

Antioxidant contents and activity in SmartFresh-treated 'Empire' apples during air and controlled atmosphere storage

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Many consumers think that vitamin C (ascorbic acid) is the major antioxidant in fruits and vegetables, but in fact it is increasingly recognized that phenolic compounds often represent the majority of health-related antioxidant activity in plant products. The phenolic compounds also contribute to appearance, as well as taste and flavor. Apples are one of the best sources of antioxidant and phenolic compounds of all fruit, and are especially beneficial to our diet because they are available year-round. Research

“Our results show that 1-MCP has little effect on the concentrations of ascorbic acid, total phenolics, flavonoids and anthocyanins of Empire apples in either air or CA storage. Total antioxidant activity in peel tissues of fruit stored in air was sometimes higher in Smartfresh-treated fruit than untreated fruit. However, the health benefits of apples are realized primarily through phenolic concentrations and these compounds are stable after harvest. The bottom line is that ‘an apple a day keeps the doctor away,’ regardless of storage technology.”

from the Cornell laboratories of Drs. Cy Lee and Rui Hai Liu, as well as elsewhere, has shown that apples provide protection against cardiovascular disease and various cancers. Of the top 25 fruits consumed in the United States, apples are the number one source of phenolics in the American diet and provide Americans with 33% of the phenolics they consume (Boyer and Liu, 2004).

For apples, the major focus on health-related properties has been on phenolic compounds, including flavonoids such as the anthocyanins that are responsible for red color of the fruit. Apple varieties vary greatly in antioxidant components and individual phenolic compounds have different antioxidant potential. Interestingly, reactions among individual compounds may be synergistic, and therefore, total antioxidant activities potentially provide a better estimate of the overall contributions of antioxidant components than individual components alone. Phenolic and flavonoid contents are consistently higher in the skin than in the flesh, and peel tissues have the highest antioxidant activity and anti-proliferation activity.

In general, the total phenolic concentrations in both peel and flesh tissues of apples remain relatively stable during storage, although individual components may vary. The advent of

SmartFresh storage technology based on 1-methylcyclopropene (1-MCP), an inhibitor of ethylene perception, has raised the question about its effects on the nutritional quality of apple fruit. Knowledge about responses of fruit in both air and controlled atmosphere (CA) storage is important. CA can prolong the impact of 1-MCP on both physical and sensory responses of apple and the two technologies generally are most effective when used in combination.

To date, studies on the effects of 1-MCP on antioxidant components are limited. MacLean et al. (2006) found that 1-MCP treatment inhibited an increase in chlorogenic acid in peel tissues of Red Delicious apples during storage, and resulted in higher flavonoid concentrations. MacLean et al. (2003) also found that total antioxidant activity was higher in peels of 1-MCP-treated than untreated Empire and Red Delicious apples during storage. The total antioxidant activity (DPPH) of Golden Smoothie flesh was unaffected by 1-MCP treatment, but total ascorbic acid concentrations were slightly lower after 30 and 90 days of air storage (Vilaplana et al., 2006).

The objective of the current study was to investigate the effects of 1-MCP treatment during air and CA storage on antioxidant components and antioxidant activity of peel and flesh tissues of the Empire apple. This research was carried out as part of a PhD program and has been published in full (Fawbush et al., 2009). Here, we highlight the main findings of importance to the New York apple industry.

Materials and Methods

The Empire apple fruit used in these experiments were all harvested on the same day from mature trees growing at the Cornell University orchards at Lansing. The IEC, flesh firmness, SSC and starch index of the fruit at harvest were 2.7 ppm, 16.6 lb-f, 12.5% and 5.7 units, respectively. Fruit were randomly sorted into experimental units for either air or CA storage experiments.

For air storage, four replicates of 100 fruit were pre-cooled overnight at 33°F and then they were either untreated or treated with 1 ppm 1-MCP (SmartFresh powder) for 24 hours. Fruit were stored for up to five months. For CA storage, replicates of 55-60 fruits were cooled overnight at 36°F, and either untreated or treated with 1ppm 1-MCP. Four replicates of fruit for each treatment and temperature were stored in steel chambers, with 2 or 3% O₂ (with 2% CO₂). Final atmosphere regimes were established within 48 hours and were maintained within 0.2% of target atmospheres. CA storage was carried out for 9 months.

Ten fruit per treatment replicates were taken at harvest for assessment of internal ethylene concentration (IEC), flesh firmness, soluble solids concentration (SSC) and starch pattern indices. IEC and firmness were measured on 10 fruit replicates after 1, 2, 3, 4 and 5 months for the air-stored fruit, and after 4.5 and 9 months for the CA stored fruit, plus 1 day at 68°F. At the last removal from storage, all remaining fruit were kept at 68°F for 7 days and then assessed for disorders.

For extraction of phenolic and other compounds, 10 fruit per treatment replicate were taken on the day of removal of fruit from air and CA storage. Total phenolics, flavonoids, anthocyanins, total antioxidant activity and total ascorbic acid were measured on apple peel and flesh using standard procedures as described by Fawbush et al. (2009).

Results

Air storage. 1-MCP prevented any increase in the internal ethylene concentrations (IEC) during storage, while the IEC in untreated fruit reached 43.9 ppm after 5 months of storage (Figure 1). Untreated fruit softened during this time to 11.4 lb-f, compared with 14.0 lb-f in 1-MCP treated fruit (Figure 1). No storage disorders were observed in air stored fruit.

At harvest, the phenolic concentrations in peel and flesh tissues were 2.82 and 1.24 g kg⁻¹, respectively (Figure. 2). Overall, the phenolic concentration was significantly higher (2.56 g kg⁻¹) in peel tissues of 1-MCP treated fruit than in those of untreated fruit (2.16 g kg⁻¹). In the flesh, total phenolic concentrations of untreated fruit were significantly higher at 0.90 g kg⁻¹ than the 0.84 g kg⁻¹ of 1-MCP treated fruit. No effect of storage time was detected for either tissue type.

Total flavonoid and anthocyanin concentrations in the peel were affected only by storage time, but the patterns of change were inconsistent (results not shown). In flesh tissues, total flavonoids were not affected by either 1-MCP treatment or storage time.

At harvest, the total ascorbic acid concentrations in the peel and flesh were 0.55 and 0.11 g kg⁻¹, respectively, and these concentrations declined in both untreated and 1-MCP-treated fruit during storage (Figure 3). There was no significant effect of 1-MCP treatment on peel concentrations, although in the flesh tissues, the total ascorbic acid concentrations were slightly lower in 1-MCP-treated tissues than untreated tissues, averaging 0.07 and 0.06 g kg⁻¹, respectively. However, effects were evident at only some time points.

The total antioxidant activity in the peel was 2.82 mmol kg⁻¹ at harvest, and overall, the activity in peel tissue from 1-MCP treated fruit was 2.64 mmol kg⁻¹, compared with 2.12 mmol kg⁻¹ in untreated fruit. However, differences between treatments were evident only at months 1 to 3 (Figure 3). Total antioxidant activity in flesh tissues was also significantly higher in 1-MCP treated fruit than untreated fruit, averaging 1.14 and 0.95 mmol kg⁻¹, respectively. Total antioxidant activity was not affected by storage time.

CA storage. The IEC was much lower, and firmness higher, in 1-MCP treated fruit than in untreated fruit (Table 1). No external or internal disorders were detected after 4.5 months of CA storage, but a high incidence of flesh browning was observed in fruit after 9 months of storage.

The total phenolics, total flavonoid and ascorbic acid concentrations in peel and flesh tissues were not consistently affected by 1-MCP treatment (Tables 2, 3 and 4). Total anthocyanin

concentrations in peel tissues were also unaffected by 1-MCP treatment (data not shown).

The total antioxidant activity in peel tissues was not affected by 1-MCP (Table 5), but the total antioxidant activity was higher in flesh tissues of 1-MCP treated than untreated fruit, being 1.40 and 1.25 mmol kg⁻¹, respectively. Interactions among all storage factors were detected, however, and overall trends were inconsistent.

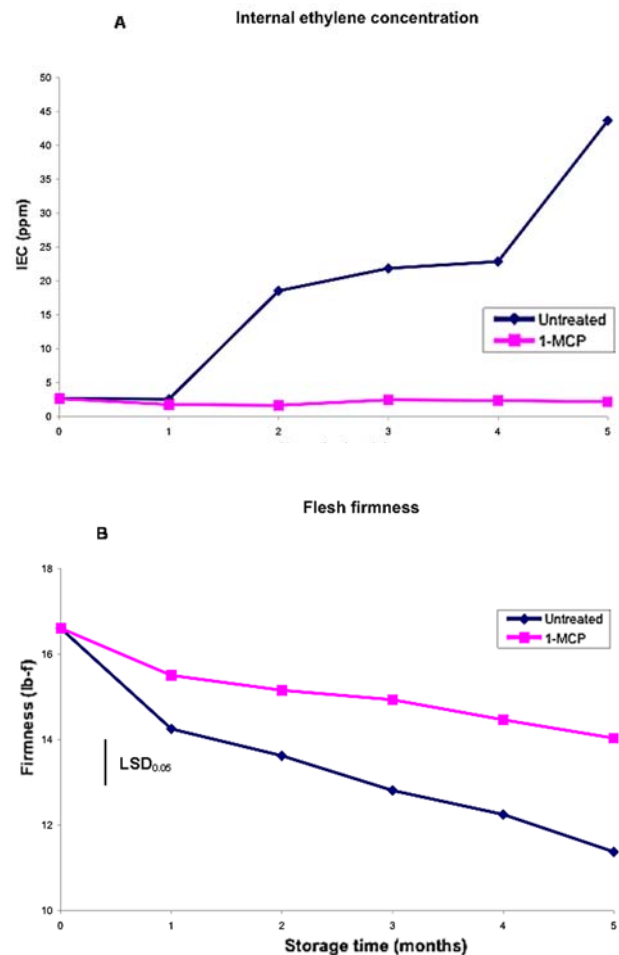


Figure 1. Internal ethylene concentrations (A) and flesh firmness (B) of Empire apples either untreated or treated with 1 ppm 1-MCP and stored at 33°F in air for up to 5 months.

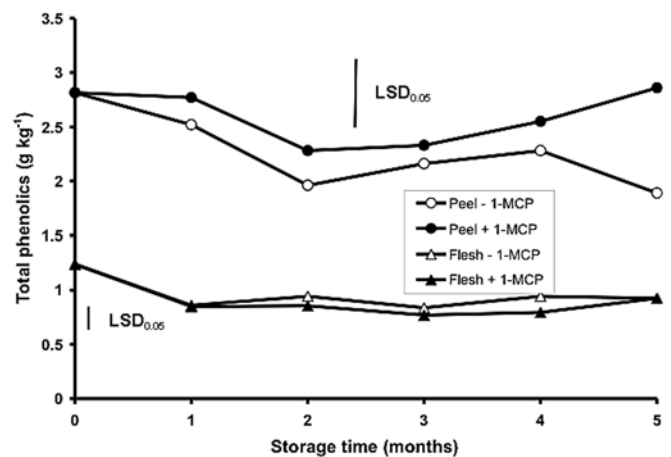


Figure 2. Total phenolic concentrations (gallic acid equivalents, mg kg⁻¹) in peel and flesh tissues of Empire apples either untreated or treated with 1 ppm 1-MCP and stored at 33°F in air for up to 5 months.

Discussion

The effects of storage conditions on antioxidants in the absence of 1-MCP treatment have been well studied, and in general, total phenolics, total antioxidant activity and radical scavenging capacity are stable or increase during storage. The results of our study also largely indicate that concentrations of total phenolics, flavonoids, anthocyanins and total antioxidant activity are

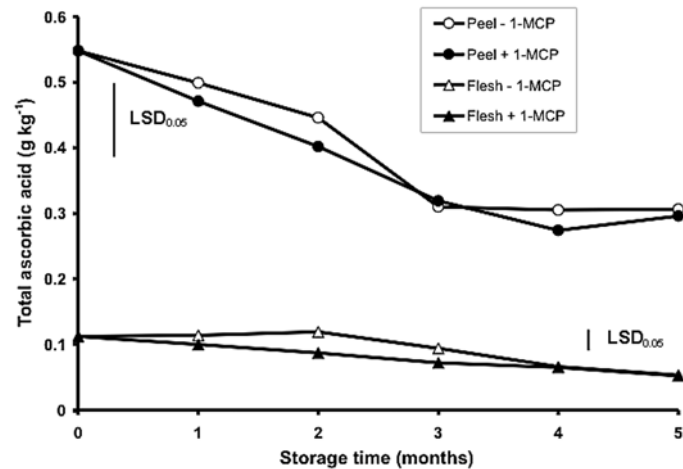


Figure 3. Total ascorbic acid concentrations (mg kg^{-1}) in peel and flesh tissues of Empire apples either untreated or treated with 1 ppm 1-MCP and stored at 33°F in air for up to 5 months.

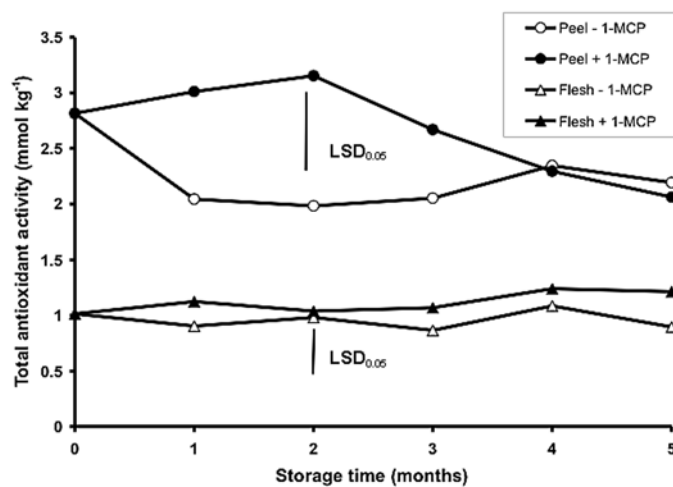


Figure 4. Total antioxidant activity (vitamin C equivalents, mmol kg^{-1}) in peel and flesh tissues of Empire apples either untreated or treated with 1 ppm 1-MCP and stored at 33°F in air for up to 5 months.

Table 1. Internal ethylene concentrations (IEC), flesh firmness and flesh browning of Empire apples, either untreated or treated with 1 ppm 1-MCP, and stored in controlled atmospheres of 2% or 3% oxygen with 2% carbon dioxide at 36°F for 4.5 and 9 months. Different letters associated with means indicate that there are differences between treatments.

Storage time (months)	1-MCP	IEC (ppm)		Firmness (lb-f)		Flesh browning (%)	
		2% O ₂	3% O ₂	2% O ₂	3% O ₂	2% O ₂	3% O ₂
4.5	-	167a	216a	13.0c	11.7d	0	0
	+	2.3c	1.5c	15.3a	15.4a	0	0
9	-	234a	170a	11.9d	9.0e	84a	60b
	+	32b	56b	14.3b	14.4b	84a	87a

Table 2. Total phenolic concentrations (gallic acid equivalents, g kg^{-1}) in peel and flesh tissues of Empire apples, either untreated or treated with 1 ppm 1-MCP, and stored in controlled atmospheres of 2% or 3% oxygen with 2% carbon dioxide at 36°F for 4.5 and 9 months. Different letters associated with means indicate that there are differences between treatments for a tissue type.

Storage time (months)	1-MCP	Peel		Flesh	
		2% O ₂	3% O ₂	2% O ₂	3% O ₂
4.5	-	2.56a	1.81g	0.96bc	1.01abc
	+	2.47b	2.27d	0.90c	1.12a
9	-	2.37c	2.12e	1.08ab	0.66d
	+	1.90f	2.15e	0.98bc	1.01abc

Table 3. Total flavonoid concentrations (catechin equivalents, g kg^{-1}) in peel and flesh tissues of Empire apples, either untreated or treated with 1 ppm 1-MCP, and stored in controlled atmospheres of 2% or 3% oxygen with 2% carbon dioxide at 36°F for 4.5 and 9 months. Different letters associated with means indicate that there are differences between treatments for a tissue type.

Storage time (months)	1-MCP	Peel		Flesh	
		2% O ₂	3% O ₂	2% O ₂	3% O ₂
4.5	-	0.91a	0.88a	0.39b	0.49ab
	+	1.01a	1.14a	0.56a	0.42b
9	-	1.12a	1.17a	0.49ab	0.44b
	+	0.92a	1.19a	0.54a	0.49ab

Table 4. Total ascorbic acid concentrations (g kg^{-1}) in peel and flesh tissues of Empire apples, either untreated or treated with 1 ppm 1-MCP, and stored in controlled atmospheres of 2% or 3% oxygen with 2% carbon dioxide at 36°F for 4.5 and 9 months. Different letters associated with means indicate that there are differences between treatments for a tissue type.

Storage time (months)	1-MCP	Peel		Flesh	
		2% O ₂	3% O ₂	2% O ₂	3% O ₂
4.5	-	0.37a	0.35a	0.11a	0.09bc
	+	0.35a	0.31a	0.10ab	0.09bc
9	-	0.38a	0.33a	0.10ab	0.09bc
	+	0.36a	0.33a	0.09bc	0.08c

Table 5. Total antioxidant activity (vitamin C equivalents, mmol kg^{-1}) in peel and flesh tissues of Empire apples, either untreated or treated with 1 ppm 1-MCP, and stored in controlled atmospheres of 2% or 3% oxygen with 2% carbon dioxide at 36°F for 4.5 and 9 months. Different letters associated with means indicate that there are differences between treatments for a tissue type.

Storage time (months)	1-MCP	Peel		Flesh	
		2% O ₂	3% O ₂	2% O ₂	3% O ₂
4.5	-	3.04a	2.66abc	1.32bc	1.52b
	+	2.55abcd	2.72abc	1.86a	1.03d
9	-	2.07cd	2.84ab	1.28bcd	1.32bc
	+	1.91d	2.37bcd	1.19cd	1.16cd

relatively stable during air and CA storage. However, the effects of 1-MCP on individual phytochemical groups were variable. In air-stored fruit, total phenolic concentrations were higher in peel tissues, but lower in flesh tissues, of 1-MCP treated fruit compared with untreated fruit. No effects of 1-MCP were found for total flavonoid or anthocyanin concentrations, except that flavonoid concentrations were higher in 1-MCP treated fruit than untreated fruit during CA storage.

Total antioxidant activity, assayed using a recently developed method (Adom and Liu, 2005), was higher in both peel and flesh tissues of 1-MCP-treated fruit compared with untreated fruit, although higher only in flesh tissues of CA-stored fruit. Higher total antioxidant activity in 1-MCP treated peel tissues of air-stored Empire and Delicious apples was also found by MacLean et al. (2003). The reasons for higher antioxidant activity in air-stored 1-MCP-treated fruit are uncertain. Both total phenolic concentrations and total antioxidant activity were higher in peel tissues, and these two factors have been strongly correlated with each other in other studies.

Ascorbic acid concentrations declined in both peel and flesh tissues during air storage, while in CA storage, patterns of change over time were inconsistent, depending on the O₂ concentration. Surprisingly little information is available concerning changes in ascorbic acid concentrations in apple fruits during storage, especially in CA, and even less is known about the effects of 1-MCP. The significance of decreased ascorbic acid concentrations during storage with and without 1-MCP-treated fruit in this study and others is uncertain. Although ascorbic acid is generally considered to be important in nutrition, it represents a minor component of the total antioxidant activity of apples. Ascorbic acid, however, is a critical component of antioxidative processes in plant cells, interacting enzymatically and non-enzymatically with damaging oxygen radical and reactive oxygen species.

Conclusion

1-MCP delays fruit ripening of Empire apples, but the effects of treatment on phytochemical groups are relatively small. The results show that eating apples, regardless of storage technology, is the right thing to do to keep the doctor away!

Acknowledgments

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Fanjaniana Fawbush was a graduate student working with Chris Watkins. **Jackie Nock** is a research support specialist who works with Chris Watkins. **Chris Watkins** is a research and extension professor in the Department of Horticulture at Cornell's Ithaca Campus who leads Cornell's program in postharvest biology of fruit crops.

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