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# Leaf and Stem Cold Hardiness Estimates of Six Selections of Chinese Evergreen Oak over Two Winter Seasons<sup>1</sup>

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### Abstract

Two of six selections of Chinese evergreen oak (*Quercus myrsinifolia* Blume) were identified through laboratory cold hardiness estimations to possess more spring cold hardiness potential than the other selections tested. The leaves and stems of all selections had similar cold hardiness levels in the fall and midwinter. Data collected over two winter seasons revealed that leaves reached a cold hardiness level of -15 to  $-18^{\circ}$ C (5 to  $0^{\circ}$ F) during the midwinter. The stems were slightly less cold hardy than the leaves and attained a cold hardiness level of -12 to  $-15^{\circ}$ C (10 to  $5^{\circ}$ F). In the spring, however, the cold hardiness levels of the leaves of two of the selections were 5 to  $9^{\circ}$ C (9 to  $16^{\circ}$ F) more cold hardy than the other selections, while their stems were 6 to  $9^{\circ}$ C (11 to  $16^{\circ}$ F) more cold hardy than those of the other selections. The data indicated that these two selections of Chinese evergreen oak may be able to avoid injury when exposed to late spring frosts better than other selections included in the study.

Index words: freeze tolerance, acclimation, deacclimation, Quercus myrsinifolia Blume

### Significance to the Nursery Industry

The results of laboratory cold hardiness tests with six selections of Chinese evergreen oak indicated that certain selections retain greater cold hardiness than others during late winter and spring. Our data indicated that by using specific selections of Chinese evergreen oak, injury to the plant caused by low temperatures can be reduced or its cultivated range of distribution can be increased.

Multiple date laboratory cold hardiness testing holds great promise to provide an index of a plant's low temperature tolerance. With prudent use, it can provide useful information about the low temperature adaptability of a particular plant selection.

# Introduction

Chinese evergreen oak is an attractive, small evergreen tree with potential use as a specimen or screening tree in

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urban landscapes (2, 4). One factor that limits the geographic distribution of this tree is its susceptibility to freeze injury. The Chinese evergreen oak is considered by Rehder (10) to be more cold hardy than two similar evergreen oak species, Quercus acuta Thunb. and Quercus glauca Thunb. Dirr (2) reported that Chinese evergreen oak is adaptable to conditions in USDA hardiness zones 7-9. One selection has survived exposure to  $-23^{\circ}$ C ( $-10^{\circ}$ F) without injury, at the U.S. National Arboretum in Washington D.C. (G. Eisenbeiss, personal communication). To potentially increase the range of adaptability, new selections must be screened for cold tolerance in the fall, midwinter and spring. Laboratory cold hardiness estimates have been successfully used for many years on stems and leaves of a variety of woody plant materials (1, 5, 6, 7, 8, 9, 11). Currently no information is available on the cold hardiness of specific selections of Chinese evergreen oak.

This study assessed the cold hardiness of leaves and stems of several seedling selections of Chinese evergreen oak during the fall, winter and spring. A second year of laboratory cold hardiness estimates, on the selections tested in the first year, were made to corroborate the first year results.

## **Materials and Methods**

Seedlings, grown from seeds collected from Savannah, Georgia in 1967, were planted in the field at the Georgia

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Experiment Station, Griffin, Georgia in 1969. Six selections which showed promising horticultural characteristics and had survived exposure to  $-22^{\circ}C$  ( $-8^{\circ}F$ ) at Griffin, Georgia were selected for use in the study. The plants were sampled on eight dates, November 13, December 3 and 18, in 1989 and January 12, February 9 and 27, March 20 and April 16, 1990. To confirm the previous year's results, more results were also determined the following year on January 16, February 12 and 25 and March 27, 1991.

Thirty-six, uniform, 10 cm (4 in) long stem tips were removed from each plant for each test day. Leaves were removed from the stems. Samples were wrapped in wet paper towels, put in plastic bags then placed on ice for transport to the lab. Within 2 hours of collection the stems and leaves were prepared for freezing.

For the freezing test the terminal 7 cm (2.75 in) of each stem was removed, wrapped in moist cheesecloth and placed into a test tube ( $25 \times 200$  mm). Whole leaves collected from the stem samples described above, were prepared in the same manner. The tubes were then submerged in ethylene glycol-water solution (1:1) in a temperature bath precooled to  $-2 \pm 0.5^{\circ}C$  (28  $\pm 1^{\circ}F$ ). Stem and leaf temperatures were measured by thermocouples placed next to the samples and recorded by a datalogger. Crushed ice crystals were applied to the wet cheesecloth to insure that samples did not undercool. The temperature of the samples was held constant at  $-2 \pm 0.5^{\circ}$ C (28  $\pm 1^{\circ}$ F) for approximately 14 hours. Samples were then cooled at a rate of  $\leq$ 4°C (7°F) per hour. Four leaves and stems of each taxon were removed from the bath at progressively lower  $3^{\circ}C(5^{\circ}F)$ temperature intervals. Controls were prepared and kept at  $4^{\circ}C$  (7°F) for the duration of the freezing test.

After thawing at 4°C (7°F) overnight, the samples were removed from the tubes and placed in disposable, round,  $100 \times 15$  mm petri dishes containing filter paper saturated with water to maintain a 100 percent relative humidity. The petri dishes were placed on their sides at room temperature  $(22 \pm 2^{\circ}C)$  (72 ± 4°F) for 10–14 d. At this time the samples were visually evaluated for injury (3, 5, 12, 13, 14). Stem segments showing brown discoloration and breakdown of cells in the cambium and phloem were rated as dead. Similarly, injured leaves were identified by tissue browning and water soaking. The controls and samples not injured by the freezing treatments remained green and showed no discoloration in the cambium, phloem or leaves. The number of stems or leaves killed at each temperature was recorded and from these data the lowest survival temperature (LST) was determined. The LST is the lowest temperature at which little or no injury is observed as described by Sakai et al. (11). In most cases, there was no variability between replicates when determining the LST for a specific cold hardiness determination. Where variability was present, the standard deviation was reported. The lack of variation between replicates can be explained by using individual taxa which are uniform by their nature, and the cold hardiness was also determined only within a 3°C (5°F) range.

# **Results and Discussion**

The leaves were consistently as or more cold hardy than the stems (Table 1 and 2). This was expected since stem tips represent the tissue most often damaged by low temperatures and for that reason, were chosen for our evaluations. Differences in the cold hardiness levels of the six selections for most sampling dates were small, but, at certain times of the year they occurred. For example, during the first year of the study (Table 1), from fall (November 13, 1989) through the midwinter sampling date (February 9, 1990), there was no more than  $3^{\circ}C$  ( $5^{\circ}F$ ) difference in cold hardiness estimates among the stems or among the leaves.

Table 1. Lowest survival temperature (C ± SD) for leaves and stems of six seedling selections of Chinese evergreen oak from November 13, 1989 through April 16, 1990.

Date collected	Seedling selection								
	1	2	3	4	5	6			
November 13, 1989									
Leaf	-9	- 9	- 12	- 9	-9	-12			
Stem	-9	- 9	-9	-9	-6	-9			
December 3, 1989									
Leaf	- 15	- 15	-15	$-17 \pm 2$	- 15	- 15			
Stem	- 15	-15	-12	-12	-12	-12			
December 18, 1989									
Leaf	- 18	-21	-18	- 18	- 18	- 18			
Stem	-15	- 18	-15	- 18	-15	- 15			
January 12, 1990									
Leaf	- 18	-21	-18	- 18	- 18	- 18			
Stem	- 15	-18	-15	- 18	-15	- 15			
February 9, 1990									
Leaf	- 18	-18	-15	-18	- 18	- 18			
Stem	-12	-12	-12	-12	-12	-12			
February 27, 1990									
Leaf	-12	-15	- 15	$-13 \pm 3$	-12	$-11 \pm 2$			
Stem	- 9	-12	-12	-9	-9	-9			
March 20, 1990									
Leaf	-6	-12	$-13 \pm 2$	$-6 \pm 2$	$-7 \pm 1$	$-6 \pm 2$			
Stem	- 3	$-11 \pm 2$	$-12 \pm 3$	$-3 \pm 3$	$-5 \pm 2$	$-3 \pm 3$			
April 16, 1990			,	2 + 1	-2 + 2	CKZ			
Leaf	CK <sup>z</sup>	-6	-0 CV/	$-5 \pm 1$	-2 - 2 CK <sup>2</sup>				
Stem	CK <sup>z</sup>	CK <sup>z</sup>	CK*	- I ± I	UN	UK			

<sup>2</sup>Only nonfrozen control survived.

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Date collected	Seedling selection							
	1	2	3	4	5	6		
January 16, 1991								
Leaf	- 18	-18	- 18	- 18	- 18	- 15		
Stem	- 12	- 15	-15	- 15	- 15	-12		
Februarv 12, 1991								
Leaf	- 18	- 18	- 18	- 18	- 18	- 15		
Stem	-12	- 15	-15	- 15	- 15	-12		
Februarv 25, 1991								
Leaf	- 18	- 18	-18	-15	- 18	-12		
Stem	$-11 \pm 2$	-12	-12	-12	- 12	-9		
March 27, 1991								
Leaf	-12	- 18	- 18	-12	$-11 \pm 2$	-9		
Stem	- 9	$-15 \pm 2$	- 15	-9	-12	-9		

 Table 2.
 Lowest survival temperature (C ± SD) for leaves and stems of six seedling selections of Chinese evergreen oak from January 16 through March 27, 1991.

The maximum cold hardiness that the leaves attained was  $-21^{\circ}$ C ( $-6^{\circ}$ F) for seedling selection 2 on December 18, 1989 and January 12, 1990 while the other selections had cold hardiness estimates of  $-18^{\circ}$ C ( $0^{\circ}$ F) on the same dates (Table 1). On these dates the stems of selection 2 and 4 had cold hardiness estimates of  $-18^{\circ}$ C ( $0^{\circ}$ F) while the other selections had estimates of  $-18^{\circ}$ C ( $0^{\circ}$ F).

As the plants began to deacclimate differences were noted in the cold hardiness estimates between the six selections. The leaves and stems of selections 2 and 3 retained higher cold hardiness levels than the other selections in the study during the spring sampling dates. On the sampling date of March 20, 1990 the leaves of the above mentioned two selections were more cold hardy, by 5 to 7°C (9 to 13°F), than the other selections while the stems were more cold hardy by 6 to 9°C (11 to 16°F). On April 16, 1990 the stems of selections 2 and 3 were at least 3°C (5°F) more cold hardy than the others; the stems, however, were completely deacclimated on this date (Table 1).

During the second year of the study few differences were found among the leaves or the stems of all the selections studied during the midwinter sampling dates (Table 2). On January 16, 1991 and February 12, 1991 the cold hardiness of the leaves of all the selections was  $-18^{\circ}$ C (0°F) except for selection 6 which was  $-15^{\circ}$ C (5°F). The stems were less cold hardy and their cold hardiness estimates ranged from -12 to  $-15^{\circ}$ C (10 to 5°F) on these same dates. As in the previous year when the plants began to deacclimate, the cold hardiness of both the leaves and stems of seedling selections 2 and 3 remained more cold hardy later in the spring than the other selections (Table 2).

The data, collected over two winter seasons, indicated that selections 2 and 3 of Chinese evergreen oak remain more cold hardy later in the spring than the other selections in the study. This could allow these two selections to survive exposure to lower temperatures than the others during the spring. The midwinter cold hardiness levels of these two selections were not greater than the other selections in the study so a great increase in the geographic distribution may not be expected. However, the significantly greater cold hardiness of the stem tips and the leaves in the spring could reduce the damage often observed on stem tips of Chinese evergreen oak after a late spring frost.

Chinese evergreen oak is a promising tree for zones 7 to 9(2). These data indicate that certain selections of the species do have more cold hardiness than others. This species

is relatively easy to root from cuttings, which can insure the propagation of clonal material with the increased cold hardiness (Mike Dirr, personal communication, 1991). Therefore, further evaluation, from a wider range of seedlings, is prudent in order to find more cold hardy Chinese evergreen oak selections. Based on evidence presented here the evaluation of cold hardiness should be taken at several times throughout the fall, winter and spring to obtain a complete understanding of the cold hardiness of specific selections of this species.

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