Effect of different distances between water level and seed bed on Jacobsen apparatus on the germination of *Pinus silvestris* L. seed

Einfluss von verschiedenen Abständen zwischen Wasserhöhe und Keimbett auf Jacobsenapparat auf die Keimung von Pinus silvestris L. Samen

Inverkan av olika avstånd mellan vattennivå och groningsbädd i Jacobsens apparat på groningen av Pinus silvestris L. frö

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ABSTRACT

In this investigation, the effect of three distances: 22, 19 and 13 cm between the water level and the seed bed on Jacobsen apparatus (referred to in the following text as "water distance"), was studied on the germination of 24 samples of fresh and old Scots pine seed. The results showed that in most of the samples there was a tendency for the germination percentage to be the highest at the shortest water distance used (13 cm), although these differences were significant only in three cases. Also the germination rate was the fastest, at least in the first few days of the test, at 13 cm water distance in several samples. For the practical seed testing work, this study has indicated that it is important to keep the distance between the water level and the seed bed on Jacobsen apparatus constant, in order to obtain comparable germination results.

Introduction

The Jacobsen apparatus, devised by Miss Ingeborg Jacobsen of the Seed Testing Station in Copenhagen, is one of the most commonly used germinators for seed testing work. During its long existence, the apparatus has undergone certain modifications. Thus, for example, the height of the water container can be different in the different models. Consequently, it is not always possible to keep the same distance between the water level and the seed bed (referred to in the following text as "water distance") in the various models. This is important, since the moisture supply to the germination substratum is adjusted by regulating the water distance. A decrease in the water distance on germination is an important practical problem, specially in comparative studies or referee tests. In view of the fact that this question has been less studied, an investigation was undertaken to elucidate it. The present paper summarizes the results of this investigation.

Material

Twenty-four samples of fresh and old seed of Scots pine (*Pinus silvestris* L.) from different parts of Sweden were used for the experiments. The details of the samples are given in Table 1.

| Sample No. | Locality | Latitude | Altitude in metres | Year of collection |
|---------------|--------------------------|-----------------|-----------------------|--------------------|
| 1 | Gävleborgs län | | 200 | 1967 |
| 2 | Västmanlands län | 60°03′ | 55 | 1967 |
| 3 | Västmanlands län | 59°30′ | 100 | 1967 |
| 4 | Kristianstads län | 55°52′ | 36 | 1967 |
| 5 | Södermanland | $58^{\circ}45'$ | 60 | 1967 |
| 6 | Kristianstads län | 56°19′ | 90 | 1967 |
| 7 | Värmlands län | 60°15′ | 270 | 1967 |
| 8 | Kopparbergs l ä n | 60°00′ | 165 | 1967 |
| 9 | Kristianstads län | 55°59′ | 110 | 1966 |
| 10 | Södermanland | 59°03′ | 55 | 1964 |
| 11 | Kalmar län | 57°45′ | 110 | 1963 |
| 12 | Norrbottens län | 66°11′ | 370 | 1962 |
| 13 | Hallands län | 56°48′ | 17 | 1961 |
| 14 | Örebro län | 59°00′ | 50 - 240 | 1961 |
| 15 | Kalmar län | 56°30′ | 0100 | 1959 |
| 16 | Kalmar län | $57^{\circ}40'$ | 100 | 1959 |
| 17 | Kalmar län | 57°36′ | 120 | 1958 |
| 18 | Norrbottens län | 67°00′ | 80 | 1956 |
| 19 | Gävleborgs län | 60°50′ | 0100 | 1956 |
| 20 | Skaraborgs län | 59°00′ | 220 | 1953 |
| 21 | Kalmar län | $56^{\circ}25'$ | 85 | 1952 |
| 22 | Gotlands län | $57^{\circ}27'$ | 40 | 1952 |
| 23 | Gotlands län | 57°55′ | 40 | 1951 |
| 24 | Norrbottens län | 65°05′ | 335 | 194546 |

Table 1: Details of the material used.

Methods

Three Jacobsen apparatuses of stainless steel of the same model were used for the experiments. They were placed in a climate chamber of the Botanical Institute of the University of Stockholm and the containers were filled with tap water to such levels that the distances between the water level and the seed bed were 22, 19 and 13 cm. These water distances were kept constant throughout the investigation. The total height from the bottom of the container to the seed bed was 25 cm in each apparatus. The temperature $(20^{\circ}C)$ and humidity (60 %) of the climate chamber were controlled automatically by special devices. The light was given from day-light tubes and its intensity (= 1 000 lux), quality and duration (8 hours daily) were kept the same for all the germinators.

The germination tests were undertaken on 4×100 seeds of each sample. Before putting the material for germination, the number of empty or otherwise defective seeds in each lot was determined by x-ray radiography (kV = 14, mA = 10, focus-film distance = 50 cm, exposure time = 3 seconds. For further details, see Kamra 1967). The germinated seeds werecounted and removed from the tests every day up to the tenth day, andevery other day thereafter up to the 21st day. A seed was considered asgerminated when the length of the root was at least equal to that of the seeditself. This criterion has been found to be reliable in comparative germinationstudies (cf. Kamra 1967, Kamra and Simak 1968, Simak and Kamra 1968).The germination percentages of all the samples were calculated uniformly onthe basis of the number of filled seeds only in each lot.

Results

The average germination percentages of the samples at the three water distances: 22, 19 and 13 cm are given in Table 2. Samples 1---8 are of fresh seed and 9-24 of old seed.

| Sample | Germination percentages at water distance of | | | |
|--------|--|-------|-------|--|
| No. | 22 cm | 19 cm | 13 cm | |
| 1 | 88 | 89 | 91 | |
| 2 | 73 | 67 | 76 | |
| 3 | 99 | 100 | 100 | |
| 4 | 91 | 88 | 92 | |
| 5 | 98 | 98 | 99 | |
| 6 | 98 | 96 | 98 | |
| 7 | 94 | 96 | 97 | |
| 8 | 64 | 68 | 72 | |
| 9 | 92 | 95 | 96 | |
| 10 | 42 | 42 | 50. | |
| 11 | 65 | 67 | 73 | |
| 12 | 53 | 56 | 59 | |
| 13 | 94 | 95 | 95 | |
| 14 | 45 | 45 | 51 | |
| 15 | 80 | 80 | 81 | |
| 16 | 86 | 86 | 88 | |
| 17 | 83 | 83 | 81 | |
| 18 | 87 | 88 | 89 | |
| 19 | 86 | 85 | 86 | |
| 20 | 42 | 44 | 47 | |
| 21 | 48 | 49 | 58 | |
| 22 | 32 | 32 | 28 | |
| 23 | 13 | 14 | 22 | |
| 24 | 50 | 53 | 59 | |

Table 2: Germination percentages of the samples at the three water distances.

The results of germination will be explained separately for fresh and old seed.

I. Fresh seed

(a) Germination percentage:

As may be observed from Table 2, the germination values of the fresh samples (Nos. 1—8) at the three water distances are similar. Of course, from the table it appears, that there is a tendency that the germination percentage

is the highest at the shortest water distance (13 cm). However, the analysis of variance showed that these differences in germination percentages of each sample at the three water distances are not significant.

In the case of samples 2 and 8, the germination percentages, in general, are rather low. However, there are certain reasons for these values, and those will be explained in the Discussion.

(b) Rate of germination:

The rate of germination of the samples is shown in Figs. 1—8. It may be seen that in the first few days the rate of germination of the samples in the series with 13 cm water distance seems to be slightly faster than that of the other two series (19 and 22 cm). However, later on the three series have similar rates of germination.

II. Old seed

(a) Germination percentage:

The germination percentages of the old seed samples (Nos. 9—24) are given in Table 2. Like in the fresh seed samples, also here there is a tendency that the germination percentage is the highest at the shortest water distance (13 cm). In 14 of the 16 samples, the germination percentages at 13 cm water distance are equal to or higher than those in the other two series. However, the analysis of variance showed that only in three cases (sample Nos. 9, 21 and 23), these differences are significant (P = 0.05).

(b) Rate of germination:

The rate of germination of the samples is shown in Figs. 9—24. As may be observed, the germination rate in some of the samples seems to be faster in the series with 13 cm water distance than in the other two series (cf. Figs. 9, 10, 11, 12, 14, 20, 21, 23 and 24). In the other samples, this difference is less marked (cf. Figs. 13, 15, 16 and 18) or the reverse is true (cf. Figs. 17, 19 and 22).



Figs. 1-8: Rate of germination of fresh seed samples. (S. = Sample. G. = Germination).





Figs. 9-24: Rate of germination of old seed samples. (S. = Sample. G. = Germination).







Discussion

From the results described above (Table 2 and Figs. 1—24), it may be observed that there are no significant differences in the germination percentages of the fresh seed samples at the three water distances used. In the old seed samples, the differences are slightly larger, but significant only in three cases.

The present investigation was carried out at the constant temperature of 20° C instead of at the alternating temperature of $20-30^{\circ}$ C (ISTA Rules 1966). This was done because of the fact that an earlier investigation (Kamra and Simak 1968), in which the germination of Scots pine seed at different temperatures was compared, showed that the germination percentages of the samples at 20° C were equal to or in some cases higher than those at $20-30^{\circ}$ C.

It will be observed from Table 2 that samples 2 and 8 did not show a very high germination percentage. This was due to poor maturity of the seed of these samples. As is well-known, the seed of Scots pine from northern Sweden has often some incompletely developed embryos (cf. Simak and Gustafsson 1954). X-ray radiographs revealed that samples 2 and 8, which were from high latitude (about 60°N) contained many seeds of embryo classes II and III mostly with well-developed endosperm (class A, according to the classification of Simak and Gustafsson 1954). Even embryos of class IV were relatively poorly developed. Müller-Olsen and Simak (1954), and Simak (1957) found that Scots pine seed of class IIA germinates 50 %, that of class IIIA 88 % and that of class IVA 99 % on Jacobsen apparatus. These facts explain the poor potential germination of samples 2 and 8.

Table 2 and Figs. 1—24 indicate that seed of Scots pine in germination behaviour reacts positively to a certain increase in the moisture content of the substratum, which was obtained by reducing the water distance from 22 to 13 cm. Baldwin (1942) has studied the relationship between the distance of the water level from the germination substratum and the moisture content of the latter. According to his Fig. 25 (p. 194), the moisture content of the germination substratum at the water distance of about 6 to 2 cm increases faster than at 6—14 cm. However, on the basis of Baldwin's Figure 25, the expected increase in the moisture content of the seed bed in the range of water distances used in the present experiment (22—13 cm), would be too little in order to affect the germination results appreciably.

Schmidt (1930) mentions that pine seed which has suffered internal damage

during storage is very sensitive to excess of moisture. According to him, this sensitivity is further increased by strong damage to seed coat and by ageing of the seed. This effect of ageing of seed is supported by the results of the present experiment, as here some of the old seed samples show larger variation in germination at the three water distances used than the fresh samples.

Not only on Jacobsen apparatus but also on other germination substrata, like for example, humus and sand, high moisture content of the medium can affect the germination of seed. Mork (1938), who studied the germination of Scots pine and Norway spruce seed at various temperatures and degrees of moisture in humus covered with sand, states: "At a moisture of 20 volume percentage, the germination percentage is low and the germination proceeds slowly. With a water content of 70 volume percentage, the germination percentage is low, too; the radicle is, as a rule, deformed, and the radicular extremities in the embryos, which grow at such a high degree of moisture, become mocous." According to him, the moisture optimum for the germination of Scots pine and Norway spruce seed lies between 35 and 50 volume percentages.

The absorption of water by seed of different embryo classes of Scots pine has been studied by Gustafsson and Simak (1956). The mechanism of water uptake by seed during germination process is discussed by Niethammer and Tietz (1961) and by Mayer and Poljakoff-Mayber (1963).

For the practical seed testing work, the present investigation has indicated, that it is important to keep the distance between the water level and the seed bed on Jacobsen apparatus constant, as it cannot be excluded, that differences in this distance can affect the results differently, especially in referee tests or other comparative studies. This factor of water distance can be expected to gain further importance, as discussed above, with shorter water distances than those used in the present experiments.

Summary

1. The present paper deals with the effect of 22, 19 and 13 cm distances between water level and seed bed on Jacobsen apparatus (referred to in the text as "water distance") on the germination of Scots pine seed. Twenty-four samples of fresh and old seed of *Pinus silvestris* L. were used for the investigation. (Table 1).

2. It was observed that in most of the samples, there was a tendency for the germination percentages to be the highest at the shortest water distance

used (13 cm). However, the analysis of variance showed that these differences in the germination percentages of each sample at the three water distances were not significant, except in three cases (sample Nos. 9, 21 and 23, cf. Table 2).

3. The germination rate in some of the samples seemed to be faster in the series with 13 cm water distance than in the other two series. In the other samples, this difference was less marked or the reverse was true (cf. Figs. 1-24).

4. For the practical seed testing work, the present investigation has indicated, that it is important to keep the distance between the water level and the seed bed on Jacobsen apparatus constant, in order to obtain comparable germination results (e.g. in referee tests, etc.).

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Zusammenfassung

Einfluss von verschiedenen Abständen zwischen Wasserhöhe und Keimbett auf dem Jacobsenapparat auf die Keimung von Föhrensamen (Pinus silvestris L.).

1. Die vorliegende Arbeit beschäftigt sich mit dem Einfluss von 22, 19 und 13 cm grossen Abständen zwischen Wasserhöhe und Keimbett auf dem Jacobsenapparat (im Text als »Wasserabstand« bezeichnet) auf die Keimung von Föhrensamen. Vierundzwanzig Proben von frischem und altem Saatgut wurden für die Untersuchung verwendet (Tabelle 1).

2. Es wurde beobachtet, dass es bei den meisten Proben eine Tendenz gab, die höchsten Keimprozente bei dem niedrigsten Wasserabstand (13 cm) zu erreichen. Doch zeigte die Variationsanalyse, dass diese Unterschiede in den Keimprozenten von jeder Probe bei diesen drei Wasserabständen nicht signifikant waren, bis auf drei Fälle (Proben Nr. 9, 21 und 23; s. Tabelle 2).

3. Die Keimungsgeschwindigkeit von einigen Proben bei 13 cm Wasserabstand schien schneller zu sein als bei 19 und 22 cm. Bei anderen Proben war dieser Unterschied weniger bemerkbar, oder die Verhältnisse waren umgekehrt (vgl. Figuren 1—24).

4. Für die praktische Samenprüfung hat diese Untersuchung gezeigt, dass es beim Jacobsenapparat wichtig ist, den Abstand zwischen Wasserhöhe und Keimbett konstant zu halten, sodass vergleichbare Keimergebnisse erzielt werden können (z.B. bei »Referee«-Testen usw.).

Sammanfattning

Inverkan av olika avstånd mellan vattennivå och groningsbädd i Jacobsens apparat på groningen av Pinus silvestris L. frö.

1. Denna uppsats behandlar inverkan av 22, 19 och 13 cm avstånd mellan vattennivån och groningsbädden i Jacobsens apparat (i texten kallat »vattenavstånd») på groningen av tallfrö. Tjugofyra prov av färskt och gammalt frö av *Pinus silvestris* L. användes för undersökningen (Tabell 1).

2. Det observerades, att det fanns en tendens hos groningsprocenten i flera prov att vara högst vid det lägsta vattenavståndet (13 cm). Emellertid visade variationsanalys av resultaten, att dessa skillnader mellan groningsprocenten för varje prov vid de tre vattenavstånden icke var signifikanta, utom i tre fall (prov nr. 9, 21 och 23, jämf. Tabell 2).

3. Groningshastigheten i vissa prov verkade vara större i serien med 13 cm vattenavstånd än i de andra två serierna. I de andra proven var denna skillnad mindre markant eller förhållandena var omvända.

4. För den praktiska frökontrollen har denna undersökning visat, att det är viktigt att hålla avståndet mellan vattennivån och groningsbädden i Jacobsens apparat konstant, för att jämförbara groningsresultat skall kunna erhållas (t. ex. i »referee»-testen, osv.).