Cold hardiness assessment of peach flower buds using differential thermal analysis (DTA) in western Colorado (dormant season 2019-20)

David Sterle & Ioannis S. Minas*

Department of Horticulture & Landscape Architecture, Western Colorado Research Center at Orchard Mesa, Colorado State University, 3170 B 1/2 Rd, Grand Junction, CO 81503

Introduction

Cold hardiness is influenced by many different factors, including variety, crop load, harvest time and postharvest conditions, and orchard weather conditions. There is a genetically determined limit to cold hardiness. However, while this is true for mid-winter hardiness, the ranking might be different at the start or end of the dormant season. Some varieties will acclimate earlier in fall and will be able to withstand colder temperatures earlier in the dormant season than varieties that have otherwise more mid-winter hardiness. Likewise, early bud-breaking varieties tend to lose their hardiness earlier in spring and might be damaged at warmer temperatures than late-breaking varieties, irrespective of their mid-winter hardiness. Also, cultural practices can have a profound influence if the genetic potential of a given cultivar is achieved. In very general terms, warm temperatures tend to reduce bud hardiness while cold temperatures tend to induce more hardiness (within limits). Hence, the weather conditions at a site will influence the ability of buds to withstand cold temperature, and the values presented in **Table 1** are in part affected by the temperature conditions at that site.

The standard cold injury assessment process is the oxidative browning method. After freezing, the buds are held at room temperature for 24 h. Following the 24-h incubation on each bud a cross and/or longitudinal sectioning is made with a single-edged razor blade to confirm the injury of the tissue manifested by brown color of the ovary (or, in the case of multi-flower buds such as in cherry, ovaries). Buds showing vibrant green tissue were judged to be viable whereas buds showing brown tissue were judged to be dead (**Figure 1 and 2**). The brown coloration is the result of oxidation of the phenolic compounds being released in the damaged tissues. Severe damage results in more pronounced, deeper browning of damaged or killed tissues. Less severe damage may produce slightly browned tissues. Shoots from the orchard need to be held for a minimum of 24 hours at 70 ºF (room temperature, 21 ºC) before cutting to maximize pistil browning. **Figure 1** shows live and dead buds of Berenda Sun and Cresthaven peach, respectively, where the dead buds are from shoots placed in a chest freezer (approx. -10 ºF or -23.3 ºC) for 50 minutes. **Figure 2** shows a bud cluster from Bing sweet cherry collected at the Western Colorado Research Center - Rogers Mesa the morning of 30 November 2006 after an overnight low of -9.9 ºF (-23.3 ºC); some

*Corresponding author: ioannis.minas@colostate.edu Update #21: 2/14/20

of the ovaries within some buds are dead while others survived.

Differential thermal analysis is a technique used to quantify cold tolerance in plants, freezing episodes called exotherms can be identified as change points, local minima or selected infection points of differential temperature [1]. When super cooled water freezes extracellularly, the heat released is referred to as a high-temperature exotherm (HTE); extracellular freezing is considered nonlethal. On the other hand, the freezing of intracellular water creates a similar, low-temperature exotherm (LTE) and is lethal [2] (**Figure 3**).

Method

Four peach cultivars including 'Suncrest', 'Sierra Rich', 'Cresthaven' and 'Red Haven' grafted on Lovell rootstock were tested. Dormant buds were randomly collected beginning in early-November of 2018. Buds were collected weekly from shoots of moderate vigor that had no obvious signs of damage, from five similar trees for each cultivar. The sample size ranged from 60 to 80 buds per cultivar. The samples were taken from the experimental orchard of 10-year old trees located at the Colorado State University Western Colorado Research Center at Orchard Mesa, CO. In addition to the above experiment 'Red Haven' dormant buds were sampled at different time points during the dormant season from different peach production areas of western CO to compare cold hardiness status of Palisade, Rogers Mesa, Cedaredge, Olathe with the regularly tested status of peach hardiness in Orchard Mesa. When samples were collected, they were placed in a container that had been previously cooled to the local air temperature. The buds were then separated and randomly assigned to 6 to 8 sets of 10 buds per cultivar (4 sets of 10 buds for the 2016 cultivar collection). One complete set of each cultivar was kept as a control and was not frozen for visual evaluation of oxidative browning to check the variability and dead material that was present in the orchard. The remaining 5 to 7 sets were then used for the differential thermal analysis (DTA). The samples were placed on three trays; each tray included eleven thermoelectric modules (TEMs) that detect temperature gradients generated by the exotherms according to the methodology described by Mills et al. (2006) [3]. Up to ten buds (depending on the size of the bud) were covered in aluminum foil and placed directly on each TEM protected by foam insulation pads. A chamber lid was tightened to the tray and then loaded into a programmable freezer (Tenney Jr Test Chamber, Model TUJR 1.22 cu.ft., Watlow F4, Temperature range: −75 ºC to 200 ºC with a resolution of 0.3 ºC, Thermal Product Solutions). The freezer was programmed for the standard cooling rate of 4 ºC/h decline which means that the temperature was held at 4 ºC for 1h and then dropped to −40 ºC in 11h, then returned to 4 ºC in 10 h, and a DTA analysis was performed. Thirty TEMs were loaded per run (150 to 180 buds). The system recorded for each TEM a voltage signal that corresponds to the temperature at which super cooled water presumably in the bud tissue freezes. The signals were sent to an output directly to an Excel spreadsheet. Exotherms were identified plotting the TEM signals (mVolts) against the temperature (ºC) (**Figure 3**). Bud exotherm output

from the DTA system was also compared with tissue browning (indicating tissue death) following the methodology described above.

Figure 1. Cresthaven peach flower buds oxidative browning symptoms due to cold damage. Left: live (L, green) fruit pistils; right: dead (D, brown) fruit pistils.

Figure 2. Bing cherry flower buds oxidative browning symptoms due to cold damage. Left: multiple-flower bud with three live pistils (L); right: multiple-flower bud with two live pistils and one dead pistil (D).

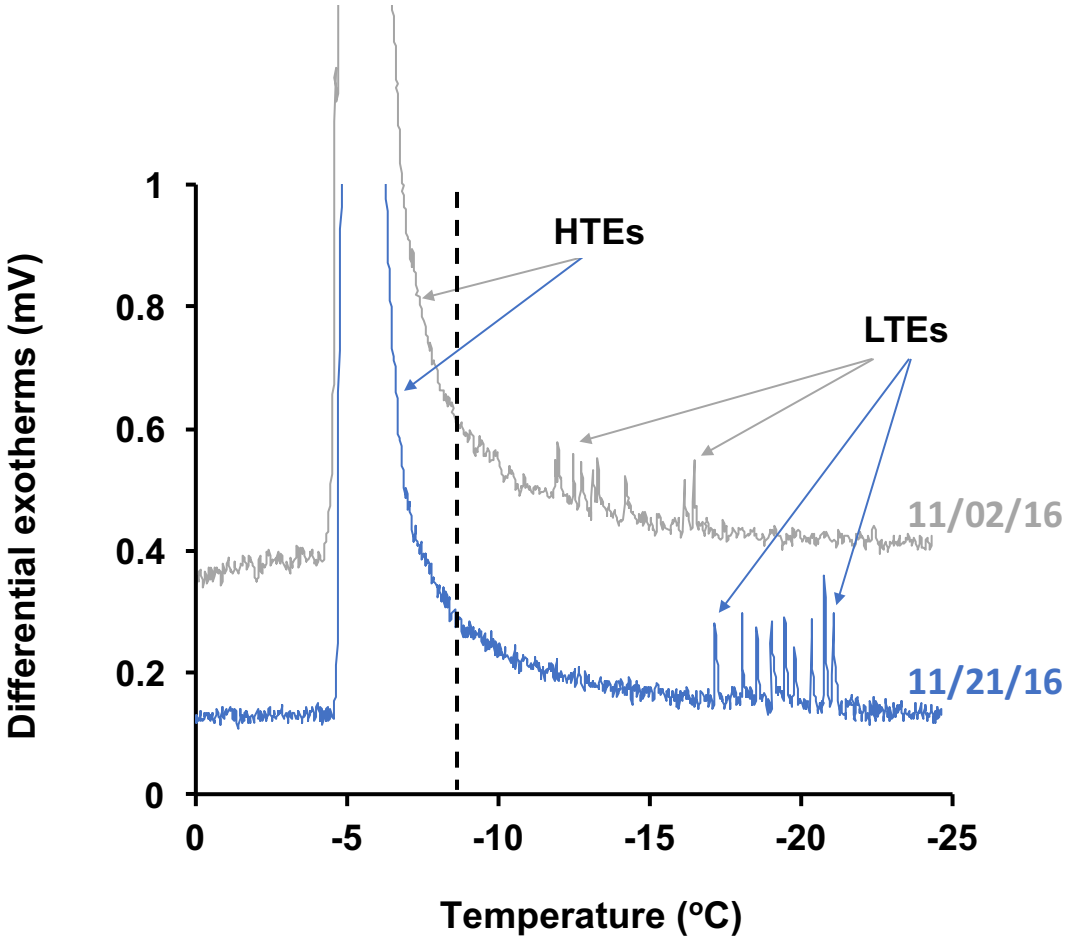

Figure 3. Differences in low temperature exotherms (LTE) for 'Red Haven' flower buds coming from trees growing at the experimental orchard at the CSU Western Colorado Research Center at Orchard Mesa near Grand Junction, CO, on November 2, 2016, and November 21, 2016. High temperature exotherms (HTEs), indicating non-lethal extracellular freezing of extracellular water, are shown to the left of the dashed vertical black line (between -5 and -8 °C). The LTEs for the two dates are shown to the right of the dashed vertical black line (below -10 °C), indicating acclimation in bud hardiness for 'Red Haven'.

Results

Table 1. Lethal temperatures (LT) in Celsius (ºC) and Fahrenheit (ºF) for 10 (LT_{10}), 50 (LT_{50}) and 90% (LT_{90}) flower buds killed, for 'Suncrest', 'Sierra Rich', 'Cresthaven' and 'Red Haven' peach cultivars grown in the experimental orchard of the Colorado State University's Western Colorado Research Center at Orchard Mesa near Grand Junction, Colorado. Most recent update in red.

		ºC			ºF		
Date	**Cultivar**	**LT_{10}**	**LT_{50}**	**LT_{90}**	**LT_{10}**	**LT_{50}**	**LT_{90}**
10/18/19	Suncrest	-14.8	-17.9	-20.3	5.4	-0.3	-4.6
10/18/19	Sierra Rich	.	.	.	.	.	.
10/18/19	Cresthaven	-16.9	-20.0	-22.2	1.5	-4.0	-8.0
10/18/19	Red Haven	-15.8	-19.2	-21.7	3.6	-2.5	-7.0
10/28/19	Suncrest	-18.5	-21.8	-24.3	-1.2	-7.3	-11.8
10/28/19	Sierra Rich	-15.0	-19.2	-22.4	5.0	-2.5	-8.3
10/28/19	Cresthaven	-18.8	-21.2	-22.8	-1.8	-6.1	-9.1
10/28/19	Red Haven	-17.3	-20.7	-23.3	0.9	-5.3	-10.0
11/1/19	Suncrest	-19.1	-22.6	-25.2	-2.3	-8.7	-13.4
11/1/19	Sierra Rich	-17.1	-20.9	-23.8	1.1	-5.7	-10.8
11/1/19	Cresthaven	-21.5	-23.2	-24.5	-6.7	-9.8	-12.0
11/1/19	Red Haven	-18.8	-22.1	-24.4	-1.9	-7.7	-11.9
11/8/19	Suncrest	-18.1	-20.4	-22.0	-0.6	-4.7	-7.6
11/8/19	Sierra Rich	-16.7	-19.2	-21.1	2.0	-2.6	-5.9
11/8/19	Cresthaven	-16.2	-19.4	-21.7	2.8	-2.9	-7.0
11/8/19	Red Haven	-16.9	-19.7	-21.7	1.6	-3.4	-7.1
11/15/19	Suncrest	-18.3	-20.6	-22.3	-1.0	-5.2	-8.1
11/15/19	Sierra Rich	-18.2	-20.6	-22.3	-0.8	-5.2	-8.2
11/15/19	Cresthaven	-18.3	-20.8	-22.6	-0.9	-5.5	-8.7
11/15/19	Red Haven	-16.6	-20.0	-22.5	2.1	-4.1	-8.6
11/21/19	Suncrest	-17.4	-20.7	-23.0	0.6	-5.2	-9.4
11/21/19	Sierra Rich	-16.7	-19.4	-21.4	2.0	-2.9	-6.4
11/21/19	Cresthaven	-18.3	-20.8	-22.6	-0.9	-5.5	-8.7
11/21/19	Red Haven	-17.7	-20.1	-21.7	0.1	-4.1	-7.1
11/29/19	Suncrest	-18.9	-21.0	-22.5	-2.0	-5.8	-8.4
11/29/19	Sierra Rich	-17.8	-20.3	-22.1	0.0	-4.6	-7.8
11/29/19	Cresthaven	-17.9	-20.7	-22.7	-0.3	-5.2	-8.8
11/29/19	Red Haven	-19.6	-21.6	-22.9	-3.4	-6.9	-9.3
12/10/19	Suncrest	-17.5	-21.0	-23.6	0.5	-5.8	-10.4
12/10/19	Sierra Rich	-16.6	-20.0	-22.5	2.2	-3.9	-8.5
12/10/19	Cresthaven	-19.1	-21.3	-22.9	-2.4	-6.4	-9.2

12/10/19	Red Haven	-18.4	-21.1	-23.0	-1.2	-6.0	-9.5
12/16/19	Suncrest	-19.6	-22.3	-24.2	-3.2	-8.1	-11.5
12/16/19	Sierra Rich	-16.9	-20.4	-23.1	1.5	-4.8	-9.5
12/16/19	Cresthaven	-20.1	-22.2	-23.6	-4.2	-7.9	-10.5
12/16/19	Red Haven	-20.4	-22.3	-23.6	-4.6	-8.1	-10.5
12/17/19	Suncrest	-20.7	-22.7	-24.0	-5.3	-8.8	-11.3
12/17/19	Sierra Rich	-20.0	-22.3	-23.9	-4.1	-8.2	-11.0
12/17/19	Cresthaven	-21.7	-23.4	-24.5	-7.0	-10.1	-12.1
12/17/19	Red Haven	-20.8	-22.8	-24.2	-5.5	-9.1	-11.6
12/19/19	Suncrest	-20.4	-22.9	-24.7	-4.6	-9.3	-12.5
12/19/19	Sierra Rich	-20.2	-22.9	-24.8	-4.3	-9.2	-12.7
12/19/19	Cresthaven	-22.0	-23.8	-24.9	-7.7	-10.8	-12.9
12/19/19	Red Haven	-20.7	-23.2	-25.0	-5.3	-9.8	-12.9
12/27/19	Suncrest	-19.9	-22.7	-24.7	-3.8	-8.8	-12.4
12/27/19	Sierra Rich	-19.2	-21.7	-23.4	-2.6	-7.1	-10.2
12/27/19	Cresthaven	-21.0	-22.6	-23.6	-5.8	-8.6	-10.5
12/27/19	Red Haven	-20.3	-22.4	-23.8	-4.6	-8.3	-10.8
12/30/19	Suncrest	-20.8	-23.3	-25.0	-5.5	-9.9	-12.9
12/30/19	Sierra Rich	-19.9	-22.5	-24.3	-3.9	-8.5	-11.8
12/30/19	Cresthaven	-21.6	-23.5	-24.8	-6.8	-10.3	-12.7
12/30/19	Red Haven	-20.3	-23.0	-24.9	-4.6	-9.4	-12.8
1/7/20	Suncrest	-20.4	-23.3	-25.3	-4.7	-9.9	-13.6
1/7/20	Sierra Rich	-19.4	-23.1	-25.8	-2.9	-9.6	-14.5
1/7/20	Cresthaven	-22.1	-24.1	-25.4	-7.8	-11.4	-13.8
1/7/20	Red Haven	-20.6	-23.7	-25.9	-5.2	-10.7	-14.7
1/9/20	Suncrest	-21.0	-23.9	-26.0	-5.8	-11.0	-14.7
1/9/20	Sierra Rich	-19.0	-22.8	-25.6	-2.3	-9.1	-14.1
1/9/20	Cresthaven	-22.4	-24.3	-25.5	-8.3	-11.7	-14.0
1/9/20	Red Haven	-21.4	-24.0	-25.8	-6.6	-11.2	-14.4
1/13/20	Suncrest	-20.5	-23.6	-25.8	-5.0	-10.5	-14.5
1/13/20	Sierra Rich	-20.5	-23.6	-25.7	-5.0	-10.4	-14.3
1/13/20	Cresthaven	-23.2	-24.8	-25.9	-9.7	-12.7	-14.7
1/13/20	Red Haven	-21.1	-24.0	-26.1	-5.9	-11.2	-15.0
1/24/20	Suncrest	-19.7	-22.0	-23.6	-3.5	-7.6	-10.5
1/24/20	Sierra Rich	-20.0	-22.0	-23.3	-4.0	-7.6	-10.0
1/24/20	Cresthaven	-21.1	-22.4	-23.3	-5.9	-8.3	-10.0
1/24/20	Red Haven	-20.7	-22.4	-23.6	-5.2	-8.3	-10.4
1/27/20	Suncrest	-21.0	-22.5	-23.6	-5.7	-8.6	-10.5
1/27/20	Sierra Rich	-18.5	-21.5	-23.6	-1.3	-6.6	-10.4

1/27/20	Cresthaven	-21.5	-22.7	-23.5	-6.6	-8.8	-10.3
1/27/20	Red Haven	-21.1	-22.6	-23.6	-5.9	-8.7	-10.5
2/4/20	Suncrest	-20.4	-23.2	-25.3	-4.7	-9.8	-13.5
2/4/20	Sierra Rich	-18.0	-21.3	-23.8	-0.3	-6.4	-10.9
2/4/20	Cresthaven	-20.7	-22.5	-23.8	-5.3	-8.5	-10.8
2/4/20	Red Haven	-20.3	-22.4	-23.9	-4.6	-8.4	-11.0
2/6/20	Suncrest	-19.6	-22.4	-24.4	-3.4	-8.4	-11.9
2/6/20	Sierra Rich	-18.2	-22.1	-24.9	-0.8	-7.7	-12.8
2/6/20	Cresthaven	-20.7	-23.3	-25.1	-5.3	-10.0	-13.2
2/6/20	Red Haven	-20.9	-23.5	-25.2	-5.6	-10.2	-13.4
2/10/20	Suncrest	-19.6	-22.5	-24.6	-3.2	-8.5	-12.3
2/10/20	Sierra Rich	-17.1	-20.4	-22.9	1.2	-4.8	-9.3
2/10/20	Cresthaven	-19.5	-21.8	-23.5	-3.0	-7.3	-10.3
2/10/20	Red Haven	-19.4	-21.8	-23.4	-3.0	-7.2	-10.1

*The data presented here is for information only, and growers should make their own assessment.

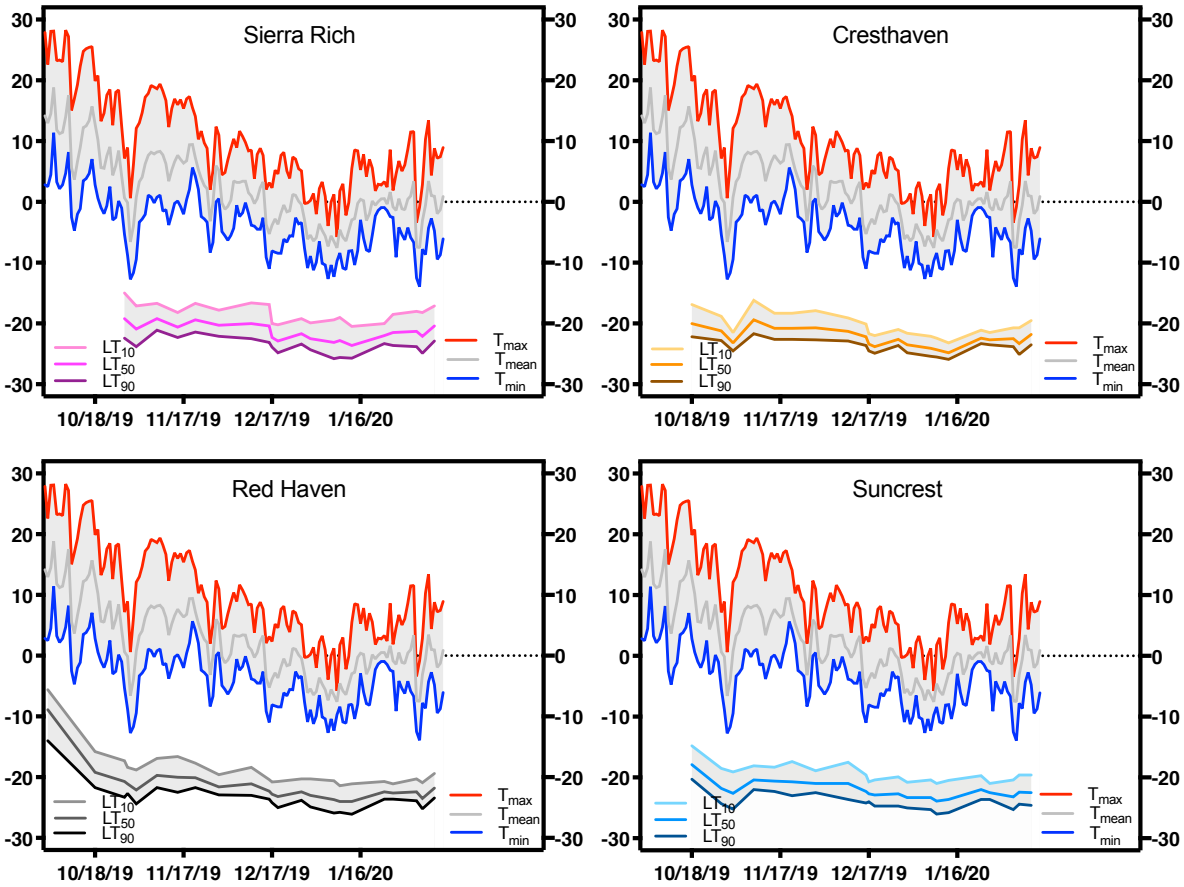

Figure 4. Seasonal patterns of temperature and cold hardiness, expressed as lethal temperature for 10, 50 and 90% of the total flower buds killed (LT_{10}, LT_{50}, LT_{90}, respectively), for peach flower buds of 'Suncrest', 'Sierra Rich', 'Cresthaven' and 'Red Haven' cultivars. Daily maximum, mean, and minimum temperatures recorded at the CSU Western Colorado Research Center at Orchard Mesa near Grand Junction, CO, 2019/20*.

*Temperature data for various locations within the Grand Valley can be found at: http://www.winecolorado.org/colorado-grape-growing/weather-station-network./
Meteorological data from other locations throughout Colorado may also be available from the Colorado Agricultural Meteorological network - CoAgMet.

Table 2. Lethal temperatures (LT) in Celsius (ºC) and Fahrenheit (ºF) for 10 (LT_{10}), 50 (LT_{50}) and 90% (LT_{90}) flower buds killed and percentage (%) of dead buds in the control sample at receiving (estimate of existing field bud damage), for 'Red Haven' peach cultivar grown in the experimental orchard of the Colorado State University's Western Colorado Research Center at Orchard Mesa near Grand Junction, CO, and in commercial orchards of Palisade, Rogers Mesa, Cedaredge, Olathe, CO. Most recent update in red.

		ºC			ºF			
Date	**Location**	**LT_{10}**	**LT_{50}**	**LT_{90}**	**LT_{10}**	**LT_{50}**	**LT_{90}**	**Dead buds in control sample (%)**
10/2/19	Orchard Mesa	-4.8	-5.9	-7.5	23.5	21.4	18.5	0.0
10/2/19	Palisade	-5.6	-8.9	-14.0	22.0	16.0	6.8	0.0
10/2/19	Roger's Mesa	-7.0	-10.0	-11.2	19.4	14.0	11.9	0.0
10/2/19	Cedaredge	-5.2	-8.0	-11.5	22.7	17.6	11.3	0.0
10/2/19	Olathe	-5.7	-9.6	-15.0	21.7	14.7	5.1	0.0
10/29/19	Orchard Mesa	-19.1	-21.3	-22.8	-2.4	-6.4	-9.1	1.7
10/29/19	Palisade	-17.7	-20.1	-21.8	0.1	-4.2	-7.3	0.0
10/29/19	Roger's Mesa	n/a	n/a	n/a	n/a	n/a	n/a	n/a
10/29/19	Cedaredge	-18.8	-21.6	-23.6	-1.8	-6.9	-10.5	5.0
10/29/19	Olathe	-17.5	-20.9	-23.5	0.5	-5.7	-10.2	1.7
12/3/19	Orchard Mesa	-19.1	-21.4	-23.1	-2.3	-6.6	-9.6	0.0
12/3/19	Palisade	-17.4	-20.7	-23.1	0.7	-5.2	-9.6	0.0
12/3/19	Roger's Mesa	-19.0	-21.7	-23.6	-2.2	-7.1	-10.6	0.0
12/3/19	Cedaredge	-20.1	-21.9	-23.1	-4.2	-7.4	-9.7	3.3
12/3/19	Olathe	-19.6	-21.8	-23.3	-3.2	-7.2	-10.0	6.7
1/17/20	Orchard Mesa	-21	-23.3	-24.9	-5.9	-10.0	-12.8	0.0
1/17/20	Palisade	-19.6	-22	-23.6	-3.4	-7.6	-10.5	0.0
1/17/20	Roger's Mesa	-18.1	-22.5	-25.8	-0.6	-8.4	-14.4	1.4
1/17/20	Cedaredge	-18	-22.7	-26.3	-0.4	-8.9	-15.4	11.4
1/17/20	Olathe	-21.3	-23.7	-25.3	-6.4	-10.6	-13.6	11.4
2/13/20	Orchard Mesa	-19.1	-22.0	-24.0	-2.4	-7.6	-11.2	0.0
2/13/20	Palisade	-20.1	-22.2	-23.6	-4.2	-7.9	-10.5	1.7
2/13/20	Roger's Mesa	-20.5	-22.9	-24.5	-4.9	-9.2	-12.1	8.3
2/13/20	Cedaredge	-20.0	-23.1	-25.3	-4.0	-9.5	-13.5	3.3
2/13/20	Olathe	-19.2	-22.4	-24.6	-2.6	-8.3	-12.3	23.3

*The data presented here is for information only, and growers should make their own assessment.

Acknowledgements

The publication of this update was supported by the Specialty Crop Block Grant Program at the U.S. Department of Agriculture in cooperation with the Colorado Department of Agriculture. Its contents are solely the responsibility of the authors and do not necessarily represent the official views of the USDA or CDA. We would like to acknowledge support and coordination for sample collection for the study of Red Haven peach cold hardiness status across the different locations of western CO by CropwoRx (Eckert, CO) for the benefit of Colorado peach growers.

COLORADO
Department of Agriculture

References

[1] P.D. Gerard, W.R. Schucany, Locating Exotherms in Differential Thermal Analysis with Nonparametric Regression, J. Agric. Biol. Environ. Stat. 2 (1997) 255–268.

[2] M.J. Burke, L. V Gusta, H.A. Quamme, C.J. Weiser, P.H. Li, Freezing and Injury in Plants, Annu. Rev. Plant Physiol. 27 (1976) 507–528.

[3] L.J. Mills, J.C. Ferguson, M. Keller, Cold-Hardiness Evaluation of Grapevine Buds and Cane Tissues, Am. J. Enol. Vitic. 57 (2006).