Increased Frost Resistance by Application of Plant Growth—Retardant Chemicals

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Frost damage to cabbage plants was markedly reduced by application of growth-retardant chemicals prior to exposure of the plants to low temperatures. Young plants were sprayed in the fall with the retardants B-Nine or Cycocel, and exposed during the winter to existing outdoor temperatures, the critical range being from 30° to −1° F. All of these plants survived and 90% quickly resumed growth when brought into a warm greenhouse the following spring. In contrast, 40 to 60% of the control plants were killed by winter frost. In the spring the surviving control plants grew very slowly and the total amount of vegetative growth was greatly reduced. The number of plants that flowered in a late-maturing variety increased when B-Nine was applied at a low dosage of 625 p.p.m. However, at 2500 and 5000 p.p.m., flowering was suppressed and all the plants formed heads instead of flower stalks.

GROWTH-retardant chemicals are receiving much attention for use in the regulation of plant growth and flowering (*5*). Acceptance has been particularly prevalent among horticulturists who use the retardants to produce short plants with intense green foliage. Flowering can be affected by these compounds and, with proper treatment and cultural management, such woody plants as azalea and rhododendron can be made to bloom at any time during the year. The flowering response is of particular interest because retardants have been used experimentally to decrease the time required for such plants as apple, pear, and holly to reach the fruit-bearing stage (*1, 3, 9, 17*).

Earlier reports from this laboratory showed that plants treated with retardants became more resistant than did untreated ones to aging (*11, 12*); certain unidentified soil-borne diseases (*11*); loss of water resulting in wilting (*10*); and death of plants due to high salt content of the soil (*10*). Others reported increased resistance to heat (*18*) and confirmed the effect on wilting and tolerance to salts (*6, 14*).

Resistance to heat, wilting, and high salt content in the soil is, of course, directly associated with the behavior of water molecules within plants. The resistance or susceptibility of plants to frost damage is governed also to a large extent by the behavior of water molecules within their various tissues (*13, 15*).

Since frost damage is one of the limiting factors in the culture of many economically important plants, the present study was initiated to determine whether some growth-retardant compounds can be used experimentally to make plants better able to withstand frost injury. Cabbage (*Brassica oleracea* var. *capitata* L.) was selected as the test plant because of its ability to develop resistance to low temperatures (*8*).

Methods

Young cabbage plants of the early-maturing variety, Early Jersey Wakefield, and others of the late-maturing variety, Late Flat Dutch, were grown in the greenhouse in composted soil in 3-inch clay pots. By October 3, 1963, the plants had developed three secondary leaves 1 to 3 inches long. Lots of 20 uniform plants of the early variety were sprayed with Cycocel, (2-chloroethyl)-trimethylammonium chloride, one lot receiving 2500 p.p.m., another 5000 p.p.m. Two other lots were sprayed with B-Nine, *N*-dimethylaminosuccinamic acid, in a comparable way at the same concentrations of this retardant. An additional lot of 20 plants was left untreated as controls.

One hundred uniform plants of the late variety were divided into groups of 20. Each group received one application of B-Nine at 0, 625, 1250, 2500, and 5000 p.p.m., respectively. All of the aqueous spray solutions used for both varieties contained 0.1% Tween 20 and were applied on October 9, 1963.

The plants were left in the greenhouse (70° to 95° F.) for 1 week after treatment and then moved out of doors. The pots were placed in sandy soil so that they were completely covered and the plant parts above the cotyledonary leaves exposed. Individual lots of five plants of each treatment and the control were arranged at random in four adjacent rows.

The plants remained out of doors to overwinter approximately 4 months (October 16, 1963, to March 5, 1964). Climatological data covering this period are shown in Table I (*20*). The plants were brought into a greenhouse maintained at 70° to 80° F. on March 5, 1964, and transferred into larger clay pots (5-inch) containing composted soil. These were arranged in randomized rows of five plants each per treatment on a greenhouse bench. Observational and other data concerning survival and

Table I. Temperatures (°F.) to Which Cabbage Was Exposed under Field Conditions

Beltsville, Md.
Data supplied by U. S. Weather Bureau (*20*)

Month	Av. Maximum Daily	Av. Minimum Daily	Av. Daily	Lowest Recorded
October	72.9	40.6	56.8	30
November	58.0	36.8	47.4	22
December	37.4	19.8	28.6	−1
January	46.7	23.9	35.3	−2
February	45.1	23.3	32.5	9
March	56.0	31.5	43.8	19

Table II. Effect of Growth Retardants Cycocel and B-Nine on Overwinter Surivival and Flowering of Two Varieties of Cabbage

Plants sprayed in greenhouse Oct. 9, 1963, and placed out of doors from Oct. 16, 1963, until March 5, 1964. Survival and flowering data obtained May 26, 1964, and final weights June 19, 1964

Compound	Spray, P.P.M.	% Survival[a]	% Flowering[b]	Av. Weight, G. Stalks	Heads
EARLY JERSEY WAKEFIELD VARIETY					
Control	0	60	60	123	
Cycocel	2500	100	100	284	
	5000	100	100	310	
B-Nine	2500	100	100	295	
	5000	100	100	305	
LATE FLAT DUTCH VARIETY					
Control	0	40	50	160	
B-Nine	625	100	100	380	
	1250	100	10		645
	2500	100	0		780
	5000	100	0		695

[a] 20 plants per treatment.
[b] Calculated on basis of plants that survived.

flowering were recorded periodically, from 2 weeks after removal to the greenhouse until June 19, 1964, when final weight measurements were made.

Results and Discussion

The rapid rate of growth after overwintering and removal to the greenhouse of the plants treated with retardants was compared with the slow rate of growth of the surviving control plants (Figure 1). The vegetative growth of many of the untreated plants arose from lateral buds because the terminal buds had been killed. This development from lateral buds resulted in a delay in over-all growth and, therefore, a reduction in average weight of the plants at the end of the experiment (Table II). Vegetative growth of the plants of both varieties treated with the retardants arose from vigorous terminal buds and ultimately resulted in plants that were larger and heavier than the untreated ones (Table II and Figure 2). Treatment with retardants resulted in markedly increased cold hardiness in both the early- and late-maturing varieties of cabbage. Since cold hardening (physiological changes that make plants resistant to frost) is generally associated with a decrease in rate of growth, it is of interest in the present study that the fast-growing type of cabbage when treated with retardants resisted low temperatures as well as did plants of the slow-growing type.

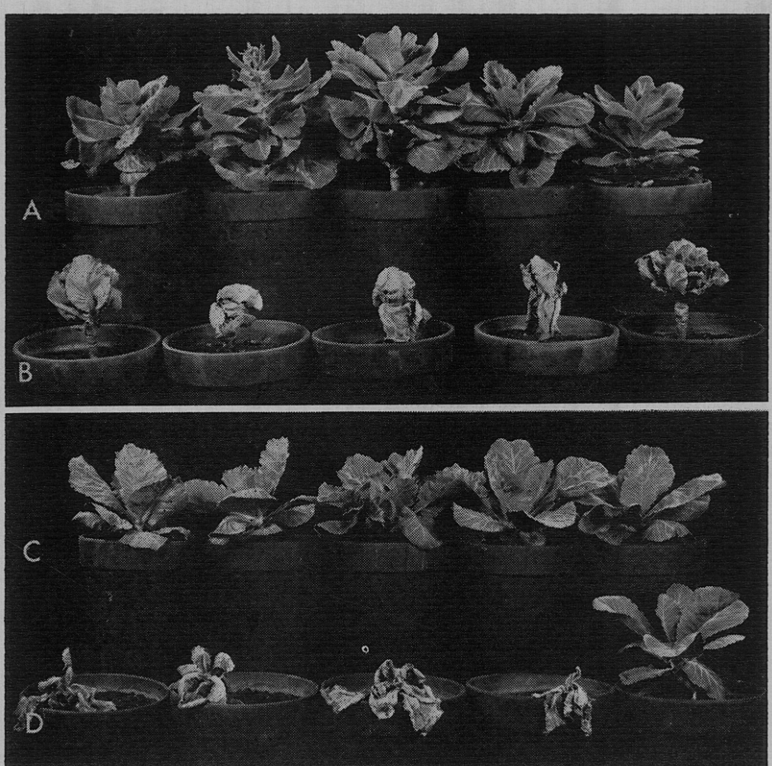

Figure 1. Effect of growth retardants on survival and rate of growth of cabbage plants after exposure to frost through winter and 2 weeks in warm greenhouse

A. Early Jersey Wakefield variety, Cycocel-treated
B. Untreated
C. Late Dutch variety, B-Nine–treated
D. Untreated

All of the Early Jersey Wakefield lots of cabbage, both the controls that survived and the ones treated with retardants, produced flower stalks. This suggests that the physiological processes associated with flowering may have been well advanced at the time of treatment. When the experiment was initiated, the plants of this variety were slightly larger than those of the late-maturing variety. Boswell showed that with cabbage large plant size at time of hardening favors bolting (*2*).

The enhancement of flowering brought about by application of retardant chemicals is significant from the standpoint of basic research and practical use. Control of flowering with a retardant was obtained with several kinds of plants other than cabbage (*5*). The fact that dosage level of the retardant chemical is critical in inducing the flowering response has not been previously reported. Application of B-Nine at the relatively low dosage of 625 p.p.m. for the entire lot of 20 plants of the late-maturing variety induced flower stalk development. These plants were not visibly different from the controls during the fall and winter months. In contrast, plants treated with 1250, 2500, or 5000 p.p.m. showed the shortened internodes and intensification of green foliar coloration generally associated with growth-retardant treatment. Two of the 60 plants included in the application of the three highest concentrations of B-Nine flowered and the others failed to flower and produced solid heads.

Research findings concerning physiological causes of frost damage throughout the world have been published by Levitt (*8*) and by Vasil'yev (*19*). There is general agreement that frost damage results from the puncturing of cell walls by relatively large, sharp-pointed ice crystals. Ice formation, however, may not in itself cause death of cells. Olien found that barley plants could tolerate a high degree of ice formation, provided the crystals themselves were small, even though numerous (*13*).

Extensive research by Rosa (*15*) in 1921 led him to conclude: "Any treatment materially checking the growth of plants increases cold-resistance. In cabbage and related plants, hardiness increases in proportion as growth is checked." Certainly, the response of plants to retardant treatment is in this

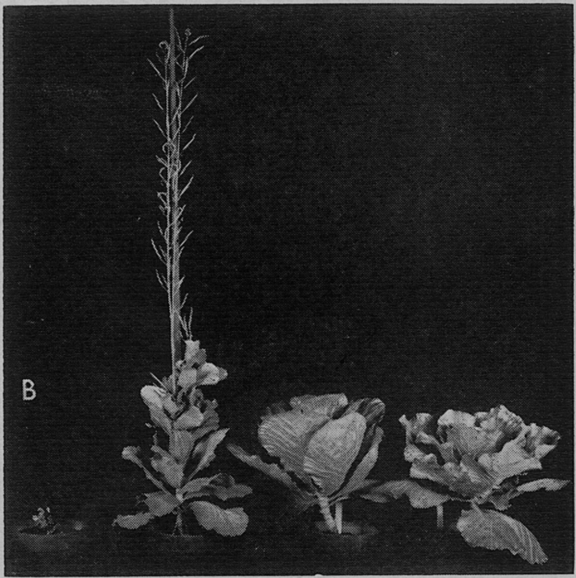

Figure 2. Effect of growth retardants on flowering of cabbage

A. Early-maturing variety
Left to right. Control, B-Nine 2500 p.p.m., Cycocel 2500 p.p.m., Cycocel 5000 p.p.m.

B. Late-maturing variety,
Left to right, B-Nine 0, 625, 1250, 2500 p.p.m.

category. Biochemical changes on the cellular level following treatment with retardants have not yet been widely studied. Studies with poinsettia, however, have shown an increase in chlorophyll content in the foliage following treatment with B-Nine. The iron, copper, and manganese content was also at a higher level (*4*). Rosa and others (*13, 15*) associated a high level of polysaccharides, particularly pentosans, with increased cold hardening. The pentosans seemed of particular importance because of their rapid and high degree of water uptake and swelling to form colloidal mixtures. Cellular contents of bean leaves of plants treated with retardants were much denser than those of controls. The cell walls were much thicker than the controls (*16*), a response that would provide a physical barrier to puncture by ice crystals.

There has been much speculation on the possibility of loading the plant system with a chemical that may serve as an "antifreeze." It has been demonstrated, for instance, that polyethylene glycol 20,000 M moves through plants from the roots to the leaves without causing adverse plant growth effects (*21*); but this phenomenon has not been studied from the standpoint of frost resistance. Recently, investigators at the Connecticut Agricultural Experiment Station showed that a new family of compounds has increased the resistance of plants to frost damage. One of these, *n*-decenylsuccinic acid, appears to be particularly promising in increasing the frost resistance of flowering peach, apple, and pear, as well as young bean plants (*7*). Kuiper reported a decrease in leaf growth and more intense green coloration following treatment with this acid. This suggests that this chemical may be of the growth-retardant type.

Acknowledgment

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