

# THE EFFECT OF DIFFERENT STRATIFICATION METHODS ON THE GERMINATION OF *ACER PLATANOIDES* AND *ACER CAMPESTRE* SEEDS

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**Abstract:** The aim of this study was to assess the effect of temperature on the germination response in six origins of *Acer platanoides* and *Acer campestre* seeds. Seeds of both species were exposed to four methods of stratification: cold (3 °C) and warm (20 °C), with and without sand-peat medium. The highest percentage of germination (54.75%) was obtained, for *Acer platanoides*, when the treatment was conducted at 3 °C. The initiation of germination with a low percentage of germinated seeds (12.75% to 22.25%) determined the initiation of secondary dormancy for the majority of *A. platanoides* seeds. In the case of the *A. campestre* seeds, the pretreatment phase in cold environment (19weeks) proved to be too short for these origins.

**Key words:** *Acer platanoides*, *Acer campestre*, dormancy.

## 1. Introduction

An important characteristic of temperate tree species is seed dormancy. Whilst most *Acer* species - including *A. campestre*, *A. monspessulanum*, *A. platanoides*, *A. pseudoplatanus*, *A. glabrum* - exhibit dormancy [8], [9], [11-13], some other species, like *A. rubrum* and *A. saccharinum*, do not require any pre-germination treatment [14].

Seeds of many tree species are released from dormant state when they are kept at relatively high humidity and generally low temperatures (from 1.5 °C to 15 °C). After dormancy removal, proper germination can usually take place at higher temperatures.

This artificial overcoming of seed dormancy by placing the seed under

generally cool and moist conditions (sand, peat etc.) for a period of time is called stratification.

Cold treatment is efficient in removing many types of seed dormancy. The embryonic dormant state which is present in seeds of *Acer platanoides*, can be removed after seed storage at high temperature, leaving only the dormant state imposed by seed coat. This is removed by further keeping the seeds at low temperatures. Seeds of other species, such as *Acer negundo* and *Acer pseudoplatanus*, shortly after maturity present a dormant state imposed by coat, which can be removed by keeping them at low temperatures [1].

The treatment of *Acer* seeds with gibberellinic acid solution had a positive

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result on dormancy breaking when applied at the same time with cold stratification. However the treatment of seeds with such a stimulator as a substitute of stratification remained without positive results [3].

Recent studies, demonstrated that seed dormancy of *Acer platanoides* could be broken if seeds were kept a period of time at a relatively constant humidity of 36% and a temperature of 0-1 °C [6]. The increase of temperature to 5-7 °C delayed the dormancy removal and a 15-18 °C temperature induced the secondary dormancy in seeds.

For the seeds of *Acer campestre* the dormant state was associated with the pericarp impermeability. Their dormancy can be removed if stratified for a month in warm condition, followed by 6-7 months of cold stratification, depending on the origin of seeds [2].

Muller et al. (1989) stated that the treatment should end when the percentage of the germinated seeds reaches 10-20%; subsequently, it is followed by germination tests.

Earlier investigations, evidenced that placing the seeds at high temperature (20° C) or at alternative temperature (20/3 °C, 8+16 hours a day) right after the germination signs begin to show, may cause the initiation of secondary dormancy for most of the seeds [5].

For nursery practice purposes it is important to identify an effective stratification method that allows for seed sowing next spring after harvesting. In this paper we assessed the impact of four different stratification conditions on the germination of *Acer platanoides* and *Acer campestre* seeds, in order to identify a suitable pre-germination treatment for seedling production.

## 2. Materials and Methods

Seeds of both species were collected in 2009 (26-29 October) from three origins from Southern Romania (Table 1).

A tetrazolium test was performed in order to assess the initial viability of seeds from each origin, according to ISTA [15] provisions (4 repetitions of 100 seeds), after the manual grading of seeds and the removal of the sterile ones at the beginning of November 2009 [4].

The 1000-seed weight was calculated based on 8 replications of 100 seeds.

In order to establish a suitable moisture for germination, seed imbibition was evaluated by gravimetric method at room temperature [7], using for each origin of the two species three repetitions of 50 seeds, as described by Drăghici and Abrudan (2011). Seeds were stored in a dark cooler at 4 °C until they were used in spring 2010.

Table 1  
*The origin of Norway maple (Acer platanoides) and field maple (Acer campestre) seed*

Seed Origin	Forestry Directorate	Forest District	Management Unit	Sub-compartment	Altitude (m)
<i>Acer platanoides</i>					
Târgovişte	Târgovişte	Târgovişte	VI Valea Bratului	152E	260
Balş B	Slatina	Balş	V Bistrita	137C	137
Balş C	Slatina	Balş	III Calui	10A	136
<i>Acer campestre</i>					
Târgovişte	Târgovişte	Târgovişte	VI Valea Bratului	152E	285
Sadova	Craiova	Sadova	III Lunca Jiului	40A	47
Balş	Slatina	Balş	III Calui	10A	135

Each treatment listed hereafter was applied to the seeds of both species:

- **Treatment 1 (T1): Cold treatment (3 °C) with medium.** Four repetitions of 100 seeds were mixed with a sand/peat medium in a volume ratio of 1:3 and were placed in plastic containers. The medium consisted of an equal proportion of sand and peat, having a relative humidity of 29% (wetting degree U6 = wet), which was measured with the ECH2O EC-5 sensors for soil moisture [10].

After weighing, the containers were kept at a temperature of 3 °C in the seed conservation centre of ICAS Braşov [4]. Medium moisture was assessed daily, by weighing and filling with water the deficit that occurred from the previous day. Once a week the seeds were removed from containers and medium aeration and homogenization were performed. At the same time, the seed moisture was measured with a thermo balance (Rumed, MB 45).

- **Treatment 2 (T2): Warm treatment (20 °C) with medium.** The seeds (4x100) of each origin were kept at 20 °C, in the sand/peat medium, placed in plastic containers. The assessment/monitoring procedure was the same as in the case of the cold treatment.

- **Treatments 3 and 4 (T3 and T4): Cold (3 °C) and warm (20 °C) treatments without medium.** Two groups of seeds (4x100 each) were kept in parallel in two climate rooms.

In the first climate room the temperature was set at 20 °C and the relative humidity of air at 95% and in the second climate room a temperature of 3 °C was maintained

at an 80% relative humidity of air. The maximum humidity allowed by the climate room was chosen in both cases because in previous tests it was observed that seeds lost a large amount of water when placed in the climate room.

After stratification, the germination tests were performed at a temperature of 3 °C.

Prior to the initiation of germination tests, the Norway maple seeds were separated from the medium and laid down on moist filter paper at 23 °C.

The germination percentage was calculated for each treatment and species.

### 3. Results and Discussion

The initial seed viability varied between 75.75% and 82.25% for *Acer platanoides* and between 53% and 53.75% for *Acer campestre* (Table 2).

The imbibitions tests showed for all origins a similar water absorption curve (Figure 1).

In the first phase very fast water absorption was recorded, followed by a period of slower absorption, in the second phase.

Based on these results, all seeds from treatments 3 and 4 (without medium, in the climate rooms) were soaked for 48 hours in water in order to reach a moisture content of around 50% before being placed in the climate rooms.

The germination tests in the case of Norway maple seeds began when the cold stratified seeds started to germinate, as follows: 12.75% for Târgovişte origin, 22.25% for Balş B origin and 14.5% for Balş C.

*The initial seed viability*

Table 2

Species	<i>Acer platanoides</i>			<i>Acer campestre</i>		
Seed Origin	Târgovişte	Balş B	Balş C	Târgovişte	Sadova	Balş
Seed viability [%]	82.25	78.25	75.75	50.75	53.75	53
The 1000-seed weight [g]	142.60	106.75	111.62	70.53	82.23	77.76

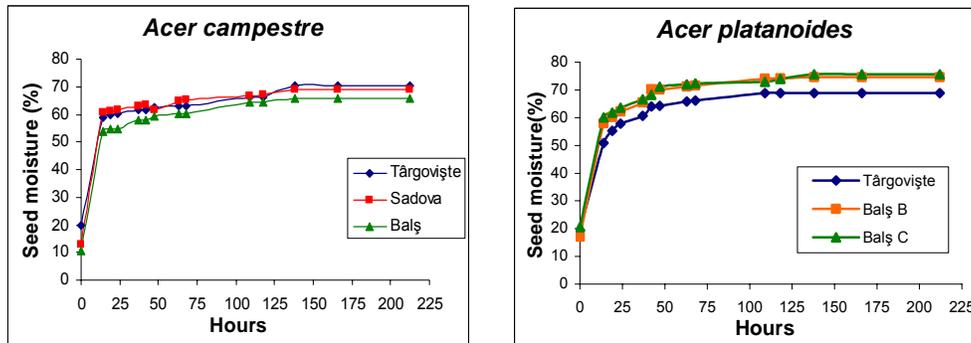


Fig. 1. Water absorption by seeds

The highest percentage of germination (Figure 2) was obtained, for *Acer platanoides*, when the treatment was conducted at 3 °C (T1). The initiation of germination tests and the placement of the Norway maple seeds at 23 °C hindered the germination process; however, a reduced number of seeds continued to germinate.

After seeds stopped germinating, the ungerminated ones were evaluated and the undegraded seeds were treated with tetrazolium.

The analysis proved that the seeds were viable, which leads to the conclusion that

the initiation of germination associated with a sudden shift of the seeds to high temperature, generated the induction of secondary dormancy.

For the *Acer campestre* seeds the pretreatment phase (T1) proved to be too short; consequently, the germination rate in May was very low (2% for Târgovişte origin, 1.75% for Balş origin and 1.75% for Sadova origin).

Seed humidity was kept relatively constant (between 57.11% and 64.52%) throughout the cold treatment with medium (T1) for both species.

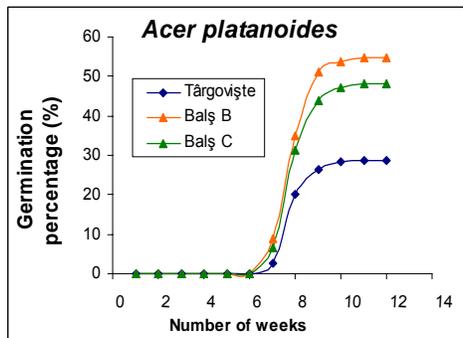


Fig. 2. Seed germination during cold treatment (3°C) with medium

Keeping the seeds in containers with sand/peat medium at high temperature (20 °C) resulted in their decay after 15 weeks of treatment, so this method proved to be ineffective.

Unfavourable results (no germination) were also recorded when applying the two treatments without sand/peat medium (in climate rooms at 3 °C / 80% air humidity and 20 °C / 95% air humidity respectively).

In both treatments, seed humidity decreased very sharply, possibly due to the air ventilation in the climate rooms.

#### 4. Conclusions

For Norway maple seeds, the initiation of the germination tests when 12%-22% of seeds germinated, triggered the inception of secondary dormancy.

The 19-week pretreatment phase in cold medium proved to be too short for the complete elimination of the deep dormancy in the case of the *Acer campestre* seeds. It was also concluded that seeds do not retain humidity constantly when placed in climate room (possibly due to air ventilation) and there was no direct relationship between the relative humidity of the air and seed humidity.

Warm treatment is not recommended for breaking seed dormancy in *Acer spp.* as the seeds decay after 3-4 months of stratification.

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