The importance of maintaining the cold chain for avocado ripening quality

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ABSTRACT

Avocado fruit (cv 'Hass') from Everdon Estate (Karkloof) were subjected to a 9 hour break in cold storage at 5, 10 or 20 days, or a delay of 24 hours before cold storage to simulate transport and shipping conditions of South African avocados to Europe. Fruit were stored at 1ºC or 5.5°C for 28 days. After storage, the respiration, ethylene evolution, hardness, mass and fruit quality were assessed. Fruit only showed significant differences in the physiological parameters on the first day after removal from cold storage, but fruit quality was severely affected by breaking the cold chain. Storage temperature also has a significant effect on fruit quality. Storage at 5.5°C resulted in only having between 8% and 30% sound fruit, while storing fruit at 1°C resulted in an increase in the percentage of sound fruit to 80% for the control. Breaking the cold chain anywhere in the shipping process reduced fruit quality. This is mitigated by storing fruit at 1°C, but it is imperative to maintain the cold chain from the pack house to the shipment's final destination.

INTRODUCTION

Transportation of fruit from the northern provinces of South Africa (Limpopo and Mpumalanga) to the port in Cape Town and eventually supermarkets in Europe requires considerable logistical management and it is inevitable that problems and delays will occur during shipment. The transportation of fruit from the northern provinces has improved from 20 to 30 years ago (Toerien, 1986), but to remain globally competitive, further improvements may be necessary. While it has been ascertained that breaking the cold chain is to be avoided (Dodd *et al.*, 2007; Eksteen, 1995; 1999; Vorster *et al.*, 1991), the effects of breaking the chain at various points has not been studied, nor has the interaction with shipping temperature, especially if lower than presently used as standard.

Successful storage of avocados (and all horticultural crops) requires the earliest possible removal of field heat and maintaining the optimal temperature throughout storage (Ginsberg, 1985). The Perishable Produce Export Control Board (PPECB) has recommendations and guidelines available for the handling of avocados due for export (Eksteen, 1995; 1999) that outline the correct handling of fruit with respect to refrigerated storage to minimize increases in pulp temperature. These documents stress the importance of fast removal of field heat and minimising delays in transport and transfers, and conclude that the post-harvest age of fruit and the product temperature are the most important determining factors in fruit market quality. The PPECB has repeatedly stressed the importance of adhering to the protocols, as outlined in their various reports in the SAAGA yearbook and at the 2007 World Avocado Congress, and concluded that there is opportunity to improve quality and reduce input costs by concentrating efforts on adhering to protocols and procedures (Dodd *et al.*, 2007). The report showed that inadequate temperature management does occur in the South African avocado industry and is costing growers money and reducing fruit quality. Dodd *et al.* (2007) also maintain that controlled atmosphere and 1-MCP treatments may be avoided with correct cold chain management.

Some research has shown that intermittent warming of fresh produce may prolong quality (Artés *et al.*, 1998; Girardi *et al.*, 2005; Schirra and Cohen, 1999), but this technique involves slight increases of ambient temperature – not prolonged warming to room temperature. Recently (Undurraga *et al.*, 2007) showed that breaking the cold chain reduces avocado quality by increasing transpiration and thereby mass loss. These authors also found that breaking the cold chain near the end of storage resulted in the fruit entering the ripening process (softening and changing colour). Research by Van Rooyen and Bower (2006) has shown that fruit stored at 1°C (and even -0.5°C) can have higher internal quality than fruit stored at 5.5°C. These ultra-low storage temperatures may mitigate some of the effects of cold chain breakage as the pulp temperature will remain below critical levels longer than fruit stored at the conventional 5.5°C.

This study aimed to compare the effect of storage temperature and the effect of breaking the cold chain at various points on aspects of ripening physiology and visual quality, in a simulated shipping trial.

MATERIALS AND METHODS

Delays can be expected at the farm or pack house, at the ports of Cape Town, during shipping and at the destination port (such as Rotterdam). These have been simulated by delaying cooling by one day, and breaking the cold chain at 5, 10 and 20 days.

Fruit

Local grade fruit were sourced from Howick in the KwaZulu-Natal Midlands and randomly placed in the ten treatment combinations (5 treatments and 2 storage temperatures) with 10 fruit per treatment and each fruit as a replication. Fruit were picked on the 2nd and 22nd of July and the 11th of August. Average moisture contents for each harvest date were 73.4, 70.4 and 67.8%.

Respiration

Carbon dioxide (CO_2) evolution, used as an indication of respiratory activity, was measured using an environmental gas monitor (EGM-1, PP Systems, Hertfordshire, UK). Fruit were incubated in a 1 L container for 12 min. Headspace CO_2 concentration (parts per million) was measured and results calculated as a rate of CO_2 evolution/production ($mL.kg^{-1}.h^{-1}$), taking into account the fruit mass, volume and head space in the jar and the ambient room CO_2 concentration.

Quantification of ethylene

Fruit were incubated for 15 min in a 1 L container which contained a 20 mL vial which was sealed on removal. Analysis was done with a DANI 1000 gas chromatograph equipped with an HT250D Autosampler. The column was stainless steel packed with alumina F1 stationary phase. A flame ionisation detector was used. The temperatures for the injector, column and detector were 160˚C, 100˚C and 180˚C, respectively. The mobile phase was nitrogen gas at 35 kPa. The autosampler injected 1 mL of sample from the 20 mL vial. Results were calculated after taking fruit mass, volume and container head space into account.

Ripeness

Ripeness was assessed using a hand-held firmness tester (densimeter; Bareiss, Oberdischingen, Germany).

Statistical analysis

Analysis was performed using Genstat v9.1 (VSN International Ltd). The statistical design was a completely randomized design with repeated measures.

RESULTS AND DISCUSSION

While the maturity of the fruit had an effect on respiration, ethylene synthesis, mass loss and the rate of softening, in order to simplify the presentation of the data, values have been averaged for the three harvest dates. Only fruit from the first harvest took longer than 5 days to ripen, hence all values on day 7 refer only to that harvest.

Breaking the cold chain at 10 or 20 days significantly reduced the initial respiration rate of the fruit at both storage temperatures, for fruit stored at 1°C (**Table 1**). Delaying cold storage and breaking it at day 20 initially reduced the respiration of the fruit stored at 5.5°C. There was a significant difference in the fruit that had cold storage delayed, at day seven, but this was

Table 1. CO_2 evolution ($mL.kg^{-1}.h^{-1}$) after cold storage at 1ºC or 5.5ºC averaged for three harvest dates and four cold chain break periods. LSD = 6.7 $mL.kg^{-1}.h^{-1}$. Asterisk (*) indicates a significant difference to that treatment's control, at the related day

Storage temp (°C)	Day	Treatment				
		Control	Delay	Break 5	Break 10	Break 20
1	0	67.6	62.8	49.0*	44.4*	51.8*
	2	80.3	82.7	80.8	76.4	85.6
	5	71.4	76.4	75.4	69.8	72.5
	7	62.8	64.5	67.0	63.4	66.5
5.5	0	86.9	79.2*	86.4	80.9	78.0*
	2	81.1	80.4	81.7	80.2	81.7
	5	76.3	74.6	74.3	72.2	79.8
	7	70.6	61.4*	77.2	69.7	67.5

Table 2. Ethylene evolution ($\mu L.kg^{-1}.h^{-1}$) after cold storage at 1ºC or 5.5ºC averaged for three harvest dates and four cold chain break periods. LSD = 21.5 $\mu L.kg^{-1}.h^{-1}$. Asterisk (*) indicates a significant difference to that treatment's control, at the related day

Storage temp (°C)	Time	Treatment				
		Control	Delay	Break 5	Break 10	Break 20
1	0	72.5	78.7	61.2	60.4	66.9
	2	93.2	104.8	95.6	93.2	102.2
	5	85.3	97.3	91.3	87.1	91.4
	7	97.0	114.7	126.5*	95.6	134.4*
5.5	0	84.8	97.7	83.9	89.0	91.0
	2	90.6	93.7	90.1	85.4	87.0
	5	113.4	129.7	103.3	125.6	126.0
	7	172.1	166.2	207.8*	171.1	225.1*

because the fruit were ripe already and had progressed into the post-climacteric stage. Further, there seems to be evidence from the respiration results, that at 5.5ºC, the climacteric had been initiated before the end of the storage period. This was not so in the fruit stored at 1ºC.

Although the fruit stored at 1ºC had an initially lower respiration rate by the second day after removal, the differences between the different temperatures (comparing within treatments) was insignificant, indicating that the lower storage temperature did not have a permanent effect on respiration.

The only significant differences in ethylene evolution (**Table 2**) were on day seven. It is suggested that the large rise in ethylene evolution from day five to day seven indicates that the fruit were starting to decay.

The fruit stored at 1ºC lost significantly less mass (**Table 3**) during storage. This was true of all treatments. The differential was maintained throughout ripening. This reduced water loss during storage resulted in improved fruit quality, as is clearly illustrated in **Figure 1**, with fruit stored at 1ºC having up to 80% sound fruit compared to only 30% for fruit stored at 5.5ºC.

The reduced storage temperature also resulted in an insignificant softening during storage (**Table 4**) which is in agreement with the findings of Biale (1941). Softening during storage has been a major problem with exporting avocados (Dodd *et al.*, 2007; Ginsberg, 1985; Undurraga *et al.*, 2007). While the fruit stored at the normally used shipping temperature of 5.5ºC had softened significantly during storage, this resulted in the fruit stored at 5.5ºC ripening sooner than the fruit stored at 1ºC. At 1ºC, the control fruit took slightly longer to ripen than the cold-interrupted fruit, which shows that the break in the cold chain did affect physiological activity, most likely the enzymes responsible for ripening.

Figure 1 clearly shows the effect of breaking the cold chain, and how reducing the storage temperature improves the percentage sound fruit. "Sound fruit" were

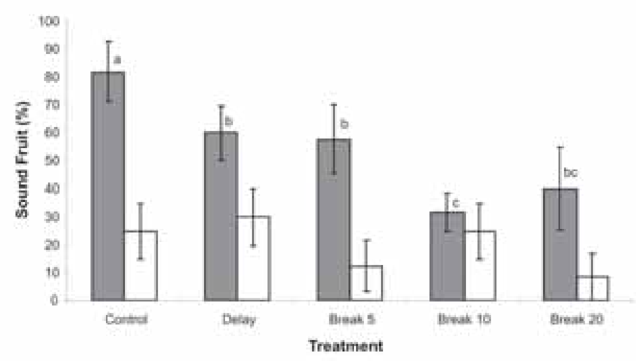

Figure 1. Percentage of sound fruit for each treatment (averaged for three harvest dates) and storage at 1ºC (solid bars) and 5.5ºC (clear bars). Standard errors shown are for the combined harvest dates. Letters (a, b, c) indicate significant differences and are only indicative for the 1ºC storage temperature

Table 3. Percentage mass loss after cold storage at 1ºC or 5.5ºC averaged for three harvest dates and four cold chain break periods. LSD = 0.9%. Asterisk (*) indicates a significant difference to that treatment's control, at the related day

Storage temp (°C)	Time	Treatment				
		Control	Delay	Break 5	Break 10	Break 20
1	0	4.6	5.1	4.6	4.4	4.5
	2	8.2	8.3	7.9	7.7	7.8
	5	11.9	11.5	11.2	11.0	11.3
	7	13.9	13.6	13.2	12.1*	13.2
5.5	0	10.3	10.0	10.7	9.8	9.7
	2	13.8	13.2	14.1	13.1	12.9
	5	17.2	16.3	17.3	16.2*	16.0
	7	18.7	17.9	18.8	17.4*	17.6

Table 4. Densimeter readings (scale 0 to 100) after cold storage at 1ºC or 5.5ºC averaged for three harvest dates and four cold chain break periods. LSD = 2.3. Asterisk (*) indicates a significant difference to that treatment's control, at the related day. Fruit are ripe when densimeter reading is below 60

Temp (ºC)	Time	Treatment				
		Control	Delay	Break 5	Break 10	Break 20
1	Start	86	86	86	87	87
	0	85	86	85	84	85
	2	74	73	73	72	73
	5	62	59*	59*	59*	58*
	7	61	58*	58	58*	55*
5.5	Start	87	88	85	88	87
	0	79	81	78	80	80
	2	64	66	64	64	63
	5	60	61	59	59	57*
	7	58	59	59	55*	58

considered to be those without shrivel or anthracnose infection and with good colouring, i.e. marketable fruit. The predominant disorder causing poor quality fruit was shrivel, with fruit with poor colour and anthracnose occurring infrequently.

At 5.5ºC, there was no difference between the treatments and the control because all were below 30% sound fruit. However, at 1ºC, the fruit that had a constant cold storage period had significantly better quality. The control had 80% sound fruit (±10.7%) while the delay, break 5, break 10 and break 20 treatments had 60, 57, 31 and 40% sound fruit (±9.8, 12.4, 6.9 and 14.9%) respectively.

Figure 2. Fruit from the first harvest date, two days after removal from cold storage. Fruit in the top row were stored at 1ºC, and fruit in the bottom row at 5.5°C. The columns, from left to right are: Delay, Break 5, Break 10, Break 20 and Control

Figure 3. Fruit from the first harvest date, five days after removal from cold storage. Fruit in the top row were stored at 1°C, and fruit in the bottom row at 5.5°C. The columns, from left to right are: Delay, Break 5, Break 10, Break 20 and Control

Figure 4. Fruit from harvest one, at ripeness. The fruit on the left were stored at 1°C, those on the right at 5.5°C. Treatment was break at 10 days

Comparing results within each treatment, 1ºC fruit were all significantly better, except for the fruit removed on day 10 where there was no significant difference. The differences in colour development are shown in **Figure 2** and **3**. Storage at 5.5ºC resulted in faster fruit colour development, and appeared to exacerbate the appearance of "mixed ripening". **Figure 4** (showing the most damaging treatment of breaking the chain at 10 days) shows the effect, at ripeness, of a lower storage temperature. The fruit stored at 5.5ºC (right) show extensive shrivel while the 1ºC fruit (left) were still of high quality.

While fruit stored at lower temperatures do colour more slowly, (**Figure 2** and **3**), the fruit quality was superior, with less mass (predominantly moisture) loss. With a reduced respiration rate during storage (**Table 1**), the fruit probably have greater energy reserves to complete ripening without developing any physiological disorders. The reduced storage temperature may help to buffer the effect of a break in the cold chain. The lower pulp temperature could keep the tissue below the critical level at which irreparable damage will occur, for longer. It will also reduce the apparent "mixed ripening".

Temperature greatly affects the respiration rate of fruit (Biale, 1941; Eaks, 1978). Avocados are climacteric fruit with a very high rate of metabolism. Once fruit are harvested, energy reserves are utilised. Climacteric ripening requires a large amount of energy to conclude successfully and if energy reserves are depleted below the amount required for successful ripening, physiological disorders (particularly rubbery pulp and shrivel) will result. Breaking the cold chain will result in respiration spikes that are likely to reduce the energy reserves of the fruit. Work still being concluded indicates that fruit stored conventionally do deplete their energy reserves and run out of soluble sugars (Blakey *et al.*, 2008).

CONCLUSION

Breaking the cold chain is known to be detrimental to fruit quality and this has been shown to be true for avocados. Although fruit recovered physiologically in terms of respiration and ethylene evolution, quality was severely affected. Breaking the cold chain resulted in severe shrivel and a large proportion of unmarketable fruit. There is also evidence that at 5.5ºC the climacteric starts during the shipping period. This is not the case for storage at 1ºC. Reducing storage temperature from 5.5ºC to 1ºC can also buffer fruit from the effects of breaking the cold chain. It is once again stressed that maintaining the cold chain is crucial in delivering quality fruit to export markets, and serious consideration to shipping at a lower temperature than presently done should be given.

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