LOW TEMPERATURE SURVIVAL OF ‘REDHAVEN’ PEACH FLORAL BUDS ON

SELECTED ROOTSTOCKS

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By

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The undersigned, appointed by the Dean of the Graduate School,

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LOW TEMPERATURE SURVIVAL OF ‘REDHAVEN’ PEACH FLORAL BUDS ON SELECTED ROOTSTOCKS

Presented by Audrey Lynn Davis

A candidate for the degree of

Master of Science

And hereby certify that, in their opinion, it is worthy of acceptance.

Dr. Michele Warmund

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ABSTRACT

The relative cold tolerance of 'Redhaven' peach floral buds grafted onto various rootstocks was evaluated at selected dates from November 2011 to March 2013. Budwood was collected from trees in coordinated rootstock trials at New Franklin, MO and Clemson, SC for artificial freezing tests in late fall, mid-winter, and early spring. Samples were cooled 3 °C/h, thawed, and 'Redhaven' floral bud T_{50} values for each rootstock were calculated from the number of dead buds per test temperature. Although winter temperatures were unseasonably warm during this study, 'Redhaven' floral buds varied in cold tolerance among the rootstocks grown in Missouri in February 2012 and March 2013. In February 2012, 'Redhaven' floral buds on trees with KV010-127 and HBOK 32 rootstocks were the most cold tolerant, but in March 2013, those on Guardian rootstock were the hardiest. For South Carolina, 'Redhaven' floral buds on trees with Lovell and Viking rootstocks were the most hardy in January 2012, which was the only sampling date in which T_{50} values differed among rootstocks. When data were pooled from both locations, mean 'Redhaven' floral bud T_{50} values were always lower in Missouri than in South Carolina at similar collection periods. Also, buds from trees on Lovell, Guardian, Bright's Hybrid #5, and HBOK 32 rootstocks were hardier than those on Controller 5 and Mirobac rootstocks.

CHAPTER 1: INTRODUCTION

Peach [*Prunus persica* (L.) Batsch] is a valuable commercial fruit crop. It is the third most important deciduous fruit tree worldwide and ranks fourth in acreage with nectarines in the United States (FAO Statistics, 2012). In 2011, there were 45,519 ha of bearing peach trees with a retail value of $588.3 million in the U.S. (USDA-NASS, 2012). In the same year, 607 ha of peaches were grown in Missouri for fresh market with a crop value of $6.1 million (USDA-NASS, 2013). With increased interest in buying high quality, locally grown fruit, Missouri peach production has expanded from 1997 to 2002 from southern areas to more northern sites. Crop loss frequently occurs on these northern sites due to low winter temperatures or spring frosts (M.R. Warmund, personal communication).

Only one full crop and one partial crop of peaches is produced in northern Missouri every four years (Warmund, 2009). Methods for frost avoidance include overhead irrigation, orchard heating, wind machines, or using helicopters to mix warm and cold layers of air during a temperature inversion (Kamas and Stein, 2011). Current practices are neither cost effective with current labor and fuel prices nor practical for small commercial plantings. However, the use of cold hardy rootstocks that influence flower bud hardiness may reduce the risk of crop loss.

Since Missouri is located in the northern range for successful peach production, with typically cold and erratic winter temperatures, it has been used as a test site for evaluating the low temperature survival of peach floral buds (Warmund et al., 2002). In contrast, winter climatic conditions in South Carolina are typically mild, but can also be erratic. Therefore, the objective of this study was to determine the relative cold tolerance of ‘Redhaven’ floral buds on peach trees grafted with several rootstocks grown in NC-140 rootstock trials in Missouri and South Carolina.

CHAPTER 2: LITERATURE REVIEW

Peach trees used for commercial fruit production are commonly propagated by budding. During June or July, when the rootstock bark lifts easily, a dormant scion floral bud is inserted into rootstock tissue (Westwood, 1993). After successful union formation, the scion grows and becomes the fruiting portion of the tree. The scion cultivar influences resistance to bacterial and fungal pathogens, fruit appearance, flesh texture, crop load, and time of harvest (Layne, 1975). The cultivar 'Redhaven' is frequently grown in commercial orchards in Missouri because it is relatively cold tolerant and moderately resistant to bacterial spot (M. Warmund, personal communication). Rootstocks influence precocity, tree size and longevity, and resistance to high soil pH, pests, and diseases (Layne, 1975). Lovell is a commonly used seedling rootstock that provides good tree anchorage, produces moderate tree vigor, and has few root suckers (Univ. Calif., 2013).

To enhance fruit yield per hectare, peach trees on dwarfing rootstocks are planted at high densities. Nanking cherry (*Prunus tomentosa* Thunb.) and sand cherry (*Prunus besseyi* Bailey) have been used as rootstocks for peach to produce dwarf trees (Layne, 1987). Only a few plum (St. Julien X and St. Julien GF 655.2) and peach seedling (Siberian C and Rubira) rootstocks are dwarfing. However, compatibility problems associated with Nanking and sand cherry, as well as insufficient cold hardiness of St. Julien X, have limited their use as dwarfing rootstocks. In addition, vigor-reducing rootstocks have not performed well in shallow soils or those with low fertility (Layne, 1987).

To maintain a viable peach industry, researchers have conducted North Central Regional Association (NC-140) peach trials to evaluate rootstocks grown under diverse environmental conditions across North America. The first trial conducted in 1984 included 'Redhaven' peach on nine rootstocks at 16 locations. In this study, tree survival was highest for Bailey, Halford, and own-rooted 'Redhaven' and lowest for Citation and Damas 1869 rootstocks (Perry et al., 2000). Trees on GF 677 had greater trunk cross-sectional area than those on Citation, GF 655-2, and Damas 1869. The most productive trees were on GF 677, Halford, Bailey, Siberian C rootstocks, and own-rooted 'Redhaven'.

In the 1994 NC-140 trial, 'Redhaven' was the scion cultivar on nineteen rootstocks at 20 sites (Reighard et al., 2004). Trees on Bailey and GF 305 rootstocks had greater survival than trees on Lovell, Higama, and Guardian across all test sites. Lovell and Guardian produced vigorous trees while Bailey and Tennessee Natural 281-1 were small. Trees on Lovell, GF 305, and Montclar had greater cumulative yield than those on Bailey and Higama.

In the 2001 NC-140 trial, 14 rootstocks were evaluated on 'Redtop', 'Redhaven', or 'Cresthaven' (Reighard et al., 2011). Ten of these rootstocks were interspecific *Prunus* hybrids and performance of these rootstocks was compared to Lovell, Bailey, Guardian, and Pumiselect. None of the rootstocks had a higher survival rate than Lovell. Cumulative fruit yield for four years was highest on peach seedling (Bailey, Guardian, and Lovell), Cadaman rootstocks and peach x almond hybrids (BH-4 and SLAP). Although dwarfing rootstocks were identified in this study, none of them performed consistently across all locations (Reighard et al., 2011).

In the 2002 NC-140 trial, eight rootstocks from Europe and Russia were evaluated with 'Redhaven' or Cresthaven scions. Pumiselect and Krymsk 2 rootstocks had low tree survival, a large number of roots, and small fruit size (Johnson et al., 2011). However, trees with Krymsk 1 and Cadaman rootstocks were more yield efficient than those with Adesoto 101, Penta, and Pumiselect rootstocks across all locations.

Although peach trees are widely adaptable to many locations, they are susceptible to diverse biotic and abiotic problems. Thus, tree performance varies by location and rootstocks may be selected to tolerate adverse site conditions. Historically, wild type peach seedlings, such as Tennessee Naturals, were used as rootstocks in North America. However, because of inherent variability in size and performance, rootstock seedlings derived from commercial peach cultivars, such as Lovell, Halford, Siberian C, Harrow Blood, Bailey, and Rubira have been propagated for commercial orchards (Layne, 1987).

More recently, other *Prunus* species and interspecific *Prunus* hybrids have been developed as rootstocks to increase peach tree uniformity and selected for resistance to a specific problem. For example, Nemaguard rootstock (*Prunus persica* x *P. davidiana*) is commonly used in Florida and California where root knot nematodes are problematic. Plums (*P. americana*, *P. blireana*, *P. cerasifera*, and *P. salicina*) and peach x plum rootstocks have been utilized for peach trees (Layne, 1987). Additionally, Nanking cherry, sand cherry and almond (*P. dulcis*) have been hybridized with peach to produce rootstocks.

Rootstocks are an important source of resistance for nematodes and peach tree short life complex, which are serious peach tree pests in warm climates. Nematodes are also associated with peach tree short life, which can cause tree mortality at about three to seven years after planting. The southern (*Meloidogyne incognita*) and javanese (*Meloidogyne javanica*) root-knot nematodes are problematic in the southern parts of the U.S. Peach trees on Nemagaurd, Shalil, S-37, Okinawa, Higama, Nemared, and Viking have shown resistance to these root-knot nematode species (Layne, 1987; Reighard, 2011). Also, HBOK 32 is resistant to *M. icognita* (race 1) isolate Beltran (Bliss et al., 2011). In cooler climates, such as southern Canada and northern U.S., root lesion nematodes (*Pratylenchus* spp.) are problematic and there are few resistant rootstocks to these roundworms. Penta, Krymsk1 and 86, and Viking are resistant to *P. vulunus*, while Guardian is quite susceptible. In the central valley of California and southeastern U.S., ring nematodes [*Mesocriconema* (= *Criconemella*, *Criconemoides*, and *Macroposthonia*) spp.] adversely affect peach production because they predispose trees to bacterial canker and winter injury (Layne, 1987). Most early European rootstock introductions (Montclar, Rubira, GF 305, Higama, Ishtara, and Myran) were hosts for ring nematodes, but Guardian and Viking rootstocks have resistance to these organisms (Reighard, 2008; Reighard, 2011). Dagger nematodes (*Xiphinema* spp.) also parasitize both peach and plum rootstocks and are a vector for the tomato ringspot virus (TomRSV), a precursor of stem pitting (Layne, 1987). Peach seedlings lack resistance to these nematodes so other *Prunus* rootstocks are used where *Xiphinema* spp. or TomRSV is problematic (Reighard, 2011). Rootstocks with cherry x plum parentage, such as Krymsk 1, are less sensitive to TomRSV, but have not been fully tested in North America.

Fungal and bacterial infections can also limit peach tree performance or contribute to tree decline. Crown rot, bacterial canker, and crown gall are important diseases caused by *Phytophthora* spp., *Pseudomonas syringae* pv. *syringae,* and

Agrobacterium tumafaciens, respectively. Rootstock susceptibly to crown rot varies with the *Phytophthora* species present in the orchard (UC Statewide IPM Program, 2002). *P. cinnamomi* is the most common among peach trees and preliminary trials indicated that Tetra rootstock had some resistance to the fungal disease (Horton and Johnson, 2011; Layne and Bassi, 2008). Peach scions on plum seedling rootstocks (Myrobalan, Myrobalan 29, and Mariana 2624) have some resistance to *P. cactorum* (Layne, 1987). When trees ≤ 7 years-old were grown in deep sandy soil, they were susceptible to bacterial canker (Texas A&M AgriLife Ext., 2013). However, Damas GF 1869 and St. Julien GF 655-2 rootstocks are resistant to bacterial canker (Bhutani and Joshi., 1995). *A. tumafaciens* infection usually occurs when tree trunks or limbs are injured during planting or cultivation or following insect feeding (Texas A&M AgriLife Ext., 2013). Several rootstocks, including *P. pumila, P. domestica, P. besseyi, P. insititia, P. mume,* and *P. umbellate* are resistant to *A. tumafaciens* (Layne, 1987).

Insect damage can also restrict peach production. While several insects feed on the scion foliage and fruit of peach trees, rootstocks are also susceptible to insect damage in the nursery before grafting. The green peach aphid (*Myzus persicae*) and peach-potato aphid (*M. varians*) are serious pests because they can transmit plum pox during feeding on rootstocks and trees may become infected with this virus during grafting (Rubio et al., 2005). Peach seedlings resistant to the two aphids include Rubira and S2678. In a study by Rubio et al. (2005), 15 *Prunus* rootstocks in an insect-proof greenhouse were inoculated with an isolate of plum pox during chip-grafting to evaluate resistance. GF677 (almond x peach hybrid), Myrobolan 29C (plum), and L2 (cherry) rootstocks did not exhibit viral symptoms, while GF305 (peach), Puebla de Soto (plum), and Real Fino (apricot) rootstocks were highly susceptible to the virus.

Rootstock selection can also influence peach vegetative growth and yield when trees are planted in high pH soils. Almond and plum rootstocks tend to enhance peach tree performance in alkaline soils where iron-induced chlorosis is c(1987) ranked Damas GF 1869 (*P. domestica* x *P. spinosa*) and GF 677 rootstocks as the most high pH tolerant, followed by St. Julien hybrids 1 and 2, Brompton, Prunier GF 43, wild peach, GF 305, St. Julien GF 655.2, and Nemaguard (Layne, 1987).

Low temperature injury is a major limiting factor in peach production. Cold episodes can result in severe limb die-back, trunk damage, root injury, floral bud injury, and tree mortality. The type of cold injury on peach trees varies during the dormant period (Weaver, 1968; Layne, 1984). For example, the lower trunk and crotches of lower limbs of trees are susceptible to injury in the early fall when temperatures drop between -6 and -9 ˚C following a warm period (Westwood, 1993). Following a low temperature event, bacterial and perennial (*Leucostoma* spp.) canker can infect the tree trunk, which eventually leads to tree mortality (Kansas State Univ., 2012). In sand, or sandy loam soils, root injury occurs when soil temperatures are -5 °C to -13 °C (Layne, 1974). When -13 °C was recorded at the Harrow Research Station, Siberian C rootstock had little or no root injury followed by Harrow Blood (Layne, 1987). Muir, Rutgers Red Leaf, and Halford had moderate root injury while Elberta, Nemaguard, Yunnan, and Shalil rootstocks were severely injured or killed (Layne, 1987). In another study, Layne (1974) evaluated cold hardiness of selected nursery tree root systems using root segments and intact and detached root systems of nursery trees in controlled freezing tests. Roots of

Tzim Pee Tao and Siberian C were more cold tolerant than those of Bailey and Rutgers Red Leaf and Elberta had the most root injury.

In colder areas of production on non-sandy soils, peach floral bud injury occurs more frequently than root injury (Warmund et al., 2002). This type of low temperature injury occurs in mid or late winter and can reduce annual yield. Several researchers used 'Redhaven' peach trees in the 1984 NC-140 rootstock trial to identify a cold hardy germplasm. Rootstocks in this trial included Bailey, Damas 1869, GF 655-2, GF 677, Halford, Lovell, Siberian C, and own-rooted 'Redhaven' trees (Perry et al., 2000). After a natural freeze (-26 °C) in New York, Brown and Cummins (1988) reported that 'Redhaven' floral buds on trees with Citation or Damas 1869 rootstocks had the highest survival while those on trees with GF 655-2 or GF 677 had the lowest survival. In the New Jersey trial, after two consecutive years of minimum temperatures at ≥ -23 °C in January, floral buds on Siberian C rootstock were hardier than those on GF 677 (Durner, 1990). In controlled freezing tests conducted in Missouri, 'Redhaven' floral buds on Damas 1869 rootstock were the hardiest followed by those on Lovell (Warmund and Slater, 1988). When 'Redhaven' floral bud hardiness was evaluated using peach trees in the 1994 NC-140 trial, T_{50} values (temperature at which 50% bud mortality occurs) of buds on Ta Tao 5 interstem/Lovell were similar to, or lower than that of Lovell trees (Warmund et al., 2002).

Peach floral buds acclimate to low temperatures in the fall, develop maximum hardiness in mid-winter and deacclimate in the spring. Acclimated floral buds avoid lethal freezing injury by supercooling during winter. When cellular water is supercooled, it remains in equilibrium until disrupted by ice nucleators at sub-zero temperatures

(Layne, 1987). As winter temperatures drop below 0 °C, cellular water in tissues at the base of a peach floral bud migrates to extracellular spaces due to differences in osmotic potentials (Quamme and Gusta, 1987). Generally, non-lethal extracellular freezing occurs between -5 to -10 °C in floral buds. However, as temperatures fall below -10 °C, ice nucleators induce intracellular ice formation that ruptures cell walls and results in lethal injury to floral primordia.

In late winter, after the chilling requirement is satisfied, peach buds deacclimate and floral tissues become susceptible to fluctuating temperatures (Westwood, 1993). From first bud swell to post bloom, average temperatures for 10% floral bud mortality range from -8 to -2 °C, respectively (Ballard and Proebsting, 1978). The first three peach bud stages (first swell, calyx green, and calyx red) are more susceptible to spring frosts and freezes than later floral stages (first pink, first bloom, full bloom, and post bloom). Generally, late flowering trees are more likely to escape cold injury than early-blooming trees.

Various peach cryoprotectant or antitranspirant products have been evaluated to prevent floral injury during frosts. Rieger (1988) sprayed Junegold peach blossoms with FrostFree® [50% propylene block copolymer of poloxyothylene, 50% propylene glycol (Plant Products, Vero Beach, Fla.)] or Protec (hydrophilic polymer) 12 hours before sub-zero temperatures were recorded in the field. However, both products were ineffective for frost protection. In another experiment, ‘Redhaven’ trees treated with FrostFree before a naturally occurring frost had higher yields than untreated trees (Reighard et al., 1991). Vaporgard contains pinolene, a terpenic polymer (Miller Chemical & Fertilizer, Hanover, Pa.) and was also tested for cold protection on Legend peaches, but results were inconclusive because field temperatures were at -2.2 °C for less than one hour (Aoun et al., 1993). Thus, such treatments may not always protect peach floral buds from low temperature injury and are an additional production cost.

In the most recent NC-140 trial, up to 18 rootstocks of diverse *Prunus* species were planted at 17 locations (NC-140, 2012). Of these rootstocks, *Prunus americana*, HBOK 10, HBOK 32, Krymsk 1, and Controller 5 rootstocks are considered dwarfing as compared to Lovell (Autio et al., 2010). Penta rootstocks produce vigorous trees tolerant of wet, heavy soils. Both Krymsk 1 and Krymsk 86 rootstocks are tolerant to heavy, wet soils. While such information is known about these rootstocks, their effect on low temperature survival of scion fruit buds has not been evaluated.

CHAPTER 3: MATERIALS AND METHODS

Peach rootstock trials were established in 2009 with 'Redhaven' as the scion cultivar budded onto several *Prunus* rootstocks (Table 1) as part of the NC-140 Regional Rootstock Project (NC-140, 2012). NC-140 trials at New Franklin, Mo. and Clemson, S.C. were used for this study. Trees at the Missouri site were planted in a Menfro silt loam soil (fine-silty, mixed, superactive, mesic typic hapludalfs) and those in South Carolina were grown in a Appling loam sand (clayey, kaolinitic, thermic typic hapludalfs). Trees at both locations were spaced 5 × 6 m apart and arranged in a randomized complete block design with eight replicate trees of each rootstock. At both locations, trees were trained to an open-vase system and blossoms were removed to promote vegetative growth in 2009 and 2010. Thereafter, fruit was harvested annually. Trees were fertilized, irrigated, and pruned following local recommendations (Doubrava et al., 2013; Warmund, 2009).

For the freezing tests, floral buds from trees on 12 common rootstocks at each location were tested, as well as tissue from trees on *P. americana* rootstocks growing in South Carolina. Dates of budwood collection were selected to compare similar phenological bud stages over a two-year period. However, bud samples were not obtained from South Carolina in Nov. 2011. November, January, and February test dates were used to assess floral bud acclimation, mid-winter hardiness, and de-acclimation, respectively. In Missouri, tissue for the first dormant period freezing tests was collected on 11 Nov. 2011, 25 Jan. and 28 Feb. 2012. During the second dormant season, tissue

Table 1. Pedigree of rootstocks used for floral bud cold hardiness evaluations in Missouri and South Carolina.[z]

Rootstock	Pedigree
Lovell	peach [*Prunus persica* (L.) Batsch]
Guardian	peach
HBOK 32	peach
KV010-123	peach
KV010-127	peach
Prunus americana	American plum (*P. americana* Marsh.)
Penta	European plum (*P. domestica* L.)
Bright's Hybrid #5	almond (*P. dulcis* Mill.) x peach
Viking	peach x flowering plum (*P. blireana* André)
Controller 5	Japanese plum (*P. salicina* Lindl.) x peach
Mirobac	Myrobolan plum (*P. cerasifera* Ehrh.) x almond
Krymsk 1	Nanking cherry (*P. tomentosa* Thunb.) x Myrobolan plum
Krymsk 86	Myrobolan plum x peach

[z] Autio et al., (2010).

was sampled on 12 Nov. 2012 and 18 Jan. and 13 Mar. 2013. For South Carolina, floral tissue was collected on 16 Jan. and 8 Feb. 2012. The following winter period, budwood were sampled on 14 Nov. 2012 and 23 Jan. and 4 Mar. 2013.

At each location, budwood was collected from six trees of each rootstock in the field trial. At each sampling date, six 5-node cuttings (approximately 15 cm-long) were collected from the middle portion of one-year-old wood from each tree quadrant at 1.2 to 1.5 m above the soil surface. Budwood from New Franklin was then transported to the laboratory in Columbia, Missouri in sealed bags and prepared immediately for freezing tests. Samples from South Carolina were collected similarly, packed on ice in an insulated cooler, and shipped by overnight mail to Columbia, Missouri where tissue was prepared for freezing tests. Immediately thereafter, a five–node cutting of each rootstock was wrapped in moistened cheesecloth and sealed in aluminum foil for each of the six test temperatures (Fig. 1). One test temperature at each date was always an unfrozen control where cuttings were wrapped in moistened cheesecloth and stored at 4 °C during the freezing test. For tissue subjected to freezing, a 0.01-mm-diameter (30-gauge) copper-constantan thermocouple was placed in contact with a floral bud on one cutting per test temperature. Each thermocouple was plugged into a digital thermometer (Omega Engineering, Inc., Stamford, Conn.) to monitor tissue temperature during the freezing test (Figs. 2 and 3). These samples were placed in a Tenney Benchmaster programmable freezer at 2.0 °C overnight and held at -2.0 ˚C for 1 h before cooling at 3.0 °C/h. Samples were quickly removed from the freezer at 3.0 °C intervals, using a range of temperatures likely to produce tissue injury (Warmund et al., 2002). The range of test temperatures ranged from -6 ˚C to -24 ˚C. After removal from the freezing chamber, samples were thawed at 4.0 °C for 24 h, and placed at 21.0 °C for 5 days before floral bud evaluation.

Five floral buds at different nodes of each sample were sectioned longitudinally with a razor blade and examined under a dissecting microscope (Leica Microsystems, Heerbrugg, Switzerland) at ≤ x 40 (Fig. 4) . The number of dead buds that exhibited pistillate browning was recorded for each sample (Fig. 5). A modified Spearman-Kärber equation was used to calculate floral bud T_{50} values (temperature at which 50% of the buds are dead) for each replication of a rootstock (Bittenbender and Howell, 1974). For each collection date, T_{50} values from each location were subjected to an analysis of variance (ANOVA), using the GLM procedure of SAS (Version 9.3; SAS Institute; Cary, NC), and means were separated by Fisher's protected least significant difference (LSD) test, $P \leq 0.05$. Next, LT_{50} data for 12 rootstocks from both sites and all sampling dates were pooled and analyzed with the MIXED procedure of SAS and means were separated by Fisher's LSD test, $P \leq 0.05$.

Fig. 1. Cuttings from trees of each rootstock placed in moistened cheesecloth and aluminum foil before a freezing test.

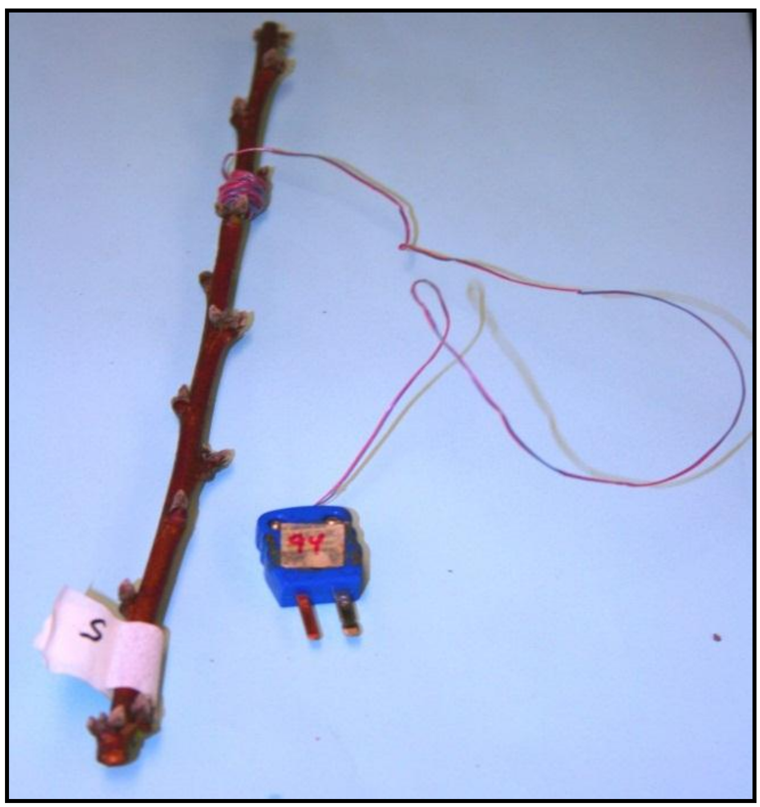

Fig. 2. Copper-constantan thermocouple in contact with a floral bud and wrapped around budwood. Thermocouple output was used to monitor floral bud tissue temperature during a freezing test.

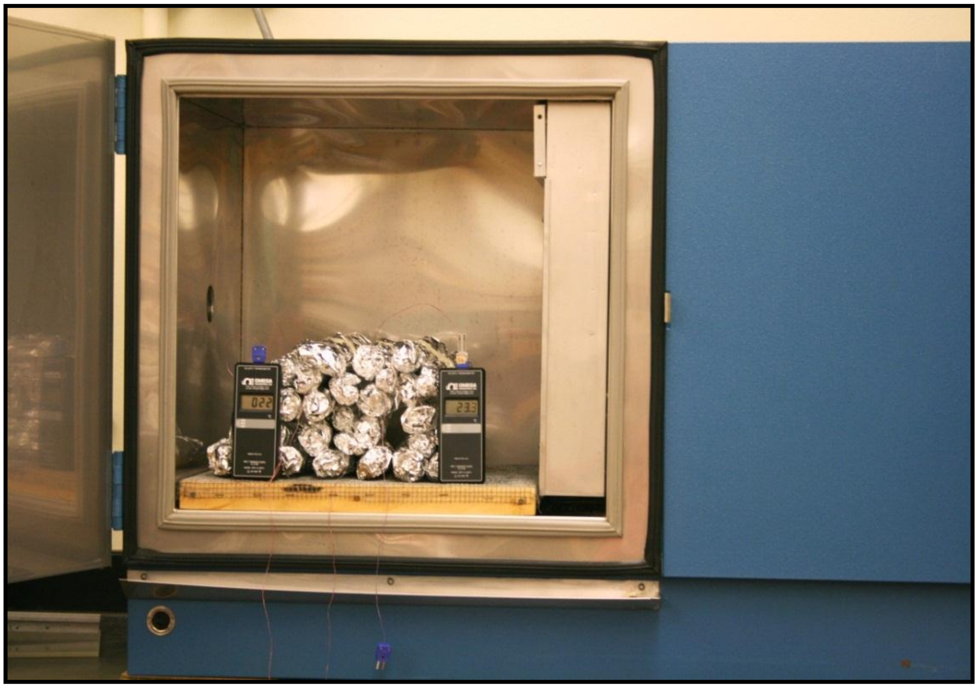

Fig. 3. Bundles of wrapped budsticks with thermocouples attached to digital thermometers in the programmable chamber before a freezing test was conducted.

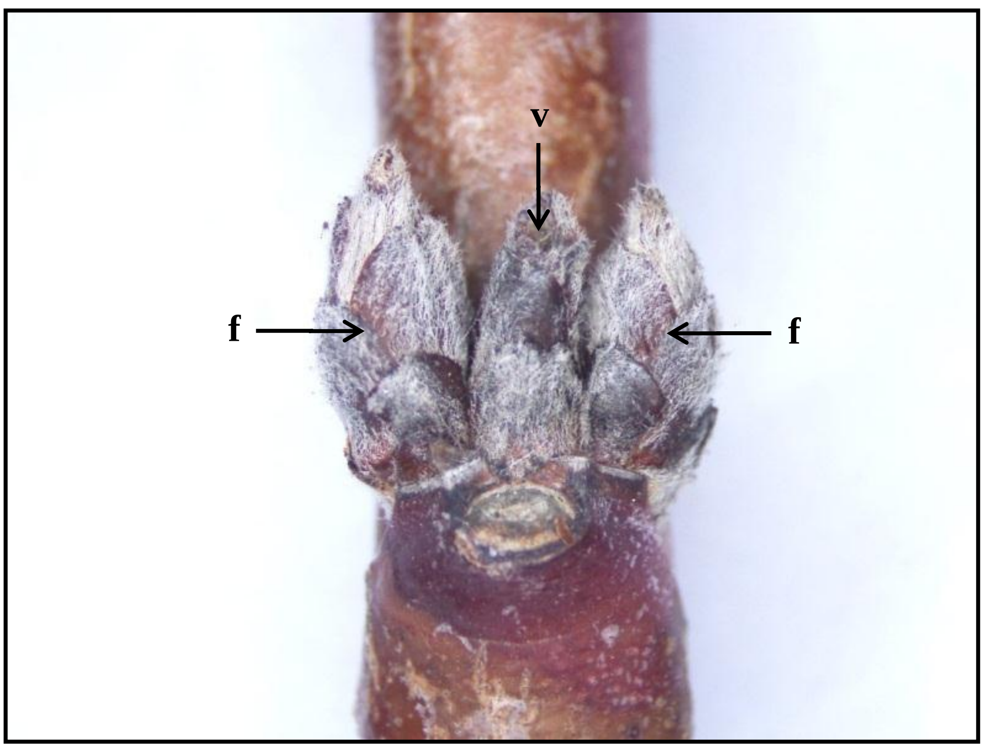

Fig. 4. Budwood node of ‘Redhaven’ peach with 2 floral buds (f) and a single vegetative bud (v).

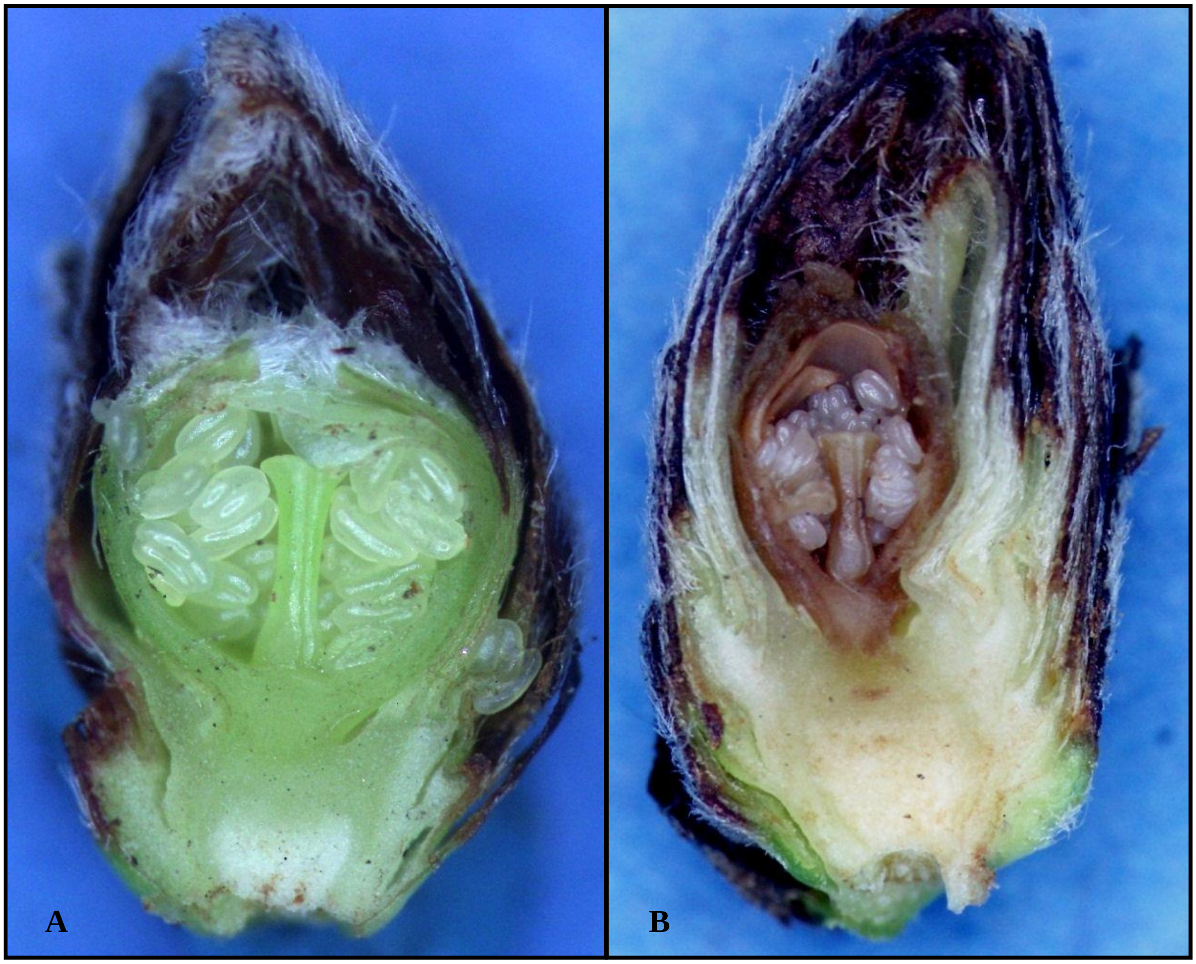

Fig. 5. Longitudinal section of (A) live and (B) dead ‘Redhaven’ peach floral buds.

Chapter 4: Results

Temperatures recorded in Missouri during this experiment were generally higher than historical average minimum daily air temperature (Fig. 6A). Over half of the daily minimum temperatures exceeded historical average temperatures by 3 ˚C from 1 Nov. 2011 to 28 Feb. 2012. March 2012 was particularly warm with 11 daily minimum temperatures at 10 ˚C above average. The average temperature of March 2012 was 14 °C, 7 °C above the historical minimum temperature (Guinan, 2013). Both regionally and statewide, March through May 2012 was the warmest spring on record since 1895 (P.E. Guinan, personal communication). The lowest temperature recorded at the Missouri NC-140 test site during the first dormant season was -13 °C on 12 Feb. 2012, which was 7 °C below the historical minimum temperature. Precipitation from November 2011 to March 2012 exceeded historical rainfall (292 mm) for these months by 107 mm (Fig. 6A).

The second test period (1 Nov. 2012 to 13 Mar. 2013) in Missouri was also unseasonably warm with the daily minimum temperatures exceeding historical averages by 3 °C on 45 of the 133 days (Fig. 6B). In November 2012, record breaking high temperatures were recorded on four days. Also, a record high of 23 °C occurred on 29 Jan. 2013 with a daily minimum temperature of 11 °C, which was 18 °C above normal. However, from 1 Nov. 2012 to 13 Mar. 2013, 41 of the 133 days were ≥ 3 °C below historical temperatures. The lowest temperature recorded during the second dormant season in Missouri was -16 °C on 2 Jan., 14 Jan., and 23 Feb. 2013. March 2013 was particularly cool with 58% of the minimum temperatures ≥ 3 °C below average. The average statewide temperature for March was the coldest in 17 years at 3.4 °C (Guinan,

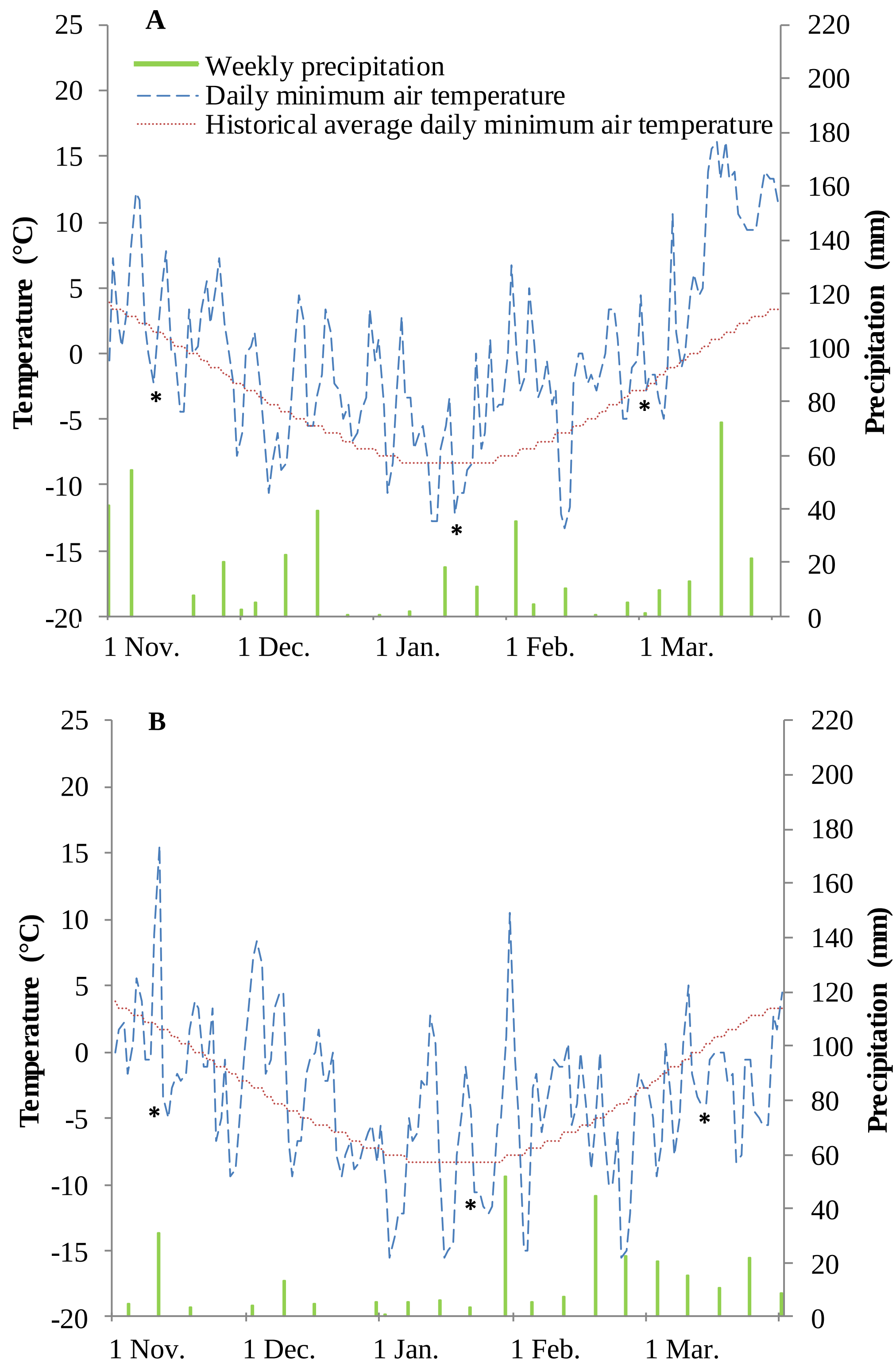

Fig. 6. Daily minimum air temperature, historical (1971-2000) average daily minimum air temperature, and weekly precipitation from (A) November 2011 to March 2012 and from (B) November 2012 to March 2013 for New Franklin, Missouri. Asterisks denote sampling dates of floral bud tissue from the field.

2013). Precipitation levels from November 2012 to March 2013 were 37 mm less than the historical annual rainfall (Fig. 6B).

Like Missouri, minimum temperatures during the two test periods were often much warmer than historical averages in South Carolina (Fig. 7A and B). From 1 Nov. 2011 to 8 Feb. 2012, temperatures exceeded historical averages by 3 ˚C for 52 of the 100 days at the South Carolina NC-140 trial site. Of these 52 days, 10 days were ≥ 10 °C above the average minimum temperature. With these above average temperatures, 2012 was the fourth warmest year on record in South Carolina (Bielenberg, 2013). Only 19 days were ≥ 3 ˚C below average daily minimum temperatures from 1 Nov. 2011 to 8 Feb. 2012. The lowest temperature recorded during the test period was -8 °C on 4 Jan. 2012, which was 7 ˚C below the average minimum temperature. Precipitation from November 2011 to March 2012 was 136 mm lower than the historical rainfall (636 mm) for these months (Fig. 7A).

The second test period from November 2012 to March 2013 in South Carolina was also unseasonably warm (Fig. 7B). From 1 Nov. 2012 to 4 Mar. 2013, temperatures frequently exceeded historical average minimum temperatures by 3 °C on 56 of the 124 days. Also during this period, 27 days were ≥ 3 ˚C below historical minimum temperatures. The lowest temperature recorded during the test period was -6 °C on 2 Feb. 2013, which was 5 ˚C below the average minimum temperature. Precipitation from November 2012 to March 2013 was 597 mm (Fig. 7B).

When floral bud data were analyzed by location, T_{50} values at the Missouri test site were similar among rootstocks when evaluated on 11 Nov. 2011 and 25 Jan. 2012 (Table 2). In November 2011, T_{50} values ranged from -10.9 to -13.1 °C. By January,

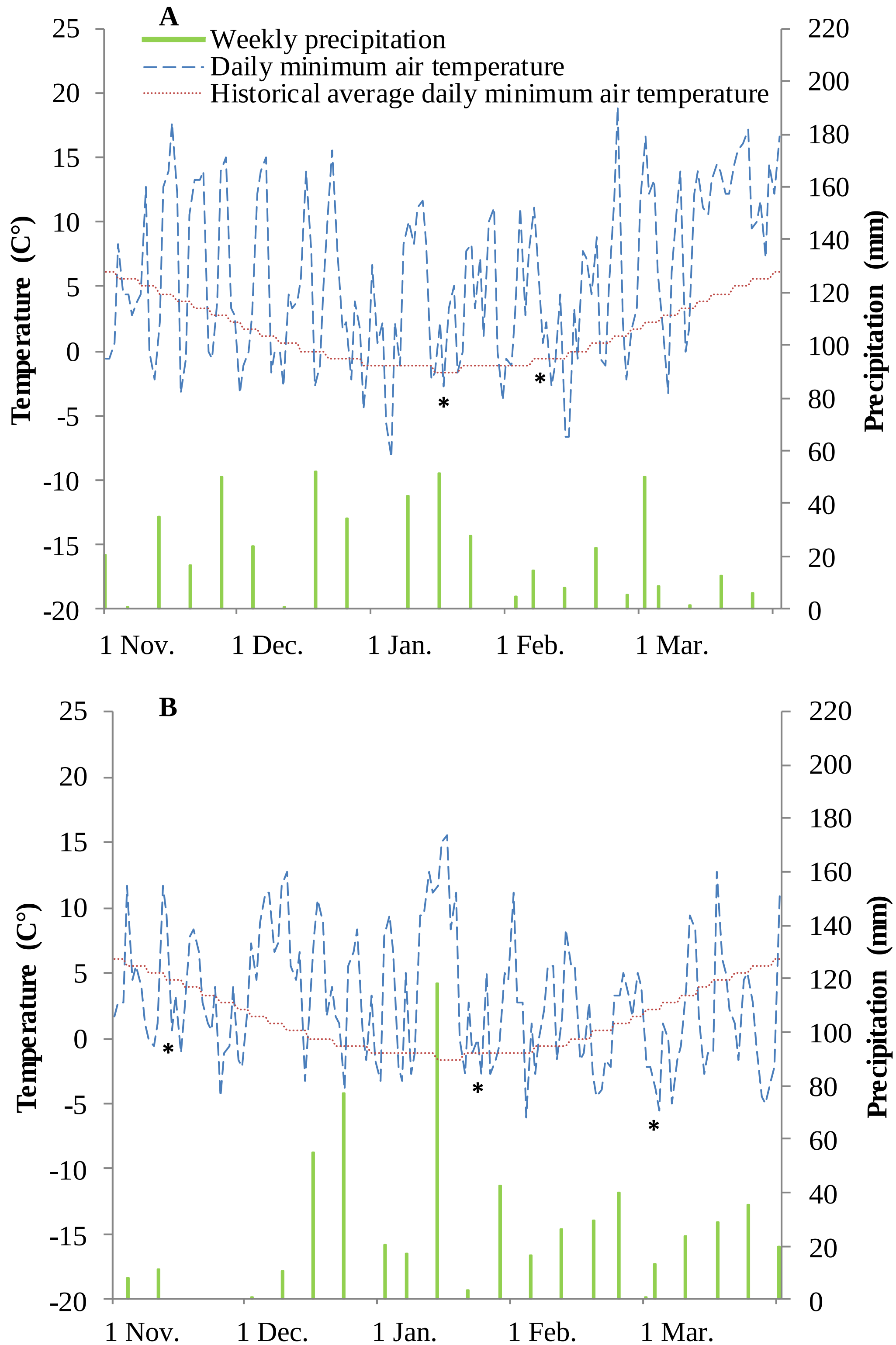

Fig. 7. Daily minimum air temperature, historical (1971-2000) average daily minimum air temperature, and weekly precipitation from (A) November 2011 to March 2012 and from (B) November 2012 to March 2013 for Clemson, South Carolina. Asterisks denote sampling dates of floral bud tissue from the field.

buds acclimated to lower temperatures and T_{50} values ranged from -18.5 to -19.6 °C. However, by February 2012, floral buds deacclimated and T_{50} values varied among rootstocks. Floral buds of trees on KV010-127 and HBOK 32 rootstocks were hardier than those on Krymsk 1, Controller 5, Mirobac, and Viking rootstocks at this test date.

Like the previous dormant period, T_{50} values of floral buds collected in Missouri were similar among rootstocks in November 2012 and January 2013. Although not statistically significant, 'Redhaven' buds on Mirobac trees had relatively high T_{50} values and Penta trees had low T_{50} values in November 2011 and 2012. Because T_{50} values only varied by 1.3 °C in January 2013 differences in floral buds hardiness among rootstocks were not detected. However, in March 2013, floral bud T_{50} values varied among rootstocks by 4.5 °C. 'Redhaven' floral buds on trees with Guardian rootstock were hardier than those on all other rootstocks except for Lovell, KV010-127, Krymsk 86, Bright's Hybrid #5, and Penta. Floral buds on trees with Controller 5 and Mirobac rootstocks generally had the highest T_{50} values.

For South Carolina, January 2012 was the only test date in which T_{50} values were significantly different (Table 3). At this date, 'Redhaven' floral buds on trees with Lovell and Viking were hardier than those on Bright's Hybrid #5, Controller 5, and Mirobac rootstocks. Floral bud T_{50} values on trees of all rootstocks ranged from -14.9 to -17.1 °C at this time. By February 2012, floral buds deacclimated and were about 5 to 7 days from budbreak. At this date, T_{50} values for buds on trees with the different rootstock varied by only 2.4 °C. Although T_{50} values were not statistically different in February 2012, floral buds on trees with *P. americana* and HBOK 32 rootstocks were generally the most cold tolerant and those on Mirobac were the most susceptible to cold injury.

Table 2. Mean T_{50} values of 'Redhaven' peach floral buds on various rootstocks sampled from Missouri at selected dates.[z]

	T_{50} value (˚C)					
Rootstock	11 Nov. 2011[y]	25 Jan. 2012	28 Feb. 2012	12 Nov. 2012	18 Jan. 2013	13 Mar. 2013
Lovell	-12.0	-19.4	-14.0 abcd	-15.4	-19.2	-14.0 ab
Guardian	-11.3	-18.8	-14.2 abc	-15.9	-19.0	-15.3 a
KV010-123	-12.1	-19.3	-14.1 abcd	-16.1	-19.0	-12.3 bc
KV010-127	-11.5	-19.2	-14.9 ab	-15.7	-18.6	-13.5 ab
Krymsk 1	-12.2	-19.3	-12.8 d	-15.2	-18.6	-12.8 bc
Krymsk 86	-11.5	-19.0	-13.6 bcd	-15.1	-18.9	-13.6 ab
Bright's Hybrid #5	---	-18.6	-14.5 abc	-16.6	-19.6	-13.8 ab
Controller 5	-11.5	-19.6	-13.5 cd	-15.1	-18.3	-10.8 c
HBOK 32	---	-18.9	-15.0 a	-16.4	-19.1	-12.7 bc
Mirobac	-10.9	-19.2	-13.3 cd	-14.5	-19.5	-11.2 c
Penta	-13.1	-18.5	-13.9 abcd	-16.4	-18.6	-13.4 ab
Viking	-12.7	-18.7	-13.2 cd	-15.4	-18.5	-12.3 bc

[z] Means represent 6 replications of each 5-node cutting for each rootstock. Mean separation within columns by Fisher's protected LSD test ($P \leq 0.05$).
[y] Bud samples not collected on this date.

Table 3. Mean T_{50} values of 'Redhaven' peach floral buds on various rootstocks sampled from South Carolina at selected dates.[z]

	T_{50} value (˚C)				
Rootstock	16 Jan. 2012	8 Feb. 2012	14 Nov. 2012	23 Jan. 2013	4 Mar. 2013
Lovell	-17.1 a	-11.2	-10.0	-13.2	-9.3
Guardian	-16.3 ab	-10.0	-10.1	-12.6	-10.0
KV010-123	-16.2 ab	-10.7	-11.4	-11.8	-9.1
KV010-127	-16.0 abc	-10.1	-10.6	-11.4	-10.9
Krymsk 1	-16.1 ab	-11.5	-10.5	-13.3	-9.3
Krymsk 86	-16.6 ab	-10.3	-10.3	-11.7	-10.1
Bright's Hybrid #5	-15.8 bc	-10.7	-11.8	-12.2	-10.0
Controller 5	-15.5 bc	-10.2	-11.0	-12.5	-9.2
HBOK 32	-16.5 ab	-12.0	-11.0	-12.9	-9.8
Mirobac	-14.9 c	-9.9	-11.2	-11.6	-9.2
Penta	-16.4 ab	-10.2	-10.9	-12.1	-9.2
Viking	-17.0 a	-11.4	-12.4	-12.0	-10.3
P. americana	-16.1 ab	-12.3	-10.4	-12.0	-9.8

[z] Means represent 6 replications of each 5-node cutting for each rootstock. Mean separation within columns by Fisher's protected LSD test ($P \leq 0.05$).

In the second dormant season in South Carolina, there was little floral bud acclimation from November 2012 to January 2013. Although differences were not significant, trees with Viking rootstock generally had lower floral bud T_{50} values than those with all other rootstocks in November 2012 and March 2013. In January 2013, floral buds on trees with Lovell and Krymsk 1 rootstocks were among the most cold tolerant and those on KV010-127 rootstocks were injured at the warmest temperatures. By 11 Mar. 2013, floral bud T_{50} values for rootstocks were only 0.5 to 3.9 °C warmer than those recorded in January for the same rootstocks, even though buds were about 5 to 7 days from calyx red stage of growth. Buds on trees with KV010-127 rootstocks deacclimated the fewest degrees from January to March, whereas those on trees with Lovell rootstock had the greatest change in hardiness.

When data were pooled to compare the scion floral bud T_{50} values of the 12 rootstocks common to both locations, rootstocks and the state x collection date interaction were statistically significant, but all other interactions were not. Floral bud T_{50} values on trees with Lovell, Guardian, Bright's Hybrid #5, and HBOK 32 rootstocks were hardier than those on trees with Controller 5 and Mirobac rootstocks across all test dates (Table 4). Mean T_{50} values were always lower in Missouri than in South Carolina at similar collection periods. As expected, the lowest mean floral bud T_{50} values were recorded in Missouri in January 2012 and 2013 (Table 5). T_{50} values among all other dates and locations differed except for South Carolina in February and November 2012.

Table 4. Mean T_{50} values of ‘Redhaven’ peach floral buds on various rootstocks sampled from Missouri and South Carolina across all collection dates.[z]

Rootstock	T_{50} value (˚C)[y]
Lovell	-14.3 a
Guardian	-14.2 a
KV010-123	-14.0 ab
KV010-127	-14.1 ab
Krymsk 1	-13.9 abc
Krymsk 86	-13.9 abc
Bright's Hybrid #5	-14.4 a
Controller 5	-13.6 bc
HBOK 32	-14.4 a
Mirobac	-13.5 c
Penta	-14.0 abc
Viking	-14.1 ab

[z] Sample dates included January 2012 to March 2013.
[y] Mean separation within columns by Fisher's protected LSD test ($P \leq 0.05$).

Table 5. Mean T_{50} value of 'Redhaven' peach floral buds on various rootstocks sampled from Missouri and South Carolina at each sampling date.

State	Date	T_{50} (˚C)[z]
Missouri	January 2012	-19.0 a
Missouri	January 2013	-18.9 a
South Carolina	January 2012	-16.2 b
Missouri	November 2012	-15.6 c
Missouri	February 2012	-13.9 d
Missouri	March 2013	-13.0 e
South Carolina	January 2013	-12.3 f
South Carolina	February 2012	-10.6 g
South Carolina	November 2012	-10.9 g
South Carolina	March 2013	-9.7 h

[z] Mean separation within columns by Fisher's protected LSD test ($P \leq 0.05$).

Chapter 6: Discussion

During the fall, plants acclimate in response to cool temperatures and shortening daylengths. The response to these signals is gradual and determines low temperature tolerance. The lowest temperature at which 50% of peach buds are killed (T_{50}) varies with the length of exposure to temperatures ≤ -2.2 °C and can vary among dormant seasons at the same location (Proebsting and Mills, 1966). The rate at which buds gain cold tolerance is 1 to 2 °C per day when temperatures are below this threshold for 24 hours (Proebsting, 1970).

Peach buds also have minimum hardiness level (MHL) during late fall and early winter, which Proebsting and Mills (1972) also described as a T_{50} value. When buds are exposed to temperatures above freezing, the T_{50} value does not rise above the MHL, until after rest completion when loss of hardiness can occur rapidly. In Washington, Proebsting and Mills (1972) reported Elberta peach had an MHL of -21.1 °C during late fall and winter. In the present study, maximum T_{50} values of 'Redhaven' buds never reached the MHL previously reported for Elberta due to the lack of cold temperatures (Figs. 6 and 7). However, in South Carolina, T_{50} values in January 2013 may have been near the MHL because buds sampled at this time acclimated only a few degrees when compared to the November 2012 sampling date. Also, T_{50} values sampled just before budbreak (4 Mar. 2013) were only slightly warmer than those of buds tested in January 2013.

'Redhaven' floral buds sampled at similar phenological stages were hardier in Missouri than in South Carolina (Tables 2 and 3). Such results are not unexpected since

New Franklin, Missouri is located in the USDA hardiness zone 6a, where the average annual extreme minimum temperatures range from -20.6 to -23.3 ˚C (USDA-ARS, 2012). In contrast, the South Carolina NC-140 trial is located in the hardiness zone 8a, with average annual extreme minimum temperatures from -9.4 and -12.2 ˚C (USDA-ARS, 2012). In spite of the higher T_{50} values in South Carolina in January 2012, differences in floral bud hardiness among rootstocks were detected, but not in Missouri. January bud T_{50} values varied among rootstocks in South Carolina by 2.2 °C, but only differed by 1.1 °C in Missouri at this time. In contrast, statistical differences in floral bud T_{50} values among rootstocks were consistently detected just before budbreak in Missouri in both years of evaluation (Table 2). The reason for different times of discrimination of floral bud hardiness among rootstocks at the two locations is likely due to minimum temperatures at each site, which may have affected dates of rest completion and rates of deacclimation thereafter (Layne, 1982; Proebsting, 1963; Warmund et al., 2002).

When T_{50} values for both locations and all sampling dates were pooled, four rootstocks, including Lovell, Guardian, Bright's Hybrid #5, and HBOK 32, conferred greater floral bud hardiness than Controller 5 and Mirobac rootstocks (Table 4). All of the rootstocks that produced cold hardy buds were derived from peach, except for Bright's Hybrid #5. This rootstock, which is derived from Titan almond x Nemaguard peach has had 100% tree survival in South Carolina, but only 38% survival in Missouri (M. Warmund, unpublished data). While the reason for this high mortality rate has not been identified, Bright's Hybrid #5 is apparently unsuitable as a rootstock for 'Redhaven' peach at the New Franklin, Missouri site. At both test sites, Controller 5, a Japanese plum x peach hybrid, and Mirobac, a Myrobolian plum x almond cross, adversely affected peach bud hardiness, but had little or no tree loss (M. Warmund, unpublished data). Controller 5 originated from California and was released as a size controlling rootstock (DeJong et al., 1993). Mirobac was selected in Spain for its resistance to high pH soils, root-knot nematodes, and root fungus (*Rosellinia necatrix*) (Pinochet, 2010). Thus, these rootstocks originate from locations that are seldom challenged by winter low temperatures, which may explain the high floral bud T_{50} values of grafted trees found in the present study.

'Redhaven' floral buds on trees with Lovell rootstock were consistently hardy among all dates at both locations in this study (Table 4). The lowest floral bud T_{50} values on trees with Lovell rootstock in Missouri (-19.4 ˚C) and South Carolina (-17.1 ˚C) occurred during January 2012. In January 2013, the average floral bud T_{50} value on trees with Lovell rootstock in South Carolina was only -13.2 °C. These values are warmer than those reported in an earlier study by Warmund et al. (2002) at the same locations. In similar freezing tests conducted in January 1999 and 1997, 'Redhaven' floral buds on Lovell rootstock had T_{50} values of -19.3 and -21.3 °C in Missouri and South Carolina, respectively. Szalay et al. (2010) also reported that 'Redhaven' grafted onto a seedling rootstock (C2630) had a T_{50} value of -23 °C. Because freezing regimes, methods of calculating T_{50} values, and rootstocks varied in these studies, direct comparisons of the maximum hardiness of 'Redhaven' floral buds cannot be determined. However, T_{50} values reported in the present study clearly do not reflect the maximum hardening capacity of 'Redhaven' floral buds on Lovell rootstock because of the unseasonably warm winter temperatures during the 2012 and 2013 dormant seasons in Missouri and South Carolina. However, when compared to other rootstocks in this study, Lovell rootstock positively influenced ‘Redhaven’ floral bud hardiness. It is also the most widely planted rootstock for peach trees in Missouri due to its consistent performance (M. Warmund, personal communication). In South Carolina, Guardian and Lovell are recommended rootstocks, but Guardian has better tolerance to peach tree short life syndrome and is resistant to most races of *Meloidogyne incognita* and *M. javanica* root-knot nematodes (Horton and Johnson, 2011).

Warm temperatures recorded during this study reflect climate change occurring in the contiguous U.S. From 1912 to 1969, winter (December to February) minimum temperatures increased at a rate of 0.07 °C per decade, but from 1970 to 2013, the rate was 0.33 °C in 47 states within the contiguous U.S. (Tebaldi et al., 2013). Missouri’s long-term (1895 to 2010) average minimum temperature from December through February is 0.2 °C and has been warming at a rate of 0.42 ˚C per decade since 1970 (Guinan, 2013; Climate Central, 2013). The average winter minimum temperature (1981 to 2010) in New Franklin, Missouri was -6.4 ˚C (NCDC, 2010b). From December 2011 to February 2013, the average winter temperature for the NC-140 test site was -2.3 °C.

In South Carolina, the average winter (December to February) minimum temperature from 1981 to 2010 is 1.1 °C and is warming at a rate of 0.26 ˚C per decade since 1970 (NCDC, 2010a; Climate Central, 2013). Clemson, South Carolina has a historical (1971 to 2000) average winter minimum temperature of -0.3 °C, but the average winter minimum temperature during this study was 3.6 ˚C (data not shown). Carbone and Schwartz (1993) predicted that with an increase of 2 ˚C to both the daily maximum and minimum winter temperatures (1961 to 1990), chilling hours during the dormant period would be reduced by approximately 400 hours. Although peach cultivars generally require 50 to 1200 chilling hours, 'Redhaven' requires 950 hours (Childers et al., 1995). Thus, with future climate change, it may become necessary to plant cultivars with lower chilling requirements than 'Redhaven'. However, once the rest requirement is satisfied, low-chilling peach cultivars can bloom at an early date, which increases the likelihood of floral mortality from sub-freezing temperatures. Therefore, rootstocks that confer cold tolerance to floral buds will be needed for sustainable peach production.

SUMMARY AND CONCLUSIONS

Floral bud hardiness of ‘Redhaven’ peach trees included in the 2009 NC-140 rootstock trial were evaluated at New Franklin, Missouri and Clemson, South Carolina during two dormant seasons. Although temperatures during this study were generally higher than historical average minimum temperatures, floral bud cold tolerance differed among the 12 rootstocks tested at each location. In Missouri, ‘Redhaven’ floral buds on trees with KV010-127 and HBOK 32 rootstocks were more cold tolerant than those on Krymsk 1, Controller 5, and Viking rootstocks when evaluated on 28 Feb. 2012. However, when buds were tested on 13 Mar. 2013 in Missouri, floral buds on trees with Guardian rootstocks were hardier than those on all other rootstocks except for Lovell, KV010-127, Krymsk 86, Bright’s Hybrid #5, and Penta. Thus, ‘Redhaven’ floral bud hardiness was discriminated among rootstocks about a week before bloom in Missouri. In contrast, differences in floral bud hardiness among rootstocks were detected in South Carolina on 16 Jan. 2012. At this date, floral buds on Lovell and Viking rootstocks were more cold tolerant than those on Bright’s Hybrid #5, Controller 5, and Mirobac rootstocks.

When data from similar rootstocks and collection periods were pooled from both locations, mean floral bud T_{50} values were always lower in Missouri than in South Carolina at similar sampling periods. Also, ‘Redhaven’ floral buds on trees budded to Lovell, Guardian, Bright’s Hybrid #5, and HBOK 32 rootstocks were most cold tolerant. These five rootstocks are derived from peach, except for Bright’s Hybrid #5 (almond x peach), which has had poor tree survival in Missouri. ‘Redhaven’ floral buds on

Controller 5 and Mirobac rootstocks were the least cold tolerant at both locations. These rootstocks originated from locations that are seldom exposed to low winter temperatures.

Site-specific floral bud hardiness evaluations from this study will be an important factor in determining local rootstock recommendations when combined with long-term yield and survival data. Additionally, bud hardiness results from differing locations may also be used by nurseries that propagate rootstocks for peach producers across the U.S. with varying minimum winter temperatures.

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