

# On-Tree water spray affects superficial scald severity and fruit quality in 'Granny Smith' apples

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## Abstract

'Granny Smith', a variety of apple is susceptible to superficial scald. Superficial scald is one of the most controversial post-harvest physiological disorders. At the moment there is no complete documented information about the pre-harvest factors responsible for its induction, symptom development and control methods. Accordingly, this study was to examine the possible link between the disorder and qualitative indicators of environmental changes, particularly humidity during early fruit growth and cell division. The study was conducted in a commercial orchard in Abyek region, Alborz-Iran in 2014. Examined trees were 7 year-olds and grafted onto seedling rootstocks. On-tree water spray was done during full bloom, 17 May till 6 June, in four levels including 0-day, 7-day, 14-day, and 21-day water spray. Fruits were harvested 160 days after full bloom (DAFB) and stored in a commercial chamber (5±2°C) in four levels including: 0-day (before storage), 30-day, 60-day and 90-day of cold storage. Superficial scald severity in the 0-day water spray samples was 91.7 % after 60- and 90-day storage, while all three water spray treatment regime significantly reduced it after 60- and 90-day storage. The 14-day water spray was significantly more effective in reducing the incidence of superficial scald which was 91 % after 60-day and 75 % after 90-day storage. The highest firmness (9.5 kg cm<sup>-2</sup>) was observed before storage. The highest SSC was observed in the 7-day water spray and before the storage. Interestingly, on-tree water spray considerably decreased superficial scald and/or delayed its severity during mid- and long-term storage.

**Key words:** Canopy humidity, cold storage, fruit growth, fruit quality, physiological disorder, scald

## Introduction

The cultivar 'Granny Smith' is a green-sour in taste with firm tissue widely accepted worldwide. However, it is very susceptible to superficial scald (Lurie and Watkins, 2012). Superficial scald, or storage scald, is a common physiological disorder that develops during cold storage of apple fruit (Emongor *et al.*, 1994). The biochemistry of scald etiology focused almost exclusively on the involvement of  $\alpha$ -farnesene and its oxidation (Lurie and Watkins, 2012). The development and severity of superficial scald in apple fruit is proportional to the amount of antioxidants in the peel and the extent of  $\alpha$ -farnesene oxidation. The extent of metabolism of these compounds is also influenced by weather, orchard management, and tree characteristics and nutrition before harvest (Emongor *et al.*, 1994). The variation in emission of plant aroma are related to temperature, humidity and rainfall in the field. Remarkably, rainfall had a significant positive influence on changes in volatiles released during all three diurnal periods, and other factors of significance were temperature and relative humidity around noon, relative humidity in the late afternoon, and temperature and relative humidity during the night (Vallat *et al.*, 2005). Temperature, sunlight and rainfall, which are uncontrollable elements of weather varied throughout the season and this had a great effect on the quality and storage behavior of apples. Scald susceptibility varies among seasons, harvests periods within seasons, and growing areas (Wilkinson and Fidler, 1973). Climatic variables have a great influence on

scald which is usually most severe in years characterized with hot and dry climate during the last few weeks of the growing season (Martin and Lewis, 1961; Smock, 1953). Fidler (1956) reported an increase in scald susceptibility with increased hot, dry weather, while the reverse was the case when the weather was cool and damp. Signs and symptoms of scald are more severe on a crop harvested after a warm dry summer than on one after a dull wet summer (Wilkinson and Fidler, 1973). Low night temperatures in the period before harvest decrease the incidence of scald, and there exist an inverse relationship between the number of days below the threshold temperature of 10 °C and the incidence of scald (Blanpied *et al.*, 1991). Superficial scald reduces the appearance, utility, and market value of apples and pears, with severely affected fruits being sold only for processing. Pre-harvest factors during fruit ontogeny greatly affect the fruit quality at harvest. Fruit response to various treatments are responsible for the development of physiological disorders and retention of fruit quality at the end of storage period (Emongor *et al.*, 1994). Over the years, many methods have been developed and tested to prevent superficial scald in apples and pears (Bordonaba *et al.*, 2013). Physical and chemical alternatives have been tested, including, fruit heating before cold-storage, 1-MCP, essential oils, *etc.* (Lurie *et al.*, 2005). Consumer awareness and dissatisfaction over the use of chemicals in food products have brought forth the necessity to control scald by harmless non-organochemical procedures (Emongor *et al.*, 1994). Unfortunately, there is no detailed information about a possible link between environmental

changes, particularly temperature and humidity with the division and cell growth. However, actual observations and practical experience have shown that the incidence of cold storage scald after the spring rains is more all through the year. Accordingly, the purpose of this research was to evaluate the effect of ambient humidity in tree canopy, around the fruit on the incidence of scald after cold storage, and also the qualitative and quantitative changes which accompany with changes in moisture content before the fruit is crowned after full bloom.

## Material and methods

### Plant material, pre-harvest treatments and storage conditions:

7-year-old 'Granny Smith' apple (*Malus domestica* Borkh.) trees grafted on seedling rootstock growing in a commercial orchard located in Abyek region (36°04' 00" N; 50°32' 59" E, 1456 m), Alborz province, Iran were studied. During the study from April 22 till October 22 the mean temperature was 28.2 °C, RH (27 %), wind speed (16.9 km/h) (IRIMO, 2014). A randomized complete block design with three replications was used. Each block contained 55 uniform trees with similar physical characteristics without any pests or diseases. On-tree water spray was applied after full bloom between May 17<sup>th</sup> and June 6<sup>th</sup>, 2014, using a tractor mounted water sprayer (TMS 600 F, Iran) with the same intensity. Water spray was applied three times a day. The water spray treatment included four levels: 0-day: without water spray (CRIRH = 1), 7-day water spray: water spray for 7 days (CRIRH = 13), 14-day water spray (CRIRH = 30), and 21-day water spray (CRIRH = 43). The water was sprayed on the canopy during the hottest hours of the day (between 12:30 to 17:30) thrice a day. For each level of treatment, a total of 10 trees were sprayed three times a day with 1.5 hours' interval. Five liter of water per tree was applied each time, a total of 15 liter/tree/day. Every-other-tree was sprayed in the same row, in order to confine the water spray to target trees. During the course of water spray, canopy humidity and temperature were recorded using a hygrometer and thermometer (PRÄZISIONS-HYGROMETR, Germany). Canopy humidity change was calculated using CRIRH formula for all treatments separately.

$$CRIRH = 1 + \sum_{i=1}^n (RH_T - RH_{T'}) / RH_{Mean.c}$$

Where:

CRIRH: The cumulative relative increase in relative humidity

$RH_T$ : Relative humidity changes in the trees under water spray

$RH_{T'}$ : Relative humidity changes in the trees without water spray

$RH_{Mean.c}$ : The mean relative humidity changes in the trees without water spray

Fruits were harvested 160 days after full bloom. Crop load and yield were calculated for the treatments, separately. Sample fruits were harvested, weight and then divided to four groups and stored at 5±2 °C and 85 % relative humidity. Superficial scald, as percentage and index, flesh firmness, soluble solids content (SSC), titratable acidity (TA), fruit size, peel color, and antioxidant activity were evaluated at 0-day (before storage), 30-day, 60-day and 90-day of cold storage.

**Scald evaluation:** Scald incidence was determined after 30, 60 and 90 days of storage plus 1-week post-storage in air at 20 °C on four replicates of 12 fruits for each period, and expressed as percentage of affected fruit. The severity of scald was expressed

using a four-point scale with for classes: 0 = no injury; 1 = slight injury (1 to 25 % of surface affected); 2 = moderate injury (25 to 50 % of surface affected) and 3 = severe (> 50 % of surface affected). A severity index was calculated as follows:

$$\text{Index} = [(1 \times \% \text{ fruit grade } 1) + (2 \times \% \text{ fruit grade } 2) + (4 \times \% \text{ fruit grade } 3)] / 4 \text{ (Lurie et al., 1990).}$$

**Flesh firmness, SSC and TA:** Flesh firmness was measured using a penetrometer (FCE- PTR 200 Extech, USA) with an 11 mm tip on opposite sides, after fruit peel removal. Results were expressed as kg cm<sup>-2</sup>. SSC was determined using a Euromex RD 635 digital electronic refractometer (Euromex, Netherlands) at 20 °C. Results were expressed as Brix degree. Total TA was measured by titrating juice with 0.1 N NaOH with a pH 8.2. Results were expressed as %. Fruit size and peel color were not affected by water spray (data not shown).

**Antioxidant capacity:** Antioxidant capacity of fruit extract was determined according to Brand-Williams *et al.* (1995), with minor modifications. Briefly, 50 µL of apple extracts was added to 950 µL of a 0.1 mM of DPPH in methanol. A control sample containing the same volume of DPPH solvent was used to measure the maximum DPPH absorbance. Upon completion of the reaction in the dark for 30 min, the absorbance at 517 nm was recorded to determine the content of remaining DPPH (Du *et al.*, 2009). The percentage of scavenged DPPH (% DPPHsc) was calculated using the following formula:

$$DPPHsc (\%) = (A_{cont} - A_{sam}) \times 100 / A_{cont}$$

**Statistical analysis:** A randomized complete block experiment and split-plot test with three replications was employed. Water spray treatment and fruit storage time were sources of variation. The data were subjected to analysis of variance (ANOVA) by using SAS software (Ver. 9.1). The least significant difference (LSD) method was used to compare the mean at  $P=0.01$  and  $0.05$ . Pearson correlations were used for quantifying the relationships among assessed factors.

## Results and discussion

**Crop load and yield:** There was no significant difference of crop load (50±3 kg/tree) and yield (20±3 Ton/ha) as affected by various water spray treatment (data not shown).

**Superficial scald:** The interaction between water spray and storage treatment on the severity and incidence of superficial scald was significant ( $P<0.01$ ) (Table 1). There was no incidence of superficial scald after 0- and 30-day storage. The percentage of superficial scald for 0-day water spray (CRIRH=1) developed after 60- and 90-day of storage was 91 %. Water spray treatment significantly postponed and decreased the incidence and severity of superficial scald after 60-day and 90-day storage; but 14-day water spray (CRIRH = 30) more significantly reduced incidence of scald; (Fig. 1 and Fig. 2).

Although scald is assumed to be the result of an oxidative process, the relationships between the endogenous levels of antioxidants and scald incidence remains still not clearly defined. Duvenage and de Swardt (1973) studied the interrelation of total polyphenol content and superficial scald onset in the 'Granny Smith' and reported that leucoanthocyanidines increased as fruit maturation

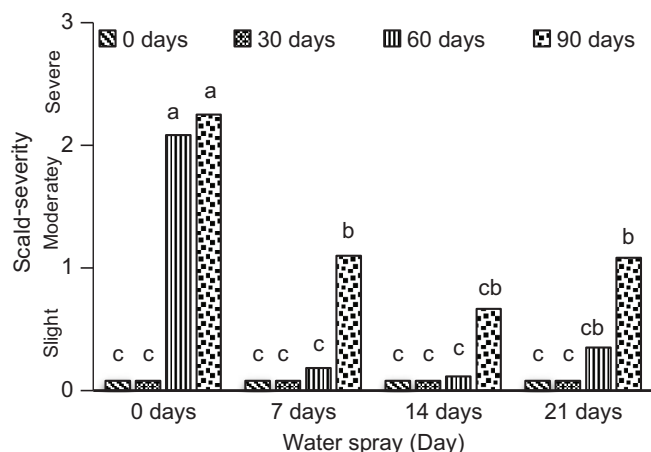


Fig. 1. Interactive influence of on-tree water spray and cold storage period on scald-severity of 'Granny Smith' apples. a–c mean significant difference at  $P \leq 0.01$ .

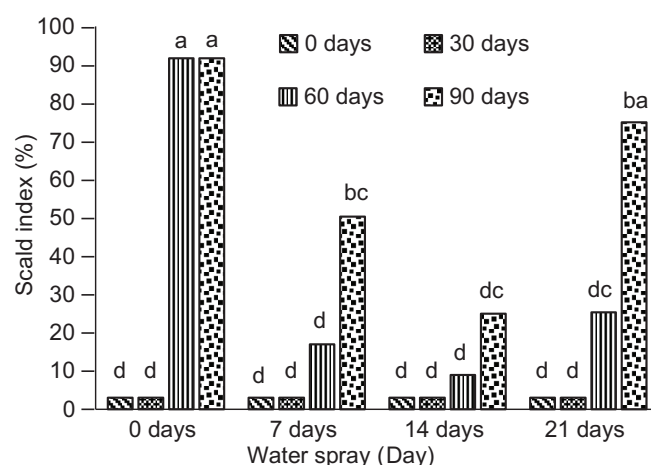


Fig. 2. Interactive influence of on-tree water spray and cold storage period on scald index of 'Granny Smith' apples. a–c mean significant difference at  $P \leq 0.01$ .

progressed, suggesting that this increase could lead to lower superficial scald susceptibility. It is therefore logical to assume the involvement of polyphenols, readily oxidizable compounds which are found in substantial amounts in the peel of apples. Weather condition such as pre-harvest air temperature, maturity stage, and fruit position in canopy may all affect the incidence and severity of scald. Superficial scald manifests when fruits are transferred from cold storage to ambient temperatures, but can also develop in storage after prolonged periods (Bauchot *et al.*, 1999). Scald is usually more prevalent on fruits harvested earlier

than those harvested late (Wang and Dilley, 1999; Wilkinson and Fidler, 1973). Low night temperatures in the period before harvest decreased the incidence of scald and an inverse relationship between the number of days below the threshold temperature of 10 °C and the incidence of scald has been noted (Ma *et al.*, 2001; Thomai *et al.*, 1998). The disorder is consistent with a two-stage event where the induction events are separated from symptom development (Yi Wang, 1990). Abiotic conditions and subsequent volatile released from plants may be subject to dramatic changes during a diurnal cycle (Loughrin *et al.*, 1994). Temperature and humidity were shown to influence the composition of released blends significantly; even in perennial plants. Here, we showed that on-tree water spray decreased canopy temperature in the critical cell division stage resulting in lesser scald incidence especially in long-term treatment (Fig. 1 and Fig. 2). Variation in emitted plant fragrance can be triggered by a changing biotic environment such as herbivore feeding (Dicke and van Loon, 2000), or by varying abiotic factors such as temperature and humidity (Gouinguene and Turlings, 2002).

**Flesh firmness, TA and SSC:** According to the results, there was no significant effect of water spray treatment on the 'Granny Smith' fruit firmness. However, the cold storage period significantly affected the firmness (Table 1). The first phase of fruit softening slowly emerged after 30-day storage. However, the softening of the fruit after 60- and 90-day storage remained unchanged. A 9.5 kg cm<sup>-2</sup> fruit firmness had the highest value and this decreased after 30-day of storage (Fig. 3). Pre-harvest water spray treatment had no significant impact on the fruit acidity. However, three different periods of fruit storage had a significant effect on fruit acid content (Table 1). The highest acidity was observed after 60-day storage and the lowest was observed before and after 90-day storage. An increasing trend was observed in the fruit total acidity in harvest time and 60-day storage; but this decreased after 90-day storage (Fig. 4). The interaction between water spray and storage term had a significant impact on the SSC (Table 1). 0-day water spray (CRIRH=1) fruits and those received 7-day, 14-day and 21-day water spray (CRIRH= 13, 14 and 43) in the three storage showed different behavior. The highest SSC were observed in 7-day water spray before cold storage. In contrast, the lowest SSC was observed in the 0-day water spray (CRIRH=1) cold-stored for 90 days. Water spray had no clear effect on SSC content during three storage periods (Table 2). Fruit softening phase could be considered irrevocable once initiated; thus, first phases softening needs to be prolonged

Table 1. Analysis of variance for 'Granny Smith' apple fruit quality characteristics

Source of variation	DF	Mean square						
		Scald-Severity	Scald-Index	FF (kg cm <sup>-2</sup> )	SSC (°Brix)	TA (%)	TAA of the peel (% DPPH <sub>sc</sub> )	TAA of the pulp (% DPPH <sub>sc</sub> )
R	2	0.34ns	320.55ns	0/11ns	2/98*	1/23ns	2/61ns	17/62ns
A	3	1.93**	1286.31**	0/77ns	1/72ns	0/47ns	10/70*	9/86ns
R×A	6	0.05ns	46.18ns	0/47ns	0/46 ns	0/89ns	4/07ns	5/80ns
B	3	4.15**	2773.43**	5/35**	11/59**	4/23**	110/50**	71/87**
A×B	9	0.69**	621.21**	0/29ns	1/57*	0/80ns	10/70**	12/37*
Error	24	0.17	165.33	0/35	0/65	0/46	2/74	5/25
CV	2	81.52	95.27	6/89	5/85	10/95	2/90	3/98

\*\* and \* Significant difference values  $P \leq 0.05$  and  $P \leq 0.01$ , ns: non-significant, FF: Flesh firmness, SSC: Soluble solid content, TA: Titratable acidity, TAA: Total antioxidant, R: Block, A: water spray, R×A: Interaction of block and water spray, B: storage periods, A×B: Interaction of water spray and storage period.



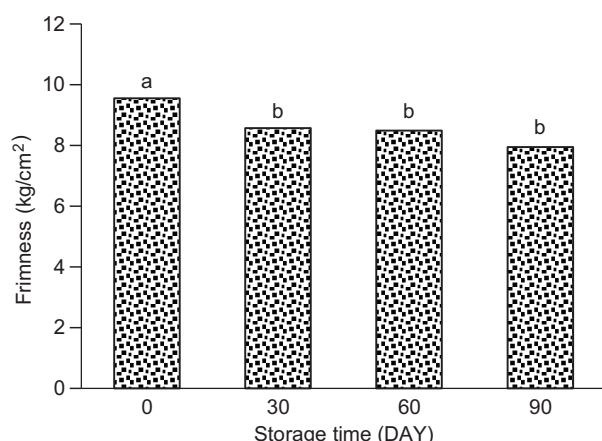


Fig. 3. The flesh firmness during storage at  $5\pm 2$  °C, different letters denote significant difference at  $P \leq 0.01$ .

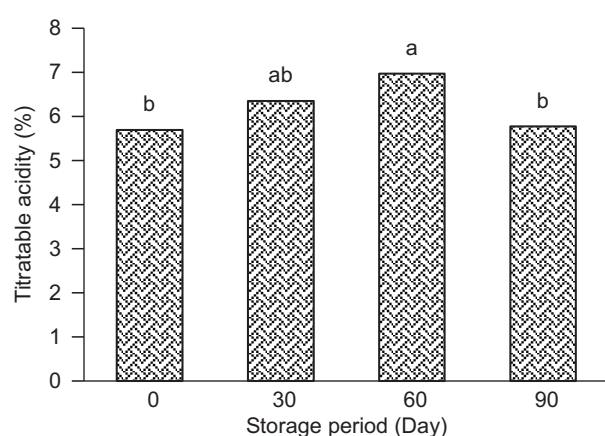


Fig. 4. The titratable acidity during storage at  $5\pm 2$  °C, different letters denote significant difference at  $P \leq 0.01$ .

if firmness is to be maintained for a long-term in storage. Several at-harvest and postharvest factors have been found to influence the duration and rate of softening in the first and second phases (Johnston *et al.*, 2002). Interestingly, second softening phase was postponed for 90 days which could be related with on-tree water spray during cell division of fruit growth (Fig. 3). Some research reports have focused that effect of environmental factors including light, temperature, and moisture had pronounced effects on fruit texture. Crouch (2003) reported a decline in apple fruit TA after harvesting. Fruits stored for six months were found to have depreciated greatly in malic acid content. TA of the fruit depended on the rate of metabolism, especially respiration which consumed organic acid, thereby reducing the acidity (Jan *et al.*, 2012). An increasing trend was observed in the fruit total acidity at harvest time and 60-day storage; but this decreased after 90-day of storage (Fig. 4). The SSC in harvested control fruit (CRIRH=1) after 90 days' storage was in a downward trend. The reaction in control fruit (CRIRH=1) during storage is contrary to the results of Ali *et al.* (2004). The results of these investigations are in conformity with the findings of Ali *et al.* (2004) who reported that reducing sugars tended to increase during storage. The change was probably due to conversion of sugar into starch (Wills *et al.*, 1980), which showed that apple may become less sweet with the passage of time.

**Antioxidant activity:** The interaction of water spray and storage treatment on peel total antioxidant capacity was significant ( $P \leq 0.05$ ) (Table 1). The highest peel total antioxidant was observed in 7-day water spray (CRIRH = 13), after 30-day

Table 2. The effect of on-tree water spray and storage period treatments on the quality of Granny Smith apple

water spray (day)	Storage Periods (day)	SSC (Brix)	Peel TAA (%DPPH <sub>sc</sub> )	Pulp TAA (%DPPH <sub>sc</sub> )
0 (control)	0	15.19 ba	58.83 bac	54.55 g
	30	14.17 bdec	59.43 bac	59.19 bdac
	60	12.69 hg	58.85 bac	60.59 bac
	90	11.86 h	58.38 bdac	58.70 ebdac
7	0	15.59 a	58.52 abc	54.78 gf
	30	13.18 feg	60.86 a	57.45 ebdgcf
	60	13.70 fdeg	56.51 dec	59.89 bac
	90	14.62 bdac	53.3 fe	53.87 hg
14	0	15.30 ab	59.17 bac	55.69 edgf
	30	13.38 feg	59.98 ba	56.77 edgcf
	60	14.02 bdec	59.48 bac	61.53 a
	90	13.57 fdeg	50.49 f	50.35 h
21	0	15.11 bac	56.33 bdec	53.61 hg
	30	13.86 fdec	59.92 ba	58.66 ebdacf
	60	12.52 hg	56.49 bdec	61.23 ba
	90	12.88 fhcg	54.61 de	54.82 egf

SSC; Soluble solid content, TAA; Total antioxidant activity. Different letter in each column shows significant difference ( $P \leq 0.05$ ).

storage; while the least was in 14-day water spray (CRIRH = 30), after 90-day storage (Table 2). The interaction of water spray and storage on pulp total antioxidant capacity was significant ( $P \leq 0.01$ ) (Table 1). The highest pulp total antioxidant capacity was observed in 14-day water spray (CRIRH = 30) and 60-day cold storage; whereas the least was in the same spray treatment but 90-day storage. Changes in total antioxidant capacity of the peel in 0-day water spray (CRIRH = 1) increased gradually after 60-day storage which slightly decreased after 90-day storage. A similar downward trend behavior was observed in all three treatment and 30-day and water spray; however, a downward trend was found after 60-day storage. Antioxidant capacity of the control fruit pulp increased during 30-day and 60-day cold storage after which decreased till 90-day, similarly for all treatments (Table 2).

A direct positive correlation was found between phenol content and antioxidative capacity which is in agreement with several scholars (Kalt *et al.*, 1999; Schmitz-Eiberger *et al.*, 2003). Apples contain many bioactive compounds, concentrated predominantly in the epidermis (Van der Sluis *et al.*, 2001; Veberic *et al.*, 2005). Pinelo *et al.* (2004) reported that antioxidant capacity of flavonoid was dependent on the variation in their antiradical activity. Owing to the fact that polyphenols belong to the most important bioactive components; a direct positive correlation between phenol content and antioxidative capacity was observed by several researchers that antioxidant capacity of flavonoid was in agreement with variations in their antiradical activity. Leja *et al.* (2003) showed that the antioxidant activity doubled during the four-month storage period. The reason for the increase in antioxidant activity is attributed to a higher content of total phenols. Ayala-Zavala *et al.* (2004) also found a slight change in total antioxidant capacity at 0 °C during storage, but increased significantly at 5 and 10 °C. Even though the overall quality was better maintained at 0 °C, the storage temperature of 10 °C significantly increased the total antioxidant capacity, total anthocyanin, and total phenolics.

In conclusion, on-tree water spray during primary fruit growth phase, cell division, resulted in higher fruit quality and lower postharvest superficial scald severity. This could be as a result

of lowering fruit temperature and increasing canopy relative humidity. The interaction between water spray and cold storage on 'Granny Smith' apple peel and total antioxidant capacity was significant. Pre-harvest water spray resulted in the higher fruit quality, especially after a long-term cold storage. This finding could be applied as a good strategy for commercial 'Granny Smith' apple production.

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