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Freezing tolerance of apple flower buds

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ABSTRACT

Flower buds are the most vulnerable parts of the dormant apple trees to frost injury. Especially during fall and spring when the processes associated with the transitional periods of hardening and de-hardening occur, tissues are more active and thus less cold hardy. The critical temperatures affecting the flower bud and the associated damage are variable and unknown for most of the current apple cultivars. The goal of this study was to identify the critical temperatures of flower buds of apple during different developmental stages from dormant to full bloom. Differential Thermal Analysis (DTA) and a controlled freezing exposure method were used to test the freezing tolerance of flower buds for three cultivars including Fuji, Gala, and Red Delicious during the late winter and early spring season of 2012, and from late fall 2013 until the early spring season of 2014. Differences in hardiness were found for the same cultivar as well as among cultivars and for different stages of flower bud development. The temperature at which the flower buds become injured using the DTA method is commonly related to the initiation of the low temperature exotherm (LTE). However, for flower buds of apples only the high temperature exotherms (HTEs) were observed within the evaluated temperature range of −44 °C to 4 °C. The inability to detect LTEs in apple buds indicates that apple flower bud hardiness cannot be estimated using the DTA method. A traditional freezing exposure method was used to determine the freezing tolerance of apples. The information generated in this study will provide a better understanding of the flower bud cold hardiness that is important for long term tree survival and orchard sustainability.

1. Introduction

Freezing tolerance is an important feature for plant survival during critical periods. It defines the lethal temperature at which freezing injury occurs (Gray et al., 1997). Cold injury can occur when the temperature is below 0 °C. Freezing injury occurs due to ice formation inside the plant tissue and the ice crystals then damage the cell membrane systems (Westwood, 1978). Species and varieties can exhibit different damage at the same temperature and phenological stage, depending on previous weather conditions, and their hardiness to the cold temperatures prior to a frost night (Levitt, 1980; Sakai and Larcher, 1987; Li, 1989; Lenz et al., 2013).

Freezes can damage fruit buds and young fruits of apple and the level of damage is directly related to the intensity and duration of the freeze as well as the developmental stage of the bud during which the freeze occurred (Proebsting and Mills, 1978b; Iezzoni, 1985; Westwood, 1993; Larsen, 2010). If the drop in temperature is gradual, trees are in better condition to resist injury and can stand surprisingly. However, a sudden the temperature drops a rapid freeze occur a prolonged freeze can kill or damage the tree. Temperatures that go from warm to extreme cold all at once are the most injurious (Rodrigo, 2000). The resistance of apple tree to freezing temperatures is variable, low temperatures resistance of flower and buds change rapidly in response to both temperature and the stage of floral development (Mexal et al., 1991; Rodrigo, 2000; Caprio and Quamme, 2005; Longstroth, 2013; Salazar-Gutierrez et al., 2014).

As apple trees begin to acclimate during the fall and deacclimate during late winter and early spring, buds become more sensitive and lose the ability to withstand cold temperatures (Howell and Weiser, 1970a; Longstroth and Perry, 1996). The response to freezing will depend on the location of the tree where the growth is taking place (Longstroth, 2013). Apple stem xylem ray parenchyma and pith cells avoid freezing by deep supercooling, whereas bark tissue and flower buds are unable to deep supercool (Palonen and Buszard, 1997). The most vulnerable parts of a dormant apple trees to frost injury are the flower buds. Temperatures that drop below freezing can cause significant damage to these buds, even prior to flowering and as early as late fall. Apple and pear buds are rarely injured during dormancy, damage can happen especially for buds approaching to full bloom if temperature

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drops suddenly and extreme low temperatures occur, flower buds de-hardened were able to re-harden in response to low temperatures (Proebsting 1963,1982). However, low temperatures during and after bud break not only injure buds but also flowers, developing fruits and even shoots (Smeeton, 1964; Rodrigo, 2000; Atkinson et al., 2013). At the more advanced stages of bud break, the buds and small flowers become increasingly susceptible to frost (Rodrigo, 2000).

Cold resistance or cold hardiness is the ability of a plant to adapt to and withstand freezing temperatures (Gusta and Wisniewski, 2013; Wisniewski and Gusta, 2014). Different types of plant tissues as buds and twigs respond differently to temperatures extremes during their rest period in winter (Ketchie, 1985). Variation in tree hardiness during this period is related to fluctuating air temperatures (Coleman, 1985). During dormancy the effect of freezing temperatures on the flower buds is different and these buds can withstand very low temperatures but, after the rest period is satisfied the flower buds start to swell and develop, becoming more susceptible to weather conditions and during this period they can be easily damaged by cold or freezing temperatures. The flower buds that survive the rest period continue with its development until bloom. It is generally accepted that subfreezing temperatures could retard bud development in apples and buds during first bloom and pink bud appear to be rather more sensitive than those at full bloom (Proebsting and Mills, 1978a). Spring bloom in apple is part of the cycle of reproductive development that begins with floral initiation in the preceding summer (Abbott, 1970; Wilkie et al., 2008). At bloom, the first symptom that is observed after freezing is the thawing of the flowers characterized by a brown discoloration at the base of the style; depending on the severity of the freeze, the damage may extend both to the style and to the ovary, resulting in death of fruit abortion (Rodrigo, 2000; Aygun and San, 2005).

There is ample evidence that flower-bud formation in apple is affected by environmental conditions and can lead the trees to yield losses (Tromp, 1984; Mexal et al., 1991; Zhu et al., 1997). The year to year variability in hardiness is traditionally related to the temporal variability in weather conditions. Different trees within the orchard or different parts within the tree could be affected due to microclimate differences, or the type of freeze that occurs, some cultivars are more susceptible than others (Glozer, 2010).

One of the most common methodologies for estimating cold hardiness is the Differential Thermal Analysis (DTA), used to quantify cold tolerance in plants (Gerard and Schucany, 1997) the successful application of this method has been demonstrated in previous studies (Mills et al., 2006; Volk et al., 2009; Ferguson et al., 2011; Salazar-Gutierrez et al., 2014), as a result temperature thresholds for acclimation and deacclimation has been reported for a wide range of species including apricot (Ashworth et al., 1981), azalea (Graham and Mullin, 1976), blackberry (Warmund et al., 1988), grape (Andrews et al., 1984; Quamme, 1986,1991), peach (Proebsting and Sakai, 1979; Quamme, 1974), pear (Montano et al., 1987), plum, sweet and sour cherry (Quamme, 1974), and sweet cherry (Salazar-Gutierrez et al., 2014). However, DTA has limited applicability to determine the hardiness of other crops (Quamme, 1976). Historical cold hardiness data are available for several tree fruit crops, including peaches, apricots, sweet cherries, grapes and plums (Proebsting (1970), Proebsting et al. (1980), apples (Ketchie and Beeman, 1973) and pears (Ketchie (1985). Proebsting and Mills (1978a) established the critical temperatures for the various stages of flower development during late winter and early spring for "Bing" sweet cherry, while Salazar-Gutierrez et al. (2014) identified critical temperatures for "Bing", "Chelan" and "Sweetheart" sweet cherries using DTA for dormant flower buds and freezing exposure methods for non-dormant flower buds. Nevertheless, the sensitivity of apples to freezing conditions and associated critical temperatures were last studied more than 25 years ago by Ballard et al. (1987) and by Mexal et al. (1991) and, very little is known about the frost susceptibility of new apple cultivars. The fluctuation in cold resistance of apple tissue in relation to temperature has not been determined in much detail and the hardiness progression through winter dormancy and early spring for apples has not been very well characterized. New knowledge of the sensitivity of flower buds during different stages of development is important for cultivar improvement through breeding programs and to provide support for crop management and frost control strategies for growers. The purpose of this study was, therefore, to identify the critical temperatures of flower buds of apples during the different development stages from dormant to full bloom.

2. Materials and methods

2.1. Plant material and sampling

Three apple cultivars were evaluated, including "Fuji", "Gala", and "Red Delicious". The samples were taken from commercial orchards located in the Yakima Valley, WA. Flower buds were randomly collected throughout the later winter and early spring season of 2013, and from late fall 2013 to early spring of 2014 to determine the hardiness of different developmental stages of the flower bud for apples (Proebsting and Mills, 1978a). The flower bud stages were characterized based on the classification defined for apples by Chapman and Catlin (1976).

Dormant flower buds were collected every week from various positions within the tree with a sample size ranging from 200 to 500 spurs. Following collection the samples were placed in a container that had previously been cooled to air temperature and were transported to the laboratory. Proebsting and Mills (1971) found that it very important for the samples to remain as close to the current air temperature as possible when transporting them from the orchard to the laboratory for further analysis.

2.2. Freezing tolerance determination

2.2.1. Evaluation of flower bud damage

A sample of the flower buds for the three cultivars that were collected was kept separate as an unfrozen check for determining variability and dead buds that were present in the field. The remaining buds were then separated into two groups one of the groups was used with Differential Thermal Analysis and the other group of samples was used with the controlled freezing method.

Following exposure to freezing temperatures the flower buds were dissected using a sharp blade under a stereomicroscope that was equipped with a photographic system (Zeiss Stemi 2000C). They were then evaluated to determine the viability or mortality of the tissue. This determination was done by visual assessment (Salazar-Gutierrez et al., 2014; Embree and McRae, 1991) as soon as browning of injured tissue was observed (Proebsting and Mills, 1971). Pictures were taken of each sample and digitally processed with Zeiss photo software.

2.2.2. DTA analysis

DTA analysis was performed on one set of samples. The method used was similar as described by Salazar-Gutierrez et al. (2014). Each set of bud samples was comprised of 12 replicates per cultivar and a total of seven to ten buds were used for each replication. The samples were placed on four trays, with each tray consisting of nine thermoelectric modules (TEMs) that detect temperature gradients generated by the exotherms (Mills et al., 2006; Salazar-Gutierrez et al., 2014). Five to ten buds (depending on the size of the buds) were covered in aluminum foil and placed directly on each TEM that was protected with foam insulation pads. A chamber lid was tightened to the tray and then the tray was loaded