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Short communication

Effect of scarification, GA and chilling on the germination of goldenrain-tree (*Koelreuteria paniculata* Laxm.) seeds

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Abstract

In contrast to scarified seeds, unscarified seed did not germinate in any of the treatments, indicating that *Koelreuteria paniculata* Laxm. seeds have hard, impermeable seed coat dormancy. Exogenous application of 100, 200 and 300 ppm GA increased germination of scarified seeds from 0 (control) to 17, 18 and 15%, respectively. Pre-chilling in distilled water (DW) for 60 days increased germination to 44%. Compared with DW-chilled seeds, the germination of seeds chilled in gibberellic acid (GA-chilled) was significantly increased after 15 days of chilling and maximum germination of seeds chilled in 100, 200 and 300 ppm GA was 60, 51 and 54%, respectively, achieved after 30 days.

Longer duration of chilling in GA appeared harmful. Germination rate was positively correlated with germination percentage. These results show that *K. paniculata* seeds exhibit both exogenous and endogenous dormancy. A combination of GA and chilling (GA-chilling) helped to alleviate seed dormancy in a relatively short period of time.

Keywords: Chilling; Dormancy; GA; Germination; Goldenrain-tree; *Koelreuteria paniculata* Laxm.

1. Introduction

Over the past 20 years, dormancy has been widely studied but the regulatory principles behind changes in several types of dormancy remain unclear. Abscisic

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acid (ABA) and gibberellins (GAs) are the hormones proposed to control primary dormancy (the form of dormancy that is acquired during seed development), ABA by inhibiting and GAs by inducing germination (Hilhorst and Karssen, 1992; Iglesias and Babiano, 1997). Moist chilling is often practiced to enhance the germination of dormant seeds (Bello et al., 1998). It is believed that cold treatment alters the inhibitor–promoter balance. Frankland and Wareing (1962) studied *Fagus sylvatica* and *Corylus avellana* and found that GAs were apparently absent from unchilled dormant seeds of *Corylus* but became detectable after 6 weeks of chilling, at that time seeds were able to germinate.

Goldenrain-tree (*Koelreuteria paniculata* Laxm.) is a woody plant, native to China. It is mainly used for landscape purposes because of its beautiful yellow flowers and green leaves. Its size and shape make it very suitable to small lawns and gardens. It is mostly propagated from seeds. Like many other temperate trees, it has pronounced seed dormancy. Unfortunately, there is limited information concerning the potential seed dormancy problems of goldenrain-tree. The seeds have dual dormancy, i.e. exogenous as well as endogenous (Dirr, 1990). The presence of hard, water-impermeable seed coat and embryo dormancy has made natural regeneration of this tree almost impossible. For this reason, it is designated as an endangered plant species in Korea. The objectives of this study were to determine the effect of exogenously applied GA and DW or GA moist chilling on seed germination and to devise an effective method for breaking seed dormancy of *K. paniculata* Laxm.

2. Materials and methods

Koelreuteria paniculata Laxm. seeds were collected from trees along the roadside in Taegu, South Korea. Seeds were collected in mid-October 1998, when the seeds had desiccated to about 12% moisture on a dry weight basis. Seeds were separated from the pods on arrival at the laboratory and dry stored in a sealed plastic box at 5°C. A fraction of the seeds were manually scarified by piercing the seeds with a needle at the cotyledon end, whilst the remaining seeds were unscarified.

The seeds were surface sterilized by soaking in 5% sodium hypochlorite (NaOCl) solution for 10 min and subsequently rinsed thoroughly with sterilized water prior to germination or chilling. In the case of scarified seeds, sterilization was carried out before piercing the seeds. For the moist chilling pre-treatment, both the scarified and unscarified seeds were sandwiched between two layers of filter paper moistened with distilled water (DW) or 100, 200 and 300 ppm gibberellic acid (GA). They were then placed in a sealed plastic box in a refrigerator at 4°C for 15, 30, 60 and 90 days.

Seeds that had not received the GA or DW chilling pre-treatment were germinated in 9 cm plastic Petri dishes with two layers of filter paper, moistened with 5 ml of sterilized DW or 100, 200 and 300 ppm GA solution. Whenever it was needed to keep filter papers moist, additional water or GA was added to the Petri dishes. Each treatment was replicated five times and 20 seeds were used in each replicate. Pre-chilled seeds were subsequently incubated for germination in the dark at 22°C. Germination was recorded every day for 30 days and seeds were considered germinated when the emerging radicle was approximately 2 mm long. Germinated seeds were discarded from the Petri dishes daily.

Final germination was calculated when no further germination took place for several days. Germination rate was calculated as $1/t_{50}$, the reciprocal of the time in days taken to complete 50% of final germination. The arcsin transformed germination proportion in degrees and germination rate data were subjected to analysis of variance using the SAS statistical software package (SAS Institute, 1988), and the LSD for all pairs comparison at $p<0.05$ was calculated using Tukey's *t*-test (Li, 1964).

3. Results and discussion

None of the unscarified seeds germinated whether untreated or pre-treated with GA or DW chilling. Similarly, scarified but otherwise untreated seeds were unable to germinate (Table 1). These results suggest that *K. paniculata* has deep exogenous and endogenous dormancy.

It is suggested that the onset of embryo dormancy is associated with accumulation of growth inhibitors and breaking of dormancy with a shift in the balance of growth regulators towards growth promoters that overcome the effect of inhibitors (Khan, 1971). There are various methods used to break seed dormancy, e.g., hormonal, temperature and/or light treatments. Endogenous GAs are widely studied in relation to the breaking of dormancy in seeds of various

Table 1
Germination and germination rate of manually scarified *Koelreuteria paniculata* Laxm. seeds in the absence and presence of GA[a]

GA in germination medium (ppm)	Germination (arcsin)	Germination rate ($1/t_{50}$)
0	0	–
100	27 (17)	0.17
200	25 (18)	0.16
300	23 (15)	0.16
LSD ($p<0.05$)	5	0.035

[a] Germination percentage is shown in parenthesis.

plant species. They have been exogenously applied as a substitute for stratification and have increased the germination of many plant species, e.g., *Leucospermum* (Brits et al., 1995), *Fagus sylvatica* (Nicolas et al., 1996) and *Helianthus* (Seiler, 1998). In the present study, it was found that a significant number of *K. paniculata* seeds germinated that had been treated with exogenously applied GA. No significant difference in germination was observed between seeds treated with 100, 200 and 300 ppm GA (Table 1).

Moist chilling is a standard procedure used to enhance the germination of dormant seeds. It has been used for various dormant seeds and has been reported successfully to alleviate endogenous dormancy. It is believed that cold treatment can only work for those seeds that contain both inhibitors and promoters as evidenced by the fact that the inhibitor–promoter balance is altered by exposing seed to moist chilling. For example, the ABA level of apple and ash increases and GA concentration increases during cold stratification of seeds (Frankland and Wareing, 1962). Our results support the previous findings (Bello et al., 1998) and it was found that the germination of DW-chilled seeds (Table 2) was significantly increased to 44 and 45% after 60 and 90 days of chilling, respectively. However, the germination of gibberellic acid moist chilled (GA-chilled) seeds (Table 2) was significantly increased after 15 days of chilling and 100, 200 and 300 ppm GA-chilled seeds attained maximum germination of 60, 51 and 54%, respectively, after 30 days of chilling. These results show that although exogenous GA and DW-chilling significantly increased germination, these treatments were not as effective as GA-chilling.

The germination rate of seeds treated with GA during germination (Table 1) and DW-chilled was low compared with that of GA-chilled seeds (Table 2). In general, it was observed that fast germination was associated with high germination percentage, i.e., there was a significant positive correlation (r=0.77, p<0.01) between germination and germination rate. According to Khan

Table 2
Germination and germination rate of manually scarified *Koelreuteria paniculata* Laxm. seeds pre-chilled in distilled water (DW) or different concentrations of GA[a]

Duration of pre-chill (days)	GA in pre-chill medium (ppm)							
	Germination (arcsin)				Germination rate ($1/t_{50}$)			
	DW	100	200	300	DW	100	200	300
15	0	38 (29)	40 (33)	33 (30)	0.00	0.30	0.31	0.00
30	14 (8)	51 (60)	46 (51)	47 (54)	0.20	0.40	0.45	0.68
60	41 (44)	39 (40)	44 (49)	39 (40)	0.47	0.56	0.50	0.40
90	42 (45)	36 (35)	33 (30)	29 (24)	0.38	0.48	0.67	0.53
LSD (p<0.05)	10				0.22			

[a] Germination percentage is shown in parenthesis.

(1977), stratification affects metabolic processes including changes in hormones, i.e., disappearance of ABA and activation of GA and initiation of germination. The present study suggests that GA and chilling affect physiological and metabolic activities of seeds resulting in early germination.

In conclusion, these results suggest that GA and moist chilling enhanced germination probably due to the inhibitor–promoter balance. However, the action of GA or moist chilling alone may not be sufficient to bring inhibitor–promoter balance and therefore, failed to increase germination to its maximum level. The combination of GA and chilling was perhaps more effective in bringing a hormonal shift that not only enhanced germination but also speeded it up. Therefore, GA-chilling may be recommended for breaking *K. paniculata* seed dormancy in a relatively short time. These results also demonstrated that *K. paniculata* require far less chilling time than that recommended by Dirr (1990) and the 60–90 days of chilling usually considered satisfactory for other woody species (Bewley and Black, 1982).

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