Seed Germination of Two Provenances of Atlantic White-Cedar as Influenced by Stratification, Temperature, and Light[1]

Laura G. Jull,[2] Frank A. Blazich,[3] and L. Eric Hinesley[3]
Department of Horticultural Science, North Carolina State University
Raleigh, NC 27695-7609

Abstract

Seeds from two provenances (Wayne Co., NC, and Escambia Co., AL) of Atlantic white-cedar [*Chamaecyparis thyoides* (L.) B.S.P.] were stratified (moist-prechilled) for 0, 30, 60 or 90 days at 4C (39F). Following stratification, seeds were germinated at 25C (77F) or 8/16 hr thermoperiods of 25/15C (77/59F) or 30/20C (86/68F) with daily photoperiods at each temperature of total darkness, ½, 1, 2, 4, 8, 12, or 24 hr. Seed germination of the Alabama provenance was greater than the North Carolina provenance for all treatments. There were no significant differences in percentage germination between 25/15C (77/59F) and 30/20C (86/68F) for any durations of stratification for either provenance. Regardless of stratification, germination was lowest at 25C (77F) for both provenances. When nonstratified seeds from the North Carolina provenance were germinated at photoperiods ≤ 12 hr, total germination never exceeded 5%, indicating an obligate light requirement. On the other hand, an obligate light requirement was not observed for seeds from the Alabama provenance since 15% of the nonstratified seeds germinated in darkness. However, for both provenances, stratification and daily photoperiods ≥ ½ hr greatly increased germination. The North Carolina provenance required 90 days stratification to maximize germination (66%), whereas the Alabama provenance needed only 30 days (80%). High germination percentages were due, in part, to rigorous seed cleaning.

Index words: *Chamaecyparis thyoides*, conifer, sexual propagation, native plants, timber species, wetlands restoration.

Significance to the Nursery Industry

Germination requirements of an Alabama and a North Carolina provenance of Atlantic white-cedar are different. Thus, stratification (moist-prechilling), temperature, and photoperiod treatments needed to maximize germination varied depending on the provenance. Stratification for 30 days followed by germination at alternating temperatures of 30/20C (86/68F) or 25/15C (77/59F) with daily photoperiods ≥ ½ hr were needed to maximize germination of seeds from Alabama. On the other hand, similar results for a North Carolina provenance at the same temperatures required 60 to 90 days stratification and a daily photoperiod ≥ ½ hr during germination. Seed viability of Atlantic white-cedar is inherently poor, thus requiring rigorous seed cleaning prior to sowing. Seeds should be left uncovered following sowing because they are relatively small and require light to maximize germination. These results provide a better understanding of how to maximize seed germination of Atlantic white-cedar, which will aid growers in production of seedling transplants for wetlands restoration and for landscape use.

Introduction

Atlantic white-cedar [*Chamaecyparis thyoides* (L.) B. S. P.], also known as southern white-cedar or swamp cedar, has a wide distribution. This evergreen tree grows in a narrow coastal belt, 80–209 km (50–130 miles) wide from the southern coast of Maine to South Carolina, with isolated stands in Georgia and eastern Florida, and a large stand located from the panhandle of Florida to Mississippi (11). This species of *Chamaecyparis* Spach (false cypress) is the only one native to the eastern United States (6). The narrow, conical form of the tree supports horizontal to pendulous branches with soft foliage. The tree reaches a height of 12–22 m (40–75 ft) with older trees devoid of branches for ¾ of their height (6, 21, 23).

Although Atlantic white-cedar is found over a large geographic area, pure stands are relatively small (21). The species occurs on wet sites in acidic, fresh-water swamps and bogs near sea level and along stream banks (6, 14). Trees usually grow on hummocks slightly elevated above the forest floor on shallow, peat-covered soils (13). Cedar wetlands provide a refuge for rare, endangered, or threatened species of plants and animals (13). Seedlings are also a favorite browse for whitetail deer, rabbits, and meadow mice (14). Excessive browsing by deer has been attributed to reduction of natural regeneration in the New Jersey Pine Barrens following clearcutting or wildfires (17).

Yield of white-cedar in pure stands is large, due to its long trunk and narrow crown (1). The wood is extremely resistant to decay, and logs of old trees, that are buried in peat bogs, are salvaged for use. Due to several desirable properties, the highly prized wood is used for a variety of purposes including house siding and outdoor furniture (16). This evergreen tree also has the potential for wetlands restoration and as an ornamental and Christmas tree (10). Understocks from Atlantic white-cedar may be used to graft superior cultivars of other species of *Chamaecyparis* (19).

Throughout its range, natural stands of Atlantic white-cedar are diminishing rapidly. Acreage of white-cedar in North Carolina alone has declined by as much as 90% within the last 2 centuries (8). Stands are diminishing due to extensive

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[2]Assistant Professor, Dept. of Horticulture, 1575 Linden Drive, University of Wisconsin, Madison, WI 53706-1590.

[3]Professor.

drainage, agricultural clearing, wildfires, and logging (2, 22). This destruction has been followed by inadequate regeneration measures and changes to hydrology of the land (3).

Reproduction of Atlantic white-cedar is primarily through natural seeding with some regrowth in areas browsed heavily by deer. However, white-cedar usually fails to regenerate naturally after logging when no measures are taken to control competing vegetation (18). At 4–5 years of age, seed production of Atlantic white-cedar begins in open stands, 10–20 years in dense stands, and usually results in fair to copious amounts of seeds being produced each year (11, 15). However, attempts at artificial regeneration have failed due, in part, to the inability to produce enough seedlings for production due to a very poor seed-to-seedling ratio (3). In general, seed germination of species of *Chamaecyparis* has been reported to be inherently low, due in part to reduced seed quality, insect damage to seeds, and various degrees of embryo dormancy (9, 12).

Due to extensive restoration efforts, there is increasing demand for transplants of Atlantic white-cedar. Seed propagation, however, is used infrequently due, in part, to lack of published protocols. Therefore, the objectives of this research were to examine the influence of stratification (moist-prechilling), temperature, and light on seed germination of two provenances of Atlantic white-cedar.

Materials and Methods

Mature cones from two provenances (Wayne Co., NC, and Escambia Co., AL) of Atlantic white-cedar were harvested from open-pollinated trees during September to November 1994. Cones collected from the Wayne Co., NC, provenance were from 3- to 4-year-old trees, whereas cones from the Escambia Co., AL, provenance were from 40- to 45-year-old trees. Cones were collected from a representative group of trees within each provenance and were dried on wire mesh racks at 21C (70F) for 2 months. Many of the cone scales closed and hardened, preventing seed extraction. To enhance seed release further, cones with closed scales were soaked overnight in water, drained, and allowed to dry for 4 days at 30C (86F). Following extraction, seeds were stored in sealed glass bottles at 4C (39F) for 6 months at a moisture content of 5.7% and 5.8% for the North Carolina and Alabama provenances, respectively. Moisture content of each provenance was determined by calculating the mean moisture content of six 100-seed samples of cleaned seeds following drying at 105C (221F) for 24 hr.

In June 1995, seeds were removed from storage and cleaned initially with the use of an air column (General Seed Blower-Model ER, Seedburo Intl. Equip. Co., Chicago, IL). To remove chaff and empty seeds, the device was operated for 10 min by using a #2 (60 mesh) screen with a setting of 21 (North Carolina seeds) or 20 (Alabama seeds), both with all ports closed. Seeds retained on the pan screen were then subjected to additional cleaning. Abnormal, damaged, undersized or discolored seeds, and other large debris not eliminated by the air column, were removed manually. Cleaned seeds [approximately 29,000 pure seeds per 28 g (1 oz)] selected for the research, were firm with a dark brown color. Cutting tests estimated germinative capacity of cleaned seeds of the North Carolina and Alabama provenances to be 92% and 89%, respectively.

Cleaned seeds were then stratified (moist-prechilled) for 0, 30, 60 or 90 days at 4C (39F). Dry sand was sieved through a 18-mesh screen [1.0 mm (0.04 in)] and the fine separate retained. One-hundred cleaned seeds were mixed with 20 ml (0.68 fl oz) moist sand [dry sand:water, (10:1 by vol)] and were placed in 476 ml (1 pt) nonvented, polyethylene freezer bags. After the designated stratification interval, 96 randomly selected bags from each provenance were removed from stratification. Seeds were separated from sand by flushing with tap water in a colander and sown in covered, 9-cm (3.5 in) glass petri dishes (100 seeds per dish). Each dish contained two prewashed (rinsed) germination blotters (Filtration Sciences Corp., Mt. Holly Springs, PA) uniformly moistened with tap water. All dishes were placed in black sateen cloth bags and allowed to imbibe overnight at 21C (70F). The following day, bags were randomized within three growth chambers [C-chambers (7)] at the Southeastern Plant Environment Laboratory, Raleigh, NC. The chambers were maintained at 25C (77F) or at 8/16 hr thermoperiods of 25/15C (77/59F) or 30/20C (86/68F). Chamber temperatures varied within ± 0.5C (0.9F) of the set point.

Within each temperature regime, seeds were subjected daily to the following photoperiods: total darkness, ½, 1, 2, 4, 8, 12, or 24 hr. Regardless of stratification and temperature, photoperiod treatments were administered the same time each day. All photoperiod treatments for the alternating temperatures of 25/15C (77/59F) or 30/20C (86/68F) began with the transition to the high-temperature portion of the cycle, with the exception of total darkness and 24 hr.

Growth chambers were equipped with cool-white fluorescent lamps that provided a photosynthetic photon flux (400–700 nm) of 27–36 $\mu mol \cdot m^{-2} \cdot s^{-1}$ (2.1–2.7 klx) as measured outside the dishes with a cosine-corrected LI-COR LI-185 quantum/radiometer/photometer (LI-COR, Lincoln, NE). All photoperiod treatments, except total darkness and 24 hr, were regulated by removal and placement of the petri dishes in black sateen cloth bags. For the 24-hr photoperiod treatment, the petri dishes remained continuously unbagged in open chamber conditions. Regardless of the photoperiod, temperatures within the petri dishes, as measured by a thermocouple, never exceeded ambient temperature by more than ± 1C (2F) of the set point. The constant-darkness treatment was maintained by keeping the petri dishes in the black cloth bags throughout the experiment, and all watering and germination counts were performed in a darkroom utilizing a fluorescent lamp equipped with a green acetate filter (Rosco Laboratories, Port Chester, NY). Germination blotters were kept moist with tap water throughout the duration of the experiment. Seeds showing signs of decay were removed immediately from the dishes.

For each provenance, all treatments were replicated four times with a replication consisting of a petri dish containing 100 seeds. Germination counts were recorded every 3 days for 30 days. A seed was considered germinated when radicle emergence was ≥ 1 mm (0.04 in). Percentage germination was calculated as a mean of four replications per treatment.

The experimental design was a split-split plot with temperatures as the main plots, stratification treatments as the subplots, and provenances and photoperiods as the sub-sub plots. Data were subjected to analysis of variance procedures and regression analysis (20). Data were also analyzed following arcsin transformation, and results were similar to the nontransformed data. Hence, results presented are based on nontransformed data. Mean separation tests were completed by least significant difference (LSD) procedures at $P = 0.05$.

Results and Discussion

Total percentage germination (total germination at the end of the 30-day germination period). Seed germination of the Alabama provenance was higher than that of the North Carolina provenance for all treatments (Figs. 1 and 2). Variation in seed germination of New Jersey provenances of Atlantic white-cedar reportedly ranges from a high of 70%–90% (11) to a low of 3%–25% (14). For each provenance, there were no significant differences between 25/15C (77/59F) and 30/

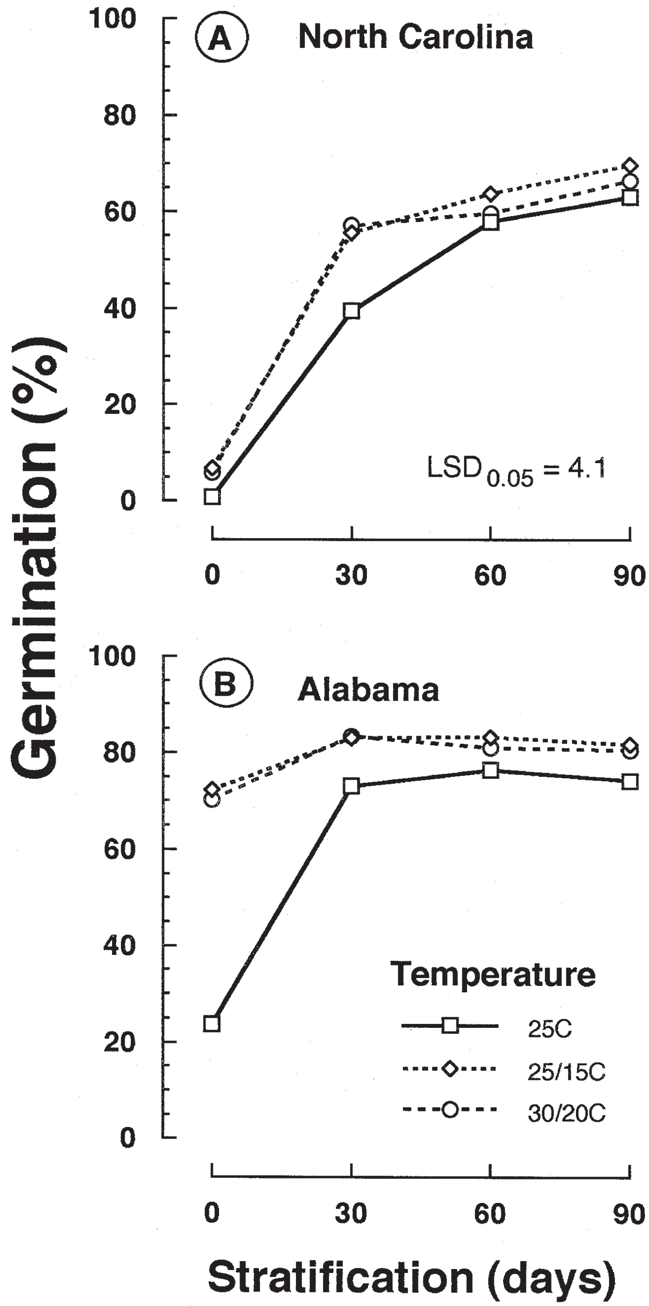

Fig. 1. Influence of temperature and stratification on total percentage seed germination of two provenances of Atlantic white-cedar combined over all photoperiods. $LSD_{0.05}$ = 4.1 for comparisons between provenances when two factors are held constant: duration of stratification and temperature. (A) North Carolina provenance stratified for 0, 30, 60 or 90 days and germinated at 25C (77F) or 8/16 hr thermoperiods of 25/15C (77/59F) or 30/20C (86/68F). (B) Alabama provenance germinated using the same treatments as in A.

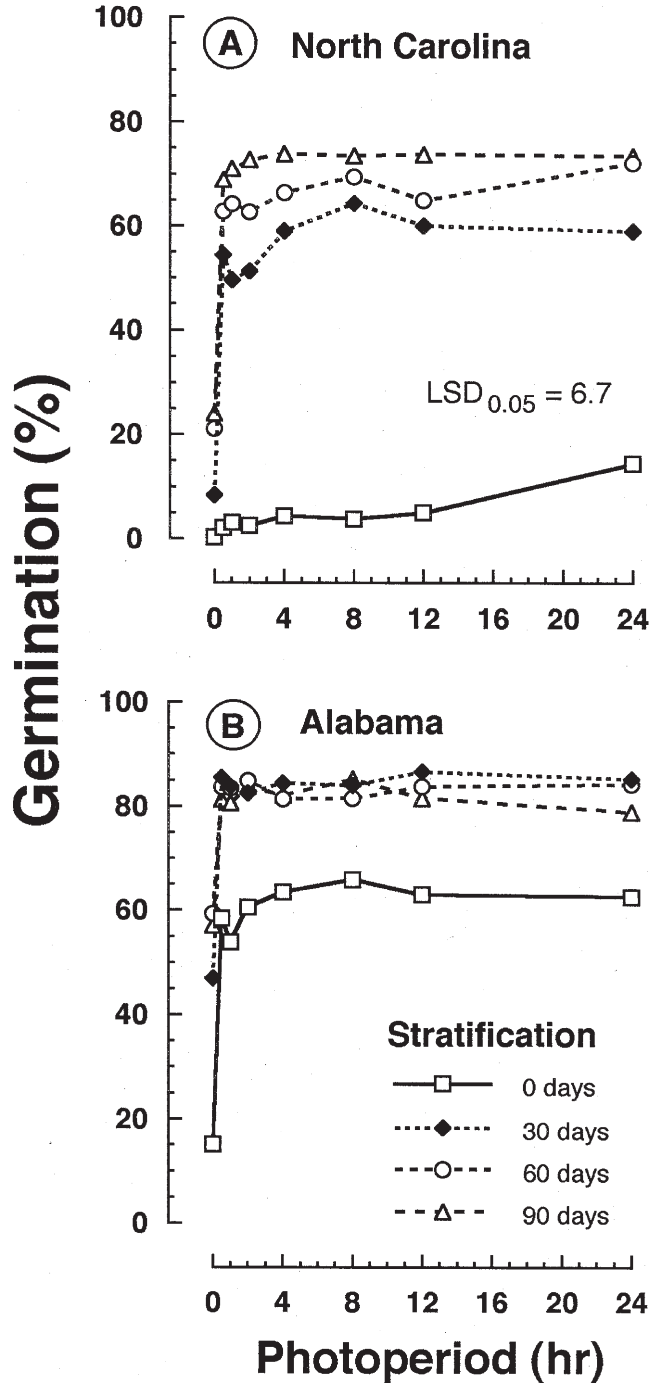

Fig. 2. Influence of stratification and photoperiod on total percentage seed germination of two provenances of Atlantic white-cedar combined over all germination temperatures. $LSD_{0.05}$ = 6.7 for comparisons between provenances when two factors are held constant: duration of stratification and photoperiod. (A) North Carolina provenance stratified for 0, 30, 60 or 90 days and germinated at photoperiods of 0, 1/2, 1, 2, 4, 8, 12 or 24 hr. (B) Alabama provenance germinated using the same treatments as in A.

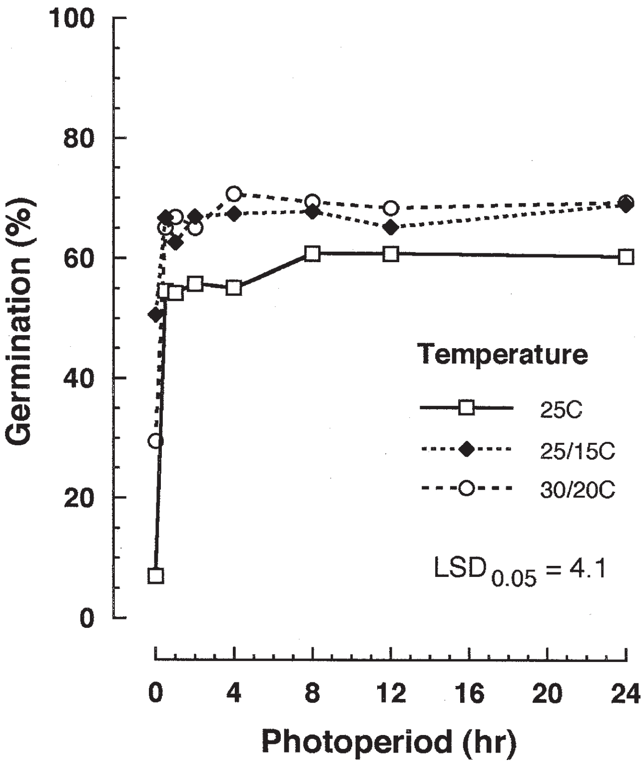

Fig. 3. **Influence of temperature and photoperiod on total percentage seed germination of Atlantic white-cedar combined over both provenances and all durations of stratification. $LSD_{0.05}$ = 4.1 for comparisons between temperatures with photoperiod held constant. Seeds were germinated at 25C (77F) or 8/16 hr thermoperiods of 25/15C (77/59F) or 30/20C (86/68F) with photoperiods of 0, 1/2, 1, 2, 4, 8, 12 or 24 hr.**

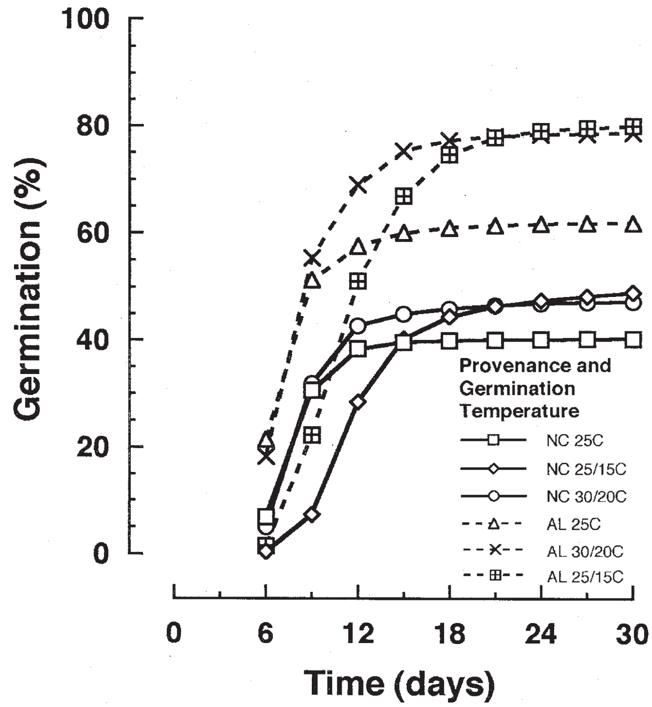

Fig. 4. **Influence of temperature on cumulative percentage seed germination of two provenances of Atlantic white-cedar combined over all durations of stratification and all photoperiod treatments. Seeds were germinated at 25C (77F) or 8/16 hr thermoperiods of 25/15C (77/59F) or 30/20C (86/68F).**

20C (86/68F) for total percentage germination at all durations of stratification (Figs. 1A and 1B). Regardless of the duration of stratification, germination was lowest at 25C (77F) for both provenances. In some cases, however, germination of the North Carolina seed was similar when stratified for 60 or 90 days and germinated at 30/20C (86/68F) or 25C (77F). Similarly, Bianchetti et al. (3) reported greater germination (40%) of stratified seeds of Atlantic white-cedar at an alternating thermoperiod of 30/20C (86/68F) versus constant temperatures of 23C (73F) (16%) or 26C (79F) (19%).

Nonstratified seeds from the North Carolina provenance germinated at photoperiods ≤ 12 hr never exceeded 4.9% (Fig. 2A). However, germination increased with increasing durations of stratification up to 90 days. In contrast, germination of nonstratified Alabama seeds exceeded 54% at photoperiods ≥ ½ hr (Fig. 2B). This provenance required only 30 days stratification for maximum germination. In another study, the effect of stratification on increasing germination of Atlantic white-cedar was marginal (3).

Little (14) reported 'a fair amount of light, probably to provide heat, is desirable for obtaining good seed germination of white-cedar.' Without stratification and with daily photoperiods of 0 to 12 hr, seed germination of the North Carolina provenance ranged from 0.3% to 4.9% indicating an obligate light requirement. With a 24 hr photoperiod, germination increased to 14.5%. On the other hand, light was unnecessary for seeds from Alabama since 15% of the nonstratified seeds germinated in darkness. However, for both provenances, stratification and daily photoperiods ≥ ½ hr greatly increased germination.

Light reportedly is an absolute requirement for seed germination of many swamp-inhabiting species (5). In the present study, seeds kept in the dark but subjected to increasing durations of stratification had increased germination, especially for the Alabama provenance (Fig. 2B). Boyle and Kuser (4) reported that increased durations of cold stratification can substitute for the light requirement. At 25C (77F), fewer seeds from both provenances germinated than those at 25/15C (77/59F) or 30/20C (86/68F), regardless of photoperiod and stratification (Fig. 3). There were no significant differences in germination at 30/20C (86/68F) or 25/15C (77/59F) for all photoperiods except 0 and 1 hr.

Cumulative percentage germination (percentage germination recorded every 3 days for 30 days). For all treatment combinations, germination was essentially complete for both provenances by day 18 (Figs. 4, 5, and 6). There was a highly significant quadratic response ($P = 0.0001$) for cumulative percentage germination for both provenances and at all germination temperatures (Fig. 4).

Without stratification, only 4.5% of the North Carolina seeds germinated compared to a maximum of 55.4% for the Alabama provenance (Figs. 5A and 5B). The North Carolina provenance required 90 days stratification to maximize germination (66.4%), compared to 30 days (80%) for the Alabama provenance. Boyle and Kuser (4) also reported similar seed germination percentages among several New Jersey provenances of Atlantic white-cedar when stratified for 30 or 60 days. However, seed decay prevented analysis of their data for a 90-day stratification treatment. There was a highly significant quadratic response ($P = 0.0001$) for the Alabama provenance at all four durations of stratification. For the North Carolina provenance, there was also a highly significant quadratic response at 30, 60, and 90 days stratification, with a significant linear response ($P = 0.0001$) at 0 days stratification.

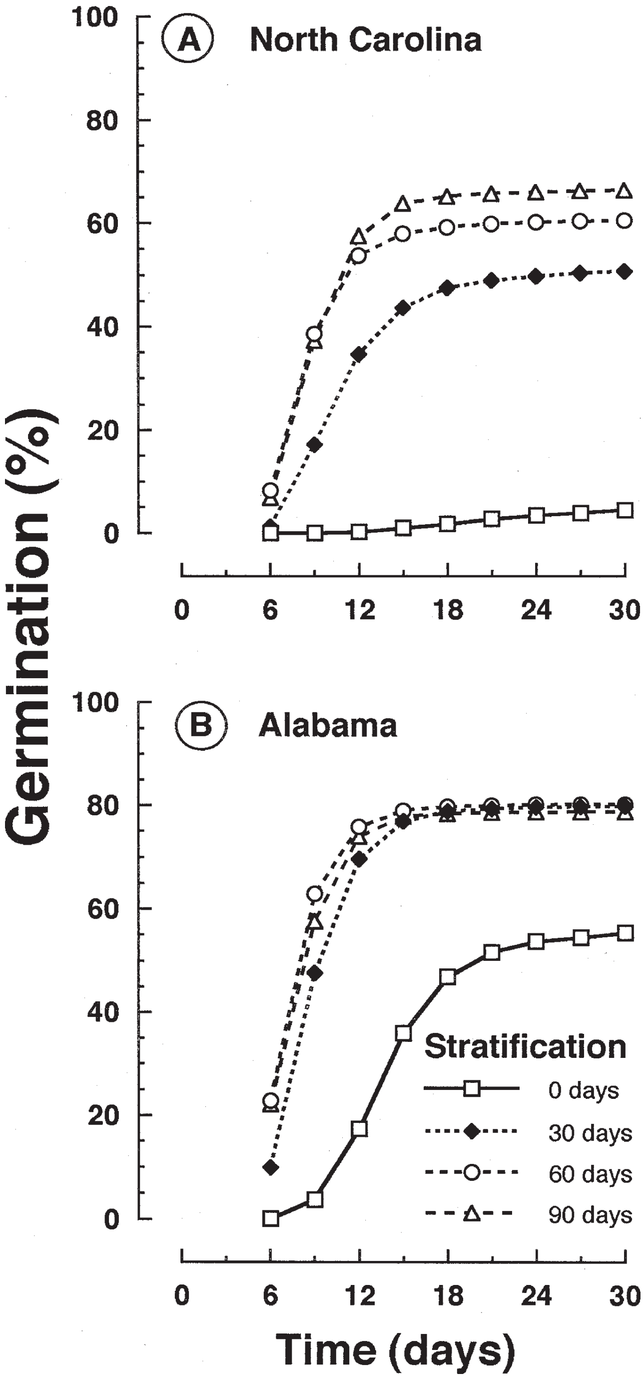

Fig. 5. **Influence of stratification on cumulative percentage seed germination of two provenances of Atlantic white-cedar combined over all photoperiods and temperatures. (A) North Carolina provenance stratified for 0, 30, 60 or 90 days. (B) Alabama provenance germinated using the same treatments as in A.**

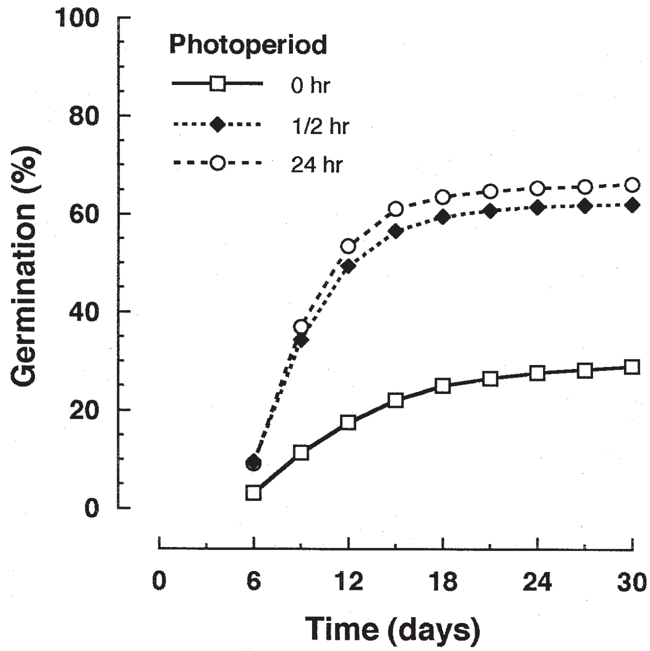

Fig. 6. **Influence of photoperiod on cumulative percentage seed germination of Atlantic white-cedar combined over both provenances and all germination temperatures and durations of stratification. Seeds were germinated under photoperiods of 0, 1/2, 1, 2, 4, 8, 12 or 24 hr. Data for the 1, 2, 4, 8, and 12 hr photoperiods were omitted since they were intermediate between the 1/2 and 24 hr photoperiod treatments.**

A significant quadratic response ($P = 0.0001$) for cumulative germination was noted for all photoperiod treatments with slightly lower significance for 0 hr ($P = 0.002$) (Fig. 6). Combined cumulative germination for both provenances in the absence of light, never exceeded 29%. Daily photoperiods of ½ hr and 24 hr yielded comparable results (62% vs. 66%). Data for photoperiods of 1 to 12 hr were intermediate between those of ½ and 24 hr. In contrast, germination of seeds from New Jersey is apparently under photoperiodic control, e.g., photoperiods of 16 hr yielded 32% germination, compared to 0.7% for 10 hr (4).

Because preliminary germination studies indicated low seed viability, rigorous seed cleaning procedures were utilized. Relatively poor germination of uncleaned seeds may explain why growers have often stated that high germination percentages of Atlantic white-cedar are difficult to accomplish. Also, seed germination requirements for Atlantic white-cedar appear to vary according to provenance and deserve further study, particularly study of provenances representing its extensive geographic range.

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