FUNGAL CONTAMINATION OF PONDEROA PINE CONES AND SEED FROM THE COEUR D'ALENE NURSERY, IDAHO

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Ponderosa pine (Pinus ponderosa Laws.) cones collected on the Rexford Ranger District (Kootenai National Forest) were recently sent to the Coeur d'Alene Nursery, Idaho for seed extraction. Several of the cones had external fungal mold. Three such cones were submitted by the nursery for identification of associated fungi, partcicularly potential pathogens. Fungi growing in cone scales were examined microscopically and cultured on potato dextrose agar. Two of the cones (designated #1 and #2) had mold restricted to the outer cone scales. The other cone (#3) had fungal growth on both outer and inner cone scales (figure 1). This cone also had been parasitized by insects (probably Conophthorus ponderosae Hopkins).

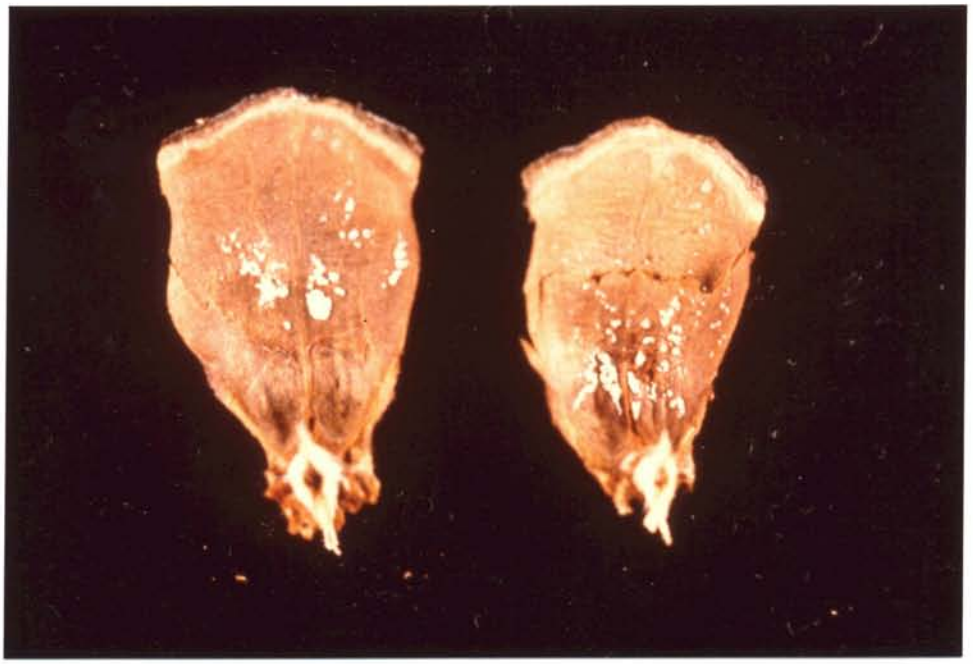

Figure 1.--Cone scales of ponderosa pine with Trichoderma viride growing on the inner scale surface.

Three major fungi were identified on cone scales. the first was Trichoderma viride Pers. ex Fr., a common soil and wood saprophyte. This fungus produced a white mold and was the organism growing on the inner scales of cone #3 (figure 1). Trichoderma was also located on the wings and seedcoats of several extracted seed (figure 2). This fungus is usually saprophytic, but may decay seed if it penetrates the seedcoat.

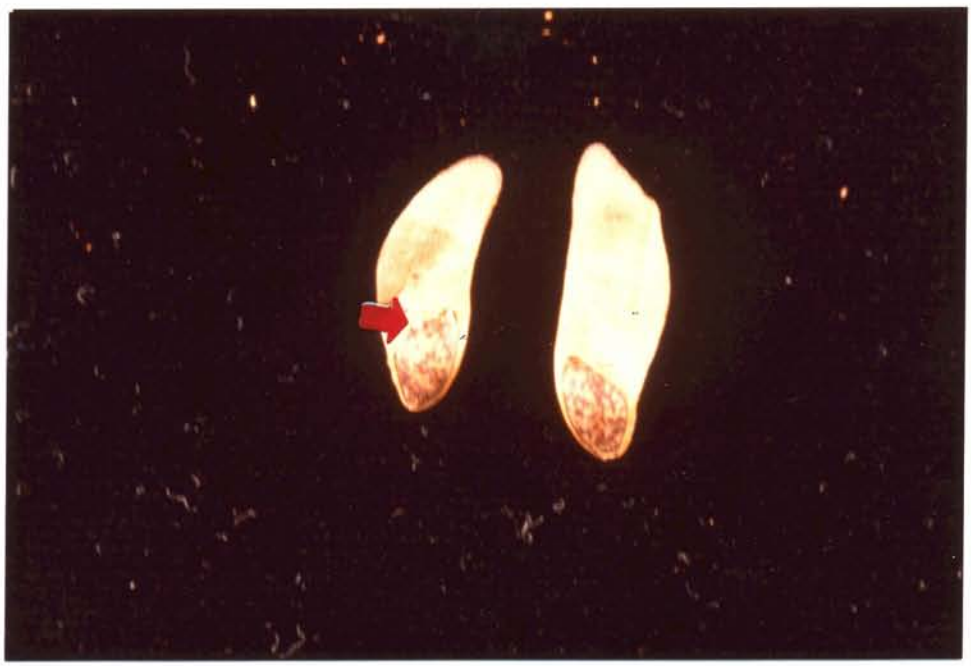

Figure 2.--Winged seed of ponderosa pine with Trichoderma viride growing at the base of the wing and on the seedcoat (red arrow).

Another fungus isolated from cones was Botrytis cinerea Pers. ex. Fr. This organism sporulated on the outer cone scales, but was not located on the inner scales near the seed. Botrytis is a common foliar pathogen of conifer seedlings and has also been reported infecting seed of many species.

The other fungus commonly growing on cones was an unidentified organism which produced a bright yellow mycelium over the outer cone scales. This organism failed to sporulate and identification could not be made. No characteristic structures other than highly vacuolate, septate hyphae were evident. Past experience with squirrel cached cones indicate that several members of the Pezizales (Ascomycetes) often colonize cones. Some produce a yellow mycelium, including members of the genera Pithya and Plectania. However, without sporulating structures, it is impossible to identify the yellow fungus growing on the cones examined.

Seed from these three moldy cones were assayed for fungal contamination. All seed that looked sound based on external appearance (those of regular size, without broken seedcoats) were carefully removed from each cone and counted. Twenty seed from each cone were placed on Fusarium selective media (developed by Nash and Snyder 1962) either directly without treatment, after being soaked for 24 hours in standard tap water, or after being aseptically dissected to expose their endosperms. The selective medium was used to more easily isolate Fusarium species, which are common conifer seedling pathogens that are often seedborne. Seed were incubated at 24° C under continuous fluorescent light for 6-12 days. After incubation, seed were examined for germination (except for the dissecting treatment) and presence of Fusarium or other potential plant pathogens.

Table 1 summarizes germination percentages of seed from the three cones after 6 and 12 days' incubation. Water-soaking (treatment 1) seemed to

improve early seed germination. However, high germination occurred after 12 days in seed from all cones regardless of treatment.

Table 1.--Percentage germination of ponderosa pine seed extracted from moldy cones.

Cone no.	No. seed extracted[1]	6 days incubation Treatments[2] 1	2	both	12 days incubation Treatments[2] 1	2	both
1	89	35	5	20	75	70	72.5
2	87	90	60	75	90	65	80
3	65	75	65	70	75	85	80

[1]Number of seed appearing sound from external examination.

[2]Treatments:
1 = Soaked in tap water for 24 hours and then placed on selective Fusarium media.
2 = Untreated seed, placed directly on selective Fusarium media.

Condition of endosperms of dissected seed is outlined in table 2. Most seed had healthy (white) endosperms, although several were empty or had abnormal (yellow or shriveled) endosperms.

Table 2.--Condition of endosperms of dissected ponderosa pine seed from moldy cones.[1]

	Endosperm Condition			
Cone no.	White	Yellow	Shriveled (black)	Empty
1	70	5	10	15
2	70	0	0	30
3	90	5	0	5

[1]Figures in table represent percentage of seed (20 seed examined per cone).

Fusarium spp. were isolated from seed obtained from cones #1 and #2, but not #3 (table 3). Percentage infection was greatest in seed from cone #1. This rate of contamination is relatively high compared to other ponderosa pine seed and those of other species we have evaluated. Amount of Fusarium in seed from cone #2 is about the level we expect to find on most seed. Treatments did not have a consistent effect on amount of Fusarium isolated. It is important to note that Fusarium was isolated from inside the seed (treatment 3) as well as externally on the seedcoat.

Table 3.--Percentage of ponderosa pine seed from modly cones contaminated with Fusarium spp.

	Treatments[1]			
Cone no.	1	2	3	All
1	45	5	5	18.3
2	5	10	5	6.7
3	0	0	0	0

[1]Treatments

1 = Soaked in tap water for 24 hours and then placed on selective Fusarium media.

2 = Untreated seed, placed directly on selective Fusarium media.

3 = Aspetically dissected to reveal endosperm and then placed dirctly on selective Fusarium media.

Two species of Fusarium were identified on seeds. The most common was F. roseum (Lk.) Sacc., which was distinguished by its slender, highly falcate macroconidia (figure 3), absence of microconidia, rare production of chlamydospores, and production of a deep red pigment in culture. The other species was F. rigidiusculum (Brick) Sny. & Hans. This species produced both macroconidia and microconidia (figure 4), but lacked chlamydospores, and produced appressed reddish colonies with yellow sporodochia in culture.

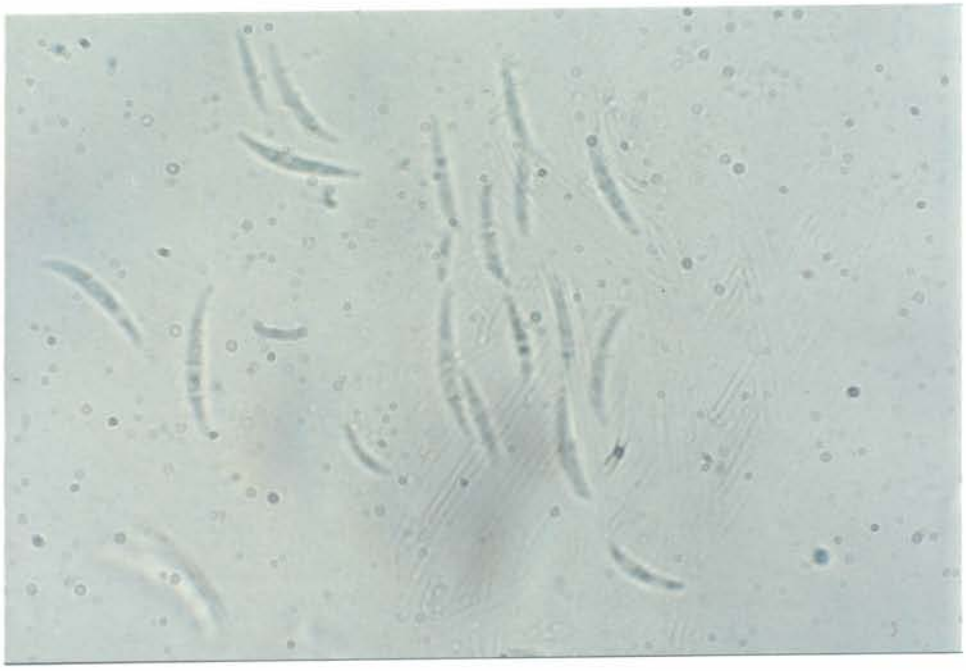

Figure 3.--Macroconidia of Fusarium roseum isolated from seed of moldy ponderosa pine cones.

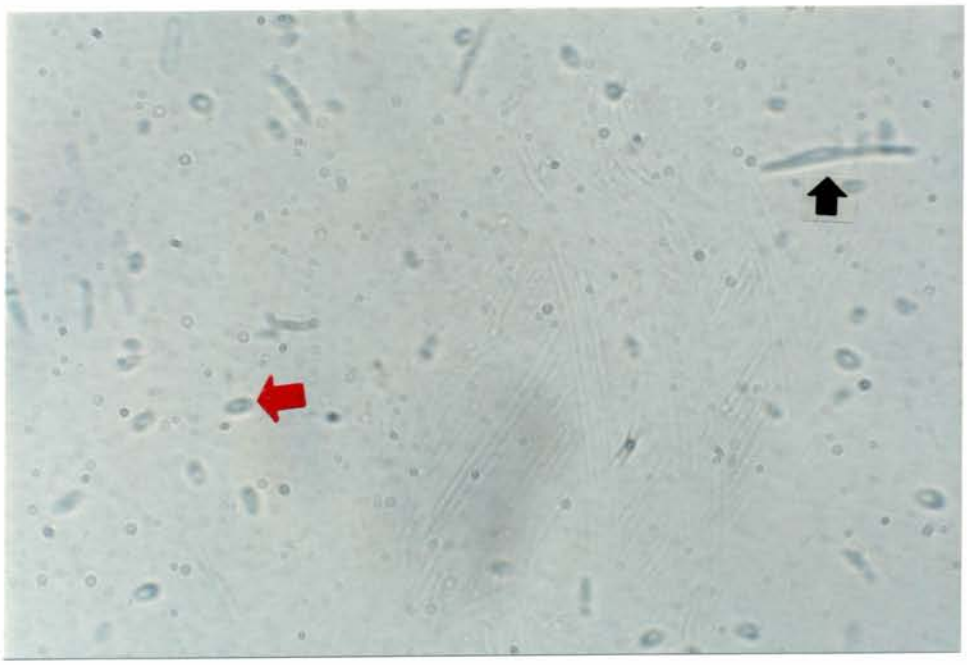

Figure 4.--Macroconidia (black arrow) and microconidia (red arrow) of Fusarium rigidiusculum isolated from seed of moldy ponderosa pine cones.

CONCLUSIONS

1. Mold noticeable on the outside of cones may not be detrimental to the seed produced within. The fungi identified on the cone scales and less commonly on the seedcoats were not pathogenic to the seed itself. Moldy ponderosa pine cones still produced many viable seed.

2. Fusarium can be expected to contaminate a certain percentage of seed, particularly if the cones come from squirrel caches. However, levels of infection should remain low (between 5-10 percent). It is not known why seed from cone #1 had such high infection rates.

3. Cones parasitized by insects may still produce viable seed, although number of seed produced may be less than nonparasitized cones. It is interesting that the only parasitized cone examined (#3) did not have any Fusarium infection of seed.

4. Comparisons should be made of seed infection with pathogenic organisms and viable seed production between moldy and nonmoldy cones.

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