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Isolation of *Pyrenochaeta terrestris* from Onion Roots[1]

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Abstract. A comparison was made of various techniques for isolating *Pyrenochaeta terrestris* (Hansen) Gorenz *et al.* from growing onion roots. Water agar was as suitable as potato dextrose agar (0.5% dextrose) and is recommended to avoid contamination. Texas L 303, Iowa Yellow Globe 51, and Trapp's Downing Yellow Globe used as trap crops were equally efficient. Maximum infection of the early planting occurred earlier than a later planting.

Since pink root of onions (*Allium cepa* L.) was first described in 1917 (4), *Pyrenochaeta terrestris* (Hansen) Gorenz *et al.*, the causal agent, has been described on a wide range of hosts. Before investigating overwintering of primary inoculum, it was necessary to find a reliable technique for isolation of the organism. This investigation proposed to compare three methods of surface disinfesting and two types of media for isolation of the pathogen.

These investigations were conducted during June, July, and August, 1960, on the S. Kennedy and Son's farm at Clear Lake, Iowa, to whom a debt of gratitude is acknowledged.

MATERIALS AND METHODS

On April 14th and June 25th, 1960, seed was planted of Iowa Yellow Globe 51 (IYG 51) and Trapp's Downing Yellow Globe (TDYG), both moderately resistant varieties, and Texas L 303, a highly resistant variety. Bulbs of various pedigrees were also planted on June 25th.

Every other day during the three months, a sample of at least 30 plants of each variety was taken at random for isolation. Bulb samples were taken every four days due to a limited quantity of available bulbs. Roots of these samples were kept moist prior to isolation. A 2-cm sample of a root from each plant was taken in the following order of preference: (1) a portion exhibiting pink coloring, (2) a portion with root cap visible, (3) a portion including a branch root, (4) any portion.

Groups of at least ten root pieces from each variety were treated as follows: one group was placed in a 1:1000 solution of $HgCl_2$ (2,3) for 3-5 minutes and rinsed in tap water; an-

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other group was placed in B.K. (50% calcium hypochlorite manufactured by Pennsalt Chemical Co.) solution (1 tsp. in 20 ml tap water) for 3-5 minutes and rinsed in tap water (2), and a third group was placed under running tap water for 1½ hours in beakers covered with cheesecloth.

Treated and washed groups were transferred aseptically to the plating media. The media used were a low sugar (0.5%) potato dextrose agar (P.D.A.) and a 2% water agar. The 0.5% dextrose P.D.A. contained 39 gms Difco P.D.A., 37.5 gms of Difco Bacto agar, 500 mls of steamed potato infusion and 3500 ml of water. Root pieces were placed directly on the low sugar P.D.A. but underneath the water agar.

Results and Discussion

In June, 27 of the 1,041 isolations from roots of plants of early-planted seed yielded fungi, but none of these were *Aspergillus niger* van Tieh. or *P. terrestris*. Root treatment with $HgCl_2$ yielded 5 successful isolations, whereas the running water method yielded 22. Sterilization with B.K. was initiated July 8th. Varieties IYG 51 and TDYG were more successful as trap crops than L 303. P.D.A. was more effective for isolating fungi than water agar. (See Table 1).

Table 1. Fungal isolations from onion roots.

	Time of isolation			
	June	July	August	Total
Successful Fungal Isolations				
Planting date				
April 14	27/1,041[a]	143/1,008	827/936	997/2,985
June 25	—	78/504	395/936	473/1,440
June 25, Bulbs	—	53/144	110/144	163/288
Efficiency of Treatments				
Method				
$HgCl_2$	5/694	63/624	434/672	502/1,990
B. K.	—[b]	79/480	441/572	520/1,152
Water	22/347	132/552	457/672	611/1,571
Efficiency as Trap Crops				
Variety				
IYG 51	11/346	85/504	417/624	513/1,474
L 303	5/348	55/504	403/624	463/1,476
TDYG	11/347	81/504	402/624	494/1,475
Bulbs	—[c]	53/144	110/144	163/288
Values of Plating Media				
Medium				
2% Water Agar	1/522	52/828	597/1,008	813/2,355
0.5% P. D. A.[d]	26/519	222/828	735/1,008	820/2,358

[a] Numerator is number of successful isolations; denominator is number of isolations attempted.
[b] B. K. sterilization started on July 8.
[c] Bulb isolations started on July 8.
[d] 0.5% dextrose.

In July, 1,656 isolations were attempted. The numbers of fungi isolated from the plant roots of early- and late-planted seed were 143 from 1,008 isolations and 78 from 504 isolations, respectively. Isolations from bulb roots were more successful–53 out of 144 attempted. Of all isolations, 274 were successful and classed as 136 *A. niger,* 47 *P. terrestris,* and 91 unknown. Most of the fungi were isolated by the running water method. Texas L 303 still was not as effective as the other two varieties as a trap crop. Water agar increased in efficiency of isolation, probably due to an overall increase in fungal microflora of the roots. (See Table 1).

In August, 2,016 isolations were attempted. Roots of plants from early-planted seed yielded 827 fungal isolates; from late-planted seed, 395 fungal isolates; and from bulbs, 110 fungal isolates. Thirty of these were *A. niger,* 30 were unidentified fungi, and 1,272 were *P. terrestris.* Only *P terrestris* was isolated after August 5th from roots of the early-planted plants, after August 11 from the roots of bulbs, and after August 21 from the roots of the late-planted plants. Delayed planting merely postponed the date on which total infection with *P. terrestris* was recorded. During this month, all methods of treating roots were equally efficient, and little difference was observed between trap crops or plating media. (See Table 1).

Of the various surface disinfesting treatments, running water appeared most effective early in the season, and the other two treatments increased in effectiveness later in the season. Low sugar P.D.A. appeared to be the better medium for early isolation of fungi, although most of these appeared to be saprophytes. Later *P. terrestris* occurred with equal facility on either medium, suggesting that water agar could be used to avoid contaminants.

All varieties were equal in August for efficiency of determining *P. terrestris* infections of the roots. Apparently, the resistance of L 303 decreased as the plants became mature in early August. At no time did the results similar to that of Davis and Henderson (1) appear, in which *Fusarium* species could be said to precede infection by *P. terrestris.*

Literature Cited

1. Davis, G. N., and W. J. Henderson. 1937. The interrelation of the pathogenicity of a *Phoma* and a *Fusarium* on onions. Phytopath. 27: 763-772.
2. Gorenz, A. M., J. C. Walker, and R. H. Larson. 1948. Morphology and taxonomy of the onion pink-root fungus. Phytopath. 38: 831-840.
3. Riker, A. S., and R. S. Riker. 1936. Introduction to research on plant diseases. John S. Swift Co., Inc., St. Louis. 117 pp.
4. Taubenhaus, J. J., and A. D. Johnson. 1917. Pink root, a new disease of onions in Texas (Abstr.). Phytopath. 7: 59.