

# Germination of Conifer Seeds Surface-Sterilized With Bleach<sup>1</sup>

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Surface-sterilizing conifer seeds with 40% laundry bleach often significantly enhanced, rather than reduced, cumulative germination percentages. Tree Planters' Notes 38(3):18-21; 1987.

Damping-off has long been a problem in forest tree nurseries. Common fungi associated with damping-off include *Pythium*, *Rhizoctonia*, *Phytophthora*, and *Fusarium*. Only tender germinants are affected, since disease incidence declines as soon as the stems begin to lignify, generally in 3 to 4 weeks (11).

Spores of damping-off fungi can be either soilborne or seedborne. Soilborne damping-off can be controlled by using sterile media and proper cultural techniques during the germination phase. Many techniques for reducing damping-off have been formulated, including lowering pH of the medium, improving its aeration, lowering relative humidity, maintaining proper moisture of the medium, delaying nitrogen fertilization until germination is complete, and drenching the growing medium with fungicide (2,7,11).

Seedborne inoculum may cause damping-off or be the source for later root disease by *Fusarium* species. The incidence of *Fusarium* root disease may vary dramatically among seedlots (3), and different levels of infec-

**Table 1**—Treatment of conifer seeds after surface sterilization with a solution of 2 parts laundry bleach and 3 parts water

Species	Length of rinse (days)	Length of stratification (days)
Western white pine <sup>1</sup> ( <i>Pinus monticola</i> Dougl. ex D. Don)	5	90-120
Ponderosa pine ( <i>P. ponderosa</i> Dougl. ex Laws.)	2	40
Shore pine <sup>1</sup> ( <i>P. contorta</i> Dougl. ex Loud. var. <i>contorta</i> )	2	40
Scotch pine ( <i>P. sylvestris</i> L.)	2	40
Austrian pine ( <i>P. nigra</i> ) Arnold	2	40
Rocky Mountain Douglas-fir ( <i>Pseudotsuga menziesii</i> var. <i>glauca</i> (Beissn, Franco)	2	21

<sup>1</sup>Species routinely sterilized at the research nursery but not included in this study.

tion may be due to seedlot collection sources (4). James (5) found that most seedlots of Douglas-fir and ponderosa pine had less than 10% seed infection with *Fusarium*. Although infection levels seem low, they may be sufficient to cause widespread disease, particularly if secondary spread is extensive and not reduced by cultural methods (5).

Methods for controlling seedborne diseases have also been evaluated (1,2,7,8,11,12). One simple method involves rinsing seed in clear, running water for 24 hrs to wash off fungal spores (6). Applying fungicides to the seed coat is another method. Captan, ETMT, and thiram are commonly used fungicides, but their adverse effects on germination and inconsistent results in controlling fungi have reduced their usage (2,7,8,11). The third method involves soaking in chlorine bleach or hydrogen peroxide to disinfect the seed coat (1,2,7,12). Problems with solution

concentration and soaking rates have produced poor fungal control or reduced germination.

Our objective was to examine the effects of bleach sterilization on the seed germination energy and capacity of several western conifers.

## Methods and Materials

Seeds were treated in a sodium hypochlorite solution consisting of 2 parts common laundry bleach (5.25% sodium hypochlorite) to 3 parts clear water. Seeds were soaked in the bleach solution for 10 min. with constant hand agitation. (Hands were protected with rubber gloves to avoid skin reactions.) Agitation is important to obtain uniform cleaning and sterilization (9). After treatment, the seed were thoroughly rinsed, with constant hand agitation, to remove all bleach that could damage the seed. The bleach solution was discarded after one use. Seed

were then rinsed in clear-running tap water for 2 days to ensure full imbibition before stratification (table 1).

Four conifer species (three species had two seed lots each) were examined for the effects of bleach on seed germination. Both treated and untreated (controls) seed were stratified and each treatment was replicated four times with 100 seeds per replicate. Analysis of variance (10) was used to detect differences in cumulative germination percentage.

**Results and Discussion**

Cumulative germination percentage was increased by sterilizing with bleach (table 2). This increase was probably due to reduced fungal propagules that would otherwise kill the germinants. Improved germination percentage may be more evident in seedlots heavily infected with pathogens and exhibiting poor germination due to fungal colonization on the seed coat. Each seedlot should have a sample tested with the bleach solution before the entire lot is treated.

This bleach seed treatment has been used routinely at the University of Idaho Forest Research Nursery as a disease preventative method. Although the bleach treatment does not eradicate all organisms on the seed coat (6,9), we believe that it helps reduce early seedling mortality when

**Table 2—Cumulative percent germination at 7 to 28 days of conifer seed sterilized with a solution of 2 parts laundry bleach and 3 parts water**

Species/treatment	Cumulative percent germination			
	7 days	14 days	21 days	28 days
Austrian pine				
Bleach	65.8*	68.6*	68.6*	68.6*
Control	53.8	59.2	62.3	62.3
Douglas-fir				
Lot 1				
Bleach	54.3	80.5*	81.5*	82.0*
Control	51.5	72.3	73.0	73.0
Lot 2				
Bleach	70.3	81.6	86.6	87.5
Control	72.5	84.3	89.0	89.6
Ponderosa pine				
Lot 1				
Bleach	68.3*	80.8**	80.8**	80.8**
Control	58.0	62.3	62.8	62.8
Lot 2				
Bleach	98.0	99.0	99.0	99.0
Control	97.5	98.5	99.0	99.0
Scotch pine				
Lot 1				
Bleach	72.5*	80.3*	80.3*	81.0*
Control	59.0	72.0	72.0	72.0
Lot 2				
Bleach	72.3**	84.3**	86.5**	86.5**
Control	52.3	58.3	59.0	59.0
Average for all lots				
Bleach	71.6	82.2	83.3	83.6
Control	63.5	72.4	73.9	74.0

Values are means of four 100-seed replications.  
 \*Significantly different from controls at  $P < 0.05$ .  
 \*\*Significantly different from controls at  $P < 0.01$ .

used with proper cultural methods, while not detrimentally affecting germination.

This treatment, used in combination with lower pH of the growing medium and low rates of applied nitrogen during the initial phase of seedling growth, has reduced early seedling mortality due to fungi associated with

damping-off and appears to reduce later losses to *Fusarium* (fig. 1). This seems consistent with the findings of James and Genz (6), who found a significantly lower incidence of disease in hypocotyls of germinated ponderosa pine seeds treated with bleach then in controls. There is still early mortality from *Fusarium*



**Figure 1**—*Fusarium cotyledon blight*, from within the seedcoat, infecting the emerging cotyledons of ponderosa pine.

cotyledon blight due to fungal propagules borne within the seed coat (fig. 2). This technique has been effective only for thick-coated seeds such as pine and Douglas-fir, which can withstand the oxidizing effects of bleach. It should not be used on seed of true firs, larch, and spruces.

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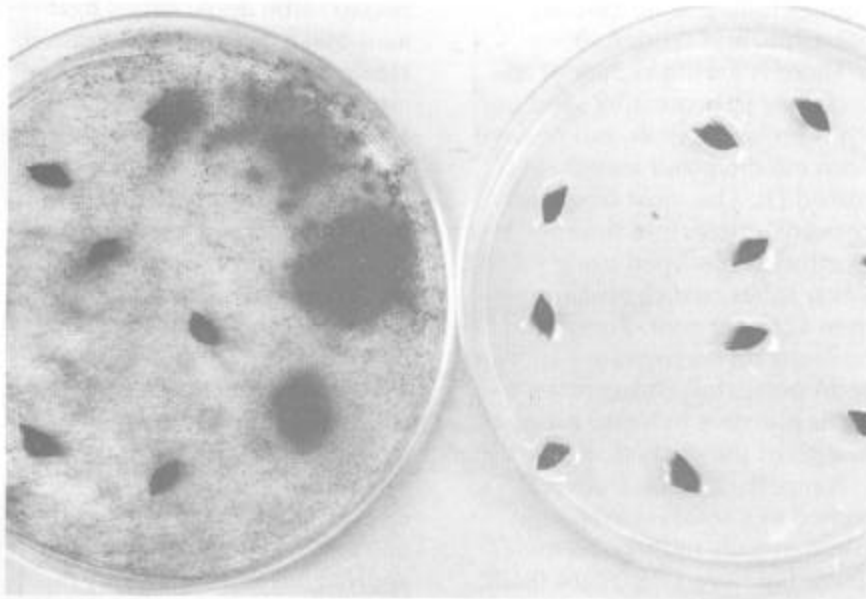


Figure 2—Agar plates with fungal growth from unsterilized (left) and sterilized Douglas-fir seed.

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