OSMOCONDITIONING OF LETTUCE SEEDS AND INDUCTION OF SECONDARY DORMANCY

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Abstract

Osmoconditioning of seeds has been developed to enhance and synchronize germination. It was shown in the present experiments that in lettuce seeds osmotic pretreatment also influenced the development of secondary dormancy. A pretreatment of lettuce seeds cv. Musette at 15 ^{o}C in polyethylene glycol (PEG) at an osmotic potential of -15 bar retarded the breaking of dormancy, prevented germination and, consequently, opened the oportunity for secondary dormancy to develop. In seeds of cv. Grand Rapids PEG postponed at 24 ^{o}C the development of secondary light-irresponsiveness. It is postulated that the decreased moisture content at the end of imbibition caused in both situations a retardation of the changes in dormancy.

1. Introduction

Osmoconditioning or priming of seeds has been developed to advance and synchronize the germination process (Heydecker and Coolbear, 1977). In combination with stimulatory compounds or factors, like growth regulators or light, the pretreatment also favours the breaking of dormancy (Negm and Smith, 1978; Kretschmer, 1982). In lettuce seeds osmoconditioning at lower temperatures causes an increase of the upper temperature limit of germination (Guedes and Cantliffe, 1980). The usefulness of osmotic solutions during seed treatments is due to the inhibition of cell-elongation growth which is characteristic for the growth of the embryo during germination. However, a prolonged inhibition of germination is also the main prerequisite for the development of secondary dormancy (Karssen, 1982). Therefore, it has been investigated in the present contribu-

tion whether osmoconditioning also influenced the development of secondary dormancy.

2. Materials and Methods

Commercial seed lots of lettuce (*Lactuca sativa* L.) cv. Musette and cv. Grand Rapids were used.

Triplicates of 50 seeds were sown in 5 cm Petri dishes on one layer of filter paper (Schleicher & Schüll No. 595) moistened with 1.5 ml of distilled water or polyethylene glycol 8000 (PEG) solution. Water potentials of PEG solutions were calculated according to Michel (1983). After pretreatment the seeds were rinsed with 100 ml of distilled water in a Büchner funnel, transfered to a filter paper moistened with distilled water in a Petri dish and further incubated. Seeds of cv. Grand Rapids were irradiated with a saturating dose of red light after the transfer to water. Germination was counted 2 days after red light (Grand Rapids) or transfer from 15 °C to 28 °C or 30 °C (Musette).

To obtain red light (620-700 nm), irradiation from 6 red fluorescent tubes (Philips, TL 20W/15) was filtered by 3 mm plexiglas (red 501, Röhm & Haas, Darmstadt, G.F.R.), the light intensity at seed level being 250 $\mu W.cm^{-2}$. All manipulations were conducted in green light obtained by filtering irradiation from one green fluorescent tube (Philips TL 40W/17) through 2 layers of yellow no. 46 and 2 layers blue no. 62 Cinemoid filters (Strand Electric, London, U.K.).

The moisture content of the seeds was determined by weighing ($\pm$ 0.1 mg) about 200 mg seeds in little vials before and after oven drying at 130 °C during 1.5 h.

3. Results

The germination of lettuce seeds cv. Musette was inhibited at temperatures exceeding 20 °C (Weges and Karssen, 1985). A pretreatment during 16 h in water at 15 °C increased the upper temperature limit to 30 °C. Stimulation of the germination at 30 °C required a longer pretreatment in water than a similar rise at 28 °C (Fig. 1).

Pretreatment in water is risky. If it exceeded at 15 °C a period of 24 h the seeds started to germinate and were vulnerable to drying back. Such a hazardous situation could be prevented by a pretreatment

in osmotic solutions. In PEG -5 bar germination during the pretreatment at 15 °C was delayed to 40 h, in -10 and -15 bar it did not occur at all (Fig. 1).

The comparison between pretreatment in water and PEG also showed a negative effect of PEG solutions with low water potentials. If seeds were pretreated in -15 bar PEG the stimulation of germination at 28 °C was clearly delayed and at 30 °C the 100% germination level was not reached anymore. It appeared at both temperatures that prolonged pretreatment at 15 °C in -10 and -15 bar PEG turned the stimulatory effect into its opposite, secondary dormancy developed.

Another effect of osmotic pretreatment was shown in experiments with lettuce cv. Grand Rapids. The light requirement of these seeds is well known and so is the observation that a delay of the light stimulus results in light irresponsiveness or skotodormancy (see Karssen 1980/81 for references). If these seeds were incubated in PEG solutions during the dark period, the induction of light irresponsiveness was strongly delayed (Fig. 2). The effect also depended on high PEG concentrations (Table 1). These concentrations reduced the moisture content of the seeds at the end of the imbibition period (Table 2).

4. Discussion

The present study has shown that the incubation of lettuce seeds in PEG not only inhibited germination, but also influenced the development of secondary dormancy. Both actions might be related, in the sense that inhibition of germination opened the opportunity for secondary dormancy to develop. In general, induction of secondary dormancy prevails at higher temperatures and breaking of dormancy at lower temperatures. At intermediate temperatures, like 15 °C, breaking of dormancy often still dominates. If germination is prevented, however, breaking of dormancy is followed by dormancy induction, which evidently proceeds at a slower rate (Totterdell and Roberts, 1979). Induction of dormancy means a narrowing of the range of suitable temperatures for germination (Karssen, 1982). Thus, secondary dormancy which developed during pretreatment at 15 °C, revealed itself more clearly at 30 °C than at 28 °C (Fig. 1).

It has to be considered that PEG not only inhibited growth but also influenced processes which preceded growth. Such an explanation is valid, particularly, for the inhibition by PEG of the induction of secon-

dary light-irresponsiveness in seeds of cv. Grand Rapids (Fig. 2). In these seeds at 24 °C light is a prerequisite for the start of growth. Since PEG was only applied during the dark period preceding red light, an effect of PEG on the inhibition of growth can be excluded. A similar effect of PEG on the processes preceding growth might also be involved in the experiment depicted in Fig. 1. It is seen there that PEG stimulated the induction of secondary dormancy at much higher concentrations than were required for the inhibition of germination.

A common explanation for the action of PEG in both experiments seems unlikely at first view, since PEG stimulated the induction of secondary dormancy in the first experiment and prevented it in the second. It has to be realized, however, that PEG inhibited also at 15 °C in cv. Musette a change in dormancy, namely the breaking of dormancy which prevailed at that temperature. Whether dormancy induction was also retarded could not be tested because secondary dormancy did not develop in the absence of PEG.

It is postulated that high concentrations of PEG in both experiments inhibited a change in the level of dormancy. This effect might be related to the reduced moisture content of the seeds which was observed at the end of the imbibition period in PEG (Table 2). PEG may influence dormancy levels through the water saturation of membranes.

The present results indicate that the use of PEG solution with a very negative water potential bears certain risks. It is recommended to apply PEG solutions at concentrations which just prevent that germination occurs during the required length of the pretreatment.

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Table 1 - The influence of a pretreatment in PEG on the germination of lettuce seeds cv. Grand Rapids. The seeds were pretreated during 4 days in PEG or distilled water, in darkness, at 24 °C. After pretreatment the seeds were rinsed and transferred to distilled water and irradiated with red light (5 min) and set to germinate at 24 °C.

PEG, bar during pretreatment	Germination, % (± S.D.)
0	17 ± 6
-7.4	43 ± 19
-9.8	67 ± 7
-12.3	87 ± 4

Table 2 - The moisture content of lettuce seeds cv. Musette after an imbibition period of 16 or 41 h in PEG or distilled water.

Imbibition medium, bar	Moisture content, 16 h	41 h
0	45.7 ± 0.2	- *
-5	39.3 ± 0.1	41.2 ± 0.3
-10	36.1 ± 0.3	39.2 ± 0.3
-15	35.2 ± 0.6	37.4 ± 1.8

* Seeds had germinated.

Figure 1 - The influence of the length of pretreatment and the incubation medium at 15 °C on germination in water at 28 °C (A) and 30 °C (B) of lettuce seeds cv. Musette. During pretreatment seeds werd incubated in water (o) or PEG at water potentials of -5 bar (Δ), -10 bar ( ) or -15 bar (∇); the seeds were kept in darkness throughout the experiment except for green safe light during the transfer. Vertical bars indicate S.D.

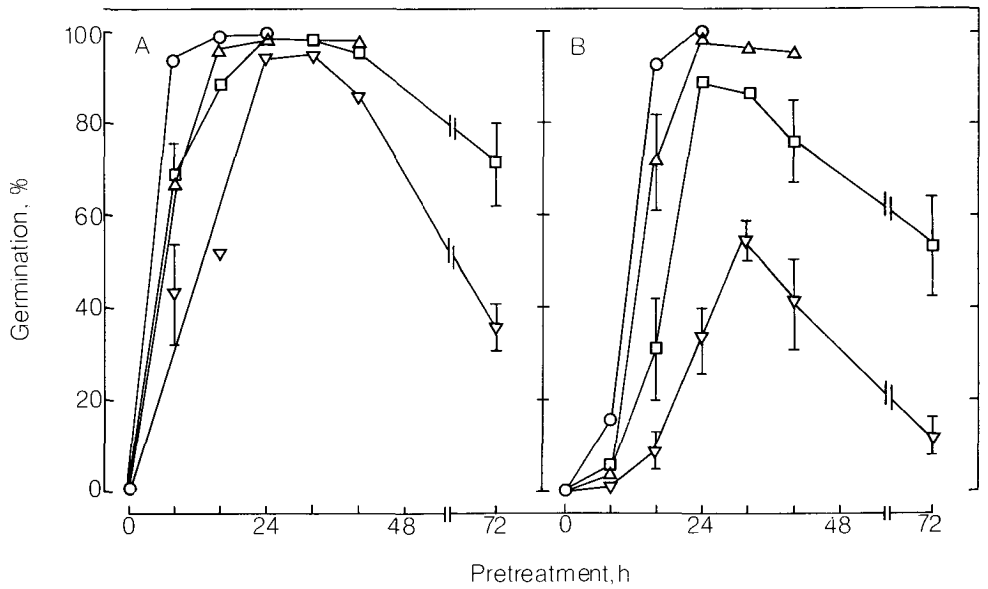

Figure 2 - The influence of the length of a pretreatment in darkness in water (o) or -12.3 bar PEG (Δ) preceding 5 min red light on the germination of lettuce seeds cv. Grand Rapids in water. The seeds were incubated before and after light at 24 °C. Vertical bars indicate S.D.