

From Forest Nursery Notes, Summer 2008

**21. Methods for *Rosa* germination.** Anderson, N. and Byrne, D. H. *Acta Horticulturae* 751:503-507. 2007.

## Methods for *Rosa* Germination

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**Keywords:** *Rosa*, leaching, stratification media, seed

### Abstract

Poor germination in *Rosa* has hindered breeding programs for years. Several methods have been proposed to increase germination of rose seed. Unfortunately, no consensus exists on the best method, or if any one method is best for all rose genotypes. In the first experiment, open-pollinated rose seeds from a *R. wichuraiana* Crep. × 'Old Blush' hybrid were leached with constant filtration and aeration at room temperature for 0, 3, 7, or 14 days. After leaching, seeds were placed in either moist milled sphagnum moss or agar and stratified ( $2.8^{\circ}\text{C} \pm 0.4^{\circ}\text{C}$ ) for 8 weeks. The combination of no leaching plus stratification in moist milled sphagnum moss gave the greatest germination. In the second experiment, fresh rose seeds from various hybrids were stratified ( $2.8^{\circ}\text{C} \pm 0.4^{\circ}\text{C}$  for 10-12 weeks) in moist filter paper, milled sphagnum moss, perlite, sand, or vermiculite. Seeds germinated best when stratified on sphagnum moss. The third experiment used 9 genotypes and compared stratification directly in the growing flats and stratification in small containers followed by transplanting small germinating seedlings combined with variations in the stratification media (sand, perlite, sphagnum moss and Sunshine Mix #4). Over all stratification media and genotypes, germination was not influenced by whether the seed was stratified directly in the seedling flat/bed or in a small container. However, the process of transplantation of the delicate germinating seed from the small container to the flat/bed resulted in greater mortality of the germinating seedlings. The stratification media affected the germination of the rose seed. Sunshine Mix #4 gave the best germination as compared to the other media types tested. As expected the germination of the genotypes varied greatly, ranging from 0.7 to 37.1%.

### INTRODUCTION

Poor seed germination is a problem for rose hybridizers. One important factor contributing to poor germination is the genetic composition of the seed. Consequently, many breeders keep careful notes on seed set and germination so they can select the parents that produce the greatest number of seedlings. In addition, seed handling procedures such as harvesting, stratification, scarification and leaching are important to maximize germination.

A common method used to increase seed germination in temperate woody perennials such as apple (*Malus* Mill.), cherry, peach, plum (*Prunus* L.), pear (*Pyrus* L.) and rose (*Rosa* L.) is stratification (Jackson and Blundell, 1963; Crossley, 1974; Gill and Pogge, 1974; Grisez, 1974; Janick et al., 1974). Most of these woody perennials require only a single, cold stratification treatment, however a few such as black jetbead (*Rhodotypos scandens* Thunb. Mak.) blackberry, and raspberry (*Rubus* L.) showed increased germination when a warm stratification treatment preceded a cool stratification treatment (Brinkman, 1974; Rudolf, 1974). Generally, roses require only a single cold stratification treatment (Semeniuk, 1969).

Woody perennials of the Rosaceae family may be stratified in a variety of media. Successful germination has been reported for *Prunus* species following stratification in perlite, vermiculite, sand, peat or a mixture of sand and peat (Grisez, 1974; Syrbu, 1977; Seeley and Damavandy, 1985). Stratification in moist perlite has been reported effective for seeds of apples, pears, apricots, cherries and quince (Seeley and Damavandy, 1985).

Rose seeds have been stratified in a variety of moist media such as agar (Basye, 1991 unpublished communication), paper towels, peat moss, perlite, sand, vermiculite, and milled sphagnum moss (Carter, 1968; Benton, 1988). Carter (1968) reported that rose seed germinated better following stratification in moist peat moss or sand as compared to stratification in moist vermiculite.

Germination of woody perennial seeds, such as blackberry and raspberry, is also reported to increase with the use of sulfuric acid as a scarification agent (Brinkman, 1974). Scarification with chemicals (Heit, 1967), physical abrasives (Benton, 1988), or enzymes (Yambe and Takeno, 1992) has also been utilized to increase rose seed germination. The use of chemicals and physical abrasives on rose seeds has given conflicting results (Julin-Tegelman, 1983; Tillberg, 1983; Benton, 1988; Cullum et al., 1990). Recent reports show that the use of macerating enzymes can stimulate germination of *R. multiflora* Thunberg seed (Yambe and Takeno, 1992).

Leaching has also been used to increase germination. In both *R. multiflora* and *R. rugosa* Thunberg, leaching increased seed germination (Svejda and Poapst, 1972; Yambe et al., 1992).

This experiment was conducted to determine the effects of leaching and subsequent stratification media selection on the germination of rose seeds.

## MATERIALS AND METHODS

During the first experiment, ripe hips of open-pollinated roses from WOB-28, a *R. wichurana* Crep. × 'Old Blush' hybrid, were collected and seeds were extracted. Within a 2-week period from harvest, cleaned seeds were divided into 3 replications per treatment with an average of 400 seeds per replication. The seeds for each replication were placed in cheesecloth bags. Bags were suspended in a 10-gallon tap-water-filled aquarium at  $23.5^{\circ}\text{C} \pm 0.5^{\circ}\text{C}$  with constant filtration and aeration. Seeds remained in the water for 0, 3, 7, or 14 days. On the first day the water was changed three times, and thereafter one time every other day. After seeds were leached, they were placed on either moist milled sphagnum moss (Mosser Lee Co., 1 part water: 2 parts moss, vol:vol) or Difco agar (0.7% w/v), then stratified ( $2.8^{\circ}\text{C} \pm 0.4^{\circ}\text{C}$ ) in closed plastic containers (6.75" x 6.75" x 2.5"). Germination initiated during the 8<sup>th</sup> week of stratification. Seeds were left in the stratification media, and considered germinated when the primary root was visible.

During the second experiment, rose seeds from several hybrids (Table 1) were cleaned and stored at room temperature over a two-week period. Subsequently, these seeds were divided into three replications per treatment. The number of seeds per replication varied from 110 to 4600 depending on the number of seed available (Table 1). Since leaching was shown to be detrimental to germination in the first experiment, this step was omitted. The seeds for each replication were placed in bags made of filter paper (#4 Brew Rite Coffee Filters, Cone Style, 5.2 cm base, 10.8 cm side). Each replication was placed into a single bag, except for *R. bracteata* and Cytology127 seed, which used three and two bags per replication respectively. The bags were then placed in closed plastic containers (6.75" x 6.75" x 2.5") and completely covered with either moist milled sphagnum moss (Mosser Lee Co., 1 part water: 2 parts moss, vol:vol), moist perlite (Hortiperl 1 part water:4 parts perlite, vol:vol), moist sand (Play Sand, 3 parts water:5 parts sand, vol:vol), moist vermiculite (Mandoval, type: fine, 1 part water:4 parts vermiculite, vol:vol), or the filter paper was simply moistened (control) and then stratified ( $2.8^{\circ}\text{C} \pm 0.4^{\circ}\text{C}$ ) for 10-12 weeks. After the stratification treatment, seeds were placed in plastic flats (10.75" x 21" x 2.5") with Sunshine Mix #4 at a depth of approximately 0.3 cm and kept moist by misting intermittently in a greenhouse environment. Seed were allowed to germinate in the plastic flats over a 5-month period, during which time the germinated seedlings were counted. The data was analyzed with an analysis of variance followed by a Duncan's Multiple Range Test. No fungicides were applied to seeds before, during or after germination.

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## RESULTS AND DISCUSSION

Although the use of agar for stratification allows easy observation of germination during stratification, the agar treatment resulted in less than 1/3 of the germination observed when rose seed was stratified in sphagnum moss (Fig. 1). In addition, agar, especially with seeds that were not leached, often supported fungal and bacterial growth, whereas sphagnum moss showed no signs of fungal or bacterial contamination. Although the interaction between the leaching treatments and the media treatments was significant, germination was always higher with the sphagnum moss as compared to an agar medium irrespective of the leaching treatment (Fig. 1). This experiment confirmed the usefulness of sphagnum moss as a stratification medium.

The results from the second experiment were a further indication that medium plays a crucial role in the germination of seed. Of the five media tested, stratification in filter paper promoted the least germination and sphagnum moss promoted the highest germination. Perlite was intermediate between sphagnum moss and the two other media (sand and vermiculite) in its ability to stimulate germination. Across all genotypes tested, sphagnum moss was better, or as good as the other stratification media tested (Table 1).

In contrast to the findings of Yambe et al. (1992) with *R. multiflora*, leaching of WOB-28 rose seed for 3 or more days decreased germination dramatically. This discrepancy may have resulted due to genotypic effects, method of leaching and/or temperature differences in the leaching water. Nevertheless, Svejda and Poapst (1972) report that leaching for 24 hours did not affect or slightly increased germination. In the first experiment, the shortest leaching period was three days and only used one genotype. Additional experiments using a wide range of genotypes with different leaching conditions and leaching times of less than three days should be conducted.

In conclusion, the combination of the two treatments of no leaching and stratification in sphagnum moss was observed to increase germination versus leaching plus stratification in agar (Fig. 1). Furthermore, in the absence of leaching, sphagnum moss was best and perlite intermediate in their propensity to promote germination when compared to vermiculite, sand or moist filter paper (Table 1).

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**Tables**

Table 1. Stratification media effects on rose seed germination in 2000<sup>z</sup>.

Cultivar	Seeds/ rep	Filter paper	Germination (percent)			
			Perlite	Sand	Sphagnum moss	Vermiculite
[90-69 x Basye's Blueberry] <sup>y</sup>	300	0.1 c	4.5 b	1.1 c	7.8 a	1.7 c
[Purple Parade x 90-69] <sup>y</sup>	590	0.1 b	27.3 a	11.8 ab	26.3 a	15.0 ab
79-333 <sup>y</sup>	110	0.0 b	17.0 ab	7.7 b	27.5 a	4.6 b
90-73 <sup>y</sup>	200	0.0 b	5.7 ab	0.5 b	12.8 a	1.7 b
Carefree Beauty	130	0.0 b	2.6 a	0.3 b	1.3 ab	1.8 ab
Cytology127 <sup>y</sup>	1060	0.2 b	3.6 a	4.0 a	4.2 a	3.0 a
<i>R. blanda</i>	510	4.9 b	10.7 ab	13.5 ab	18.4 a	16.7 a
<i>R. bracteata</i>	4600	1.8 c	36.8 ab	19.9 bc	41.5 a	18.1 bc
Overall	-	0.9 c	13.5 ab	7.4 b	17.5 a	7.8 b

<sup>z</sup>Mean separation in rows by Duncan's multiple range test. Means within a row followed by the same letter are not significantly different at the 5% level.

<sup>y</sup>The genotypes 90-69 and 90-73 are siblings from the cross between Basye's Blueberry and 86-7. Thus these tetraploid roses are complex hybrids that include parentage from commercial roses and various wild diploid (*R. wichuraiana* Crep. × *R. rugosa rubra* Hort. from 86-7) and tetraploid (*Rosa carolina* L. and *Rosa virginiana alba* Mill.) species (Ma et al., 2000). 79-333 is a complex tetraploid hybrid with *R. carolina* and several commercial hybrid teas and polyanthas in its parentage. Cytology 127 is a tetraploid form of *R. bracteata* Wendl.

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Fig. 1.

**Figures**

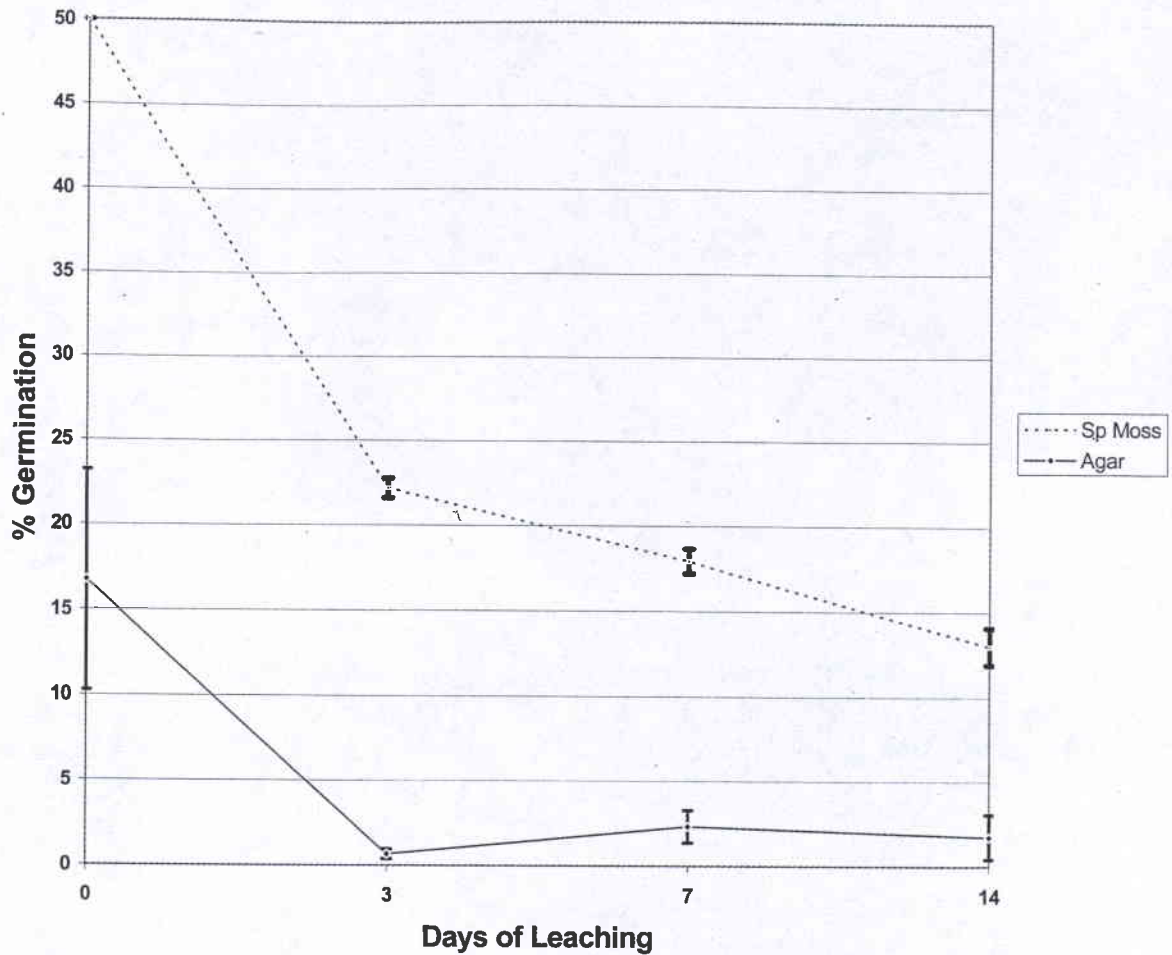


Fig. 1. Leaching and stratification media effects on WOB 28 rose seed germination. The bars above and below the data points indicate standard deviation (95% level).

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