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Storage of stratified seeds of European ash (*Fraxinus excelsior* L.)*Abstract*

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The effect of dehydration and storage of after-ripened seeds of European ash on its later germinability has been investigated.

It was found that the partial drying of samaras to 12% moisture content after a warm-followed-by-cold stratification (15/3°C, 16+16 weeks) and then storage for 4, 8 or 12 weeks at 3°C or -3°C in sealed bottles has not lowered their germinative capacity. Also there was no change in germinability of seeds after partial drying of samaras to 12.3% of moisture content following the warm phase of the warm-followed-by-cold stratification and storage in this state at -3°C for 12 weeks.

Additional key words: germination, seed moisture content, drying.

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INTRODUCTION

European ash (*Fraxinus excelsior* L.) is characterized by having dormant seeds of the type A₁-B-W₃ (Nikolaeva et al. 1985). These symbols indicate: A₁ — weak inhibitive action of the pericarp on seed germination, B — morphological dormancy, W₃ — deep physiological dormancy.

According to phenological observations (Marinov 1977), the growth of European ash embryo in the seed stops in the milky-waxy stage and in the Bulgarian conditions this corresponds to the end of August. The magnitude of the embryo at the time corresponds more or less to 0.5—0.6 of the length of the endosperm. The placing of isolated embryos or seeds with water available at a temperature not lower than 9—10°C (Nikolaeva and Vorob'eva 1978) results in the growth of the embryo being continued. The optimal temperature for the growth of the European ash embryo varies between 15—16°C (Judin 1973) or 17—20°C (Wciślińska 1977). On the other hand Steinbauer (1937) has found that embryos in seeds of *Fraxinus nigra* Marsh. (a species that has similar thermal requirements for after-ripening) grew fastest at 20°C and much more slowly at 25°C and 30°C, while isolated embryos grew fastest at 30°C. Judin (1973) has shown that both the embryos of ash left in seeds and those

isolated from them grew fastest at 15°C and much more slowly at higher and lower temperatures. Discrepancies concerning the optimal temperature for the growth of ash embryos as observed by various authors are most likely caused by differences in the geographic and edaphic conditions (Varasova 1956, Judin 1967, Marinov 1977, Nikolaeva and Vorob'eva 1978, Rypak 1978/79). The range of occurrence of European ash covers almost the whole Europe, in the west from the Cantabrian Mts. and Ireland to East Europe 48° Long. E. The northern limit of the range includes Norway, Sweden and Finland and then in the northeasterly direction the USSR, while in the south as a rule it does not cross 36° Lat. N. The extent of the range of occurrence of the species (discounting outliers in the East) covers approximately 4000 km from West to East and 2600 km from South to North (Meusel 1978).

Nikolaeva and Vorob'eva (1978) have recognized in the USSR three zones differentiated in the size of the embryo relative to the endosperm (K): western, $K = 0.4-0.5$, central-eastern, $K = 0.6$, and southern, $K = 0.7-0.9$. Seeds with relatively longer embryo require a shorter period of warm stratification (Marinov 1977) or even they do not require the warm phase at all (Nikolaeva and Judin 1974, Nikolaeva and Vorob'eva 1978).

The morphological and physiological dormancy of ash seeds results in the need for a warm-followed-by-cold stratification of fully mature seeds (USDA Forest Service 1948, Villiers and Wareing 1964, Wcislińska 1977, Nikolaeva et al. 1985). The optimal thermal conditions for the stratification of European ash seeds in Poland have been developed by Suszka (1980). The proposed thermal regime is very similar to that recommended by Nikolaeva and Vorob'eva (1978) for ash seeds from the western parts of the USSR and it consists of 16 weeks at a temperature of 15–20°C followed by 16 weeks at 0–3°C.

For various reasons, the earlier planned Spring sowing of stratified seeds may be delayed, which could be associated with an undesirable germination of a large number of seeds. The germination of seeds during stratification can be withheld by freezing them at –3°C together with the stratification medium (Suszka 1987) even for a period of 48 weeks. This method is very successful because after defrosting of the medium seeds start to germinate immediately or they continue doing so if they had already started. Thus studies were undertaken aimed at testing the possibility of withholding the germination process in seeds during stratification in some other ways then by freezing them together with the stratification medium.

MATERIALS AND METHODS

For the study use was made of fully mature ash (*Fraxinus excelsior* L.) seeds collected on October 7th 1985 from a single tree in Poznań. After collection the samaras were partially dried to 10.3% of moisture content and stored in sealed

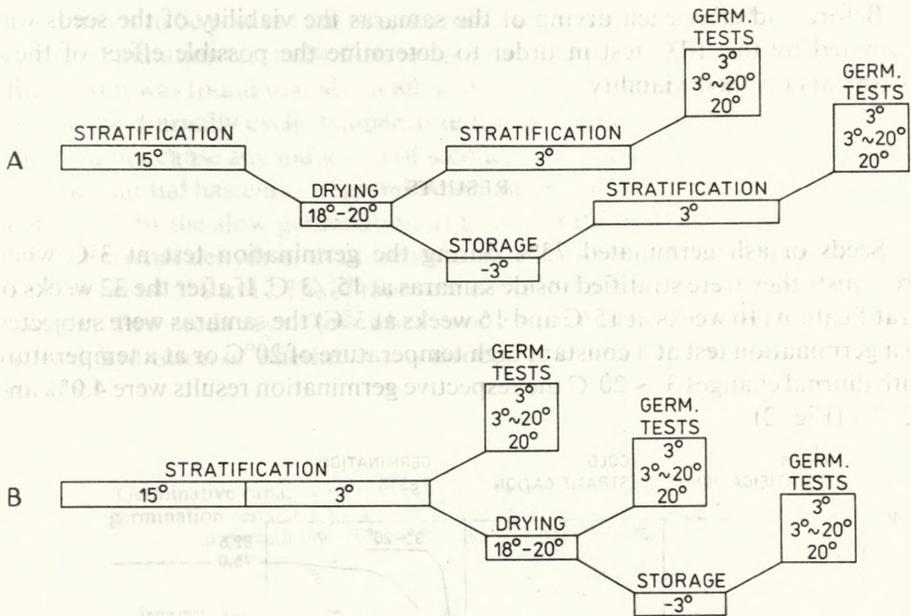


Fig. 1. Design of the experiment. The samaras were partially dried to 12.3% of moisture content after the warm phase of stratification (A) or to 12% after warm-followed-by-cold stratification (B)

bottles at -3°C until February 5th 1986. The viability of seeds after storage was 91.0%. The viability was tested by the method of staining embryos with a solution of tetrazolium chloride (TTC) according to the ISTA (1976) rules.

Seeds left in samaras after storage have been stratified in moist sand and peat (1:1) mixture at $15^{\circ}/3^{\circ}\text{C}$ for 16 weeks at each temperature (i.e. 16 + 16 weeks). After the 32 weeks of stratification the seeds were subjected to a germination test at 3°C , $3^{\circ}\sim 20^{\circ}\text{C}$ in the daily cycle of 16 + 8 h (Suzka 1978 and 1980) or at 20°C (Fig. 1A), in 4 replicates with 50 seeds each.

After the warm stratification at 15°C part of the seeds were partially dried at room temperature (about 20°C) in an artificial air current, to a moisture content of 12.3%. Immediately after dehydrating the seeds, they were stratified at 3°C for 16 weeks or stored in bottles at -3°C for 12 weeks and only then were they stratified at 3°C for 16 weeks. After termination of stratification the seeds were subjected to a germination test as above (Fig. 1A).

The partial dehydration of the seeds and their storage in the partially dried state has been conducted also after the warm-followed-by-cold stratification at $15^{\circ}/3^{\circ}\text{C}$ (Fig. 1B). In this case the samaras were dried to 12% of moisture content and two storage temperatures were used, 3° and -3°C for three storage periods of 4, 8 or 12 weeks in each of these temperatures. Both after storage and immediately after partial drying the seeds were subjected to the germination tests (Fig. 1B).

Before and after each drying of the samaras the viability of the seeds was estimated by the TTC test in order to determine the possible effect of these treatments on their viability.

RESULTS

Seeds of ash germinated 75% during the germination test at 3°C when previously they were stratified inside samaras at 15°/3°C. If after the 32 weeks of stratification (16 weeks at 15°C and 16 weeks at 3°C) the samaras were subjected to a germination test at a constant high temperature of 20°C or at a temperature with diurnal changes 3° ~ 20°C the respective germination results were 4.0% and 82.5% (Fig. 2).

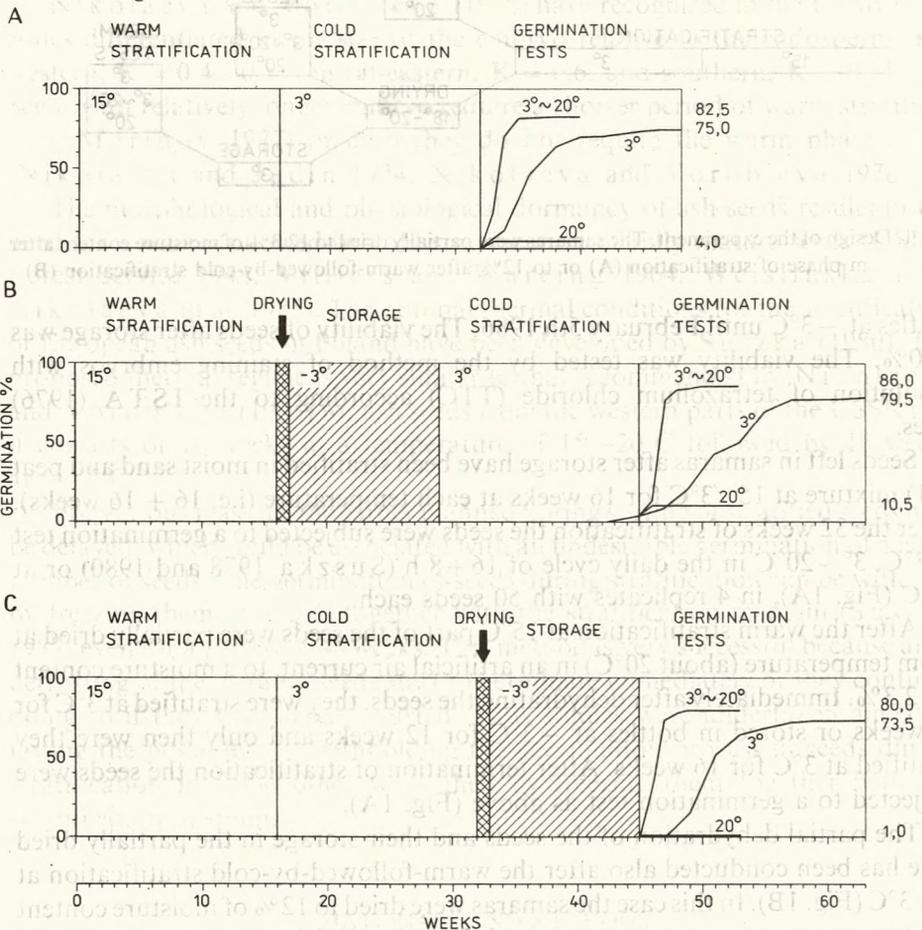


Fig. 2. Course of germination of European ash (*Fraxinus excelsior* L.) seeds during germination tests at 3°C, 3° ~ 20°C and 20°C after warm-followed-by-cold 15°/3°C stratification (A), interrupted by partial drying and storage of samaras at -3°C for 12 weeks after the warm phase (B) of stratification or after the full warm-followed-by-cold stratification (C)

It needs to be pointed out that the majority of ash seeds which were subjected to the germination test at 20°C have been induced to secondary dormancy. In the final test it was found that about 80% of nongerminating seeds were viable. The use of the diurnally cyclic temperature for the germination test (3° ~ 20°C) not only did not cause any induction of secondary dormancy but it also contributed to a substantial hastening of germination and reduction of the germination time compared to the slow germination at a temperature of 3°C (Fig. 2).

The partial dehydration of samaras after 16 weeks of warm stratification at 15°C from 64.5% to 12.3% of moisture content did not affect the germination of the stratified seeds regardless whether the germination was conducted shortly after dehydration or whether it was done after storage of the seeds for 12 weeks at -3°C.

Table 1

Germinative capacity of European ash (*Fraxinus excelsior* L.) seeds during germination tests at 3°C, 3° ~ 20°C and 20°C, either stored or not stored in a partially dried state following stratification at 15°/3°C

Temperature of germination test °C	Seeds not stored after partial drying %	Seeds stored after partial drying at					
		3°C			-3°C		
		Storage duration, weeks					
		4	8	12	4	8	12
3°	84.5	85.0	75.5	81.0	84.5	81.0	73.5
3° ~ 20°	81.5	76.5	82.0	82.5	82.5	82.5	80.0
20°	1.5	3.5	3.5	2.0	2.5	3.0	1.5

Both in the case of seeds partially dried after the warm phase of stratification (16 weeks) or partially dried after the full warm-followed-by-cold stratification (16+16 weeks), no quantitative changes were observed in the germinative capacity of the seeds. No changes were there to be observed also when the germination test was tried after 4, 8 or 12 weeks of storage at 3°C or -3°C in the partially dried state (Table 1).

DISCUSSION

The withholding of the germination process in seeds of ash by lowering their moisture content to 12% proved a successful method when the dehydration took place either after the warm phase of stratification or after the full warm-followed-by-cold stratification. No differences have been observed between the germinative capacity of seeds that were partially dried during stratification and seeds that were not dried.

The partial drying of ash seeds/samaras proved equally successful in withholding germination of ash seeds as their freezing together with the stratification medium at -3°C (Suszka 1987). It appears however that the storage of such

seeds in a partially dried state is a much more favourable and less cumbersome method than the freezing of medium with the seeds, in view of the smaller mass and volume of the seeds compared to the whole stratification medium with the seeds. Also of importance is the fact that dried, after-ripened seeds could be stored at 3°C with the same results as at -3°C. From the investigations of Vlasov et al. (1973) it is known that samaras of European ash, dried to about 7% of moisture content after collection, could be stored sealed at 10–11°C without decrease of germinability after 2.5 years of storage. This fact eliminates the necessity to use a storage temperature below 0°C for dried seeds both before and after stratification. Besides it is much easier to remove dry seeds from storage and sow them in the nursery than to defrost the whole medium and then to sow wet seeds together with the medium into the ground.

The lowering of the moisture content of the seeds during stratification, in order to arrest germination is a known procedure used for various species of forest tree seed. It is possible to partially dry seeds of coniferous trees such as *Pseudotsuga menziesii* (Vanesse 1967, Hedderwick 1968, Danielson and Tanaka 1978, De Matos Malavasi et al. 1985), *Pinus ponderosa* (Danielson and Tanaka 1978), *Pinus taeda* (Barnett 1972) and firs (Edwards 1980). Of deciduous trees it is possible for the seeds of beech (Suszka 1975, Muller and Bonnet-Masimbert 1985). After partial drying the seeds can be stored from several weeks to several months without loss of the benefits of stratification (after-ripening).

According to Pustovoitova and Oknina (1966) and De Matos Malavasi et al. (1986) the partial drying of stratified seeds before sowing can not only inhibit the metabolic processes, active during stratification, but it can even enhance them after the addition of water later on. As a consequence of this, these authors have reported an improved vigour of the seedlings. However not all seeds react so well to partial drying of seed as do the seeds of European ash. I have found that in the case of cherry plum (*Prunus cerasifera* var. *divaricata* Bailey) in the latter phase of dormancy breaking the seeds of this species are more sensitive to germination withholding by partial drying, than by lowering temperature below 0°C (Tylkowski 1987). One needs to expect however that there are species of woody plants the seeds of which sustain partial drying during stratification well. This topic will be continued in the further investigations of the author.

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Przechowywanie stratyfikowanych nasion jesionu wyniosłego (*Fraxinus excelsior* L.)

Streszczenie

Badano wpływ częściowego odwodnienia i przechowywania stratyfikowanych nasion jesionu wyniosłego na ich późniejszą zdolność kiełkowania. Stwierdzono, że podsuszenie skrzydłaków do 12% wilgotności po stratyfikacji ciepło-chłodnej 15°/3°C (16+16 tygodni) i przechowywanie ich przez 4, 8, lub 12 tygodni w 3°C lub -3°C w szczelnie zamkniętych butelkach nie wpłynęło na obniżenie zdolności kiełkowania nasion. Również częściowe odwodnienie skrzydłaków do 12,3% wilgotności po cieplej fazie stratyfikacji ciepło-chłodnej i ich przechowywanie w stanie poduszonym w -3°C przez 12 tygodni, nie wpłynęło na zmianę poziomu kiełkowania nasion.

Хранение стратифицированных семян ясеня обыкновенного (*Fraxinus excelsior* L.)*

Резюме

Исследовали влияние частичного обезвоживания и хранения стратифицированных семян ясеня обыкновенного на их дальнейшую способность прорастания.

Подтверждено, что подсушивание крылаток до 12% влажности после тепло-холодной стратификации 15/3°C (16+16 недель) и хранение их в течение 4, 8 или 12 недель в 3°C или -3°C в герметически закрытых бутылках не повлияло на снижение способности прорастания семян. Также частичное обезвоживание крылаток до 12,3% влажности после теплой фазы тепло-холодной стратификации и их хранение в подсушенном состоянии в -3°C в течение 12 недель не повлияло на изменение уровня прорастания семян.

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