# Changes in Physiological Characteristics of Kiwifruit Harvested at Different Maturity Stages after Cold Storage

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# Summary

Changes in the nutritional composition, along with some other kiwifruit (*Actinidia deliciosa* (A.Chev.) C.F.Liang & A.R.Ferguson) characteristics, at different harvest stages were evaluated after four months cold storage. Various physiological characteristics in kiwifruit harvested at seven maturity stages (~6.5, 7, 7.5, 8, 8.5, 9 and 10 °Brix, respectively) and after four months storage at 0°C were determined. Fruit weight loss increased significantly during storage, depending on the harvesting stage. After four months cold storage, the earlier harvested fruits retained weight better than late ones. Flesh firmness decreased significantly after storage independently to the harvesting stages. At the end of storage period, fruits harvested at later stages showed a significant higher level of ascorbic acid and of phenolic compounds compared to the earlier harvested fruits. Overall, kiwifruit harvested with 6.5 °Brix retained weight better than the other harvesting stages and showed a higher antioxidant capacity after four months cold storage.

# Key words

antioxidant capacity; ascorbic acid; flesh firmness; total soluble solids

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# Introduction

In recent years, food scientists and nutrition specialists have come to the conclusion that consumed fruits and vegetables reduce risks of certain diseases, including cancer and cardiovascular diseases (Liu et al., 2000; Martin et al., 2002). It is currently accepted that the consumption of kiwifruit has a preventive effect against certain cancers and cardiovascular diseases. Many different cancers, especially cancers of the digestive system (mainly stomach cancer), lung, and liver, have been treated with kiwifruit prescriptions due to its cytotoxic and antioxidant activities (Rush et al., 2002). Therefore, the beneficial effects of fruits and vegetables may be related to the antioxidant properties (Imeh and Khokhar, 2002; Moyer et al., 2002; Rush et al., 2006).

Fruit is one of the major dietary sources of various antioxidant phytocompounds for humans. Kiwifruits, as well as citrus fruit, are excellent sources of the ascorbic acid (Nishiyama et al., 2004). In kiwifruits, the ascorbic acid content is higher than that determined in orange; strawberry, lemon and grapefruits. Esti et al. (1998) have observed that the ascorbic acid content of kiwifruit depends on genotype, ripening degree, storage and the analysis method utilized. Imeh and Khokhar (2002) underlined the important role of the pre- and post-harvest factors on the chemical composition of plant foods.

Maturity stage is an important factor that influences the compositional quality of fruit and vegetables. In fact, several biochemical, physiological and structural modifications take place at different maturity stages and these changes determine the final quality during ripening. Furthermore, storage conditions can influence the quality indices and nutritional content of fresh fruit (Lee and Kader, 2000; Ayala-Zavala et al., 2004). Tavarini et al. (2008) found that harvest time and storage conditions strongly influenced the qualitative and nutritional characteristics of kiwifruit.

For a good fruit quality, the standard practice for kiwifruit *Actinidia deliciosa* 'Hayward' is the harvest when the minimum of total soluble solids (TSS) is 6.28 °Brix and the fruit is stored at 0°C for up to six months (Tavarini et al., 2009). A rapid loss of starch and an increase in soluble solids content provides one of the main criteria for harvesting (Walton and De Jong, 1990). In addition, fruit firmness is very high, up to 60 N, at harvest time, but rapidly decreases when the fruit is ready to eat at 5–8 N (Beever and Hopkirk, 1990). This can be used as another maturity index for harvesting. Softening of 'Hayward' kiwifruit was proposed to be the consequence of solubilization of insoluble materials such as cell wall materials and starch (Bonghi et al., 1996).

Therefore, the aim of this research was to evaluate the influence of harvesting stages on several kiwifruit quality traits and to determine the proper harvesting stage necessary for maintaining fruit quality during the storage time.

# Materials and methods

# Materials

This experiment was conducted in 2009 in a commercial kiwifruit (*Actinidia deliciosa*, cv. 'Hayward') orchard of Citrus Research Institute of Iran located at Ramsar in Mazandaran province. The vines were trained as a T - bare system. The maturity indices were evaluated at seven different harvesting stages, when fruit TSS reached approximately 6.5, 7, 7.5, 8, 8.5, 9 and 10 °Brix. TSS was measured directly in the field on 20 fruit samples, randomly collected from the whole canopy in different sites in the orchard. Fruits were stored at 0°C for four months and analysed for their quality indices immediately after harvesting and after four months storage. Weight loss, flesh firmness, flesh color, TSS, titratable acidity (TA), ascorbic acid, phenolic compounds and antioxidant capacity were evaluated.

## Flesh color

Fresh-cut kiwifruit surface color was directly measured with a CR-400 Minolta chroma meter (Minolta, Inc., Tokyo, Japan). Color was measured using the Hunter *L value* ( $L^*$ ), Hue angle ( $H^\circ$ ) and Chroma ( $C^\circ$ ) coordinates in three points of each fruits in the CIELAB system. Ten fruits were evaluated for each treatment. The colorimeter was calibrated with standard black and white calibrations tiles.

# Weight loss

Fruits were weighed at the beginning of the experiment just after harvest, and thereafter every 30 days during the storage period. Weight loss was expressed as the percentage loss of the initial total weight. For each measurement, 10 fruits corresponding to each treatment were used.

# Flesh firmness

The measure was performed on two opposite faces of the equatorial zone using a digital penetrometer installed on a driving column equipped with an 8 mm probe (Model FTO 11). Measurements were carried out on a flat surface of the fruits.

#### TSS and TA

TSS was determined using a refractometer (CETI-Belgium, Kontich-Antwerp, Belgium). Measurements were carried out at the same sites as flesh firmness and were expressed as °Brix. TA was determined by titration of 5 mL of filtered juice by 0.1 N NaOH up to pH of 8.3.

#### Ascorbic acid

The ascorbic acid concentration was measured based on the reduction of the dye 2, 6-dichlorophenol-indophenol (DIP) by ascorbic acid (Kim and Yook, 2009). Kiwifruit fresh tissue (3 g) was mixed with 20 mL (3%) metaphosphoric acid and homogenized. Ascorbic acid was determined by titration of 15 mL filtrated juice by DIP containing bicarbonate sodium and expressed by mg ascorbic acid  $\cdot 100 \text{ g}^{-1} \text{ FW}.$ 

#### Phenolic compounds

Phenolic compounds were determined by the Folin-Ciocalteau method, based on colorimetric oxidation/reduction reaction of phenolic compounds (Singleton et al., 1999). Polyphenols were extracted from 0.5 g fine powder of fruits with 3 mL 85% methanol. To 0.5 mL extract, 0.25 mL Folin-Ciocalteau reagent, 2.25 mL distilled water and 2 mL of 7% Na<sub>2</sub>CO<sub>3</sub> was added. Then, the samples were shaken for 2 h. The absorbance of samples was measured at 765 nm by a UV-visible spectrophotometer (model PG Instrument +80, Leicester, UK). Gallic acid was used for calibration curve. Results were expressed as mg GAE-100 g<sup>-1</sup> FW.

Harvest time (°Brix)	L		Н		С	
	At harvest	Storage 4 months	At harvest	Storage 4 months	At harvest	Storage 4 months
6.5	56.92 abc	55.26 abc	109.9 a	112.2 a	28.89 a	26.73 a
7.0	57.04 abc	52.57 cde	110.5 a	111.9 a	30.00 a	27.34 a
7.5	56.79 abc	54.57 abc	109.5 a	111.6 a	26.69 a	26.75 a
8.0	56.94 abc	53.59 cde	109.6 a	111.7 a	29.93 a	25.43 a
8.5	55.74 abc	52.01 e	110.6 a	111.5 a	29.90 a	26.82 a
9.0	60.34 a	52.97 cde	110.0 a	112.9 a	28.31 a	25.98 a
10.0	58.60 ab	52.48 de	109.7 a	113.0 a	27.16 a	25.42 a

Table 1. The changes in pulp *hue* (*H*), *lightness* (*L*) and chroma (*C*) value of kiwifruit harvested in different maturity stages at harvest and after four months cold storage

\*Means within columns followed by the same letters are not significantly different at  $P \le 0.05$  with Duncan's multiple range test

#### Antioxidant capacity

Antioxidant activity in the extract of flesh was measured by the scavenging of 2, 2-diphenyl-2-picrylhydrazyl hydrate (DPPH) (Sigma-Aldrich Chemie, Steinheim, Germany) free radical with a UV-visible spectrophotometer (model PG Instrument +80, Leicester, UK) at 515 nm as described by Brand-Williams et al. (1995). The antioxidant activity was expressed as the percentage of decline of the absorbance, relative to the control, corresponding to the percentage of DPPH that was scavenged. The percentage of DPPH, which was scavenged (%DPPHsca), was calculated using;

%DPPHsca= ( $A_{cont} - A_{samp}$ ) ×100/ $A_{cont}$ 

where  $\rm A_{cont}$  is the absorbance of the control and  $\rm A_{samp}$  the absorbance of the sample.

## Statistical analysis

SAS statistical software was used for data analysis. The means were compared by Duncan's multiple range test (DMRT) at confidence levels of 99%. The mean and standard error of means were calculated.

# **Results and discussion**

# Flesh color

Flesh color value (L, H and C) of kiwifruits at seven different harvesting stages and after four months cold storage are summarized in Table 1. The results showed that no significant differences were found for pulp color H and C value at harvesting stages and at the end of cold storage, but L significantly declined independently to the harvesting stages after four months storage at 0°C. In fact,  $L^*$  value measures luminosity that varies from zero (black) to 100 (pure white). Krugera et al. (2010) found that overall color values such as  $L^*$ ,  $a^*$  and  $b^*$  and also chroma and hue decreased after cold storage for all harvesting stages compared to the fresh fruit.

# Weight loss

Weight loss of kiwifruit harvested at seven maturity stages after four months cold storage is shown in Figure 1. Fruits weight loss significantly increased during storage, dependently to the harvesting stage; the lowest value was found in fruits harvested with 6.5 °Brix, although no significant difference was found



Figure 1. Weight loss of kiwifruits harvested in seven different maturity stages (°Brix, 6.5, 7, 7.5, 8, 8.5, 9 and 10) after four months cold storage. The means followed by the same letters are not significantly different at  $P \le 0.01$ . The vertical bars are standard errors of the means (n=3)

with 7 and 7.5 °Brix. Data showed that during four months cold storage, early-harvested fruits retained weight loss better than late harvested ones. The mean water content of kiwifruit at harvest is typically 80–90% of the fresh weight (Burdon and Clark, 2001). Postharvest water loss can cause rapid deterioration in produce quality through shrivelling. However, before shrivel becomes apparent, postharvest water loss may also alter metabolism and in some instances, hastens fruit ripening (Burdon et al., 1994). Therefore, reducing water loss from fruit during storage or ripening helps to maintain fruit quality. Our study showed that delay in the harvesting time make fruits more susceptible to water loss than early harvest, near the commercial mature time (6.5 °Brix).

# Flesh firmness

Firmness of kiwifruit at seven harvesting stages immediately after harvest and after four months of cold storage is presented in Figure 2. The flesh firmness significantly decreased with progress of fruits maturation at harvesting stage. The highest value was found in fruits harvested with 6.5°Brix. Furthermore, flesh firmness decreased significantly during cold storage indepen-



Figure 2. Firmness of kiwifruits harvested in seven different maturity stages (°Brix, 6.5, 7, 7.5, 8, 8.5, 9 and 10) and after four months cold storage. The means followed by the same letters are not significantly different at  $P \le 0.01$ . The vertical bars are standard errors of the means (n=3)

dently of the harvesting stages. Thus, no significant difference was found after four months storage at 0°C. The flesh firmness is widely used for defining postharvest quality of kiwifruit (Bonghi et al., 1996). Fruit firmness decreased slowly from 81.8 N at harvest to 20.7 N after 12 weeks of storage at 0°C (Yin et al., 2009).

Crisosto and Kader (1999) reported that late harvested kiwifruits retain their flesh firmness during storage better than early harvested fruits. But in this study, there was no significant difference among harvest times at end of storage. In comparison with data obtained from Tavarini et al. (2008), it is important to underline that the first harvest has shown more firmness just after harvesting, which means that fruits were near the physiological maturity stage (8 °Brix). Furthermore, during maturation and storage period, a decrease in flesh firmness was observed as a result of polygalacturonase activity. This activity depends on storage conditions and the genetic features of cultivars as reported by Villarreal et al. (2008). Our study showed that softening of *Actinidia deliciosa*, cv. 'Hayward' fruit after four months does not depend on its maturity.

# TA and TSS/TA

Delaying harvest from standard maturity stage (6.5-7 °Brix) decreased TA content during harvesting stages (Figure 3); the lowest value being registered at fruits harvested at 8 °Brix. TA declined significantly during storage independently of the harvesting stages. The decrease in acidity during storage of fruits observed by Fisk et al. (2006) and Krupa et al. (2011), who reported a lower TA after storage, is confirmed in this study. They also observed that the decrease in TA was in concomitant with a higher TSS, but it was not statistically significant, which is in agreement with previous finding.

The harvesting stages, cold storage and the interaction between two variables have a significant influence on TSS/TA of kiwifruits (Figure 4). Long-term storage significantly increased TSS/TA of kiwifruits, being dependent on harvesting stages. Generally, early harvesting stages have shown lower TSS/TA than later ones; the highest value was registered in fruits harvested with 9 and 10 °Brix after four months cold storage. Burdon et al. (2004) have shown that TSS content of ripe kiwifruit was



Figure 3. Titratable acidity (TA) in kiwifruits harvested in seven different maturity stages (°Bbrix, 6.5, 7, 7.5, 8, 8.5, 9 and 10) and after four months cold storage. The means followed by the same letters are not significantly different at  $P \le 0.01$ . The vertical bars are standard errors of the means (n=3)

found to reflect the eating quality of the ripe fruit. As it was previously reported, fruits harvested at higher °Brix value are generally considered more acceptable to consumers. Crisosto and Crisosto (2001) have shown that California - grown "Hayward" kiwifruit has acceptable flavour if the fruit contains >11.6% TSS. The increase in TSS content is due to the hydrolytic change in starch and conversion of starch to sugar, which is an important index of ripening process in fruits (Arthey and Ashurst, 2005). The increase in TSS/TA of fruit during storage was correlated with increase of TSS followed by declining of TA content. Arthey and Ashurst (2005) explained that activity of glycolytic enzymes caused starch degradation, and net starch to sucrose conversion. Therefore, the results of this study are consistent with finding of Langenkamper et al. (1998) who reported increase of the activity of these enzymes during the maturity time, regardless of storage temperature. Therefore, early harvest time lowers TSS/ TA compared to late harvest time.

#### Ascorbic acid

The harvesting stages, cold storage and their interaction significantly influenced ascorbic acid content (Figure 5). The data showed that ascorbic acid increased significantly with harvesting delay up to 8°Brix and thereafter declined; but after four months cold storage, late harvested fruits showed significantly higher ascorbic acid content compared to early harvests. The higher value was found in fruits harvested with 10 °Brix. Previous reports had shown that ascorbic acid in kiwifruit ranges between 25 and 155 mg ·100<sup>-1</sup> g of fresh weight (FW) of fruit (Kabaluk et al., 1997). Generally, ascorbic acid content declines when fruits become overripe with the degradation of fruit tissues concurrently (Kalt, 2005). However, Lee and Kader (2000) reported that ascorbic acid content increased with ripening in apricot, peach and papaya, but decreased in apple and mango. Tavarini et al. (2008) reported that ascorbic acid of kiwifruit harvested at 10 °Brix had not changed at end of a long storage period. In the present study, ascorbic acid content of kiwifruit significantly increased after four months cold storage. We also found that fruits harvested later showed a significantly higher content of ascorbic acid compared to early harvests.



Figure 4. TSS/TA of kiwifruits harvested in seven different maturity stages (°Brix, 6.5, 7, 7.5, 8, 8.5, 9 and 10) and after four months cold storage. The means followed by the same letters are not significantly different at  $P \le 0.01$ . The vertical bars are standard errors of the means (n=3)



Figure 5. Ascorbic acid of kiwifruits harvested in seven different maturity stages (°Brix, 6.5, 7, 7.5, 8, 8.5, 9 and 10) and after four months cold storage. The means followed by the same letters are not significantly different at  $P \le 0.01$ . The vertical bars are standard errors of the means (n=3)

# Phenolic compounds

Harvesting stages, cold storage separately, and in interaction significantly influenced phenolic compounds of kiwifruit (Figure 6). Phenolic content changed significantly in fruits collected at seven maturity stages and stored four months at 0°C. The data showed that phenolic content increased significantly with harvesting delay up to 8 °Brix and thereafter declined. In contrast, at the end of storage period fruits harvested later than 8 °Brix showed a significantly higher phenolic content than early harvests. Generally, phenolic content may either increase or decrease in fruits and vegetables depending on the storage conditions (Kalt, 2005). Tavarini et al. (2008) found that the change of phenols content in kiwifruits during storage depends on fruits maturity at harvesting time. They reported that in fruits collected approximately at 8 °Brix and stored for two months at 0°C a significant rise of phenolic compounds was observed. The same change was reported for total phenols in the peel of two apple cultivars during long-term cold storage (120 days at 1°C) (Leja et al., 2003). Gil et al. (2006) found that during nine days



Figure 6. Content of phenolic compounds of kiwifruits harvested in seven different maturity stages (°Brix, 6.5, 7, 7.5, 8, 8.5, 9 and 10) and after four months cold storage. The means followed by the same letters are not significantly different at  $P \le 0.01$ . The vertical bars are standard errors of the means (n=3)



Figure 7. Antioxidant capacity in kiwifruits harvested at seven different maturity stages (°Brix, 6.5, 7, 7.5, 8, 8.5, 9 and 10) and after four months cold storage. The means followed by the same letters are not significantly different at  $P \le 0.01$ . The vertical bars are standard errors of the means (n=3)

of storage, no significant changes occurred in phenolic content in kiwifruits and no difference was determined between slices and whole fruits.

#### Antioxidant capacity

Harvesting stages, cold storage, and their interaction significantly influenced antioxidant capacity (Figure 7). The highest values of antioxidant capacity in kiwifruits has been recorded when fruits were harvested with 6.5 °Brix and stored four months. These results suggested that the cold storage could negatively influence the antioxidant capacity of kiwifruits, which could be dependent on harvest time. This is in agreement with the results obtained by Tavarini et al. (2008), who reported that antioxidant capacity decreased significantly after four months of cold storage and that cold storage has a negative effect on antioxidant capacity of kiwifruits fruits harvested with 8.5 and 10 °Brix. In this study, we also found a negative effect of storage when fruits were harvested later than commercial maturity stage (6.5 °Brix). Shivashankara et al. (2004) reported that antioxidant capacity of Irwin Mango fruits unchanged up to 20 days of storage period and decreased thereafter. In 'Cortland' and 'Delicious' apples water-soluble antioxidants and anthocyanins declined but lipid-soluble antioxidants generally increased during storage time (Barden and Bramlage, 1994). The antioxidant capacity in the peel of two apple cultivars was determined during 120 days storage at 1°C and total antioxidant activity increased, irrespective of the storage conditions (Leja et al., 2003). Ferreyra et al. (2007) reported that antioxidant activity at commercial maturity decreased notoriously in strawberry fruit, in relation to the decrease in ascorbic acid and phenolics.

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