VOICE: Have you considered similar studies with *C. kousa*?

ELWIN ORTON: Yes, we have. However, there are no pink or red forms of *C. kousa*. We did interspecific crosses between *C. florida* and *C. kousa* at the suggestion of Hans Hess with the idea of producing a pink *C. kousa*. We have also incorporated *C. nuttallii*. The major thrust of our work now is interspecific hybridization. I have imported supposedly pink forms of *C. kousa* from Japan but they have all produced only white flowers. I have chased down many claims of pink flowered *C. kousa* dogwoods. Every year I get calls from people who have pink-flowered *C. kousa* and they are right. If you look at them during a 24-48 hour period when the flowers are senescing they will have a pink color. In some cases I have seen the color last for 7 days.

AL FORDHAM: If the flowers abort, you will also get pink bracts.

PROPAGATION OF TRIFOLIATE MAPLES BY SEED

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In the section *Trifoliata* of the genus *Acer* are 4 species: *A. griseum*, *A. mandshuricum*, *A. maximowiczianum* (*A. nikoense*) and *A. triflorum* (10). The 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 these maples, making them excellent ornamentals.

Schizocarps of *A. griseum*, *A. maximowiczianum* and *A. triflorum* have a ligneous pericarp which delays germination for several years (3,6). Two to 5 years can elapse between good fruiting, with most fruits producing few seeds exhibiting double dormancy (6,15). Trifoliate maples are not easily rooted by cuttings but can be grafted; however, they need a rootstock of a similar species. Thus, trifoliate maples are rarely seen in cultivation.

Dormant seeds of *Acer* have germinated following gibberellin or kinetin treatments (12,13). Radicle elongation of *Acer pseudoplatanus* was promoted by kinetin while gibberellin treatment promoted cotyledon unrolling and growth (9). This paper reports results from experiments conducted to deter-

mine the role of growth regulators, the seed coat, and light on germination of trifoliate maple seeds.

MATERIALS AND METHODS

Fruits of *Acer griseum* and *A. maximowiczianum* were collected during October, 1979, from trees in the U.S. National Arboretum, Washington, D.C. *Acer mandshuricum* and *A. triflorum* fruits were supplied by the Arnold Arboretum, Jamaica Plain, Massachusetts. Samaras were stored in paper bags at 10°C in the dark.

Seeds were extracted from schizocarps by breaking the hard pericap 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 folded 12.5 cm Whatman No. 2 filter paper inserted into a 25 x 150 mm 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 diluting 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 1. Naked embryos excised 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 $\mu Em^{-2}s^{-1}$) 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.

Experiment 2. Germination is promoted in some kinds of seed 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.

Experiment 3. Dormancy is caused in many kinds of seed 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° in the lighted environment.

Experiment 4. Seed coats can prevent imbibition 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.

Experiment 5. The influence of GA_3 on overcoming embryo dormancy of other trifoliate 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.

RESULTS

Experiment 1. 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 embryos 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.

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.

		Length (mm)		
Growth regulator	Concn (mg/liter)	Radicle	Cotyledons	Epicotyl
Control	0	5.4a[z]	14.6ab	0 a
GA_3	10	13.4c	27.3e	4.8d
	100	5.5a	16.9c	0 a
Kinetin	10	9.9b	16.0bc	1.9c
	100	23.6e	19.6d	3.7c
BA	10	20.0d	17.2c	2.7b
	100	5.4a	13.3a	0 a
IAA	10	5.2a	117.0c	0 a
	100	5.4a	14.6ab	0 a
NAA	10	5.5a	17.0c	0 a
	100	5.3a	13.3a	0 a

[z] Mean separation within colums by Student-Newman-Keuls Test, 5% level.

Experiment 2. 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. Based on the bioassay, there were no apparent inhibitory substances in the seed coats of *A. maximowiczianum* preventing lettuce seed germination. Ninety percent germination occurred in both treatments.

Experiment 4. 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. *Acer 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 resulted in germination. However, embryos treated with GA_3 germinated in 7 days while non-treated controls required 14 days.

DISCUSSION

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 seed germination in a large number of plant species (11), inducing germination of many light-requiring seeds and substituting for the effect of light. Results from the current experiments demonstrate that both light and GA_3 were required for germination of *A. griseum*, *A. maximowiczianum* and *A. triflorum* seeds, but not for *A. mandshuricum*. Thus, light cannot substitute for GA_3 during germination of the first three maples. However, the promotion of *A. mandschuricum* seed germination in the light without GA_3 demonstrates that basic physiological differences exist among these species.

Seed coats can prevent germination by restricting water flow (4,13), reducing oxygen uptake (1), preventing embryo enlargement (2,5,14), or by chemical inhibition (7,8). The current experiments showed that seed coat laceration of *A. max-*

imowiczianum seeds and treatment with GA_3 were insufficient to promote germination and that application of seed coat leachate to lettuce seeds did not suppress their 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 seed 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.

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RALPH SHUGERT: Have you tissue-cultured *A. griseum* and *A. maximowiczianum*.

DENNIS STIMART: We have tried them; however, both have a bacterial contaminant in the conducting vessels.