Fungi Associated with Tip Dieback of Ponderosa Pine Seedlings at the Clifty View Nursery, Bonner's Ferry, Idaho

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Ponderosa pine seedlings (1-0) were received from the Clifty View Nursery to determine possible cause(s) of tip dieback and necrosis. Seedlings were examined under the binocular microscope (10-70x) for fungal fruiting structures and several fungi were identified directly from necrotic tissues. Isolations were made onto water agar and potato dextrose agar (PDA) from surface sterilized portions of necrotic tips. Associated fungi were maintained in axenic cultures and identified using several standard taxonomic guides.

Associated Fungi:

1. Sirococcus strobilinus - this fungus was isolated from or found colonizing only 10 percent of the seedlings received. Those seedlings with Sirococcus were easily identified from purple-brown lesions commonly associated with a crooked tip. Extensive pycnidial production was found on necrotic stems and needle tissues (figure 1). Pycnidia appeared as small black dots and were particularly prevalent at the base of necrotic needles. Pycnidia yielded elongate, hyaline, one-celled conidia, typical of Sirococcus.

2. Phoma eupyrena and Phoma herbarum - these fungi were commonly found sporulating on and were isolated from necrotic tissues both at the tips and at the base (near the expected soil line) of affected seedlings. These fungi are common soil inhabitants and are usually considered saprophytic. However, P. eupyrena has been implicated in tip dieback and mortality of 1-0 red fir and Douglas-fir seedlings at the Humboldt Nursery in California and 1-0 lodgepole pine seedlings at the Bessey Nursery in Nebraska.

3. Diplodia pinea - this fungus was infrequently isolated from tip blighted seedlings. Diplodia is often associated with a tip blight of mature trees; most damage has been recorded in the Great Plains on Austrian pine and ponderosa pine. The fungus apparently sporulates on pine cone scales and is disseminated by rain splash to nearby branch tips. Occurrence of this fungus has been reported in several nurseries and it was recently found on 1-0 ponderosa pine at the Coeur d'Alene Nursery in Idaho.

4. Epicoccum nigrum - this fungus was commonly isolated from necrotic stem and needle tissues. It is a common soil-borne saprophyte, probably colonizing tissues previously killed by other fungi.

Chlorothalonil Tolerance Test

Four isolates of Sirococcus strobilinus were screened for possible tolerance to chlorothalonil. Isolates tested included two from the Clifty View Nursery (designated 83-8 and 83-9) and two from red pine in Minnesota (designated 83-10 and 83-11). All tested isolates were initially grown on PDA for 16 days. Square plugs of mycelium 5 mm in diameter were transferred from the edge of PDA cultures to the center of test plates (PDA amended with 50 ppm active ingredient of chlorothalonil). Inoculated test plates were incubated in the dark at 22°C. At the time test plates were inoculated, an equal number of standard PDA plates were also inoculated. Growth on fungicide-amended media was expressed as a percentage of growth on PDA after 7 and 16 days' incubation. Results are summarized below:

Table 1.--Linear growth of Sirococcus strobilinus on PDA amended with chlorothalonil (50 ppm a.i.).

Isolate	Linear growth percentage[3] 7 days	16 days
83-8[1]	0	3.6
83-9[1]	0	30.4
83-10[2]	40.0	68.8
83-11[2]	60.0	68.8

[1]Isolated from 1-0 ponderosa pine seedlings at the Clifty View Nursery.
[2]Isolated from 1-0 red pine seedlings from Minnesota.
[3]Expressed as the percentage of linear growth on standard PDA.

Both isolates from the Clifty View Nursery had limited growth on chlorothalonil-amended media (figures 2 and 3). However, growth was much less than for isolates from Minnesota (figure 4).

To determine if isolates could adapt to chlorothalonil once exposed to the fungicide, another test was performed. Sirococcus was reisolated from the fungicide-amended cultures in the first test, grown on standard PDA for 16 days and placed again on chlorothalonil-amended media (50 ppm a.i.). These plates were again incubated for 16 days at 22°C in the dark and then measured. Results indicated that isolate 83-8 grew 7.5 times farther the second time on fungicide-amended media than the first time it was exposed (figure 5). Isolate 83-9 and the two Minnesota isolates grew about the same distance as in the first test (figure 6).

Conclusions

1. Not all tip dieback of ponderosa pine seedlings at the Clifty View Nursery is due to Sirococcus strobilinus. Other possible causes include Phoma eupyrena, P. herbarum, and Diplodia pinea.

2. Isolates of Sirococcus from the Clifty View Nursery that we examined are not very tolerant to chlorothalonil. However, total toxicity to the fungicide was not evident. We suspect that sustained heavy doses of the fungicide may select for more tolerant strains of the fungus. That tolerant strains do exist was evident from the test with two isolates from Minnesota (table 1). We were also able to show that tolerance can probably develop if Sirococcus is continually exposed to chlorothalonil. Therefore, further screening for buildup of fungicide tolerance may be necessary.

Selected References

Robak, H. 1956. Some fungi occurring on died-back tops and branches of Picea abies and Abies spp. in western Norway. Friesia 5: 366-389.

Skilling, D. D. and J. T. O'Brien. 1973. How to identify Scleroderris canker and red pine shoot blight. USDA Forest Service. North Central Forest Exp. Sta. Leaflet. 6 pp.

Smith, R. S., Jr. 1973. Sirococcus tip dieback of Pinus spp. in California forest nurseries. Plant Dis. Reptr. 57: 69-73.

Srago, M. D. 1978. Nursery disease problems - Sirococcus strobilinus. In Gustafson, R. W. (ed.). Proceedings 1978 Nurseryman's Conference & Seed Processing Workshop. Western Forest Nursery Council and Intermountain Nurseryman's Association. pp. B-134-B-137.

Figure 1.--Tip dieback of 1-0 ponderosa pine seedling from the Clifty View Nursery caused by Sirococcus strobilinus. Pycnidial production is evident at the base of necrotic needles.

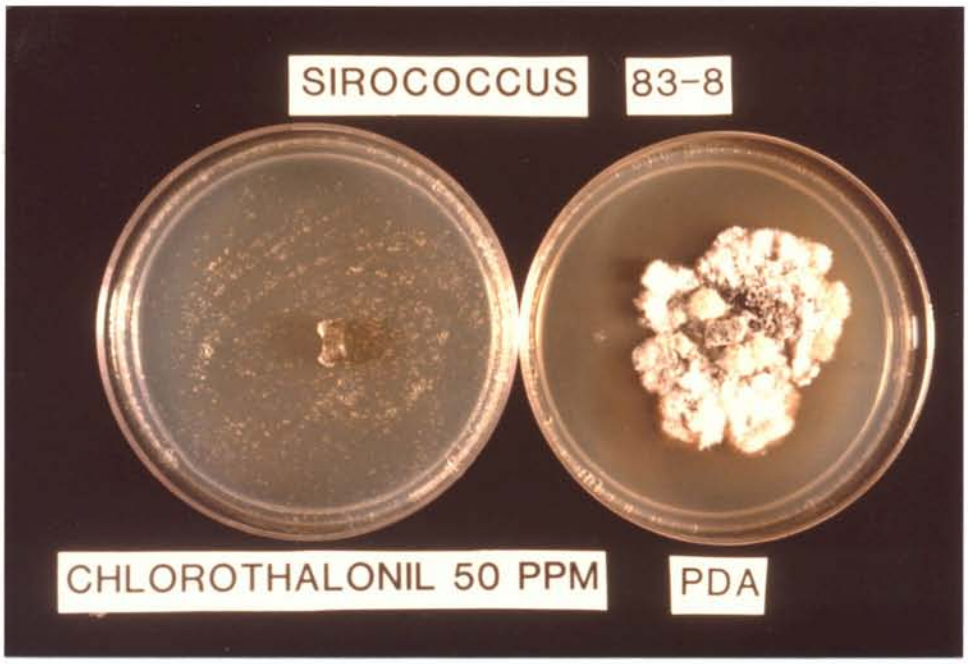

Figure 2.--Growth of Sirococcus strobilinus (isolate 83-8) on chlorothalonil-amended PDA and standard PDA after 16 days.

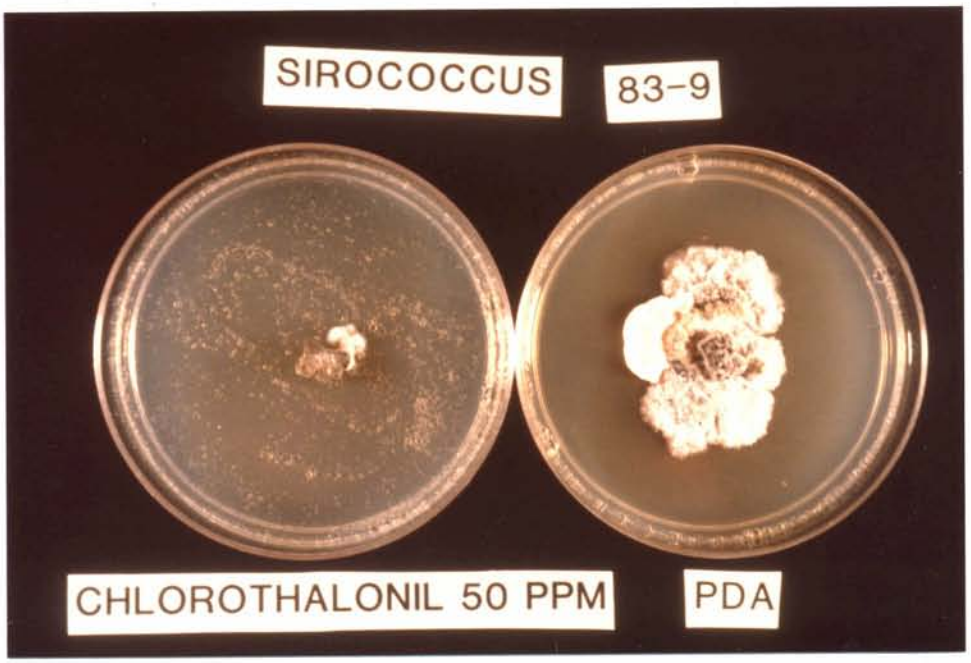

Figure 3.--Growth of Sirococcus strobilinus (isolate 83-9) on chlorothalonil-amended PDA and standard PDA after 16 days.

Figure 4.--Growth of Sirococcus strobilinus (isolate 83-10) on chlorothalonil-amended PDA and standard PDA after 16 days.

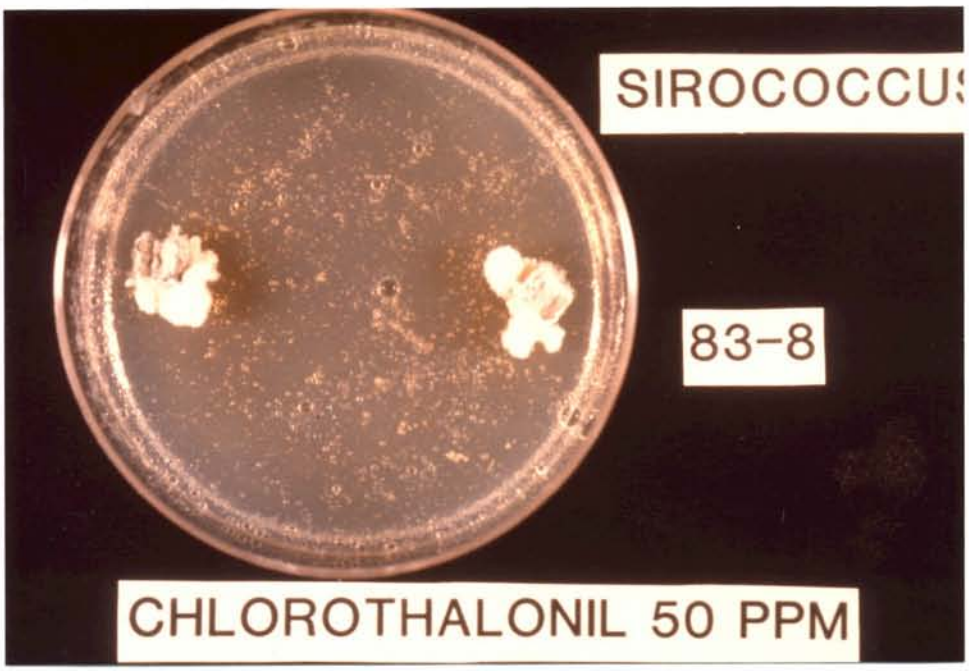

Figure 5.--Growth of Sirococcus strobilinus (isolate 83-8) on chlorothalonil-amended PDA after 16 days - second exposure to the fungicide medium.

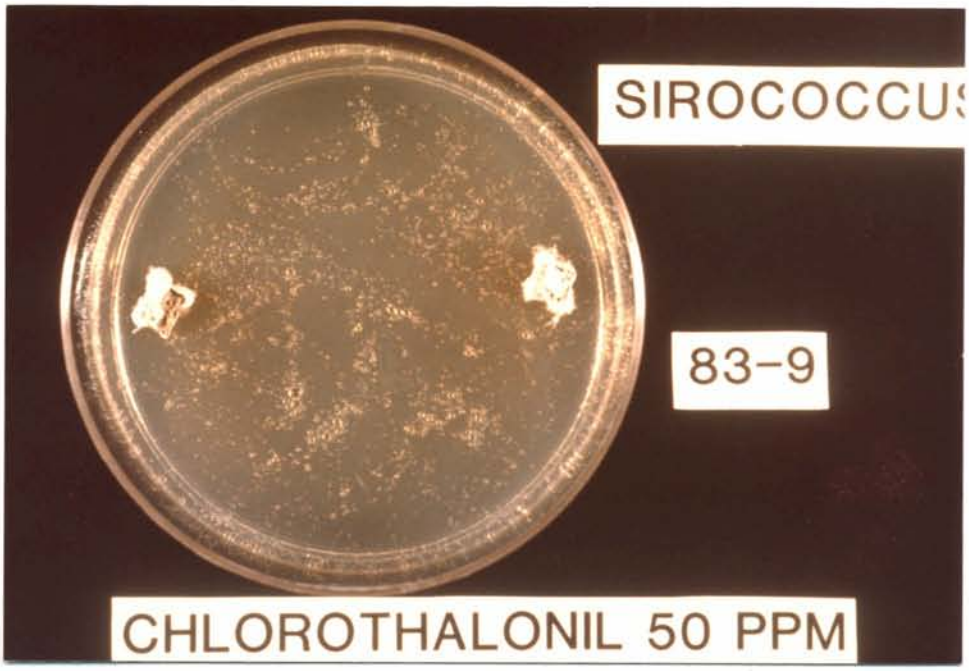

Figure 6.--Growth of Sirococcus strobilinus (isolate 83-9) on chlorothalonil-amended PDA after 16 days - second exposure to the fungicide medium.