Factors Regulating Germination of Trifoliate Maple Seeds

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Abstract. Excised embryos from Acer griseum (Franch.) Pax., A. mandshuricum Maxim., A. maximowiczianum Mig. (A. nikoense Maxim.) and A. triflorum Kom. germinated within 21 days when incubated in a lighted environment on wick cultures moistened with a solution of 10 mg/liter gibberellic acid (GA$_3$). Cotyledon and epicotyl growth of A. triflorum were greatest when treated with GA$_3$. Embryos of A. triflorum, when incubated with 6-benzylaminopurine (BA) or 6-furfurylamino purine (kinetin) developed the longest roots while those treated with indoleacetic acid (IAA) or naphthaleneacetic acid (NAA) remained tightly coiled and dormant. Excised embryos incubated in darkness or intact seeds placed in all treatments failed to germinate. In A. maximowiczianum 3 sites of dormancy delay germination: pericarp, seed coat, and embryo.

Acer griseum, A. mandshuricum, A. maximowiczianum (A. nikoense) and A. triflorum belong to the subsection Trifoliate (11). All species are characterized by short tree stature at maturity and autumn foliage of red, scarlet, or orange. A cinnamon-brown or yellow-brown flaking bark of A. griseum and A. triflorum respectively, further enhance the horticultural qualities of trifoliate maples, making them excellent ornamentals.

Samaras of the trifoliate maples, especially A. griseum, A. maximowiczianum and A. triflorum, have a ligneous pericarp which delays germination for several years (4,7). In addition, 2 to 5 years will elapse between good fruiting years with most fruits producing few seeds exhibiting double dormancy (7,16). They are not easily rooted from cuttings and must be grafted, but unfortunately need a rootstock of a similar species. Thus, trifoliate maples are rarely seen in urban areas.

Seeds were extracted from samaras by breaking the pericarp with pruning shears and a budding knife. Softening the seed coat in water at 25°C for 30 to 40 min facilitated embryo removal under a dissecting microscope. Seeds or embryos received treatments while being incubated on wick cultures which consisted of a 25 x 150mm glass tube. The seeds or embryos were placed into an upper fold of the filter paper with the embryonic radicle pointing down.

Control and growth regulator solutions of auxins, cytokinins or gibberellic acid were prepared by adding 5 drops of 1N KOH to the crystals and diluted to volume with deionized water. Ten ml of solution were added to each culture before the tubes were capped. Cultures were incubated at 25°C for 21 days. Each treatment consisted of 10 replications. Germination was defined as expansion of the embryonic radicle, cotyledons, and plumules.

Experiment I. Naked embryos excised in solution culture. Commun. Soil Sci. & Plant Analysis 9:781-783...

Table 3. Mineral element composition of Taxus media 'Anderson' terminal 5 cm shoots grown at 4 boron levels.

<table>
<thead>
<tr>
<th>Variable</th>
<th>N (%)</th>
<th>P (%)</th>
<th>K (%)</th>
<th>Ca (%)</th>
<th>Mg (%)</th>
<th>Na (ppm)</th>
<th>Mn (ppm)</th>
<th>Fe (ppm)</th>
<th>B (ppm)</th>
<th>Cu (ppm)</th>
<th>Zn (ppm)</th>
<th>Al (ppm)</th>
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<tbody>
<tr>
<td>B (ppm)</td>
<td></td>
<td></td>
<td></td>
<td></td>
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<td></td>
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<tr>
<td>0.5</td>
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<td>.23</td>
<td>2.1</td>
<td>.84</td>
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<td>30</td>
<td>130</td>
<td>39</td>
<td>48</td>
<td>6.4</td>
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<td>20</td>
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<td>5.0</td>
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<td>.77</td>
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<td>121</td>
<td>38</td>
<td>67</td>
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<td>58</td>
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<td>.23</td>
<td>2.1</td>
<td>.59</td>
<td>.18</td>
<td>34</td>
<td>92</td>
<td>31</td>
<td>171</td>
<td>6.5</td>
<td>50</td>
<td>9</td>
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<td>.24</td>
<td>2.1</td>
<td>.66</td>
<td>.18</td>
<td>39</td>
<td>100</td>
<td>33</td>
<td>357</td>
<td>6.4</td>
<td>52</td>
<td>15</td>
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<tr>
<td>LSD 5%</td>
<td>NS</td>
<td>NS</td>
<td>NS</td>
<td>.05</td>
<td>NS</td>
<td>NS</td>
<td>9</td>
<td>3</td>
<td>16</td>
<td>3</td>
<td>3</td>
<td>4</td>
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</table>

Literature Cited
from seeds of *A. maximowiczianum* were treated with solutions of IAA, NAA, BA, kinetin or GA$_3$, at 0, 10, or 100 mg/liter. Treatments were placed in 8 hr of darkness with irradiation from Cool White fluorescent lights (160 μE m$^{-2}$ sec$^{-1}$) for 16 hr on a 24 hr cycle. The purpose of this experiment was to determine if auxins, cytokinins or GA$_3$ at different concentrations could stimulate germination.

Germination of *A. maximowiczianum* naked embryos occurred when incubated in all solutions of kinetin or GA$_3$ and BA at the lowest dilutions (Table 1). Most significant increases in root length occurred on embryo placed on wick cultures wetted with solutions of BA (10 mg/liter) or kinetin (100 mg/liter). Cotyledon and epicotyl growth were greatest on embryos cultured in solutions of GA$_3$ (10 mg/liter). Excised embryos remained tightly coiled and dormant when incubated in water, dilutions of IAA or NAA, or solutions of GA$_3$ or BA at 100 mg/liter.

**Experiment 2.** Germination is promoted in many seeds by light. To establish the role of light in germination, seeds and naked embryos of *A. maximowiczianum* were treated with solutions of 0 or 10 mg/liter GA$_3$ and incubated in either darkness or in the lighted environment previously described.

Excised embryos of *A. maximowiczianum* were induced to germinate in the lighted environment when incubated in the GA$_3$ solution. Seeds in the same solution or seeds and naked embryos incubated in water remained dormant. No growth was detected on treated seeds and extracted embryos when placed in continuous darkness.

**Experiment 3.** Dormancy is caused in many seeds by water soluble inhibitors present within seed coats. Seed coats of *A. maximowiczianum* were therefore assayed for nonspecific water soluble inhibitors. Air dried seed coats (250 mg) were finely ground and extracted with 5 ml of deionized water for 24 hr at 25°C. The leachate was filtered and a 4 ml aliquot placed on filter paper in a 90 mm Petri plate. One hundred seeds of *Lactuca sativa* L. cv. Grand Rapids were then sown on the filter paper and percent germination determined after 48 hr at 25°C in the lighted environment.

Based on the bioassay, there were no apparent inhibitory substances in the seed coats of *A. maximowiczianum* preventing lettuce seed germination. Ninety percent germination resulted in both treatments.

**Experiment 4.** Seed coats can prevent inhibition and prolong seed dormancy. To study this, seed coats of *A. maximowiczianum* were lacerated on one side and the cut surface was placed in contact with filter paper moistened by solutions of 0 or 10 mg/liter GA$_3$. Germination results were collected after 21 days of incubation in the lighted environment.

Laceration of *A. maximowiczianum* seed coats failed to initiate embryo germination. Solutions of 0 or 10 mg/liter GA$_3$ were ineffective in promoting the unfolding of embryonic radicles or cotyledons of lacerated seeds.

**Experiment 5.** The influence of GA$_3$ on overcoming embryo dormancy of other triticate maples was investigated. Since limited seeds were available of *A. griseum*, *A. mandshuricum*, and *A. triflorum*, only treatments of 0 or 10 mg/liter GA$_3$ were given to naked embryos. Incubation was in the previously described lighted environment for 21 days.

Germination of *A. griseum* and *A. triflorum* embryos excised and treated with 10 mg/liter GA$_3$ germinated within 21 days. The non-treated control embryos did not germinate since they failed to unfold their cotyledons or radicles. Treatments of *A. mandshuricum* naked embryos with either 0 or 10 mg/liter GA$_3$ were given to naked embryos. Incubation was in the previously described lighted environment for 21 days. However, embryos treated with GA$_3$ germinated in 7 days while non-treated controls required 14 days.

Treating *A. maximowiczianum* embryos with GA$_3$ promoted cotyledon expansion, cytokinins promoted longest root growth and auxin treated embryos remained dormant. Thus, in some species of *Acer* it appears that termination of seed dormancy, cotyledon unfolding and expansion are promoted by gibberellins, root development is enhanced by cytokinins, and auxins are ineffective in promoting germination.

Gibberellins can stimulate germination in a large number of plant species (12), inducing germination of many light-requiring seeds and substituting for the effect of light. Results from the current experiments demonstrate that both light plus GA$_3$ were required for germination of *A. griseum*, *A. maximowiczianum* and *A. triflorum*, but not for *A. mandshuricum*. Thus, light cannot substitute for GA$_3$ during germination of the first three maples. However, the promotion of *A. mandshuricum* germination in the light without GA$_3$ demonstrates that basic physiological differences exist among these species.

Seed coat integuments can prevent germination by restricting water flow (5, 14), reducing oxygen uptake (1), preventing embryo enlargement (2, 6, 15), or by chemical inhibition (8, 9). The current experiments showed that seed coat laceration of *A. maximowiczianum* seeds and treatment with GA$_3$ were insufficient to promote germination and that application of seed coat leachate to lettuce seeds did not induce germination. This suggests oxygen, water deficiency, or mechanical restriction to embryo expansion by the seed coat are probably of minor importance in the regulation of embryo dormancy however, the seed coat appears to have an active role in preventing germination. Thus, it appears that 3 sites of dormancy delay germination of *A. maximowiczianum*: 1. the ligneous pericarp surrounding the seed probably enforces mechanical restriction to embryo growth; 2. the seed coat; and 3. a physiologically dormant embryo.

### Table 1. Lengths of radicles, cotyledons and epicotyls of *Acer maximowiczianum* embryos incubated in solutions of GA$_3$, kinetin, BA, IAA, or NAA after 21 days.

<table>
<thead>
<tr>
<th>Growth regulator</th>
<th>Concentration (mg/liter)</th>
<th>Radicle Length (mm)</th>
<th>Cotyledons Length (mm)</th>
<th>Epicotyl Length (mm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>0</td>
<td>5.4a</td>
<td>14.6ab</td>
<td>0 a</td>
</tr>
<tr>
<td>GA$_3$</td>
<td>10</td>
<td>13.4c</td>
<td>27.3e</td>
<td>4.8d</td>
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<tr>
<td>Kinetin</td>
<td>100</td>
<td>5.5a</td>
<td>16.9c</td>
<td>0 a</td>
</tr>
<tr>
<td>BA</td>
<td>100</td>
<td>9.9b</td>
<td>16.0bc</td>
<td>1.9b</td>
</tr>
<tr>
<td>IAA</td>
<td>100</td>
<td>23.6e</td>
<td>19.6d</td>
<td>3.7c</td>
</tr>
<tr>
<td>NAA</td>
<td>100</td>
<td>20.0d</td>
<td>17.2c</td>
<td>2.7b</td>
</tr>
</tbody>
</table>

$^a$Mean separation within columns by Student-Neuman-Keuls Test, 5% level.

### Literature Cited

Vegetative Growth Control of Hibiscus rosa-sinensis Hedges with Chlormequat

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Additional index words: maleic hydrazide, daminozide, ancyridol, growth retardant, landscape maintenance, growth inhibitor

Abstract. 2-Chloroethyl-trimethylammonium chloride (chlormequat) was the most effective of several growth retardants in retarding new growth of field-grown Hibiscus rosa-sinensis L. without serious side effects. A reduction by one-third to one-half of non-treated growth occurred with 1000 and 3000 ppm (active ingredient), respectively. Over a 3½ year period of repeated shearings, chlorotic leaves appeared on chlormequat-treated plants within a week of treatment, but the leaves gradually regreened over the 6 week observation period. A concentration series with chlormequat at 0, 300, 500, 1000, 1500, and 3000 ppm was conducted in 1977. Each rate was applied to five 2-plant experimental units 4 weeks after shearing when regrowth was 13 cm long. Measurements were taken at 50 and 100 days following treatment. Average shoot length of the control was 52.3 and 103.7 cm at 50 and 100 days, respectively. Growth subsequent to chlormequat treatment was decreased by 30 to 50% of the control at concentrations between 1000 and 3000 ppm (Fig. 1).

The effect of long-term application of chlormequat was evaluated over a 3½ year period from 1971 to 1974. The experiment compared 1500 ppm application at each shearing with a 3000 ppm rate applied with each shearing or at alternate shearings. The growth retarding effect of 3000 ppm chlormequat was carried over from previous applications when sheared growth was untreated.

Chemical growth retardation has been found effective for a number of hedge species (3, 5, 6, 7, 8, 9). Since chlormequat was identified as a potent retardant for potted hibiscus (1, 2, 4, 10), it seemed reasonable to determine if it would be as effective on much larger outdoor hedge plants.

Rooted cuttings of a clone of a single pink form of H. rosa-sinensis were planted at the Waimanalo Experiment Station on Oahu in 1969, and sheared regularly to develop a dense, compact hedge. Fertilizer (16N-6.9P-13.3K) was applied to five 2-plant experimental sections from the top of each treated section were taken at 50 and 100 days following treatment. Average shoot length of the control was 52.3 and 103.7 cm at 50 and 100 days, respectively. Growth subsequent to chlormequat treatment was decreased by 30 to 50% of the control at concentrations between 1000 and 3000 ppm (Fig. 1).

The effect of long-term application of chlormequat was evaluated over a 3½ year period from 1971 to 1974. The experiment compared 1500 ppm application at each shearing with a 3000 ppm rate applied with each shearing or at alternate shearings. The schedule of treatments was:

1. Sheared only. Shearing was carried out 2 or 3 times a year as growth exceeded 0.5 m. Other treatments were...