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Responses of 'Honeycrisp' Apples to Short-term Controlled Atmosphere Storage Established During Temperature Conditioning

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Abstract. **'Honeycrisp' apples are susceptible to bitter pit, a physiological disorder that impacts peel and adjacent cortex tissue. 'Honeycrisp' is also susceptible to chilling injury (CI) that can be prevented by holding fruit at 10 to 20 °C after harvest for up to 7 days. This temperature conditioning period reduces CI risk but can enhance bitter pit development. Previous research demonstrated a controlled atmosphere (CA) established during conditioning can reduce 'Honeycrisp' bitter pit development without inducing other physiological disorders. The objective of this research was to evaluate the duration of CA needed to reduce bitter pit development. Experiments were conducted in 2014, 2016, and 2017 with fruit obtained from commercial orchards in Washington State and, in 2017 only, Ontario, Canada. Half the fruit were treated with 42 μmol·L^{-1} 1-methycyclopropene (1-MCP) for 24 hours at 10 °C immediately following harvest. The untreated fruit were held at the same temperature (10 °C) in a different cold room. Following 1-MCP treatment, all fruit were conditioned at 10 °C for an additional 6 days, then fruit was cooled to 2.8 °C. During conditioning, fruit were held in air or CA (2.5 kPa O_2, 0.5 kPa CO_2) established 1 day after harvest, for 1 to 8 weeks, then in air. All fruit were removed from cold storage after 4 months and then held 7 days at 20 °C. Fruit from most orchards/years stored in CA developed less bitter pit compared with fruit stored continuously in air. CA during conditioning also reduced poststorage peel greasiness but CA for 2 weeks or longer enhanced cortex cavity development in some orchard lots. Treatment with 1-MCP did not reduce bitter pit but enhanced development of peel leather blotch and core browning for some orchards/years. 1-MCP–treated fruit slowed the loss of soluble solids content, titratable acidity, and reduced internal ethylene concentration. Results suggest the potential for postharvest management of bitter pit development in 'Honeycrisp' apples by CA established during conditioning with minimal development of other postharvest disorders.**

'Honeycrisp' apples (*Malus ×domestica*) are a high-value cultivar with a desired crisp texture and a distinct flavor profile. These characteristics make this cultivar popular among consumers (Luby and Bedford, 1992; Mann et al., 2005; Yue and Tong, 2011). Increased 'Honeycrisp' production (U.S. Apple Association, 2018) has resulted in a need for strategies to improve the storage performance to maintain quality and reduce fruit losses. One challenge is controlling bitter pit, a physiological disorder that is associated with fruit calcium content (Miqueloto et al., 2014; Rosenberger et al., 2004). Economic losses resulting from bitter pit can be considerable, and incidence is often unpredictable annually, between orchards in the same region, or even tree-to-tree. Bitter pit symptoms are characterized as depressed brown lesions in and just beneath the peel within the first 5 mm, especially in the distal portion of the fruit (Amarante et al., 2006, 2013; Cobb, 1895; Ferguson and Watkins, 1983, 1989; Freitas et al., 2010; Garman and Mathis, 1956; Jaeger, 1869). Bitter pit may be visible at harvest, but typically develops during the initial storage period. Factors increasing susceptibility of 'Honeycrisp' to bitter pit are hot, dry growing conditions (Watkins, 2009), young trees (less than 5 years old) (Rosenberger et al., 2004), excessive vegetative vigor (Wünsche and Ferguson, 2005), large fruit size (Telias et al., 2006), fruit nutrient content (Rosenberger et al., 2004; Telias et al., 2006; Torres et al., 2017), fruit maturity at harvest (Prange et al., 2011), and crop load (Robinson and Watkins, 2009; Serra et al., 2016). Bitter pit can be more severe in fruit picked immature compared with fruit harvested more mature (Le Grange et al., 1998; Perring and Pearson, 1986; Prange et al., 2011; Volz et al., 1993).

Practices that can reduce 'Honeycrisp' bitter pit development include field-applied calcium (Biggs and Peck, 2015; Peryea et al., 2007; Rosenberger et al., 2004), optimal crop load (Delong et al., 2006; Robinson and Lopez, 2012) and harvest at optimal maturity (Prange et al., 2011). After harvest, bitter pit development can be reduced by calcium dips (Reid and Padfield, 1975) as well as CA storage (Hewett, 1984; Sharples, 1982; Webster and Forsyth, 1979). A short period of CA that creates low oxygen stress established during temperature conditioning also can reduce bitter pit development (Pesis et al., 2010) as can a nonstress CA that is continued throughout cold storage (Mattheis et al., 2017).

The ethylene action inhibitor (1-MCP) prevents ethylene-mediated ripening processes in apple fruit (Fan et al., 1999a, 1999b). This treatment also impacts development of fruit physiological disorders including senescent breakdown, CO_2 injury, CI, and superficial scald (Blankenship and Dole, 2003; Contreras et al., 2014; DeEll, 2010; DeEll et al., 2015; Fan et al., 1999b; Watkins, 2008; Watkins and Nock 2012). Reduction in bitter pit following 1-MCP treatment and/or CA established during 'Honeycrisp' conditioning has been reported (Mattheis et al., 2017; Mirzaee et al., 2014). However, other studies reported inconsistent effects of postharvest 1-MCP treatments on apple bitter pit development (Gago et al., 2015; Mirzaee et al., 2015). Postharvest 1-MCP application to 'Honeycrisp' apples can affect fruit quality by limiting soluble solids content (SSC) and titratable acidity (TA) loss during air storage (DeEll, 2010). Postharvest applications of 1-MCP also can increase CA-related internal disorders such as CO_2 injury, cavities, and internal browning (DeEll, 2010; Watkins and Nock, 2012).

'Honeycrisp' is a chilling sensitive cultivar and can develop soft scald and soggy breakdown when cooled immediately after harvest (Watkins and Rosenberger, 2000). However, the use of a temperature conditioning period at a relatively warm storage

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temperature (10 to 20 °C) after harvest for up to 7 d followed by storage at 3 °C can reduce the development of CI during cold storage (Contreras et al., 2014; Delong et al., 2004, 2006, 2009; Watkins et al., 2004). Impacts of delaying cooling can be cultivar specific and are known to reduce (Argenta et al., 2000; de Castro et al., 2007) or enhance disorder development (DeEll et al., 2016; Neven et al., 2000; Watkins et al., 2004).

Storage temperature and CA gas composition affect apple physiological disorder development (Watkins and Liu, 2010). Critical O_2 and CO_2 concentrations that can lead to injury vary with cultivar (Gran and Beaudry, 1993) and temperature can influence fruit response to CA as respiration rate increases with temperature. 'Honeycrisp' apples can be sensitive to injury during CA (Contreras et al., 2014; DeEll et al., 2015; Watkins and Nock, 2012), with delayed establishment of CA known to reduce injury development risk (DeEll et al., 2016).

Commercially, conditioning is a typical 'Honeycrisp' management practice regardless of how long fruit will be stored. As some fruit are marketed soon after conditioning is completed, the duration of CA established during conditioning that is necessary to reduce bitter pit development is a relevant commercial issue. The objective of this study was to determine if a short period of CA initiated during conditioning with or without previous 1-MCP treatment affects the development of bitter pit and other physiological disorders as well as fruit quality of 'Honeycrisp' apples.

Materials and Methods

Plant material and postharvest treatments. 'Honeycrisp' apples were obtained in Washington State (WA) at commercial harvest from two orchards in 2014 (lots A and B), two in 2016 and 2017 (lots C and D), and an additional lot in 2017 (lot E) in Ontario, Canada (ON) (Table 1). Apples without external disorders were selected the day of harvest and placed onto pressed fiber trays (WA) or into plastic containers (ON). All fruit were held at 10 °C and some were exposed to 42 $\mu mol \cdot L^{-1}$ 1-MCP (AgroFresh, Inc., Spring House, PA) for 24 h in an 800-L gas-tight metal cabinet (WA) or an air-tight treatment tent (ON; DeEll and Lum, 2017). Control fruit were not held in the same areas where 1-MCP application was performed. After 24 h, fruit was removed from the treatment chamber or tent and moved to the same cold room as controls. All fruit were held at 10 °C for 6 additional days, then the storage temperature was reduced to 2.8 °C. During conditioning, fruit were stored in air or in CA (2.5 kPa O_2, 0.5 kPa CO_2) established the day after harvest. WA fruit were stored in 0.14-m^3 CA chambers for up to 8 (2014) or 2 weeks (2016, 2017). The CA system was operated as described previously (Mattheis et al., 2017). ON fruit were stored in boxes in air or CA as described in DeEll and Lum (2017). After CA, all fruit were held in air for 4 months followed by 7 d at 20 °C after removal from cold storage.

Harvest maturity and fruit quality assessment. Fruit maturity and quality were evaluated from a random sample of 10 (ON) or 16 (WA) fruit on the day of harvest. Nondestructive (weight, fruit size, peel background color) and destructive (firmness, starch score, SSC, TA, internal disorders) assessments were as previously described (DeEll and Lum, 2017; Mattheis et al., 2017). Internal ethylene concentration (IEC) of ON fruit was determined by withdrawing a 3-mL gas sample from the core of each fruit using a syringe and injecting the gas sample into a Agilent 7820A gas chromatograph (Agilent Technologies Canada Inc., Mississauga, Ontario, Canada) equipped with a 0.25-mL sample loop, flame ionization detector, and 25 m × 0.53 mm CarboBOND capillary column (Agilent Technologies Canada Inc.). The injector, column, and detector temperatures were 150, 80, and 250 °C, respectively. High-purity helium was used as the carrier gas at a flow rate of 0.46 $mL \cdot s^{-1}$ with a typical run time of 1.5 min. For WA fruit, ethylene in a 0.5-mL gas sample removed from the fruit core was analyzed using a G1530a gas chromatograph (Agilent Technologies, Wilmington, DE) fitted with a 500 mm × 3.2 mm glass column packed with Porapak Q (Supelco, Bellafonte, PA) and a flame ionization detector (FID). The injector, oven, and FID temperatures were maintained at 100, 35, and 300 °C, respectively. The N_2 carrier, H_2, and airflow were 5.0, 0.5, and 5 $mL \cdot s^{-1}$, respectively.

Quality parameters and external and internal disorders including bitter pit were rated on each fruit after 4 months of storage plus 7 d at 20 °C. Physiological disorders (bitter pit, leather blotch, lenticel breakdown, senescent browning, core browning, internal browning, cavities, greasiness, soft scald, and soggy breakdown) are reported as incidence. Bitter pit was determined as surface lesions <5 mm in diameter with underlying brown, corky tissue. Irregularly shaped peel areas of rough, brown tissue >5 mm were rated as leather blotch.

Experimental design and statistical analysis. The experimental design was a 2 × 5 (2014) or 2 × 3 (2016, 2017) factorial with two treatments (control, 1-MCP) and weeks of CA (0, 1, 2, 4, 8 weeks in 2014; 0, 1, 2 weeks in 2016 and 2017). Data were analyzed using SAS 9.4 (SAS Institute Inc., Cary, NC). All data were subjected to testing of normality and assumptions for analysis of variance (ANOVA). Storage disorders were expressed as the percentage of fruit affected per replicate and percentage data were transformed before analysis. When interactions between factors were significant ($P < 0.05$), data for each storage treatment replicate were combined across repeated measures to determine differences among treatments using Fisher's least significance difference. In the case of fruit quality and disorder incidence after 4 months in storage, a two-way ANOVA (PROC GLM) was conducted to determine treatment and storage atmosphere effect and possible interactions between treatment × atmosphere.

Results and Discussion

Fruit maturity and quality at harvest varied with lot and harvest year (Table 1). Both WA lots in 2014 and WA lot D in 2017 had the highest (yellowest) peel ground color and WA lots in 2016 and the ON lot in 2017 had the lowest (greenest). Firmness, SSC, TA, and starch index values were typical for 'Honeycrisp' at harvest in Ontario and Washington State (Moran et al., 2010; Serra et al., 2016). IEC greater than 1 $\mu L \cdot L^{-1}$ for all lots indicated that the fruit were physiologically mature despite relatively green peel background color for some lots. Harvest timing in relation to maturity can influence the susceptibility of apples to many physiological disorders, including bitter pit (Ferguson and Watkins, 1989). In addition, 'Honeycrisp' large fruit size can increase bitter pit risk (Serra et al., 2016). Although each orchard lot had reached commercial maturity, fruit quality varied among lots. 'Honeycrisp' fruit maturity and quality at harvest affects storage life and postharvest fruit quality (Prange et al., 2011; Serra et al., 2016). In this

Table 1. 'Honeycrisp' fruit maturity and quality at harvest. Fruit from commercial orchards in Ontario, Canada (ON) and Washington State (WA).

Lot	Location	Yr	IEC $\mu L \cdot L^{-1}$	Wt g	PBC 1–5	Firmness N	SSC %	TA %	Starch 1–6
A	WA	2014	12.5 b	273 b	3.3 a	60.4 cd	14.4 a	0.53 c	4.6 ab
B	WA	2014	4.9 bc	243 d	3.6 a	62.6 c	13.5 b	0.52 c	5.0 a
C	WA	2016	20.8 a	323 a	1.2 c	60.8 cd	14.3 a	0.55 c	5.0 a
D	WA	2016	2.9 c	237 d	1.5 c	59.1 d	12.1 c	0.48 d	4.0 b
C	WA	2017	12.3 b	272 bc	2.4 b	59.8 cd	13.4 b	0.62 b	5.2 a
D	WA	2017	2.0 c	244 cd	3.4 a	75.5 a	14.2 a	0.79 a	4.6 ab
E	ON	2017	7.6 bc	226 d	1.2 c	66.7 b	11.9 c	0.62 b	4.2 b

Values are means, n = 16 (weight, starch pattern index, firmness, SSC Ontario lot, IEC); n = 8 (SSC, TA, WA lots), n = 4 (TA Ontario lot). Means within columns followed by different letters are significantly different, Fisher's least significant difference ($LSD_{0.05}$).
Wt = weight; PBC = peel background color; SSC = soluble solids content; TA = titratable acidity; IEC = internal ethylene concentration.

study, all the fruit was harvested 1 or 2 d before the commercial harvest date. Among lots and harvest years, fruit weight varied significantly, ranging from ≈210 g to 350 g. Mean starch rating was between 4.0 and 5.2 for all lots and SSC was between 12.1% and 14.4%.

Bitter pit occurred in all lots, and incidence in fruit continuously stored in air and not treated with 1-MCP ranged from 6% (lot D 2016) to 61% (Lot C 2016) (Table 2). Incidence was mostly expressed within 1 month of harvest (data not presented) similar to a previous report (Mattheis et al., 2017). Bitter pit incidence was highest in fruit with average weight at harvest exceeding 270 g, consistent with previous literature (Robinson and Watkins, 2009; Serra et al., 2016; Telias et al., 2006) linking 'Honeycrisp' fruit size and bitter pit susceptibility. Bitter pit incidence was affected by CA or 1-MCP treatment alone in four of the seven lot/years or one of the five lot/years, respectively. The CA × 1-MCP interaction was significant in two of the five lot/years. Results in 2014 indicated CA longer than 2 weeks did not enhance bitter pit reduction, therefore subsequent experiments were limited to 1 or 2 weeks CA. Overall, mean bitter pit incidence for fruit stored continuously in air was 27% for control fruit and 20% for 1-MCP fruit (Table 3). Fruit stored in CA had less bitter pit (16% controls, 13% 1-MCP). The 1-MCP treatment was not significant for the combined data set, but CA during conditioning reduced bitter pit development (Fig. 1). Overall, there was no difference in bitter pit between fruit stored 1 or 2 weeks in CA.

The 1-MCP and/or CA established during conditioning affected development of other physiological disorders. Leather blotch developed on several lot/years on controls as well as 1-MCP–treated fruit. CA during conditioning of lot A enhanced leather blotch development compared with fruit continuously stored in air; this was the only result of this type. Leather blotch incidence was observed only on 1-MCP–treated fruit from lot C in both years, and was enhanced by 1-MCP treatment of lot E. Consistent with Mattheis et al. (2017), leather blotch incidence was lower compared with bitter pit as fruit with leather blotch also had bitter pit symptoms. Factors influencing leather blotch development are not completely understood; however, leather blotch incidence on 1-MCP–treated 'Honeycrisp' fruit is consistent with previous research in which symptoms developed from bitter pit lesions, but bitter pit was not always

Table 2. Mean disorder incidence of 'Honeycrisp' apples by orchard lot and harvest year after cold storage. Fruit was obtained from four orchards in Washington State, two lots in 2014, and two different lots in 2016 and 2017, and one lot in Ontario, Canada, in 2017. Control and fruit treated with 42 μmol·L^{-1} 1-methycyclopropene (1-MCP) after harvest were held 7 d at 10 °C then at 2.8 °C. Fruit were held in air or a controlled atmosphere (CA) (2.5% O_2, 0.5% CO_2) for up to 8 weeks in 2014 or 1 or 2 weeks in 2016 and 2017. Fruit were evaluated after 4 mo. in cold storage plus 7 d at 20 °C.

	Weeks in CA					*P* value
Lot A	0	1	2	4	8	Weeks
Bitter pit	41 a	31 ab	14 b	17 b	19 b	*
Leather blotch	0 b	17 a	14 a	11 ab	14 a	*
Lenticel breakdown	0	0	0	0	0	NS
Senescent breakdown	0	0	0	0	0	NS
Core browning	0	0	0	0	0	NS
Internal browning	0	0	0	3	0	NS
Cavities	0 b	0 b	0 b	3 b	8 a	*
Greasy peel	0	3	0	0	0	NS
Soft scald	0	0	0	0	6	NS
Soggy breakdown	0	0	0	3	3	NS

	Weeks in CA					*P* value
Lot B	0	1	2	4	8	Weeks
Bitter pit	17 a	6 ab	3 b	3 b	3 b	*
Leather blotch	0	0	3	3	0	NS
Lenticel breakdown	0	0	0	0	0	NS
Senescent breakdown	0	0	0	0	0	NS
Core browning	0	0	0	0	0	NS
Internal browning	0	0	0	0	0	NS
Cavities	0b	3 ab	6 ab	6 ab	14 a	*
Greasy peel	100 a	0 b	0 b	0 b	0 b	***
Soft scald	3	3	11	11	17	NS
Soggy breakdown	0 b	6 ab	8 ab	14 ab	19 a	*

Lot C	Control weeks in CA			1-MCP weeks in CA			*P* value		
2016	0	1	2	0	1	2	Treat.	Weeks	T × W
Bitter pit	61	39	34	48	40	33	NS	**	NS
Leather blotch	0	0	0	26	31	26	***	NS	NS
Lenticel breakdown	0	0	0	0	0	0	NS	NS	NS
Senescent breakdown	0	0	0	0	0	0	NS	NS	NS
Core browning	0	0	0	0	0	0	NS	NS	NS
Internal browning	16	8	11	14	3	0	*	**	NS
Cavities	0	0	5	0	0	1	NS	NS	NS
Greasy peel	93	85	23	85	65	46	NS	**	NS
Soft scald	0	0	0	0	0	0	NS	NS	NS
Soggy breakdown	0	0	0	0	0	0	NS	NS	NS

Lot D	Control weeks in CA			1-MCP weeks in CA			*P* value		
2016	0	1	2	0	1	2	Treat.	Weeks	T × W
Bitter pit	6	0	3	3	5	6	NS	NS	NS
Leather blotch	0	0	0	0	0	0	NS	NS	NS
Lenticel breakdown	0	0	0	0	0	0	NS	NS	NS
Senescent breakdown	0	0	0	0	0	0	NS	NS	NS
Core browning	0	0	0	0	0	0	NS	NS	NS
Internal browning	0	0	0	0	0	0	NS	NS	NS
Cavities	0	0	0	0	0	0	NS	NS	NS
Greasy peel	0	2	2	0	0	0	NS	NS	NS
Soft scald	0	0	0	0	0	0	NS	NS	NS
Soggy breakdown	0	0	0	0	0	0	NS	NS	NS

Lot C	Control weeks in CA			1-MCP weeks in CA			*P* value		
2017	0	1	2	0	1	2	Treat.	Weeks	T × W
Bitter pit	25	14	18	18	8	14	NS	NS	NS
Leather blotch	0 b	0 b	0 b	1 b	3 b	11 a	***	**	**
Lenticel breakdown	5	4	8	4	4	0	NS	NS	NS
Senescent breakdown	0	0	0	0	0	0	NS	NS	NS
Core browning	0	0	0	0	0	0	NS	NS	NS
Internal browning	0	1	0	0	0	0	NS	NS	NS
Cavities	0	0	0	0	0	0	NS	NS	NS
Greasy peel	99 a	58 b	39 b	14 c	18 c	25 c	***	*	***
Soft scald	0	0	0	0	0	0	NS	NS	NS
Soggy breakdown	0	0	0	0	0	0	NS	NS	NS

D	Control weeks in CA			1-MCP weeks in CA			*P* value		
2017 harvest	0	1	2	0	1	2	Treat.	Weeks	T × W
Bitter pit	9 b	9 b	11 b	21 a	8 b	3 b	NS	NS	*
Leather blotch	0	0	0	1	1	0	NS	NS	NS
Lenticel breakdown	1	1	2	2	3	3	NS	NS	NS
Senescent breakdown	1	0	1	0	0	0	NS	NS	NS
Core browning	0	0	0	0	0	0	NS	NS	NS
Internal browning	0	0	0	0	0	0	NS	NS	NS
Cavities	1	0	4	2	1	3	NS	NS	NS
Greasy peel	29	27	8	14	8	3	**	*	NS
Soft scald	1	0	0	1	0	1	NS	NS	NS
Soggy breakdown	4	0	2	1	2	3	NS	NS	NS

E	Control weeks in CA			1-MCP weeks in CA			*P* value		
2017 harvest	0	1	2	2	1	2	Treat.	Weeks	T × W
Bitter pit	28 a	7 b	10 b	2 b	6 b	2 b	***	**	***
Leather blotch	1	5	9	10	15	6	NS	NS	NS
Lenticel breakdown	6	4	7	6	3	2	NS	NS	NS
Senescent breakdown	1	0	1	0	0	0	NS	NS	NS
Core browning	0	0	0	1 6	10	13	***	NS	NS
Internal browning	0	0	0	0	0	1	NS	NS	NS
Cavities	0	0	2	0	4	1	NS	NS	NS
Greasy peel	84 a	53 b	0 d	33 bc	14 cd	0 d	***	***	***
Soft scald	0	0	0	1	1	0	NS	NS	NS
Soggy breakdown	1	0	2	0	1	1	NS	NS	NS

NS, *, **, ***Nonsignificant or significant treatment effects at $P < 0.05$, 0.01, or 0.001, respectively (Fisher's least significant difference).

accompanied by leather blotch (Mattheis et al., 2017). Figure 2 illustrates the sequential development of leather blotch symptoms on a fruit treated with 1-MCP compared with an untreated fruit with bitter pit symptoms that did not develop into leather blotch. The progression of lesion size from that typical of bitter pit to a larger lesion with appearance different compared with bitter pit suggests initial metabolic events resulting in these disorders may be similar. The progression of bitter pit to leather blotch supports a hypothesis that the absence of ethylene action results in enhanced bitter pit symptom progression resulting in larger lesions. Results for lot A also may be consistent with this hypothesis, as low oxygen reduces ethylene action (Burg and Burg, 1967). although DeEll et al. (2016) observed blotch increased as CA was delayed.

Incidence of peel lenticel breakdown and senescent, core, and internal browning were low in most lots and not affected by 1-MCP or CA. In lot C/2016, internal browning was reduced in 1-MCP–treated fruit held in CA. Core browning was higher in 1-MCP–treated fruit compared with controls in ON fruit, but CA during conditioning was not a significant factor for this disorder. Watkins and Nock (2012) previously reported less core browning in 1-MCP–treated fruit relative to controls in which orchard susceptibility to core browning was high. Core browning in ON fruit had a defined and translucent appearance (Fig. 3).

Previous research indicates that conditioning and the delay of CA storage can reduce the incidence of internal CO_2 injuries, such as cortex browning, but the delay may aggravate other disorders, such as lenticel breakdown, bitter pit/blotch, and greasiness (DeEll et al., 2016). The general lack of internal browning disorders in this study may indicate low susceptibility

Table 3. Mean disorder incidence of 'Honeycrisp' apples after cold storage using pooled data from fruit obtained from two orchards in Washington State in 2016 and 2017, and one orchard in Ontario, Canada in 2017. Control and fruit treated with 42 μmol·L^{-1} 1-MCP after harvest were held 7 d at 10 °C then at 2.8 °C. Fruit were held in air or a CA (2.5 kPa O_2, 0.5 kPa CO_2) for 1 or 2 weeks beginning the day after harvest. Fruit were evaluated after 4 months in storage plus 7 d at 20 °C.

	Control weeks in CA			1-MCP weeks in CA					
	0	1	2	0	1	2	Treatment	Weeks	T × W
Bitter pit	27	15	16	20	14	13	NS	*	NS
Leather blotch	1	1	1	8	10	9	*	NS	NS
Lenticel breakdown	2	2	3	2	2	1	NS	NS	NS
Senescent breakdown	1	0	1	0	0	0	NS	NS	NS
Core browning	0	0	0	2	1	2	**	NS	NS
Internal browning	4	2	3	3	1	0	NS	NS	NS
Cavities	1	0	2	1	1	1	NS	*	NS
Greasy peel	62	44	23	27	24	17	***	***	NS
Soft scald	1	0	0	1	0	0	NS	NS	NS
Soggy breakdown	1	0	1	1	1	1	NS	NS	NS

NS, *, **, ***Nonsignificant or significant treatment effects at $P < 0.05$, 0.01, or 0.001, respectively (Fisher's least significant difference).
CA = controlled atmosphere; 1-MCP = 1-methycyclopropene.

of the fruit used to these disorders, as well as the relatively short time fruit was held in CA. Also, CO_2 concentration during the CA period was low (0.5 kPa), which also may have reduced the potential for injury,

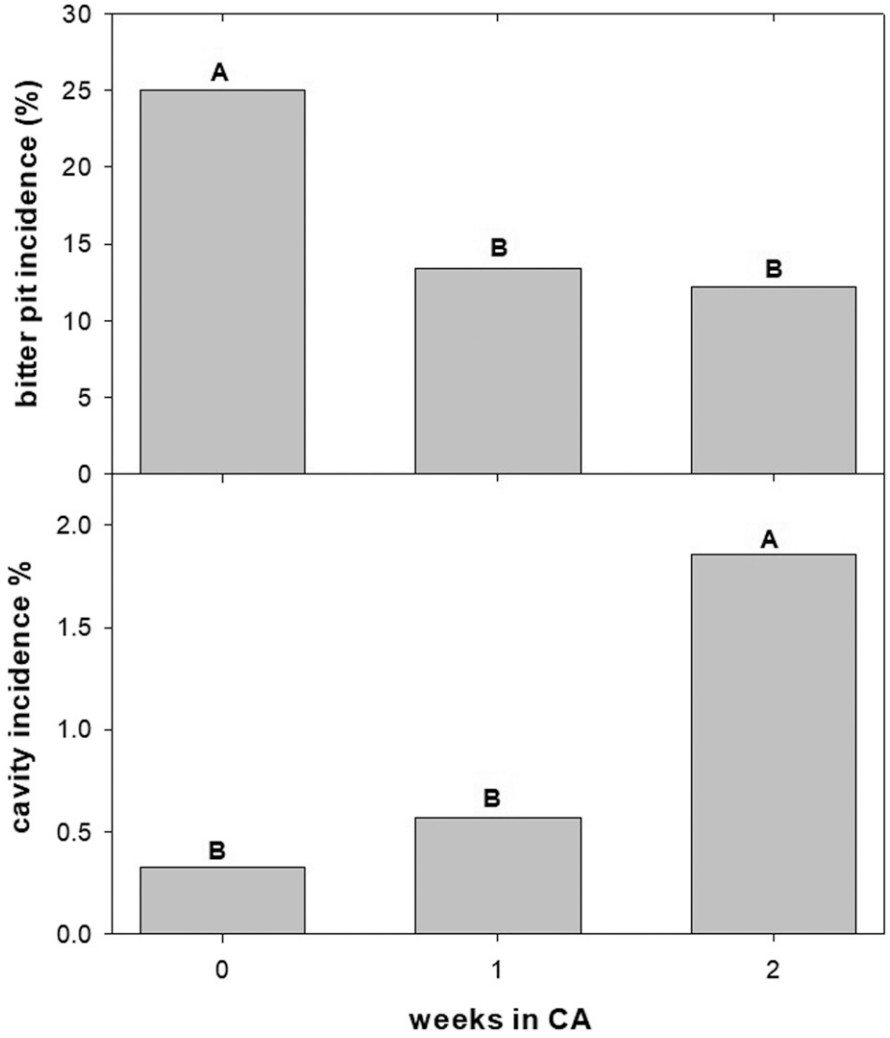

Fig. 1. Bitter pit and cortex cavity incidence of 'Honeycrisp' apple fruit after 4 months cold storage at 3 °C plus 7 d at 20 °C. Fruit from seven orchard lots were held at 10 °C for 7 d after harvest, then at 3 °C in air. Fruit from each lot was also held in a controlled atmosphere (CA: 2.5 kPa O_2, 0.5 kPa CO_2) established 1 d after receipt for 1 or 2 weeks. Bar values are means (n = 156), different letters above the bars indicate values are significantly different, Fisher's protected least significant difference, $P < 0.05$.

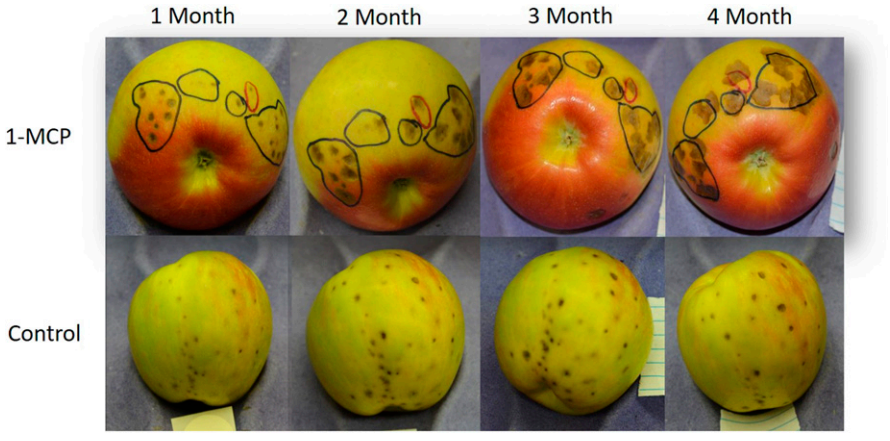

Fig. 2. Peel bitter pit and leather blotch on 'Honeycrisp' apples. All fruit were held at 10 °C after harvest for 7 d then storage temperature was reduced to 3 °C; 1-methycyclopropene (1-MCP) treatment (42 µmol·L^{-1}) was performed the day of harvest for 24 h. Photographs of the same fruit were taken monthly through 4 months.

Cavity development increased with CA duration up to 8 weeks in both 2014 lots. In the combined data set, cavity incidence was highest in fruit stored 2 weeks in CA. The higher incidence could reflect a fruit response to the establishment of CA soon after harvest, as delayed CA can reduce cavity development (DeEll et al., 2016). Conditioning at relatively warm temperature before establishing CA (Contreras et al., 2014) also reduces 'Honeycrisp' cavity development. Although cavity incidence overall increased in fruit held 2 or more weeks in CA, 2 or more weeks CA during and after conditioning did not enhance bitter pit reduction, therefore a single week of CA for bitter pit reduction may avoid the risk of enhanced cavity development. Even with 2 or more weeks in CA, the increase in cavity incidence was usually similar or less than the decrease in bitter pit incidence from the same treatment. The relatively low incidence of cavities as well as other internal browning known to be associated with CA storage (Contreras et al., 2014; DeEll et al., 2016; Watkins and Nock, 2012) may indicate fruit used in these studies was not highly susceptible to this type of injury. It also may indicate the CA setpoints with relatively high CA O_2 (2.5 kPa) and low CO_2 (0.5 kPa) concentrations are not causing enough stress during the week of conditioning and the first week of low temperature to result in internal injury.

For five of the seven lot/years, peel greasiness was significantly impacted by atmosphere and/or 1-MCP. These results are consistent with previous research for 'Honeycrisp', in which 1-MCP treatment and/or CA reduced greasiness (DeEll et al., 2016; Delong et al., 2006; Watkins and Nock, 2012; Mattheis et al., 2017).

Incidence of chilling disorders soft scald and soggy breakdown was low for all lots. All fruit in the experiment were conditioned for 7 d at 10 °C, a protocol known to reduce chilling disorder development (Delong et al., 2004; Watkins et al., 2004). The use of 1-MCP or CA during conditioning did not affect chilling disorder development, consistent with Mattheis et al. (2017).

Fruit quality after 4 months in storage. The 1-MCP and/or CA established during conditioning did not affect fruit firmness, but effects on SSC, TA, and IEC were observed

Fig. 3. 'Honeycrisp' apple core browning in fruit stored 4 months. Fruit were held 7 d at 20 °C after removal from cold storage.

(Tables 4 and 5). The absence of 1-MCP and CA effects on firmness has been reported previously (DeEll et al., 2015; Delong et al., 2006; Watkins and Nock, 2012). Although the impact of 1-MCP and/or CA on SSC varied with orchard lot/year, consistent trends were not apparent and only 1-MCP was a significant factor in the combined data set. The 1-MCP fruit overall had higher SSC values compared with untreated fruit, consistent with previous reports (DeEll et al., 2015; Mattheis et al., 2017). TA was not affected by 1-MCP and/or CA in the combined data set; however, TA in most lots were affected by either 1-MCP alone or the 1-MCP × CA interaction. Where present, these effects resulted in higher TA values compared with control fruit stored in air. This result is consistent with previous reports (Delong et al., 2006; Mattheis et al., 2017; Watkins and Nock, 2012). IEC was lower in 1-MCP–treated fruit compared with controls in all lots and also in the combined data set where weeks in CA was also significant. For the one lot/year (C/2017) in which the 1-MCP × CA interaction was significant, the lowest value was for fruit treated with 1-MCP and held 2 weeks in CA, similar to a previous report (Mattheis et al., 2017) in which the IEC was lowest for fruit treated with 1-MCP and placed in CA during conditioning. Reduced ethylene production by 1-MCP–treated fruit and/or CA is consistent with previous literature for 'Honeycrisp' and other cultivars (Contreras et al., 2014; DeEll et al., 2015; Mattheis et al., 2017; Mirzaee et al., 2014; Watkins, 2008; Watkins and Nock, 2012). The impacts of 1-MCP and/or CA established during conditioning on firmness, SSC, TA, and IEC are consistent with previous reports (Contreras et al., 2014; DeEll et al., 2015, 2016; Delong et al., 2006; Mattheis et al., 2017; Mirzaee et al., 2014; Watkins, 2008; Watkins and Nock, 2012).

Previous literature indicates low temperature and air storage limit the breakdown of fruit texture and firmness of 'Honeycrisp' apples but can promote the development of off-flavors (Wargo and Watkins, 2004); however, CA storage slows the breakdown of traits that are highly associated with eating quality, such as TA and SSC (Mann et al., 2005; Watkins et al., 2005; Watkins and Nock, 2012). In these experiments, fruit stored in CA during conditioning maintained higher fruit quality than fruit conditioned in air. Similar results were reported by Mattheis et al. (2017) where fruit were stored in CA for either 1 or 9 d during conditioning of 'Honeycrisp'. That the short-duration fruit were held in CA is enough to result in higher SSC and TA for some orchard lots through 4 months cold storage is a potential benefit of this protocol in addition to reduced bitter pit development.

Conclusions

CA established and held 1 week during conditioning of 'Honeycrisp' apples reduced bitter pit development. Longer CA durations up to 8 weeks did not appreciably enhance the effects on bitter pit and in some orchard lot/years were associated with increased cavity development. Other peel or internal disorders were not affected by CA during conditioning. 1-MCP with or without CA during conditioning did not impact bitter pit but was associated with leather blotch and core browning. In the lots used for this study, the most effects of CA established during conditioning on bitter pit incidence were observed in orchard lot/years where fruit was predisposed to bitter pit development. The effect of CA and 1-MCP slowing ripening during early storage had some effects on fruit quality after 4 months. As minimizing bitter pit development after harvest remains a commercial challenge, a short-duration CA established during fruit conditioning may be an additional tool to limit postharvest bitter pit losses for 'Honeycrisp' apple.

Table 4. Mean fruit firmness, soluble solids content (SSC), titratable acidity (TA), and internal ethylene concentrations (IEC) after 4 mo. of storage for two lots from two production seasons (C, D) of 'Honeycrisp' apples from Washington State and one lot (E) from one production season in Ontario, Canada.

			Control weeks in CA			1-MCP weeks in CA			*P* value		
Lot	Yr		0	1	2	0	1	2	Treat.	Weeks	T × W
C	2016	Firmness (N)	55.9	57.9	59.8	58.8	60.8	58.8	NS	NS	NS
		SSC (%)	14.3	14.3	14.1	14.9	14.2	14.2	NS	*	NS
		TA (%)	0.39 c	0.46 ab	0.49 a	0.47 ab	0.45 ab	0.43 bc	***	NS	NS
		IEC (μL·L^{-1})	284	248	231	131	113	108	***	NS	NS
D	2016	Firmness (N)	57.9	57.9	58.8	57.9	53.9	55.9	NS	NS	NS
		SSC (%)	12.3 ab	12.6 ab	11.9 b	12.9 a	12.1 b	12.3 ab	***	**	***
		TA (%)	0.39 ab	0.34 b	0.37 ab	0.41 a	0.40 a	0.42 a	NS	***	***
		IEC (μL·L^{-1})	219	165	199	64.2	47.7	41.1	***	NS	NS
C	2017	Firmness (N)	59.8	62.8	60.8	60.8	61.8	62.8	NS	NS	NS
		SSC (%)	13.2	13.8	13.4	13.5	14.0	14.0	**	**	NS
		TA (%)	0.45	0.49	0.46	0.50	0.50	0.50	NS	NS	NS
		IEC (μL·L^{-1})	199 a	112 bc	208 a	153b	112 bc	88.7 c	***	*	***
D	2017	Firmness (N)	73.6	77.5	77.5	75.5	81.4	78.5	NS	NS	NS
		SSC (%)	13.9	14.1	14.9	14.4	14.8	14.8	*	**	NS
		TA (%)	0.54 c	0.53 c	0.60 b	0.61 b	0.69 a	0.67 a	*	***	**
		IEC (μL·L^{-1})	134	93.6	105	59.8	35.6	36.7	***	NS	NS
E	2017	Firmness (N)	68.6	69.6	69.6	69.6	68.6	68.6	NS	NS	NS
		SSC (%)	12.0	12.7	12.3	12.7	12.9	12.5	**	**	NS
		TA (%)	0.43	0.44	0.44	0.49	0.46	0.40	NS	NS	NS
		IEC (L·L^{-1})	138	110	117	38.3	36.8	38.5	***	NS	NS

CA = controlled atmosphere; 1-MCP = 1-methycyclopropene.
NS, *, **, ***Nonsignificant or significant treatment effects at $P < 0.05$, 0.01, or 0.001, respectively (Fisher's least significant difference).

Table 5. Mean quality parameters of 'Honeycrisp' apples (lots C–G) treated with 1-methycyclopropene (1-MCP) compared with an untreated control and conditioned for 0, 1, or 2 weeks in controlled atmosphere (CA). Fruit were rated after 4-month storage in air.

	Control weeks in CA			1-MCP weeks in CA			*P* value		
	0	1	2	0	1	2	Treat.	Weeks	T × W
Firmness N	63.2	65.1	65.3	64.5	65.3	64.9	NS	NS	NS
SSC %	13.2	13.5	13.4	13.8	13.7	13.6	*	NS	NS
TA %	0.56	0.59	0.62	0.65	0.65	0.63	NS	NS	NS
IEC μL·L^{-1}	179	136	156	74.7	59.8	55.5	***	**	NS

IEC = internal ethylene concentration; SSC = soluble solids content; TA = titratable acidity.
NS, *, **, ***Nonsignificant or significant treatment effects at $P < 0.05$, 0.01, or 0.001, respectively (Fisher's least significant difference).

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