



How That Trends in North Iran Citrus Cultivars Production Correlate with Evastating Freeze Events

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ABSTRACT

Cold and Freezing stresses cause harmful and devastating damage to many agricultural and horticultural crops. In early December of this year (2016), the occurrence of frost stress led to damage to citrus orchards in the north of the country, especially in the east of Mazandaran province. Investigating and calculating the damage to the orchards after the occurrence of cold and/or frost stress is one of the first actions of the Ministry of Jihad Agricultural and its subordinate organizations. However, the exact estimation of the orchards is one of the problems in this area. that considering the harvest season of the main citrus crops of the province hadn't yet started (Thomson orange) and the fruits were still on the tree (half ripe), the amount of frost damage increased. Due to the sustainability of the tangerines, especially Unshiu, the amount of damage to the fruits was little or few. In addition, the result of evaluating frost damage to citrus trees showed that the amount of leaves and fruit damage in the young trees was much more than in the older trees. The amount of fracture of trunks and main branches caused by snowfall was much higher in the hillsides and foothills compared to the plains. According to the calculated observations, it was found that the amount of damage to Thompson oranges in the cities of Babol, Ghaemshahr and Sari was higher than in the other regions due to a decrease in the air temperature to -4 degrees C and a longer glacial period. The fracture of the trunk and main branches of the citrus tree in the cities of Ghaemshahr and Babol was more than in the other regions. Also, the dryness of Thomson orange leaves in the cities of Babol, Ghaemshahr and Sari was more than in the other regions. The leaves and fruits of Page tangerines were damaged to different degrees, but Unshiu tangerines weren't severely damaged in this event.

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1. Introduction

Cold and intense freeze is detrimental to many crops so losses equivalent to billions of Iranian Rials were inflicted upon the farmers-especially citrus growers in North Iran- in some years including the freeze period between 2007-2013 (Rafie-Rad *et al.*, 2023). Freezing stress during the last days of November in 2016 also led to heavy losses to citrus orchards in the North Iran-especially East of Mazandaran province- and the Kiwi orchards in the West of Mazandaran. Therefore, the occurrence of cold and freezing periodically in the Northern provinces of Iran is possible. Estimation of loss in orchards is among the first actions done by Agricultural Jihad Ministry and its subordinate

organizations after cold stress or freeze. Nonetheless, the exact estimation of this loss is a challenge due to various definitions of freezing stress type and damage. Different levels of loss to orchards were reported during the freezing stress of 2016 in citrus orchards located in the North of Iran (especially in Mazandaran province).

2. Low-temperature stress

Low temperature is categorized into cold (0-15°C) and freezing (lower than 0°C) and the basis for this classification is the plant tolerance (Raza *et al.*, 2023). Freezing stress occurs when the temperature falls to lower than the water freezing point. In plant sciences,

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freezing is the temperature at which ice nucleus starts to form. This critical temperature may vary from one to a few degrees below 0°C, depending on the plant species, tissue type, climate conditions before stress, stress duration, etc. (Jiang et al., 2021).

3. Freezing process in plants

Plants and plant parts freeze when they are unable to prevent ice nucleus formation and ice growth. Decrease in the freezing point (1-2 °C) which occurs due to the presence of soluble materials such as sugars and during supercooling to prevent freezing in colder and temperate climates is negligible (Dami and Zhang, 2023). Thus, plants (Citrus cultivars) form different types of ice nuclei at moderate temperatures (Hamilton et al., 2023).

4. Formation of ice nuclei in plant tissues

Water molecules gather around either by themselves (homogenous ice nucleation) or by exogenous stimulation (heterogeneous ice nucleation) to form stable ice nuclei (Primo-Capella et al., 2021). Homogenous ice nucleation is unlikely at a few degrees below zero. In contrast, it is difficult to prevent heterogeneous ice nucleation in humid climates (Tajvar

et al., 2011). Materials involved in heterogeneous ice nucleation include 1) ice nucleus active bacteria; 2) other biological structures and molecules; and 3) organic and non-organic residues. Ice nucleation factors may be on the plant surface (exogenous) or in some cases, inside of plants (endogenous) (Mohammadian et al., 2012). Potential heterogeneous ice nucleation factor must be connected with water. Therefore, these factors will be rendered ineffective when the plant surface is dry. Snow and blizzard may also start nucleation (Livanov, 2023).

5. Cold injury and freezing in citrus

Various oranges (Thomson in particular) with 83940 ha cultivation area consist 76.38% of the total citrus cultivation area in Mazandaran province (Primo-Capella et al., 2021). Also, orange production in this province is 1.27 million tons which consists of 71.53% of the total citrus production of this province. Various types of tangerines with 23400 ha cultivation area (21.29 %) and 460000 tons of production are ranked second (Table 1). Sour orange, lemon, lime and grapefruit are ranked next regarding cultivation area and production (Statistics of horticultural products in Iran, 2015).

Table 1. Cultivation area, production and yield of various citrus types in Mazandaran province (Statistics of horticultural products in Iran, 2015)

Crop	Cultivation area (ha)			Percentage of total	Production (ton)	Percentage of total	Yield (kg/ha)
	Non-fertile	fertile	total				
Orange	3370	80570	83940	76.38	1267136	71.53	16610
Tangerine	1700	21700	23400	21.29	460000	25.97	22632
Sour orange	132	1460	1592	1.47	32500	1.83	22430
lemon	0	500	500	0.45	5000	0.28	11250
Lime	15	80	95	0.09	2000	0.11	34000
Grapefruit	0	70	70	0.06	1000	0.06	16000
Other citrus types	0	290	290	0.26	4000	0.22	11538
Total	5217	104670	109887	100	1771636	100	18099

6. Freezing process in citrus

Citruses are sub-tropical and ever-green plants and thus, sensitive to freezing. The specific temperature at which a certain tissue of a plant freezes and the extent of this injury depends on several factors including species, cultivar, freezing point, duration of cold, plant conditions before stress, and plant age (younger trees are considerably less tolerant compared with more mature trees) (Tajvar et al., 2011). Occasionally, fruit trees bear heavily one year and sparsely the next. This is called biennial bearing. The spring-flowering buds of most hardy fruit trees formed during the previous

spring or summer. Therefore, an especially heavy crop in one year may prevent adequate bud formation for the following year, or may seriously weaken the tree. In any case, trees are less tolerant to cold when they bear higher yields (Juurakko and Walker, 2021). Trees of various species show different sensitivity to freezing stress. Generally, as the temperature falls to -1.7 °C for 30 minutes or longer, some freezing symptoms may appear on citrus trees (Sanie Khatama et al., 2022).

In General, fruits are more sensitive to freezing stress compared with shoots and leaves. The temperature at which citrus fruits are injured depends

on the species and degree of fruit maturity (Lo'ay and Dawood, 2019). Determination of a specific critical temperature for citrus fruits is difficult (Salehi Sardoei et al., 2022), as it is affected by several factors. Duration of the low-temperature period, the growth potential of the tree, meteorological conditions before freezing, fruit maturity and the rate of decline in the temperature affect the extent of injury to the fruit of oranges and other citruses (Medda et al., 2022). Mature fruits contain higher soluble solids that act as antifreeze agents (to lower the temperature at which the liquid freezes) and therefore, are more tolerant compared with the immature fruits. Pulp of frozen orange fruits have a lighter color. The position of the fruit in the crown also affects the freezing degree. Fruits situated on the outer layers of the crown are more sensitive and are injured faster compared with the ones grown on the inner parts (Moellering et al., 2010). Fruit size also affects the extent of freezing injury, so smaller fruits get colder faster than larger ones. Also, thick peels are weaker heat conductors. Therefore, due to this protective role of the peel, the internal temperature declines much slower than the peripheral temperature of the fruit (Ghasemi-Soloklui et al., 2023). When the temperature declines rapidly, the internal part of the fruit may be about 1 °C warmer than the peripheral temperature. Therefore, a decrease in the internal fruit temperature so as to be equal to the peripheral temperature may be delayed by 1 to 1.5 hours. The supercooling phenomenon makes it more difficult to determine the temperature at which the citrus fruit is damaged (Salehi Sardoei et al., 2023b). Supercooling is a phenomenon in which the internal parts of the fruit can fall to temperatures lower than freezing without ice nucleation (Felgitsch et al., 2019). In general, citrus fruits readily freeze when exposed for a few hours at -2.2 to -3.3 °C (Rafie-Rad et al., 2023). Grapefruit fruits required more time to freeze compared with those of orange. Navel oranges may get cold down to 3 degrees below their freezing point before freezing commences. The temperature at which the fruits of a certain orange cultivar and even on the same tree start to freeze slightly varies. Experiments have shown that the freezing point of mature Navel oranges ranges from -2.2 to -2.8 °C (Salehi Sardoei et al., 2023a). Freezing in semi-mature Washington Navel oranges starts from -1.1 to -2.2 °C and is based on the internal temperature of the fruit. Freezing in Green Navel oranges starts

from -1.4 to -1.6 °C (internal temperature of the fruit). The freezing temperature of Valencia oranges is similar to that of Green Navel, and therefore is more sensitive (Sanie Khatama et al., 2022). Critical temperatures at which citrus fruits freeze for some selected fruits is presented in Table 2 along with comments (Sanie Khatama et al., 2022; Geisel and Unruh, 2003).

Table 2. Critical temperatures of freezing and injury for some citrus fruits (Salehi Sardoei, 2022)

Fruit	Maturity stage	Critical temperature
Oranges	Green	-1.4 to -1.9 °C
Oranges and grapefruits	Semi-mature	-1.7 to -2.8 °C
Oranges and grapefruits	Mature	-1.7 to -3.9 °C
Lemons	Mature	-0.8 to -1.4 °C

The critical temperature depends on relative humidity and stress duration. Fruits can tolerate lower temperatures in drier environments and shorter stress periods.

The decline in the internal temperature of the fruit to reach its peripheral temperature may be delayed by 1 to 1.5 hours.

The freezing temperature in the mature Navel oranges ranges from -2.2 to -2.8 °C.

Freezing in the semi-mature Washington Navel oranges starts from -1.1 to -2.2 °C based on the internal temperature of the fruit.

Freezing in the green Navel oranges starts from -1.4 to -1.9 °C (internal temperature of the fruit).

The freezing temperature of Valencia oranges is similar to that of Green Navel, and therefore, Valencia is more sensitive.

7. Freezing injury symptoms in citrus

Freezing injury symptoms depend on the weather conditions after stress (Rafie-Rad et al., 2023). Warmer and drier days following freezing injury lead to faster fruit deterioration. Slightly injured Valencia oranges may almost completely recover at harvest, whereas Navel oranges rarely recover. Frozen oranges may remain on the tree when there is no rainfall, but will fall immediately following rain. Often, the only injury symptom a few days after freezing is the much lighter color of the frozen fruit in comparison with unfrozen parts (Salehi Sardoei et al., 2024). Freezing injury lead to juice loss and dried juice vesicles and thus, results in puffiness. Severely injured fruits may fall rapidly or gradually during freezing stress (Salehi Sardoei, 2022). However, the outer appearance of fruits may not show a remarkable change as the freezing injury is mostly mechanical (Rafie-Rad et al., 2023). Most frozen oranges remain longer on the tree compared with grapefruits (Fig. 1). Loss of juice in the injured fruits occurs during a period of a few weeks. The total reduction of juice depends on the extent of injury and weather conditions after freezing (Zekri et al., 2020). In general, it is revealed that citrus peels freeze at a lower temperature compared with their pericarp.

Therefore, the freezing injury may occur inside the fruit (pericarp) without any visible injuries outside (peel). The bitterness of the fruit is due to the oxidation of phenolic compounds- especially limonene- by polyphenol oxidase enzymes after low-temperature stress (Salehi Sardoei, 2022).



Figure 1. Branch breaking due to freezing stress (photographed by the author)

Freezing stress lead to bruised and curled leaves (Salehi Sardoei et al., 2022) (Fig. 2). Freezing stress symptoms in citrus leaves are not visible in the early stages of stress. However, leaves show symptoms including bruising, wetness and softness (Rafie-Rad et al., 2023). In case the injury is minimal, leaves may recover. Severe stress renders the leaves dry (Salehi Sardoei, 2022). Lower temperatures and longer freezing periods can result in leaf abscission and shoot and main stem necrosis (Salehi Sardoei, 2022) (Fig. 3).



Figure 2. Bruised leaves due to freezing stress (photographed by the author)



Figure 3. Damaged grapefruit and lime seedlings freezing stress (photographed by the author)

8. History of low-temperature stress in citrus orchards of Mazandaran province and the extent of inflicted loss

The most damage during freezing stress in the United States is observed in orchards in California. The blizzard of 1990 in California inflicted a loss of 500 million US dollars to citrus industries, and 450000 ha of citrus orchards were damaged (Attaway, 1997). The Blizzard of 1998 in California resulted in 700 million US dollars of loss.

Besides the freezing stress of 2016, citrus orchards in the Northern provinces of Iran- especially Mazandaran province- witnessed two freezing stress events in January 2008 and February 2014. According to reports, freezing stress during 6-12 January 2008 destroyed a third of orange crops or lowered their quality in a manner that the fruits could not be consumed fresh. Freezing stress of 2013- which occurred between February 2-8- destroyed 10% of citrus crops in the North of Iran. It is highly probable that since most of the fruits had been harvested, this freezing stress led to rather low damage (10%), as citrus- oranges in particular- are mostly harvested in

January. Also, a comparison of minimum temperatures in major citrus production regions of Mazandaran province such as Sari and Ramsar during 2011 and 2014 stresses showed that the minimum temperature of these two regions in 2014 was lower (Tables 3 and 4). Thus, variations in the estimation of losses were due to the difference in the timing of occurrence, and harvest of most fruits in 2014, rather than lower damage inflicted by freezing stress in that year.

Table 3. Minimum temperature (°C) in some regions of Mazandaran province in January 2008 (Iranian Meteorological Organization, 2008)

	6 Jan 2008	7 Jan 2008	8 Jan 2008	9 Jan 2008	10 Jan 2008	11 Jan 2008	12 Jan 2008
Sari	-0.5	-3	-2.5	-3.2	-2.6	1.8	-0.2
Babolsar	-0.4	-2.4	-2	-1	-0.6	-1	0.2
Ramsar	-0.4	-1.5	-2.5	-2.4	-2	-1	0

Table 4. Minimum temperature (°C) in some regions of Mazandaran province in February 2014 (Iranian Meteorological Organization, 2014)

	2 Feb 2014	3 Feb 2014	4 Feb 2014	5 Feb 2014	6 Feb 2014	7 Feb 2014	8 Feb 2014
Sari	-1.6	-5.4	-5.4	-3.4	-3.4	0.8	-2
Babolsar	-1.2	-3.6	-3.6	-1.4	-0.6	-3.2	1.4
Ramsar	-1.2	-3.6	-3.6	-1.8	-3.4	0	-1.6

9. Occurrence of low-temperature stress in the province and the necessity of estimating its inflicted loss

Entry of a cold front and heavy snowfall during 22-23 September 2016 led to freezing stress in Mazandaran province, during which temperature declined to -4 °C in the Eastern regions of Mazandaran province (Table 5). Snowfall, cold and freeze were rare at this time of the year, which surprised the growers and led to severe injuries to citrus trees and fruits. Since orange is mostly harvested during early December, the main citrus product of the province (Thomson orange) had not been harvested yet and thus, the extent of loss soared. After this stress, different levels of loss were reported across the province.

The level of leaf desiccation and damage to citrus fruits in younger trees was much higher than those of older ones. The extent of damage to the bark and main branches in the foothills was considerably higher compared with those located in plains. The extent of injury to Thomson orange fruits in Sari, Ghaemshahr and Babol townships was higher than in other townships due to the decline of temperature down to -4 °C and longer periods of blizzard. Damage to barks

and main branches was more severe in Ghaemshahr and Babol townships. The leaf desiccation level of Thomson oranges in Babol, Ghaemshahr and Sari was higher than that of other regions. Leaves and fruits of Page tangerines showed different levels of injury, whereas Unshiu tangerines were not significantly damaged during this event.

Table 5. Minimum temperature (°C) in some regions of Mazandaran province in September 2016 (Iranian Meteorological Organization, 2016)

	22 Sep 2016	23 Sep 2016	24 Sep 2016	25 Sep 2016	26 Sep 2016
Behshahr (Amirabad port)	-4	-4	-3	-	-
Sari (Dashtnaz)	-3	-4	-2	-	-
Ghaemshahr (Gharakhil)	-3	-4	-1	-4	-1.2
Nowshahr	0	0	-2	-	-
Ramsar	3	0	-1	-	-

10. Conclusion

According to the results obtained from studies conducted on citrus fruits during the last freezing stress, it was revealed that in case the temperature becomes lower than the critical threshold for freezing the fruits for at least a few hours, there is no scientific rationale to keep the fruits on the tree. Otherwise, fruits will lose their juice rapidly and fall after the temperature is increased during the following days and weeks. They also will lack the necessary marketability. The best method for short or middle-term storage of fruits under these conditions is to harvest the fruits and their storage in a cool warehouse at 4-5 °C with 90-95% relative humidity. In case these cool warehouses are not available, fruits should be harvested earlier and sent to process industries before they lose their juice and turn bad. Tangerines are mostly harvested in late January in Mazandaran province, whereas this time for oranges is early December, and since the main citrus product of the province (Thomson orange) had not been harvested yet and fruits were on the trees (semi-mature), the loss due to freezing increased. About half of the tangerines (Page and Unshiu) had been harvested before the snowfall and the other half were harvested after. Since tangerines-especially Unshiu- are tolerant, the extent of loss to their fruits was low or negligible.

Conflict of Interests

All authors declare no conflict of interest.

Ethics approval and consent to participate

No human or animals were used in the present research.

Consent for publications

All authors read and approved the final manuscript for publication.

Availability of data and material

All the data are embedded in the manuscript.

Authors' contributions

All authors had an equal role in study design, work, statistical analysis and manuscript writing.

Informed Consent

The authors declare not to use any patients in this research.

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