium* species were consistently isolated from apparently healthy red clover roots (4).

In contrast, isolates of *F. roseum* 'Avenaceum' caused necrosis and formed distributive hyphae in the root cortex at the 4-cm inoculation site. Formation of distributive hyphae is typical of the pathogenic development of *Fusarium* species that cause cortical root or crown rot in other hosts (3,12,15). In the interaction of red clover with *F. roseum* 'Acuminatum' and 'Avenaceum,' formation of distributive hyphae appears to be necessary for pathogenic development, and necrosis was always associated with distributive hyphae formation. In the case of *F. roseum* 'Acuminatum,' penetration of the epidermis and limited colonization of the root cortex can occur without obvious visible disease symptoms. Only when distributive hyphae were formed did necrosis ensue.

Four day-old plants did not differ in reaction to isolates of *F. roseum* 'Acuminatum' and *F. roseum* 'Avenaceum,' confirming research evidence that seedling inoculations often do not predict the pathogenicity and virulence of isolates on older plants (4,6). The pattern of penetration and colonization of seedlings inoculated with isolates of both *F. roseum* 'Acuminatum' and *F. roseum* 'Avenaceum' is similar to previous reports of red clover seedlings inoculated with isolates of *F. roseum* 'Avenaceum' (1,16).

As previously reported (11), necrosis was more frequent and severe when roots of 5-wk-old plants were inoculated at the root tip in comparison to other sites. In addition, our results indicated that fungal penetration and colonization occurred more rapidly at the root tip than at the 4-cm inoculation site. This root tip area corresponds to the meristematic zone and area of cell elongation. Penetration by all isolates occurred at highest frequency when roots were inoculated at the tip. The 2- and 3-cm inoculation sites, where necrosis was less frequent and less severe, are in the area of cell differentiation where there was abundant root hair formation. Only isolate 959 of *F. roseum* 'Avenaceum' caused a higher frequency of necrosis in areas of secondary root formation than at the 2- and 3-cm inoculation sites. In some cases, necrosis was associated with the region around the lateral root, but hyphae were not observed entering through openings around the emerging lateral roots.

Because of the interaction between stress factors and Fusarium root rot severity, the ability of *F. roseum* 'Acuminatum' to colonize root tissue without causing disease symptoms is of interest. Stresses such as winter injury (4,20), increased frequency of clipping (6,14,17), low nutritional levels (2), and insect damage (10) increased the incidence and severity of root rot in red clover. Future investigations of the role of plant stress on the development of *F. roseum* 'Acuminatum' in red clover roots are needed.

LITERATURE CITED

1. Chi, C. C., Childers, W. R., and Hanson, E. W. 1964. Penetration and subsequent development of three *Fusarium* species in alfalfa and red clover. Phytopathology 54:434-437.
2. Chi, C. C., and Hanson, E. W. 1961. Nutrition in relation to the development of wilts and root rots incited by *Fusarium* in red clover. Phytopathology 51:704-711.
3. Christou, T., and Snyder, W. C. 1962. Penetration and host-parasite relationships of *Fusarium solani* f. *phaseoli* in the bean plant. Phytopathology 52:219-226.
4. Elliott, E. S., Baldwin, R. E., and Carroll, R. B. 1969. Root rots of alfalfa and red clover. W.Va. Agric. Exp. Stn. Bull. 385 T. 32 pp.
5. Fisher, N. L., Burgess, L. W., Toussoun, T. A., and Nelson, P. E. 1982. Carnation leaves as a substrate and for preserving cultures of *Fusarium* species. Phytopathology 72:151-153.
6. Fulton, N. D., and Hanson, E. W. 1960. Studies of root rots of red clover in Wisconsin. Phytopathology 50:541-550.
7. Hoagland, D. R., and Arnon, D. E. 1938. The water culture method for growing plants without soil. Calif. Agric. Exp. Stn. Circ. 347. 39 pp.
8. Kendall, W. A., and Leath, K. T. 1974. Slant-board culture methods for root observation of red clover. Crop Sci. 14:317-320.
9. Kovacikova, E., and Kudela, V. 1982. *Fusarium* species associated with root rot of red clover. Zentralbl. Mikrobiol. 137:407-414.
10. Leath, K. T., and Byers, R. A. 1977. Interaction of Fusarium root rot with pea aphid and potato leafhopper feeding on forage legumes. Phytopathology 67:226-229.
11. Leath, K. T., and Kendall, W. A. 1978. Fusarium root rot of forage species: Pathogenicity and host range. Phytopathology 68:826-831.
12. Malalaskera, R. A. P., Sanderson, F. R., and Colhoun, J. 1973. Fusarium diseases of cereals. IX. Penetration and invasion of wheat seedlings by *Fusarium culmorum* and *Fusarium nivale*. Trans. Br. Mycol. Soc. 60:453-462.
13. Miller, P. M. 1955. V-8 juice agar as a general-purpose medium for fungi and bacteria. Phytopathology 45:461-462.
14. Moody, A. R., Kilpatrick, R. A., and Rich, A. E. 1967. Effect of cutting height on development of root rot in clover. Plant Dis. Rep. 51:826-828.
15. Pearson, N. C. 1931. Parasitism of *Gibberella saubinettii* on corn seedlings. J. Agric. Res. 43:569-596.
16. Siddiqui, W. M., and Halisky, P. M. 1968. Histopathological studies of red clover roots infected by *Fusarium roseum*. Phytopathology 58:874-875.
17. Siddiqui, W. M., Halisky, P. M., and Lund, S. 1968. Relationship of clipping frequency to root and crown deterioration in red clover. Phytopathology 58:486-488.
18. Sundheim, L. 1970. Pathogenicity of *Fusarium* species on red clover roots. Ann. Acad. Sci. Fenn., Ser. A IV Biol. 168:63-65.
19. White, N. H., and Baker, E. P. 1954. Host pathogen relations in powdery mildew of barley. I. Histology of tissue reactions. Phytopathology 44:657-662.
20. Ylimaki, A. 1967. Root rot as a cause of red clover decline in leys in Finland. Ann. Agric. Fenn. 6:1-59.