Harvest Date and Crop Load Effects on a Carbon Dioxide–related Storage Injury of ‘Braeburn’ Apple

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Abstract. The incidence and severity of “‘Braeburn’ browning disorder” (BBD), a CO₂-induced disorder of ‘Braeburn’ apple (*Malus ×domestica* Borkh.), vary markedly with production district and orchard block. We investigated the effects of harvest date, blush, and crop load on fruit maturity, minerals, skin permeance, and BBD incidence. Incidence of BBD was higher in late- than in early-harvested fruit and in fruit on light than on heavily cropping trees, but blush intensity did not influence susceptibility to the disorder. Fruit maturity factors were affected by region, harvest date, blush type, and crop load, but no consistent relationships between these factors and BBD occurrence were found. Concentrations of Ca, Mg, and/or K were influenced by harvest date, blush type, and crop load, and skin permeance to gas exchange was affected by growing region and blush type, but not by harvest date or crop load. However, no physiological or mineral factor measured in this study was strongly correlated with susceptibility of fruit to BBD. Fruit from orchards that have a history of susceptibility to BBD, or subjected to adverse harvest date, regional, crop load, microclimate, and seasonal influences, should be segregated at harvest, and should not be held in controlled-atmosphere (CA) storage. Crop load should be managed to reduce the occurrence of biennial bearing.

A disorder known as “‘Braeburn’ browning disorder” (BBD), a CO₂-related disorder, can develop in the flesh of ‘Braeburn’ apples during storage (Elgar et al., 1998; Lau, 1998). The disorder has characteristics typical of other CO₂ injuries, such as brownheart and cavity formation in the flesh (Lidster et al., 1990). Symptoms can be induced by elevated CO₂, and can be aggravated by depressed O₂ partial pressures in storage (Elgar et al., 1998; Lau, 1998), and by methyl bromide fumigation (Lay-Yee, 1993). However, BBD can develop in fruit either prior to harvest or during air storage (personal observations).

Susceptibility of ‘Braeburn’ apples to BBD has damaged commercial confidence in the cultivar. Occurrence can be erratic as a result of unknown preharvest factors that influence

susceptibility; identification of these factors would increase confidence among marketers. Little information is available in the literature about preharvest effects on either external or internal CO₂ injuries in apple fruit. Most of these injuries are associated with incorrect storage conditions, such as poor ventilation during air storage (Carne, 1950), but the disorder is often associated with controlled-atmosphere (CA) storage (Blanpied and Smock, 1961; Bramlage et al., 1977; Wilkinson and Fidler, 1973). Research therefore has focused on identification of safe atmospheres and defining postharvest strategies, such as keeping CO₂ concentrations low during the early stages of storage when the risk of injury is highest, treatment with diphenylamine, or delaying the period between harvest and transfer to CA storage (Elgar et al., 1998; Johnson et al., 1998; Smock and Blanpied, 1972; Watkins et al., 1997).

Preliminary studies indicate that the incidence and severity of the disorder are higher in fruit grown in colder or more southern regions, or in colder or higher altitude districts within a region in New Zealand, and that large variations in incidence can occur among both orchards within regions, and trees within orchards (unpublished observations). Several possible factors contributing to greater fruit susceptibility to the disorder were identified, including late harvest, light crop load, and low blush. The objective of this study was to assess the importance of these factors on BBD incidence, using fruit from four orchards in each of two districts.

Plant material. ‘Braeburn’ apples from each of four orchards in each of the Central Hawke’s Bay and Central Otago regions of New Zealand were harvested during the first, middle, and final week of the commercial season (4 weeks) of 1994 for each district (29 Mar., 12 Apr., and 26 Apr. for Central Hawke’s Bay, and 5 Apr., 19 Apr., and 3 May for Central Otago harvests, respectively). Within each orchard, five trees with either high or low crop load were identified by visual assessment prior to harvest, and trunk girth and number of fruit on each tree were recorded. At each harvest, 64 fruit (average fruit weight 180 g; range 160–200 g) were graded into two blush categories (intense blush on >50% of the surface vs. no blush or weak blush on <50% of the surface), packed into standard (18.5 kg) export apple cartons, and transported to the Mt. Albert Research Centre in Auckland that same day.

Harvest quality. Internal ethylene concentration (IEC), background skin color, flesh firmness, soluble solids concentration, and starch pattern indices were determined for 10 fruit from each orchard, crop load, and blush category. Opposite segments from each of five apples were combined to provide two samples for each replicate, and frozen for later determination of titratable acidity. For ethylene determinations, 1-mL samples of internal gas were drawn into a syringe through a hypodermic needle inserted into the core cavity of each fruit, and ethylene content was measured using gas chromatography (Philips PU4500 gas chromatograph, Pye Unicam Ltd., Cambridge, England; 1.5 m × 6 mm alumina F1 80/100 mesh column; injection, detector, and column temperatures 160, 200, and 130 °C, respectively). Background color was assessed visually on industry color cards ranging from 1 (green) to 7 (yellow). Flesh firmness was determined with a semi-automated firmness tester (EPT-1; Lake City Technical Products Inc., Canada) on pared surfaces on two opposite sides of each fruit. Soluble solids concentration of each fruit was determined with a refractometer (Atago model PR-1; Atago Co. Ltd., Tokyo) using the juice expressed during firmness determination. Titratable acidity was determined on a composite sample of each replicate with an auto-titrator (Mettler DL40 RC; Mettler Instruments AG, Zurich, Switzerland). Acids were measured by titrating 20 g of diluted, blended apple tissue against 0.1 N NaOH to a pH 8.1 endpoint, in triplicate, and results are expressed as meq/g fresh weight. Starch content was determined by dipping the cut half of each fruit into an I₂/KI solution for 30 s, allowing the fruit surface to develop color over several min. The resulting patterns were compared with the ENZA ‘Braeburn’ starch pattern chart (New Zealand Apple and Pear Marketing Board, Hastings, 1992).

Skin permeance of four fruit from each orchard, harvest, crop load, and blush category was measured using a slightly modified version of the nonsteady state ethane efflux method (Banks, 1985) as described by Yearsley (1996). Briefly, 1 mL of compressed pure

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ethane was injected into 1.16-L jars containing individual fruit of known weight and left overnight to equilibrate at 20 °C. Fruit were removed from the jars in a fume hood, exposed briefly to a fan to disturb the boundary layer of the fruit, and sealed in a jar of the same volume with a magnetic stirrer operating. One-milliliter gas samples were withdrawn from the jars every 15 s for 2 min to determine the time course of ethane efflux from the fruit. Permeance was calculated from Fick's First Law of Diffusion (Banks, 1985).

Concentrations of Ca, Mg, and K in six pooled samples of five fruit from each orchard, crop load, and blush type at early and late harvests were determined by atomic absorption. Fruit samples were dried at 70 °C, ground in a laboratory mill (Cyclotec, Tecator AB, Hoganas, Sweden), and digested in HNO₃:HClO₄ prior to analysis by atomic absorption (GBC AA spectrophotometer; GBC Scientific Equipment Pty. Ltd., Dandenong, Victoria, Australia).

Storage procedures. Fifty fruit from each orchard, crop load, and blush category were packed into cartons, placed into 330-L plastic chambers, and cooled overnight. On the day after harvest, the chambers were sealed with stainless steel lids and a controlled atmosphere of 2 kPa O₂: 2 kPa CO₂ was flushed through each chamber at a flow rate of 18 L·h⁻¹ (greater than two air changes per day) and mixed continuously with a fan. Equilibrated atmospheres were obtained within 3 d of harvest. Chambers were monitored twice weekly by analyzing 1-mL gas samples for oxygen and carbon dioxide using an oxygen sensor (Citicel C/S; City Technology Ltd., London, U.K.) and an infrared detection cell (Servomex 01514/701 infrared transducer; Servomex PLC Crowborough, East Sussex, U.K.), respectively.

The fruit were held under CA storage for 12 weeks at 0 ± 0.5 °C. Flesh firmness and soluble solids content of 10 fruit per category were subsequently measured after 1 d at 20 °C. After a further 6 d at 20 °C, the remaining 40 fruit from each category were sliced several times to evaluate the presence and severity of internal and external disorders. Flesh browning and cavities were assessed separately. Flesh browning was described as slight, moderate, or severe, depending on the degree of browning in the flesh of the fruit and whether the disorder was visible through the skin. Cavities appeared either in the core or flesh of the fruit. Symptoms were described as "slight" if the cavities were confined to the core of the fruit and did not affect its edibility; "moderate" if the cavities were more extensive, but still largely confined to the core; and "severe" if the cavities had progressed to the cortical flesh. Overall disorder severity was rated as a combination of incidence of flesh browning and cavities, taking into account those instances where individual fruit had symptoms of both disorders. Flesh browning, cavity, and overall disorder scores were determined by multiplying the incidence at each severity by a weighted score (1 = slight, 2 = medium, 3 = severe) and dividing by the total number of

fruit, to give an average value for each treatment.

Statistical analyses. Analysis of variance was used to make inferences from the randomized complete-block designs. Percentage data were arcsine transformed to obtain homogeneity of variance. Relationships between measured variables were examined using pairwise scatterplots. No significant relationships were found that justified further regression analysis.

Results

Crop load estimations. High crop load trees had an average of 7.2 fruit/cm² trunk cross-sectional area (TCA), almost double the load factor of low crop load trees at 3.4 fruit/cm² TCA (Table 1). Trees from Central Hawke's Bay had higher crop loads than did those from Central Otago, although average crop loads varied considerably among orchards.

Maturity at harvest. Only starch pattern indices and titratable acidity concentrations were affected by region, with fruit from Central Otago having higher starch pattern indices (less starch) and lower titratable acidity than those from Central Hawke's Bay (Table 2). All factors assessed indicated that fruit were more mature at later than at early harvest, but effects of blush and crop load on maturity were not always consistent. High blush fruit were more mature, on the basis of IEC, background color, soluble solids, and titratable acidity, but also had a slightly lower starch pattern index (more starch) and were firmer than low blush fruit. Crop load did not affect starch pattern index or soluble solids. Although fruit from low crop load trees were more mature on the basis of IEC and background color than those from high crop load trees, they were firmer and had higher titratable acidity.

Interactions between region, harvest date, crop load, and blush were detected for IEC, background color, soluble solids, starch pattern index, and flesh firmness, but these interactions were not consistent across the various physiological factors (data not shown).

Mineral analyses. Calcium, Mg, and K concentrations in the fruit did not differ between regions (Table 2). Fruit from the last harvest had higher Mg and K concentrations

than fruit from the first harvest, but Ca was not affected by harvest date. High blush fruit had lower Ca and K concentrations than did low blush fruit, but Mg concentrations were unaffected. Calcium concentrations were higher, and Mg and K concentrations lower, in fruit from high crop load trees than in those from low crop load trees.

Skin permeance. Skin permeance was lower in fruit from Central Otago than in those from Central Hawke's Bay, and in high blush than in low blush fruit, but was not affected by harvest date or crop load (Table 2). Because BBD is a population-based phenomenon, outliers within the population may be more useful in explaining treatment effects; while treatment means may be similar, the susceptible fruit might be associated with lower permeance, and, consequently, a higher internal CO₂ concentration. Box plot analyses showed a slightly greater number of outliers in the fruit population from late-harvested trees (Fig. 1), which would support the possibility that skin permeance is involved in fruit susceptibility to BBD. However, the greater range in permeance in low blush fruit than in high blush fruit was not supported by differences in BBD incidence.

Flesh browning and cavity incidence. Because BBD appears in two forms, as flesh browning and as cavities (Elgar et al., 1998), we assessed each disorder individually. No difference in flesh browning incidence or severity was detected between fruit from Central Hawke's Bay and Central Otago (Table 3), perhaps because the variation between orchards within a district was high. Incidence of cavities was greater in Central Otago fruit (Table 3). Incidence and severity of flesh browning increased as harvest was delayed, and were higher with low crop loads (Table 3). The incidence, but not severity, of cavities declined as harvest was delayed, and both were higher in fruit from low crop load trees. There was no effect of blush intensity, although the disorder generally was found on the shaded side of blushed fruit (data not shown).

Firmness and soluble solids concentration were not affected by region (Table 3). Late harvested fruit were softer and had greater soluble solids. Low blush fruit and fruit from high crop load trees were also softer, but had lower soluble solids concentrations.

Table 1. Crop load of 'Braeburn' trees in orchards in Central Hawke's Bay and Central Otago, New Zealand.

Orchard	Nominal crop load	Crop load (fruit/cm ² TCA ^a)	
		Central Hawke's Bay	Central Otago
1	High	9.8	4.0
	Low	4.2	2.5
2	High	8.2	6.6
	Low	2.7	3.3
3	High	9.9	5.3
	Low	5.4	2.4
4	High	7.8	6.2
	Low	2.9	4.2
Average	High	8.9	5.5
	Low	3.8	3.1
SE	Region (R)	0.75***	
	Crop load (C)	0.32***	
	R × C	0.69***	

^aTCA = trunk cross-sectional area.

***Significant at $P \leq 0.001$

Table 2. Main effects of growing region, harvest time, crop load, and blush type on physiological factors of 'Braeburn' apples measured at harvest.

Factor	Internal ethylene concentration ($\mu\text{L}\cdot\text{L}^{-1}$)	Starch pattern index (0-8)	Background color (1-7)	Flesh firmness (N)	Soluble solids (%)	Titratable acidity (meq/g FW^z)	Skin permeance ($\text{nmol m}^{-2}\cdot\text{s}^{-1}\cdot\text{Pa}^{-1}$)	Fruit mineral concn ($\mu\text{g}\cdot\text{g}^{-1}\text{FW}^z$)		
								Ca	Mg	K
Region										
Central H.B. ^y	0.33	2.3	3.1	94	11.2	0.17	0.211	24	32	1000
Central Otago ^x	0.37	3.5	3.0	93	10.8	0.16	0.186	29	34	1060
	NS	***	NS	NS	NS	*	***	NS	NS	NS
Harvest										
1	0.16	2.1	2.3	98	10.4	0.18	0.206	27	31	990
2	0.46	2.8	2.8	95	11.1	0.16	0.195	---	---	---
3	0.43	3.8	4.0	88	11.6	0.15	0.194	26	35	1070
	***	***	***	***	***	***	NS	NS	***	***
Blush type										
Low	0.24	3.1	2.6	93	10.6	0.17	0.206	28	33	1050
High	0.45	2.7	3.5	95	11.5	0.16	0.191	26	33	1010
	***	***	***	***	***	*	**	***	NS	*
Crop load										
Low	0.39	2.9	3.1	95	11.0	0.17	0.198	25	34	1090
High	0.30	2.9	3.0	92	11.0	0.16	0.199	28	32	970
	***	NS	*	***	NS	***	NS	***	***	***

^zFW = fresh weight.^yCentral Hawke's Bay, New Zealand.^xCentral Otago, New Zealand.NS, *, **, ***Nonsignificant or significant at $P \leq 0.05$, 0.01, or 0.001, respectively, within each factor.

Fig. 1. Box plot analysis of effects of harvest date, crop load, and blush on skin permeance to gas exchange in 'Braeburn' apple fruit. The box represents the 25th to 75th percentile around the median line, the vertical lines outside the box represent the 99% confidence limits, and the points represent data outside these limits.

Discussion

Fruit from both regions had similar BBD occurrence as expressed as flesh browning, although cavity incidence and severity were highest in fruit from Central Otago. However, variability of disorder incidence was high among orchards. Such variability is a general feature of fruit susceptibility to CO_2 injuries (Elgar et al., 1998; Johnson et al., 1998; Lau and Looney, 1978; Volz et al., 1998; Watkins et al., 1997).

Despite variations among orchards, BBD incidence, expressed as flesh browning, was consistently higher in later harvested fruit and in fruit from trees with low crop loads. In contrast, cavity incidence, though not severity, was slightly higher in the early harvested fruit than in fruit from subsequent harvests. Harvest date has been implicated in fruit susceptibility to CO_2 injury, higher levels of external and internal injury being associated with early and late harvest, respectively (Meheriuk, 1977; Smock and Blanpied, 1963). Crop load has not previously been shown to influence CO_2 injury, but fruit from low crop load trees are generally more susceptible to other storage disorders (Ferguson and Watkins, 1992; Sharples, 1964; Volz et al., 1993).

Maturity, as affected by harvest date and crop load, was not consistently related to BBD occurrence. Measurements of IEC and background color indicated that fruit from late harvest and low crop load trees were the most mature; although late harvested fruit had higher starch indices and soluble solids concentrations, neither factor was affected by crop load. Later harvested fruit were softer than early harvested fruit, and fruit from high crop load trees were slightly softer. In contrast, blush type, which did not affect BBD occurrence, affected all maturity factors. Therefore, even though later harvest of fruit and low crop load appear to be major factors contributing to fruit

Table 3. Main effects of growing region, harvest time, crop load, and blush type on incidence and severity scores for flesh browning, cavities and total disorders, and flesh firmness and soluble solids, of 'Braeburn' apples. Fruit were stored under controlled-atmosphere (2 kPa CO₂ : 2 kPa O₂) conditions at 0 °C for 12 weeks and assessed after 1 d (firmness and soluble solids) and 7 d (disorders) at 20 °C.

Factor	Flesh browning		Cavities		Total disorder		Firmness (N)	Soluble solids (%)
	(%)	(score)	(%)	(score)	(%)	(score)		
Region								
Central H.B. ^z	21	0.8	29	0.6	45	1.4	92	12.8
Central Otago ^y	32	1.0	45	1.0	64	2.0	91	11.7
	NS	NS	**	*	***	NS	NS	NS
Harvest								
1	13	0.4	44	0.9	50	1.2	94	11.9
2	23	0.8	35	0.8	49	1.5	90	12.3
3	44	1.6	33	0.8	64	2.4	90	12.5
	***	***	**	NS	**	***	***	***
Blush type								
Low	25	0.9	37	0.8	54	1.7	90	11.7
High	28	0.9	37	0.8	56	1.7	92	12.8
	NS	NS	NS	NS	NS	NS	***	***
Crop load								
Low	39	1.3	45	1.0	72	2.4	92	12.4
High	14	0.4	29	0.6	37	1.2	90	12.1
	***	***	***	***	***	***	***	***

^zCentral Hawke's Bay, New Zealand.

^yCentral Otago, New Zealand.

NS, *, **, *** Nonsignificant or significant at $P \leq 0.05$, 0.01, or 0.001, respectively, within each factor.

susceptibility to BBD, collectively these data do not establish critical maturity factors that might be used to predict susceptibility.

Greater incidence of external and internal CO₂ injury in 'Golden Delicious' apples was associated with higher fruit N, Mn, and Zn, and lower K and Mg, but not with Ca (Lau and Looney, 1978). In our study, with more limited mineral analyses, Ca concentrations were not affected by harvest date, but Mg and K were higher at the last than at the first harvest. Fruit from trees with low crop loads had lower Ca and higher Mg and K concentrations than those from trees with high crop load. Light cropping can have a profound effect on fruit storage quality and Ca concentrations are lower in fruit from trees with low crop regardless of fruit size (Ferguson and Watkins, 1992). Although differences in mineral concentrations between treatments were small, more widespread analyses of these minerals may be warranted to further explore possible relationships with BBD incidence. The confounding effect of crop load on relationships between minerals and BBD incidence should be considered.

Low permeance to gas exchange has been implicated in greater susceptibility of fruit to low O₂ injury (Park et al., 1993). Less is known about possible relationships between permeance and high CO₂ injury. Johnson et al. (1998) detected a close association between low permeance and internal, but not external, CO₂ injury in 'Bramley's Seedling' apples, however, no similar relationships were found in 'Fuji' apples (Park and Lee, 1991; Volz et al., 1998). 'Braeburn' has a low, but variable, permeance compared with many other apple cultivars (Rajapakse et al., 1990; Yearsley et al., 1996, 1997), which could contribute to its sensitivity to BBD. We were not able to detect significant treatment differences associated with skin permeance that might be related to BBD incidence, although further study is required to examine if the number of outlying

fruit with low permeance might contribute to increased injury in certain fruit within the population. However, other cultivars that are susceptible to internal CO₂ injury, such as 'Fuji' (Volz et al., 1998), do not have markedly low skin permeance (Park and Lee, 1991). Thus, susceptibility of fruit to high CO₂ is analogous to susceptibility to low O₂, where differences in fruit tissue thresholds to injury appear to exist in different cultivars (Park et al., 1993).

None of the physiological factors measured in this study were consistently associated with BBD occurrence. Because of the high variation among orchards, we also analyzed all data sets by pairwise scatter plot comparisons in an attempt to detect relationships between factors measured in this study and BBD that might be missed by comparison of treatment effects. However, no factor was related significantly to BBD (data not shown).

The primary factors affecting BBD development are unknown but are probably associated with tissue tolerance to CO₂ concentrations in the fruit. While maturity, mineral content, and skin permeance are all potential factors contributing to susceptibility of 'Braeburn' fruit to BBD, they probably have secondary rather than primary roles. For example, bitter pit and breakdown are usually described as disorders associated with low Ca in the fruit (Ferguson and Watkins, 1989; Marmo et al., 1985), but their incidence can be low even in susceptible fruit when harvested at certain maturity stages (Watkins et al., 1989).

From a practical standpoint, we conclude that late harvested fruit and/or fruit from low cropping trees grown in at-risk regions have a moderate to high risk of developing BBD during storage. If an orchard is found to be of high risk due to factors of region, site history, harvest date, crop load, and seasonal influence, then we recommend that fruit from light cropping trees should be segregated by the

grower at harvest. Growers should manage crop load to avoid light cropping trees within a block, and reduce the likelihood of biennial cropping by avoiding overcropping or overthinning of flowers and fruitlets.

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