Four or 5 weeks of cold treatment significantly promoted shoot emergence off the basal plate (Fig. 2). Emergence began within 17 days after planting and 80% of the shoots had emerged within 24 days. One or 2 weeks of cold treatment did not significantly promote or retard shoot emergence. Three weeks of 10° C gave the greatest difference between the date of first shoot emergence and the date when 80% of the shoots had emerged (Fig. 2).

On the other hand, leaf emergence from the scale bulblet was retarded as cold treatment duration increased. This was true for both the beginning and for the 80% leaf emergence date. This retardation of leaf emergence was most pronounced when bulbs were cold treated for more than 3 weeks.

It is well known that cold treatments promote shoot emergence from Easter lily bulbs, and this has long been employed for programming the bulbs by flower forcers. This was the case in the present experiment: 4 to 5 weeks at 10° C promoted shoot emergence. On the other hand, cold treatments retarded leaf emergence from scales bulblets developed from scales detached from the intact bulb after cold treatment. These differences in reponse suggest that scales, once separated from the whole bulb, behave differently from the remaining bulb core as related to leaf and shoot emergence.

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Deep Supercooling of Winter Flower Buds of *Cornus florida* L.¹

Akira Sakai

The Institute of Low Temperature Science, Hokkaido University, Sapporo, Japan 060

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Abstract. Differential thermal analysis on winter flower buds of Cornus florida revealed that numerous small low temperature exotherms occurred between -15 and -23° C. These exotherms resulted in the death of the winter bud florets. Only a single exotherm near -20° C was observed in individual florets. These observations indicate that the florets of the winter flower bud survive subfreezing by supercooling and that the temperature of the exotherms may be correlated with the minimum temperature occurring at the northern range of the species.

American flowering dogwood, a deciduous shrub or tree 3 to 6 m in height, is one of the most popular ornamental trees in Europe and America. In the winter flower bud, 4 conspicuous petal-like, notched involucral bracts enclose a dome shaped cluster of tiny greenish flowers about 10 mm in diameter (Fig. 1). The inflorescense consists of 20 to 30 tiny sessile flowers clustered on a common receptacle. The size of each individual flower (floret) is about 1 mm in diameter near the apex and 6 to 7 mm long.

Although most plant tissues survive subfreezing temperatures by tolerating extracellular ice formation, recent work has shown that reproductive tissues of some species survive by avoiding freezing or supercooling. Graham (5) and Graham and Mullin (6) observed low temperature freezing events or exotherms near -30° C in dormant azalea flower primordia in midwinter. Similar freezing avoidance has been observed in flower primordia of several Prunus species (8). George et al. (3, 4) employed differential thermal analysis (DTA), nuclear magnetic resonance spectroscopy and cryomicroscopy to show that low temperature exotherms in floral primordia of azalea result from the freezing of supercooled tissue water. These data suggest that supercooling may be a more important freezing resistance mechanism in the flower buds than originally presumed. Supercooling has also been found to be important for the winter survival of living xylem in many hardwoods (1, 2).

This study was designed to determine whether the flower buds of *C. florida* survive subfreezing by supercooling during winter.

Flower buds of *C. florida* were collected in midwinter in Tokyo. The occurrence of exotherms in excised flower buds was investigated by differential thermal analysis (DTA). A copper-constantan thermocouple was inserted near the florets through involucral bracts. Another thermocouple, connected in series with the first, was inserted into another flower bud which had been previously dried in an oven. Flower buds with the inserted thermocouple were placed in a thermos flask



Fig. 1. A dome shaped cluster of florets from a winter flower bud of *Cornus florida* (8 x) with involucral bracts removed (F = Floret).

measuring 7.3 cm in diameter and 20 cm in height and held at 10° C. The flask was immediately transferred to a freezer held at about -42° C. The bud temperature was determined by an additional thermocouple and recorded. Winter buds were cooled at the rate of 0.3° C/min in the temperature range between 0 and -10° C, and 0.1° C/min between -10 and -30° C.

After thawing, flower buds were individually placed in polyenthylene sacks and kept at 20°C for 3 days to evaluate viablility. Browning was used as a criterion for rating injury.

Three types of exotherms were observed by DTA. A large first exotherm (Fig. 2-exotherm A) at about -6°C resulted from the freezing of involucral bracts. In flower buds with bracts removed, exotherm A did not appear on the DTA profile. Second numerous small exotherms (Fig. 2-exotherm B) following exotherm A were probably due to freezing of the receptacle on which the florets are attached. A number of small but more distinct spike-like exotherms totaling about 25 occurred between -13 and -23°C. The number of exotherms generally corresponded to the number of florets on the receptacle of the flower bud. An excised floret was found to produce only one exotherm near -20°C (Fig. 3) which resulted in the death of the floret. A

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Fig. 2. DTA profile of winter flower bud of *Cornus florida*. A = Large first exotherm resulting from the freezing of involucral bracts; B = Exotherms probably due to freezing of the receptacle; DR = Differential response. Exotherms between -13 and -23° C resulted from freezing of individual florets.



Fig. 3. DTA profile of an excised floret.

separate freezing test revealed that the survival rate of florets decreased gradually below about -15° C and were all killed at about -23° C. These observations indicate that in the winter flower bud of *C. florida*, supercooling plays an important role in winter survival. The same result was obtained with the winter bud of Japanese flowering dogwood (*Cornus kousa*).

Exotherms of dead excised floral primordia of azalea were observed to be similar to those of living primordia (4). However, in winter flower buds of C. florida which were precooled to -25° C or below, their DTA supercooling temperature, no distinct low temperature exotherms were observed (Fig. 4). This suggests that the barriers to ice growth from the receptacle to individual



Fig. 4. DTA profile of winter flower bud which had sustained freezing injury by precooling below -25°C.

florets were lost during injurious freezing.

Spring buds collected in Tokyo on March 31 revealed no low temperature exotherms and had a water content of 66% on a fresh weight basis. For comparison, the water content of winter buds was 38%.

In Cornus florida, the living bark, vegetative bud and xylem survived freezing to $-30^{\circ}C$ (10). Thus, the freezing resistance of flower buds may be the principal factor limiting the northern natural ranges of the species. More work is needed to determine the morphological and/or physiological details which allow supercooling in individual florets of *C. florida* winter buds.

There is considerable interest in intraspecific differences in freezing resistance among ecotypes and climatic races of widely ranging species (9). Climatic stresses are prime factors in the natural selection pressures which have led to the evolution of adapted ecotypes and climatic races. C. florida is a widely ranging species in North America, found from central Florida northwards to Massachusetts, southern Maine southern Ontario, southern Michigan and westward to southeastern Kansas (7). Isolated provenances were even found in northern Mexico. An extensive study of freezing resistance of trees from different provenances of C. florida may help explain its wide distribution.

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