

Short day treatment of Norway spruce - stops height growth and promotes the development of freezing tolerance

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Figure 1. Short day treatment in an open field using black curtains. Photo Anders Lindström.

Conifers are adapted to survive in regions where conditions change profoundly between seasons. During fall, they enter a dormant phase and while resting they develop freezing tolerance to survive a harsh winter. In spring they resume growth as a response to increasing temperatures. As night length increases again towards the end of summer and night temperatures drop, trees start developing freezing tolerance.

Norway spruce is particularly sensitive to changes in night length. This reaction can be utilized in forest tree nurseries by covering the seedlings with black curtains (Figure 1) to extend night length for 2-4 weeks in late summer. Short day (SD) treatment of Norway spruce seedlings in July stops height growth and promotes bud set and development of freezing tolerance, which makes seedlings

less frost-sensitive. Scots pine also reacts to changes in night length but normally SD treatment is not necessary. It is often sufficient to transfer pine seedlings from the greenhouse to outdoor growing conditions well in advance of the longer nights in late summer.

Background

As forest tree nurseries started to switch production from bareroot to containerized seedlings during the late 1960s, Ingegerd Dormling at the phytotrone in Stockholm performed a series of experiments forcing Norway spruce seedlings to grow continuously over a number of years. The initial objective was to promote earlier and faster flowering of the species. Dormling showed that if each growth cycle was terminated with 3 to 4 weeks of long night treatment (to obtain dormancy), followed by additional weeks of exposure to low temperatures (0 to 5°C), Norway spruce seedlings could complete four growth cycles per year.

During the 1970s and 80s Dormling *et al.* performed several extensive studies to investigate the required number of hours in darkness to obtain dormancy for Norway spruce seedlings of different origins. The term “critical night length“ is well

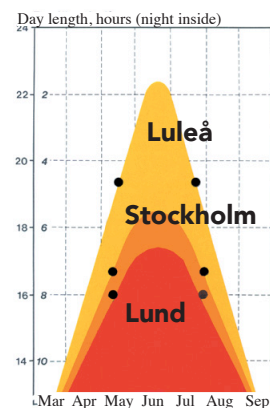


Figure 2. Day- and night lengths from mid-March to mid-September in Luleå, Stockholm and Lund. The critical night length for each local provenance, CN (indicated by small black circles) is reached in May and surpassed again in early August (from Dormling & Lundkvist 1983, Plantaktuellt nr 2, 2001).

established and defined as the period required so that “50% of the population starts to set bud”. This is obtained in late July/early August, for local Norway spruce provenances in Sweden. Therefore, for local material, the critical night length for bud set is shorter for the more northern Norway spruce in Luleå (approximately 3 to 4 hours) compared to southern Norway spruce growing in Lund (approximately 8 to 9 hours) (Figures 2 and 3). In addition to latitude, the altitude also determines when trees start terminal bud formation and start developing freezing tolerance. Higher altitudes correspond to a shorter critical night length (Figure 3).

Studies of the development of freezing tolerance for the same species were also performed in Norway during the 1970s by Ola Heide and others, in which the effects of temperature were included. Ketil Kohman studied the critical night length of Norway spruce in the 1990s. Martin Sandvik performed a series of trials with the intention of improving the way

seedlings were cultivated in Norwegian forest tree nurseries, focusing on increased survival after winter storage. In addition, in Finland, researchers have studied how to use controlled night length as a way to improve seedling storability. Researchers in North America have investigated the impact of photoperiod and temperature on the development of freezing tolerance and how conifers survive temperatures as low as -40°C .

The production of containerized seedlings increased in Sweden during the 1970s and 80s. To meet the need for safe winter storage of this stock type, extensive research was conducted to investigate how light and temperature conditions during fall affect spruce and pine seedlings. During the 1980s, Gunnel Rosvall-Åhnebrink studied how growth cessation, bud set, dormancy, and the development of shoot freezing tolerance related to nursery practices in containerized seedlings. Since then, this research has advanced further in the regions where containerized seedlings are cultivated.

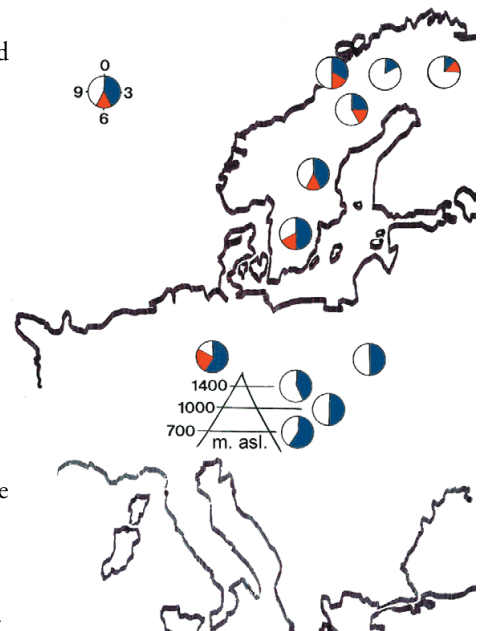


Figure 3. Critical night length CN (the night length required for apical bud set among 50% of the seedlings) for Norway spruce of a specific origin. The blue sector shows the CN resulting from an abrupt switch from continuous light to the specified night length during cultivation in a phytotrone. Blue and red sectors combined represent CN for a more gradual natural increase in night length (from Dormling & Lundkvist 1983, *Plantaktuellt* nr 2, 2001).

Night length controls growth cessation and development of freezing tolerance in Norway spruce

Night length is the most important environmental factor for inducing growth cessation and development of dormancy. Initially, long nights combined with warm temperatures favor the development of freezing tolerance but later during the fall, cold nights are required to reach maximum hardiness. Growth cessation begins towards the end of July as the natural night length increases. Thereafter, seedlings develop buds, a process that requires light and warm temperatures during late summer. After bud set, the seedling shifts to root growth during the first half of September and responds to the cold nights and increasing night length by entering a resting phase, dormancy.

Shoot growth will not occur until the seedling has reached dormancy release. All growth conditions eventually result in dormancy release but the process is promoted by low temperatures around 0°C . Normally, dormancy release is obtained during late fall or early winter. Following dormancy release, seedlings may reflush if they become exposed to favorable growth conditions. During normal years these conditions occur in late spring when the risk of damaging subzero temperatures is low.

When seedlings have reached dormancy, freezing tolerance in shoots and roots

gradually increases. Normally, shoots become freezing tolerant before roots, especially in bareroot seedlings growing in warm soils during fall. Roots are generally more sensitive to low temperatures than shoots. One way to protect containerized seedlings from damaging low root temperatures during fall and winter is to move them off raised pallets and place them directly on the ground or to put them into frozen storage. Another rather common method used in northern nurseries is to cover the seedlings with a protective layer of artificial snow. When the seedlings are stored outdoors, the maximum level of freezing tolerance of shoots and roots is reached in January, followed by dehardening, a process that is accelerated by warm temperatures.

There are a number of different objectives when performing SD treatment of Norway spruce seedlings. Some nurseries use this treatment to stop shoot elongation (see Figures 4 and 5) in order to avoid oversized seedlings, whose tops may be damaged during packing. SD treatment may also increase the root-shoot ratio. Another purpose is to promote the development of freezing tolerance for early frozen storage or autumn planting.

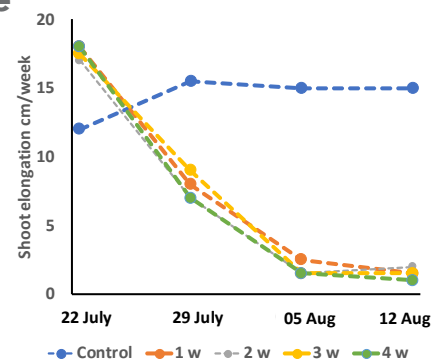


Figure 4. Mean weekly apical shoot elongation for seedlings cultivated and treated with short days (SD) in a greenhouse. SD treatments (with a night length of 13 h) started on July 15 and were performed for 1, 2, 3, and 4 weeks. Seedlings exposed to natural night lengths were used as the control. One week of SD treatment was sufficient to stop height growth (from Wallin et al. 2017).

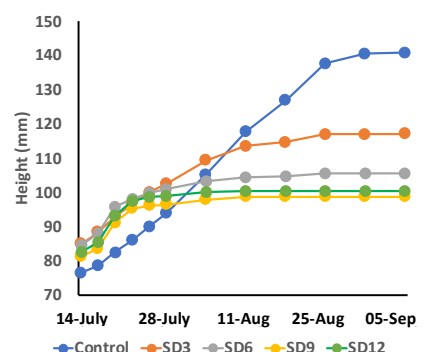


Figure 5. Height growth of Norway spruce seedlings of a central Swedish provenance after short day (SD) treatments (with a night length of 13 h) lasting 0 (=control), 3, 6, 9 or 12 days (SD3-12) in mid-July. With this material, height growth was significantly reduced after as little as 3 days of SD treatment (from Wallin 2018).

Storability tests

It is important to measure storability status before seedlings are transferred to freezer storage. This is usually performed from the middle of September to mid-November. Since the mid-1980s, nurseries in Sweden, Norway and Finland have used a method based on analysis of dry matter content of shoots. Unfortunately, the dry matter content (or water content) has turned out to be a rather imprecise measure of storability. This inaccuracy could partly be explained by the fact that as freezing tolerance increases, dry matter content changes rather slowly. Freezing tests are a more reliable method to assess the seedlings' ability to endure long-term frozen storage. Swedish researchers have shown that if seedlings can withstand freezing to -25°C they can also be successfully stored in darkness at -3°C for 6-7 months.

Based on these results a test was developed: the electric conductivity (EC)-method. In this test, the upper two centimeters of the shoots are cut off and slowly frozen to -25°C and then the ion leakage from damaged cells is measured. High conductivity values indicate shoot damage caused by low temperatures, which means that seedlings are not yet storable. Even when the shoots have been classified as storable, care must be taken to avoid root damage caused by freezing too rapidly. One option is to keep the seedlings in cold storage (temperature just above 0°C) to allow further hardening of the roots for a couple of weeks before the seedlings are transferred to the freezer (-3°C). A com-

mercial gene activity test for determination of seedling storability has also been developed.

Short day treatment and bud burst

SD treatment affects the timing of bud burst in spring. Several studies have shown that a longer period (21 or 28 days) of SD treatment results in earlier bud burst the following spring compared to shorter treatments (7 or 14 days) as well as no treatment. Therefore, longer SD treatments may increase the risk of spring frost damage.

This problem can be avoided by using frozen stored SD treated seedlings for planting at sites with increased risk of spring frost, since such seedlings are frost tolerant for a couple of weeks after thawing, the alternative is simply to postpone spring planting. However, when planting SD treated seedlings in the fall, special attention needs to be directed towards risks associated with early bud burst in spring.

Another issue to consider is that SD treated seedlings may have a second bud flush in fall. According to Norwegian studies, this risk decreases with longer periods of SD treatment and with late termination of the treatment (see figure 6).

Studies of gene activity

Since the 1980s, several studies have investigated the development of freezing tolerance of seedlings. For one-year-old Norway spruce seedlings, a study by

Wallin et al. (2017) illustrates how the gene activity in cells changes during fall. It shows that different gene systems are probably involved in controlling growth cessation, bud set, dormancy, and shoot freezing tolerance. SD treatment promotes dormancy induction, and in a subsequent step, the development of shoot freezing tolerance.

The results show a clear correlation between longer (21 or 28 days) SD treatment and increased activity of dormancy-related genes (Figure 7). Freezing tests were performed on shoots from one-year-old seedlings to confirm this association between longer SD treatment and gene activity. Seedlings exposed to a longer (21 or 28 days) SD treatment developed greater freezing tolerance and became storable earlier compared to the shorter (7 or 14 days) treatments and untreated control seedlings. However, a short SD treatment (7 days) also had a positive effect on the development of dormancy and freezing tolerance. Besides giving a distinct signal for shoot growth cessation (see figure 4) the short SD treatment (7 days) also gives the seedlings a small, but nonetheless measurable, advantage during the development of freezing tolerance in fall compared to control seedlings.

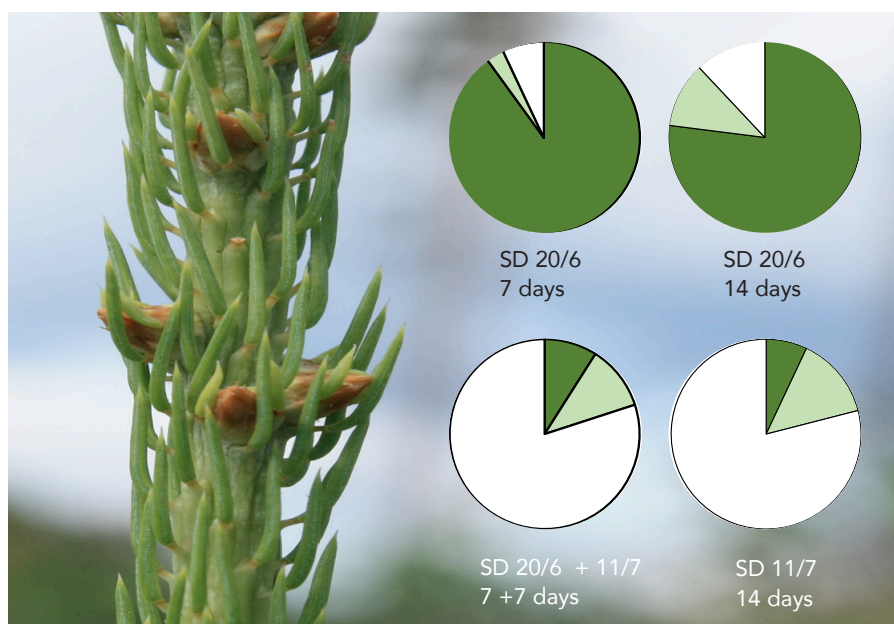


Figure 6. Proportion of seedlings with a second bud flush during fall following short day (SD) treatments with different starting dates and durations. Dark green: both apical and lateral buds. Light green: only lateral buds. White: no reflusing. No second bud flush during fall was observed for non-SD treated control seedlings (based on Floistad & Granhus 2019).

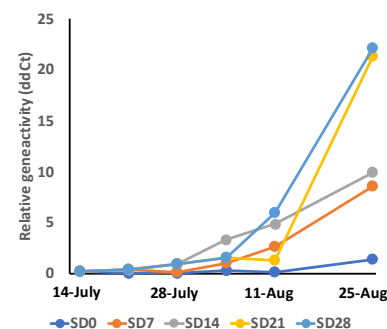


Figure 7. Gene expression (ddCt) for one of the genes indicating dormancy induction in seedlings transferred to outdoor growing conditions after termination of short day (SD) treatment. A longer SD treatment (21 or 28 days, SD21 and SD28) resulted in increased gene activity. These seedlings reached dormancy earlier than the seedlings subjected to a shorter treatment (7 or 14 days, SD7 and SD14). SD0 represents control seedlings exposed to natural night length. For the same material, gene activity was lower in seedlings remaining in the greenhouse after treatment, indicating that both longer nights and lower night temperatures are required to obtain sufficient hardiness (from Wallin et al. 2017).

SD treatment in practice

At nurseries located in southern or mid-Sweden, SD treatments normally start during late June to mid-July (Figure 8). The seedlings are placed under automatically operated black curtains that completely cover the seedlings for about 11-14 hours a day. A rule of thumb is to give the seedlings approximately **5 hours longer night than the critical night length for the specific provenance**. This blackout treatment continues for 2-4 weeks, either in a separate field outside the greenhouse or, which is less common, in the greenhouse. When the treatment is completed, the seedlings are transferred to an open field to be exposed to natural night length.

Several studies from Sweden, Norway, and Finland have shown that the timing of start and termination of SD treatment strongly affects the result. If the treatment starts in late June, it is very important to keep the seedlings under the black curtains for at least 4 weeks so that the treatment ends when the natural night length is the same or longer than the critical night length for the specific provenance. Otherwise, there is a risk of a second bud flush by the end of August.

If the SD treatment is terminated too early, the seedlings have only partially entered dormancy. The apical bud may have become dormant while the lateral buds have only reached a lower level of dormancy and could therefore easily resume growth. A second bud flush during fall is sometimes referred to as reflushing or prolepsis. If the main purpose of early SD treatment is to suppress shoot elongation, it could be combined with a second treatment later in the season. This split SD treatment both induces growth cessation and prevents a second bud flush in autumn (see figure 6). The interruption in SD treatment gives the seedlings an opportunity to further increase their root collar diameter after termination of shoot elongation.

It is important to provide seedlings with favorable growth conditions following bud set in autumn. In this phase, seedlings require a great deal of light combined with low night temperatures to develop freezing tolerance. The curtains used for SD treatment may increase night temperatures and if this is combined with limited access to light, SD treatment during late August may delay the natural

development of cold hardiness. If SD treatment begins in late July, a short treatment (2 to 3 weeks), may be sufficient to make the seedlings storable earlier. The reason is that the SD treatment then starts at approximately the same time as the critical night length occurs naturally. Nursery location in relation to the origin (provenance) of the seedling material is of course something to consider when applying SD treatment programs. When cultivating provenances from a more northerly location than the nursery, the seedlings will often manage without treatment, while southern provenances grown at the same location will require SD treatment to be able to achieve sufficient freezing tolerance in time. For the southern provenances, it is of particular importance to avoid light leakage since even very low levels of light will stimulate further growth. More northern provenances are not as sensitive to low light levels under the curtains.

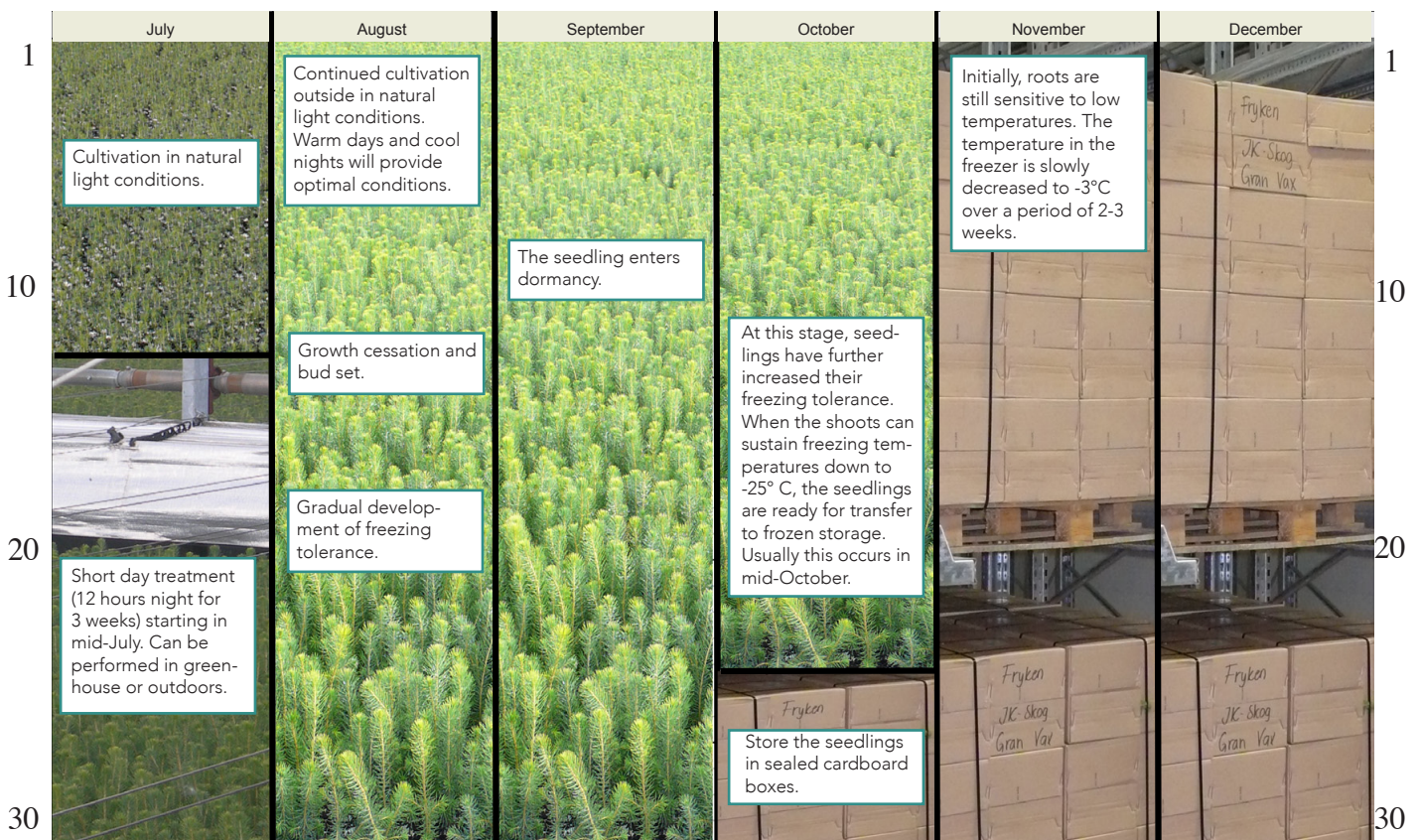


Figure 8. An example of a standard cultivation program for Norway spruce originating from approximately 60°N and a critical night length of 7 hours.

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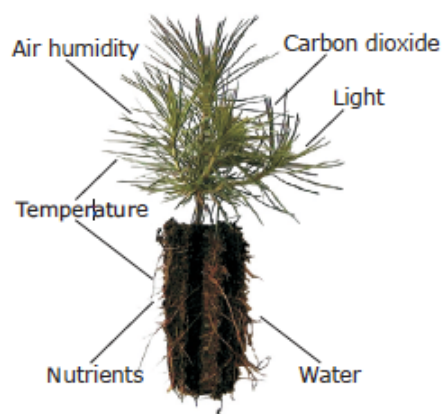
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PLANTskolan is an educational material about forest plant production and regeneration, originally produced for the magazine *PLANTaktuellt*. The series contain, with this issue, 17 lessons. All previous lessons are written in Swedish.

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