ETHREL FOR BREAKING DORMANCY IN SEEDS OF SOME WOODY PLANTS¹

DAVID F. HAMILTON²
Purdue University
Lafayette, Indiana 47907

Abstract. Seeds of some woody plants were treated with (2-chloroethyl) phosphonic acid (Ethrel) to determine if ethylene would replace the cold requirement for germination. In the first phase of the experiment, seeds were soaked without a cold treatment in Ethrel solutions and in the second phase seeds were soaked after a cold treatment.

Soaking seeds in Ethrel increased germination of most species, although the effective concentration and length of treatment varied among species. A cold treatment in addition to the soak in Ethrel did not further increase germination.

REVIEW OF LITERATURE

The physiological effects of ethylene on plants are dramatic and commercially important (2,9). One source of ethylene for plant and seed treatment is (2-chloroethyl) phosphonic acid (Ethrel; also occasionally cited as 2-chloroethanephosphonic acid, AmChem 66-240, CEPA, and Ethephon). In 1946 Kabachnik and Rossiikaya (6) reported the chemical synthesis of "2-chloroethanephosphonic" acid and in 1963 Maynard and Swan (8) described the formation of ethylene from this compound. When Ethrel, (2-chloroethyl) phosphonic acid, disintegrates, it releases ethylene and also chloride and phosphate ions (8, 12, 14). When the presence of hydroxyl ions is increased and the pH rises above 4, disintegration of the chemical takes place. The pH of the cytoplasm of plant cells is generally greater than 4, so the plant growth activity of Ethrel has been attributed primarily to its ability to release ethylene to plant tissues (11, 12). Ethrel formulations provide a convenient way to apply ethylene without the need of gas-confining chambers (3).

The effect of Ethrel treatment on woody plant seed germination is not known; however, Ketring and Morgan (7) found that germination of Virginia type peanut seeds from apical and basal stem locations soaked for 16 hr in Ethrel concentrations of 72 to 145 ppm was 100% compared to 13% with untreated apical and 60% with basal seeds. Soaking delinted cotton seed in 10 to 100 ppm Ethrel accelerated and increased germination percentage in greenhouse trials (1). Iyer, Chacko, and Subramiam (5) reported that pretreatment of dormant strawberry seeds with 1000, 2500 and 5000 ppm of Ethrel for 24 hr induced 30, 50, and 90% germination in 4 weeks whereas 20% of untreated controls germinated. In

¹Ethrel was supplied by AmChem Products, Inc., Ambler, Pennsylvania

²Department of Horticulture

laboratory studies Ethrel has also stimulated germination of dormant witchweed (*Striga lutea* Lour.) seed placed in 1 to 1000 ppm germination solutions. Germination was also stimulated by placing seed in a non-sterilized Eustris loamy sand into which Ethrel had been incorporated at rates of 10 and 1000 mg/kg of soil (4).

The objective of this study was to determine if treatment with Ethrel would replace the cold requirement of dormant embryos in seeds of woody plants.

MATERIALS AND METHODS

To study the influence of ethylene evolution from (2-chloroethyl) phosphonic acid on germination, seeds of five tree species were treated with Ethrel at various concentrations for different time periods. The following species were used: Taxodium distichum L., Elaeagnus angustifolia L., Sheperdia argentea Nutt., Cercis canadenis L., and Robinia pseudoacacia L., The seed of Taxodium, Elaegnus and Sheperdia are often difficult to germinate because they have dormant embryos, while Cercis often germinates poorly because the seed has a double dormancy (13). Robinia normally germinates readily and was used to see if Ethrel treatment decreased seed germination. Seed of Cercis was scarified in H₂SO₄ for 30 min before treatment with Ethrel, and the resinous seed coats of Taxodium were removed by hand.

With each species four replications of 5 seeds each were soaked in an Ethrel solution. Ethrel solutions were made with distilled water of pH 5.5. The following concentrations were used: 50, 300, 600, and 900 ppm of Ethrel, and a distilled water control. Different seeds were soaked at each concentration for 0.5, 6, 12, 18 and 24 hrs. After removal from the Ethrel solution, seeds were planted in flats of peat-perlite mix (1:1 v/v) in the greenhouse. Water was applied as needed to keep the peat-perlite mix moist.

In one phase of the study Ethrel treatments were given to seeds before any cold stratification. In the second phase seeds were stratified for half the maximum number of cold days recommended in the Woody Plant Seed Manual (13).

Seedlings were counted periodically for eight weeks, but only the mean percentage germinating is reported here.

Analyses of variance (Steel and Torrie, 10) were then performed on the individual experiments to determine significant variables. Significance of means was determined by Duncan's multiple range test.

RESULTS AND DISCUSSION

Seeds of the various species tested did not respond identically to Ethrel treatment. Robinia pseudoacacia, which normally germinates readily without a cold treatment, showed no response to Ethrel

(Table 1). However, seeds of Elaeagnus angustifolia not given a cold treatment but soaked in Ethrel germinated significantly better than seeds soaked in distilled water (Table 2). Concentrations of 300 and 600 ppm for 6 to 18 hrs gave maximum germination. Soaking seeds for 24 hrs at these same concentrations did not stimulate germination as much, nor did treatment with 900 ppm Ethrel. However, germination was still greater than that of the seeds soaked in distilled water. Germination was not further stimulated by giving seeds a cold treatment before soaking in Ethrel (Table 2).

Table 1. Effect of Ethrel on germination of seed of Robinia pseudocacia.*

Ethrel concentration (ppm)	Percentage of Seeds Germinating Length of Ethrel Soak (hrs)								
	0.5	6	12	18	24				
distilled water	60a	55a	65a	80a	70a				
50	60a	65a	65a	65a	65a				
300	75a	65a	70a	75a	70a				
600	75a	70a	70a	65a	80a				
900	65a	70a	55a	∂5a	70a				

^{*}Only seeds of *Robinia* not given a cold treatment were soaked in Ethrel.

Table 2. Effect of Ethrel on germination of seed of Elaeagnus angustifolia.

Ethrel	(ppn	n) N	lo col	d trea	atment		45	days		
				Leng	tn of E	Cthrel S	soak	(nrs)		
	0.5	6	12	18	24	0.5	6	12	18	24
distilled water	25a	35a	35a	20a	20a	40a	25a	40a	35a	50a
50	35a	40a	30a	55a	45a	25a	25a	25a	30a	45a
300	30a	100b	75b	85c	50b	30a	30a	80b	75b	35a
600	20a		80b		50b	25a	35a	70b	90b	45a
900	_	60c			25a	30a	25a	40a	45a	35a

^{*}Percentages not followed by the same letter are significantly different at the 5% level.

^{**}Percentages not followed by the same letter are significantly different at the 5% level.

Germination of seeds of *Taxodium distichum* was significantly increased following an 18 and 24 hr soak at 50 ppm. As the length of soak increased at 600 and 900 ppm, seed damage was apparent. Percentage germination was significantly less than the seeds soaked in distilled water. Germination was not increased by giving the seeds a cold treatment before soaking in Ethrel (Table 3).

Sixty percent germination of seeds of *Sheperdia argentea* not given a cold treatment was obtained with a 50 ppm Ethrel 24 hr soak (Table 4). Germination of seeds under other Ethrel treatments did not differ significantly from seeds soaked in distilled water. No stimulation of germination occurred in seeds given a cold treatment and then soaked in Ethrel (Table 4).

Table 3. Effect of Ethrel on germination of seed of Taxodium distichum.

Ethrel concentration (ppm)		Percentage of Seeds Germinating*									
		No	cold	treat	ment	30 days at 5° C					
	Length of Ethrel Soak (hrs)										
	0.5	6	12	18	24	0.5	6	12	18	24	
distilled water	30a	0a	40a	25a	40a	25a	50a	35a	0a	35a	
50	30a	45a	35a	60b	65b	35a	40a	40a	65b	65b	
300	20a	35b	40a	40a	35a	40a	40a	35a	40c	45a	
600	20a	10a	10b	25a	10c	30a	35a	40a	25c	15c	
900	15a	10a	0b	25a	15 c	5b	15a	40a	25c	10c	

^{*}Percentages not followed by the same letter are significantly different at the 5% level.

Table 4. Effect of Ethrel on germination of seeds of Sheperdia argentea.

Ethrel concentration (pp		Percentage of Seeds Germinating*										
		n) No	o cole	d trea	tment	45 days at 5° C						
	Length of Ethrel Soak (hrs)											
	0.5	6	12	18	24	0.5	6	12	18	24		
distilled water	20a	0a	20a	10a	0a	10a	20a	15a	0a	0a		
50	20a	25b	0b	0a	60b	5a	0b	10a	5a	5a		
300	20a	10ab	0b	5a	0a	10a	0b	0a	0a	15a		
600	0b	0a	0b	15a	5a	0a	10ab	15a	0a	0a		
900	0b	0a	0b	15a	0a	5a	0b	0a	5a	10a		

^{*}Percentages not followed by the same letter are significantly different at the 5% level.

Seeds of *Cercis canadenis* soaked in Ethrel but given no cold treatment germinated significantly better when soaked for 6 or 24 hrs at 300 and 600 ppm. The poor germination percentages obtained for 12 and 18 hr soaks at these same concentrations is probably from unrelated factors. Damage to the seeds is apparent at 900 ppm as germination significantly decreased. Giving seeds a cold treatment before soaking in Ethrel did not further increase germination at 300 and 600 ppm (Table 5).

Results from this experiment indicate that (2-chloroethyl) phosphonic acid may be useful to the plant propagator. Elimination of all or even reduction of the cold requirement of seeds would not only decrease the time required to start seedlings, but would also release space used for cold storage of seeds. Treating seeds in this manner would not require expensive equipment.

It is apparent that stimulation of seeds by ethylene does not occur at the same levels in all species. Additional study is needed to determine injury levels of seeds to ethylene. Although (2-chloroethyl) phosphonic acid is known to break down above a pH of 4 releasing ethylene (12), it would also be helpful to know the rate of release and the length of time ethylene is released.

Table 5. Effect of Ethrel on germination of seeds of Cercis canadenis.

Ethrel concentration (No e			_	ls Germinating* 30 days at 5° C							
		Length of Ethrel Soak (hrs)										
	0.5	6	12	18	24	0.5	6	12	18	24		
distilled water	0a	15a	5a	0a	0a	65a	65a	70a	90a	55a		
50	40a	55a	0a	35b		45b	55a	75ab	85a	60a		
300	50b	100c	0a	10a	85c	75a	90b	70a	25b	65a		
600	45b	95c	0a	5a	85c	70a	85b	90b	85a	90b		
900	50b	55b	5a	5a	45b	45a	50a	80ab	75a	55a		

^{*}Percentages not followed by the same letter are significantly different at the 5% level.

LITERATURE CITED

- 1. Amchem Products, Inc. 1969. Ethrel. Technical Service Data Sheet H-96. Ambler, Pa. (cited by deWilde, 1971).
- 2. Crocker, W. 1948. Growth of Plants. Reinhold, New York. 139-171.

- 3. deWilde, R. C. 1971. Practical applications of (2-chloroethyl) phosphonic acid in agricultural production. *Hort Sci.* 6(4): 12-18.
- 4. Ehley, G. H., and J. E. Dale. 1970. Ethylene, 2-chloroethylphosphonic acid, and witchweed germination. Weed Sci. 18:586-589.
- 5. Iyer, D. P., E. K. Chacko, and M. D. Subramaniam. 1970. Ethrel for breaking dormancy of strawberry seeds. *Curr. Sci.* 39: 271-272.
- 6 Kabachnik, M. I., and P. A. Rossiishaya. 1946. Organophosphorus trichloride. (In Russian). *Izvest. Akad. Nauk* (S.S.S.R., 7421 f, abstract in English. (cited by deWilde, 1971).
- 7. Ketring, D. L., and P. W. Morgan. 1970. Physiology of oil seed. I. Regulation of dormancy on Virginia-type peanut seeds. *Plant Physiol.* 45: 268-273.
- 8 Maynard, J. A., and J. M. Swan. 1963. Organo-phosphorous compounds. I. 2-chloroalkylphosphonic acid as phosphorylating agents. *Australian J. Chem.* 16: 596-608.
- 9 Pratt, H. K., amd J. D. Goeschl. 1969. Physiological roles of ethylene in plants. Ann. Rev. Plant Physiol. 20: 541-584.
- 10 Steel, R. G. D., and J. H. Torrie. 1960. Principles and Procedures of Statistics. McGraw-Hill Book Co., Inc., New York.
- 11. Warner, H. L., and A. C. Leopold. 1967. Plant growth regulation by stimulation of ethylene production. BioScience 7:722.
- 12. Warner, H. L. and A. C. Leopold. 1969. Ethylene evolution from 2-chloroethylphosphonic acid. *Plant Physiol.* 44: 156-158.
- 13. Woody Plant Seed Manual. 1948. U.S.D.A. Forest Service Misc. Publ. No. 654. Washington, D.C.
- 14. Yang, S. F. 1969. Ethylene evolution from 2-chloroethylphosphonic acid. *Plant Physiol.* 44: 1203-1204.

RALPH SHUGERT: Thank you, Phil, for presenting Dave Hamilton's paper to us.

Our next speaker is Dr. John Ahrens and he is going to talk on a subject which has come up in our meetings on several occasions. His talk is entitled "Rooting Cuttings from Plants Treated with Herbicides".