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Enhancing the export performance of Australian mandarins by improving flavour quality

Dr Andrew Macnish
The Department of Agriculture and Fisheries (DAF)

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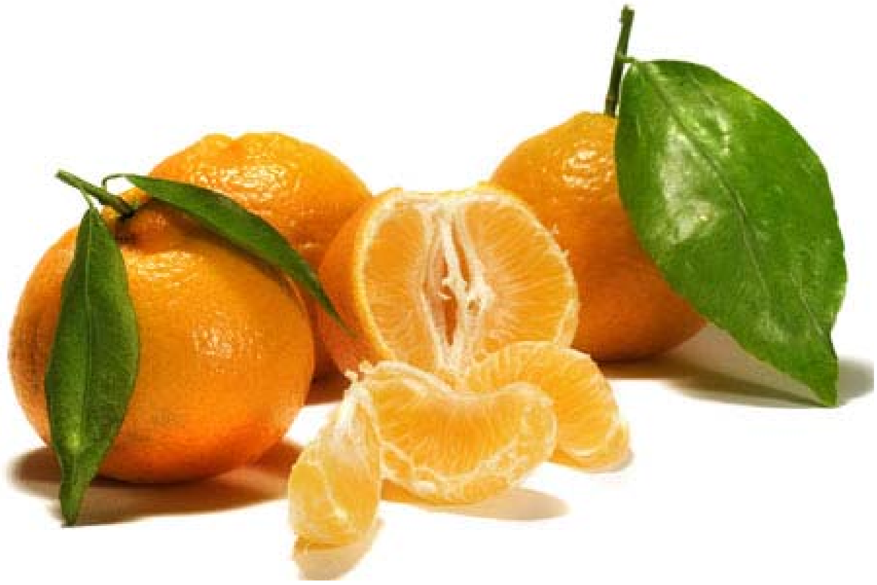

Enhancing the export performance of Australian mandarins by improving flavour quality

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Project Number: CT12023

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Summary

Mandarins are becoming increasingly popular with consumers for their easy-to-peel skin. While this has prompted increased plantings in Australia and a greater focus on export marketing, little is known about consumer attitudes towards the flavour quality of mandarins. The purpose of this project was to develop a better understanding of the flavour profile and consumer expectations of Australian mandarins in the Chinese market. China is Australia's fastest growing market for mandarins.

As a first step, we characterised the sensory profile of 'Murcott' mandarins following export. Fruit from Mundubbera, Queensland were subjected to simulated shipment (21 days at 1.5, 5, 10°C) and shelf life (8 days at 20°C) prior to evaluation by a trained Australian sensory panel. Key findings were:

- Coating mandarins with carnauba wax increased peel shine relative to non-coated fruit.
- The wax coating did not affect fruit juice soluble solids, acids or BrimA (a new measure of fruit juice flavour quality derived from soluble solids and acids content) but was associated with higher juice ethanol content during shipment and shelf life than the non-coated mandarins.
- Panellists distinguished that juice from wax-coated fruit exhibited considerably more "over-ripe" and "musty/mouldy" off-notes than the non-wax-coated fruit juice.
- The intensity of juice flavour off-flavours increased with export temperature and time.

As a second step, we demonstrated that an enzyme-based assay could accurately measure the ethanol content in 'Murcott' fruit juice. The assay could potentially be used to monitor off-flavours in mandarin shipments and help predict consumer acceptance and/or practices to improve fruit quality.

As a third step, additional 'Murcott' mandarins were subjected to simulated shipment and shelf life as described above prior to evaluation by Chinese consumers residing in Australia. Key findings were:

- Consumers preferred the flavour of wax-coated fruit over non-coated fruit and apparently were not influenced by the ethanol content of the fruit juice.
- Consumers detected a loss in flavour quality of stored fruit versus freshly-harvested samples which suggests the presence of flavour defects other than ethanol.

As a final step, we tracked a shipment of 'Murcott' mandarins from Mundubbera, Australia to Beijing, China and monitored fruit performance and consumer reaction. We also randomly sampled Australian 'Murcott' mandarins from Chinese retailers and documented fruit flavour quality. Key findings were:

- Fruit pre-treated with a carnauba wax maintained a superior peel shine, lost 33% less weight but developed a higher ethanol content than non-coated mandarins during marine shipment.
- Chinese consumers surveyed in Shanghai and Beijing preferred the appearance and lower acid flavour of the non-coated fruit over wax-treated fruit and did not comment on ethanol flavours.
- Australian mandarins sampled from Chinese retailers had acceptable appearance, total soluble solids and acids but exhibited moderate ethanol content.

This study highlighted that Australian mandarins can lose considerable flavour quality during export to China. While the fruit ethanol content was closely related to flavour deterioration in mandarins, our data suggest that other metabolites may be important in the perception of flavour loss by Chinese consumers. Further research is needed to identify the specific contributors to flavour loss with a view to optimising flavour in Australian export mandarins to better meet Asian consumer expectations.

Keywords

China; consumer preference; ethanol; mandarin; off-flavour; postharvest; surface coating; temperature

Introduction

Mandarin fruit are becoming increasingly popular with consumers worldwide owing to their convenient easy-to-peel skin and sweet, fruity flavour. This rise in consumption has prompted increased plantings of mandarins in Australia (ABS, 2013). China, Indonesia, New Zealand, Thailand, and the UAE are currently the main export markets. China is Australia's most valuable and fastest growing market for mandarins with exports worth $8.6 million in 2013/14 (QGSO, 2015).

Despite their increasing popularity, mandarins are more perishable than other citrus species such as oranges, grapefruit and lemons (Kader and Arpaia, 2002). High rates of respiration, water loss, decay and flavour loss are among factors limiting the storage life of mandarins. Mandarins are also inherently sensitive to anaerobic stress and to developing 'winey' alcohol off-flavours during postharvest storage and shelf life (Shi et al., 2005). Mandarin fruit possess a peel that is less permeable to gas exchange than that of other species (Shi et al., 2005, 2007). In addition, mandarin juice exhibits relatively high activities of pyruvate decarboxylase and alcohol dehydrogenase, the two enzymes that produce ethanol off–flavours (Davis et al., 1973; Shi et al., 2005). Taken together, these biological features may explain the higher susceptibility of mandarins to lose flavour quality.

It is customary to coat mandarins after harvest with waxes or resins to reduce water loss and enhance surface shine, but it can also further restrict gas exchange through the peel (Hagenmaier and Baker, 1993). The resulting decrease in O_2 diffusion into fruit and increase in internal CO_2 levels can trigger anaerobic respiration and the accumulation of fermentation off-flavours in the fruit juice (Cohen et al., 1990; Hagenmaier, 2002; Tietel et al., 2011). Considerable research has been directed at developing and evaluating different surface coatings that afford citrus fruit with a glossy peel shine but still allow sufficient gas exchange to minimise off-flavour development. Numerous studies have established that polyethylene, beeswax and carnauba wax-based coatings are more permeable to gas exchange than the high-gloss shellac and wood resin-based coatings (Hagenmaier, 2000; Hagenmaier and Shaw, 2002; Porat et al., 2005). Findings from the HAL project CT12000 indicated that mandarin fruit coated in shellac resin developed considerably more ethanol flavours than carnauba wax-coated fruit during simulated export handling (Hofman et al., 2013).

Given the relatively high rate of respiration by mandarins, the accumulation of off-flavours is a particular issue when fruit are handled and marketed at non-refrigerated temperatures and/or for extended durations. For example, findings from the HAL project CT12000 showed that ethanol off-flavours in commercially picked and packed mandarins were low after a 30-day simulated export shipment at 1.5°C but increased substantially during subsequent shelf life at 25°C (Hofman et al., 2013). It is well established that exposure of fruit to relatively warm temperatures further increases rates of respiration and therefore the potential for elevated concentrations of CO_2 to accumulate within fruit tissues. Additionally, ethanol fermentation is a metabolically-active reaction whereby warmer fruit temperatures can increase enzyme activity required to produce ethanol. Anecdotal evidence suggests that alcohol flavours can be present in Australian mandarins supplied to lucrative export markets such as China (Terry Campbell, pers. comm.).

Although Australian mandarins are popular with consumers, and an increasing number of trees are being planted, very little is known about the effects of flavour on consumer acceptance and purchasing behaviour. Previous international research using trained sensory panels has established that mandarin fruit acidity and flavour acceptability scores decrease substantially during postharvest storage (Tietel et al., 2010). Moreover, wax-coated fruit showed a greater rate of flavour deterioration in association with an increase in off-flavours. Findings from a Queensland Government Global Market Initiative study in Thailand also suggested that local consumers preferred the flavour of non-coated 'Murcott' mandarins over conventionally wax-coated fruit due to their lower levels of off-flavours (Macnish et al. 2014). While use of the more permeable coatings such as carnauba wax can potentially minimise off-flavour development in mandarins, consumer perceptions of their reduced surface shine still need to be determined.

The purpose of this project was to develop a better understanding of the flavour profile in Australian mandarins on the export market, and consumer attitudes towards off-flavours in general, with a focus on China. A range of practical strategies could be applied to consistently enhance the flavour quality of mandarins and thereby encourage repeat purchases and increased consumption.

The objectives of this 1.5-year project were to:

1. Determine the Chinese consumer reaction towards the appearance and eating quality of Australian mandarins following export.
2. Complete preliminary work to identify biochemical tests for quantifying mandarin flavour and correlating to consumer acceptance.
3. Survey Australian mandarins on retail shelves in China to document the fruit flavour profile, including the presence of off-flavours, and consumer reaction to these fruit.
4. Work with supply chain partners to improve specific practices to ensure fruit are delivered with enhanced flavour quality to consumers.

Methodology

Objective 1. Consumer expectations of mandarin quality

'Murcott' mandarins were harvested at commercial maturity from a collaborating grower in Queensland in September 2013. 'Murcott' is a commonly exported mandarin variety. The fruit were processed for export and coated with or without a proprietary carnauba wax product as per commercial practice. Fruit were packed into cartons and transported to the Maroochy Research Facility in Nambour, Queensland. A random selection of fruit were allocated to controlled environment rooms operating at 1.5, 5 and 10°C for 3 weeks to simulate a range of domestic and export handling regimes including. The fruit were then transferred to 20°C for shelf life evaluation. Different sub-samples of fruit were removed every 2 days over 1 week for extraction of juice. The rationale behind these treatments was to generate a broad spectrum of eating quality responses. The fruit juice was frozen and transported to the Health and Food Science Precinct in Coopers Plains, Queensland, for sensory and consumer analysis.

A descriptive sensory panel of 10 participants completed intensive training to familiarise themselves with the visual and eating characteristics of mandarins. As part of the sensory training, the panellists generated a list of key external and internal quality attributes to describe mandarins, including "on-notes" (sensory attributes expected in the product) and "off-notes" (potential sensory defects). The panelists also established a scale for flavour intensity, including off-flavours. Juice samples from the stored fruit were thawed and presented to the expert sensory panel for evaluation. The panellists rated each sample and quantified the flavour attributes. The panel generated a sensory map to identify key sensory attributes of 21 samples. Due to a staff availability issue, we were not able to complete a consumer test on the samples collected during the 2013 season.

Additional 'Murcott' mandarins were harvested at commercial maturity in August 2014 from the same collaborating grower as above with a view towards completing consumer testing. The fruit were processed and subjected to simulated export handling as described above. Based on responses from the 2013 season, nine juice samples that represented fruit from a range of storage regimes were selected and evaluated by a trained sensory panel as outlined above. Following on from the sensory testing phase, five samples were selected to be evaluated by consumers. These products were chosen to best represent the range of variability of flavour quality. One hundred and two consumers of Chinese ethnicity residing in Brisbane were recruited. Preference was given to consumers who had resided in Australia for less than 3 years. The consumer group was comprised of 54 males and 48 females with an average age of 32 years. The consumers were invited to evaluate five mandarin juice samples for the degree of like or dislike using a 9-point hedonic scale where 1 = dislike extremely and 9 = like extremely. The sensory and consumer data were then statistically linked in order to identify the key sensory attributes leading to the decrease in acceptability, and to quantify the impact of these sensory attributes. A more detailed methodology is provided in Appendices 1 and 2.

Objective 2. Biochemical markers to rapidly detect mandarin flavours

Sensory and consumer panels are expensive. Accordingly, an accurate biochemical test based on measuring the juice components contributing to the flavour profile was investigated with a view to reducing the cost of monitoring commercial practices that lead to fruit flavour loss on the export market. Juice from the same fruit used for the sensory panel and consumer test was collected and frozen at -20°C pending chemical analysis. The total soluble solids content (°Brix) of juice samples was measured using a digital refractometer. The organic acid (e.g. citric acid) content of juice was determined by titration with NaOH using an automatic titrator. The BrimA (calculated as °Brix – 4*titratable acidity*16.5), a new measure of fruit juice flavour quality, was also determined. The ethanol concentration of fruit juice (well-established compound responsible for off-flavour) was quantified via an enzyme-based assay (Porat et al., 2005; Obenland and Arpaia, 2011). The overall aim of this work was to quantify and correlate flavour intensity levels as determined by biochemical markers to the sensory panel and consumer expectations. Further details on the employed methodology are provided in Appendices 1 and 2.

Objective 3. Australian mandarin flavour in the Chinese market

To document the fruit flavour profile and consumer reaction to fruit following export and distribution, Australian 'Murcott' mandarins were randomly sampled from retail shelves in Shanghai and Beijing, China during September 2014. Fruit were sampled over several days and from a range of retail outlets. The fruit were analysed in-country for biochemical markers of flavour quality as described above. The Chinese in-country collaborator assisted with these assessments.

We also followed a commercial consignment of 'Murcott' mandarins from a grower in Queensland through importers and wholesalers to retailers in China. The fruit were harvested at commercial maturity, processed for export and coated with or without a proprietary carnauba wax product as described above. The fruit were packed into vented cartons lined with or without plastic modified atmosphere bags, palletised, cooled, and loaded into a refrigerated truck for dispatch to markets. Key supply chain parameters (e.g. temperature, time) were audited for the duration of the supply chain. Representative fruit were sampled at container opening in Shanghai, China, and from the retail shelf, and evaluated as above. Observed commercial practices were documented to identify potential improvements to maximise flavour quality. The consumer acceptance of Australian mandarin appearance and flavour quality was also determined for fruit sampled upon arrival in Shanghai and after a 3-day shelf life in Beijing. In-store consumer tests were completed on fruit samples from the four coating and packaging treatments described above. Groups of 100 consumers of were surveyed in a major supermarket in Shanghai and at a leading green grocer in Beijing that both sold imported mandarin fruit. Freshly peeled fruit segments were tasted. Representative non-peeled fruit from each treatment were also displayed beside each segment for assessment of appearance. The consumer was asked to score samples on a 9-point hedonic scale where 1 = dislike extremely and 9 = like extremely. Additional details on the methodology are described in Appendix 3.

Outputs

- Recommendations on the preferred mandarin appearance and flavour profile for the China market that includes fruit exhibiting relatively low peel shine and citric acid flavour.
- Recommendations to maintain mandarins at 1.5-2.1°C and to minimise the time fruit remain in the system during export handling, distribution and marketing to reduce flavour quality loss.
- Recommendations for the use of an ethanol enzyme assay to rapidly quantify mandarin juice ethanol off-flavours with a view towards predicting potential remaining fruit flavour life.
- Recommendations on the selection and use of high permeability surface coatings that permit sufficient gas exchange through the mandarin peel and minimise ethanol off-flavour development.
- An oral paper presentation to the "Citrus Australia Queensland Growers Pre-season Field Day and Forum" in Gayndah, Queensland on 13 February 2014 by Andrew Macnish that included discussion of the mandarin sensory work. The presentation was attended by 25 citrus growers and industry representatives.
- An oral paper presentation to the 29th International Horticulture Congress in Brisbane, Queensland on 20th August 2014 by Andrew Macnish entitled "Comparison of surface coatings to maintain mandarin fruit quality after export". The presentation was attended by scientists and citrus industry representatives and growers.
- An interview to ABC radio was aired in Queensland on 17 December 2014 in which Andrew Macnish discussed some of the results from the Chinese export market study including Chinese consumer perceptions of Australian mandarins and steps that could be taken to improve fruit quality and better meet customer expectations.
- An oral paper presentation to the "Citrus Australia Technical Forum" in Mildura, Victoria on 17 February 2015 by Andrew Macnish on the mandarin sensory work and fruit quality survey in China. The presentation was attended by about 80 citrus growers and service providers.

Outcomes

Objective 1. Consumer expectations of mandarin quality

The general purpose of this activity was to develop a better understanding of the flavour profile in Australian 'Murcott' mandarins subjected to simulated export handling, and consumer attitudes towards flavour attributes including off-flavours.

We found that fruit harvested at commercial maturity exhibited high initial flavour quality that comprised of satisfactory total soluble solids content, titratable acidity, BrimA and very low (≤ 0.1 g/L) ethanol off-flavours. Coating mandarins with a relatively permeable carnauba wax imparted considerable peel shine that was maintained throughout simulated shipment (1.5, 5, 10·C for 21 days) and shelf life (20·C for up to 8 days) relative to non-coated fruit. In general, the wax coating did not affect fruit soluble solids content, titratable acidity and BrimA during simulated shipment and shelf life. However, the wax-coated fruit typically developed significantly (up to 8-fold) higher juice ethanol concentrations during shipment and shelf life than the non-coated mandarins. This response was more pronounced in fruit harvested during the 2013 season than in 2014. The increase in juice ethanol to relatively high levels (> 1.0 g/L) was also associated with storage or display of fruit at elevated temperature (e.g. 20°C).

A trained sensory panel was able to distinguish differences in the flavour of the wax-coated and non-coated fruit during the 2013 season. The juice from fruit coated with carnauba wax exhibited considerably more "over-ripe" and "musty/mouldy" off-notes than juice from fruit without a wax coating. The intensity of juice flavour off-notes from wax-coated fruit increased with storage temperature (1.5°C to 10°C) and time (day 0 to day 8 of shelf life). The juice from non-wax-coated fruit was described as possessing a distinct and "citrus peel" and "fruit punch" smell and taste. While the juice soluble solids, titratable acidity and BrimA did not vary among the samples, elevated juice ethanol content was associated with over-ripe, musty, mouldy off-notes. On average, juice samples containing ≥ 0.8 g/L ethanol were described by the largely Caucasian panel as having discernible off-flavours. Of interest, when the study was repeated during the 2014 season and extended to a consumer test, conflicting taste preferences were observed. A consumer group of 102 ethnic Chinese showed the greatest degree of liking for juice from the freshly harvested control fruit. They also strongly preferred the flavour of wax-coated fruit that had been stored at 10°C and juiced after a 6-day shelf life. Non-wax-coated fruit stored at 1.5°C prior to juicing were the least preferred sample.

Objective 2. Biochemical markers to rapidly detect mandarin flavours

The purpose of this activity was to identify biochemical tests for rapidly quantifying mandarin flavour quality attributes that closely relate to consumer acceptance.

Previous reports suggested that alcohol off-flavours in mandarins were more accurately determined by measuring the concentration of ethanol in fruit juice rather than quantifying ethanol volatiles produced by intact fruit. We utilised a multi-step enzyme-based assay (Ethanol kits; Megazyme International, Ireland) to measure the ethanol content in 'Murcott' mandarin fruit juice over the range of 0-5 g/L. The kits were relatively easy to use, accurate and permitted rapid throughput. The kits cost about $5 to analyse each juice sample. The assay was used to successfully monitor ethanol off-flavours in mandarin shipments from Australia to China that correlated with a decrease in sensory quality.

Objective 3. Australian mandarin flavour in the Chinese market

The aim of this activity was to survey Australian mandarins on retail shelves in China and document the fruit flavour profile. We also tracked a mandarin consignment from Australia to China and monitored supply chain conditions, fruit out-turn quality and consumer reaction to these fruit.

Four brands of Australian 'Murcott' mandarin fruit were randomly sampled from five retail (green grocer, supermarket) outlets in Shanghai and Beijing during September, 2014. The fruit were sampled from refrigerated (10°C), air-conditioned (20-22°C) and ambient (25-30°C) display environments. All fruit displayed considerable peel shine which suggested they were coated with wax or resin during packing in Australia. The total soluble solids content of the sampled fruit juice was similar (i.e. 11.4-12.7° Brix) for all four brands tested. The titratable acidity of samples varied from 0.52% to 0.79% citric acid equivalents. The variation in °Brix and titratable acidity for each sample was also reflected in the BrimA and all fruit sampled at retail exceeded the minimum BrimA standard of ≥110. Six out of the seven fruit samples exhibited a high juice ethanol content of > 1 g/L.

The consignment of mandarin fruit that we tracked from Australia to China was maintained at a relatively constant 0.6°C and 93-97% relative humidity during the 21-day marine shipment. Thereafter, the fruit were exposed to fluctuating temperatures of 2-12°C during handling in Shanghai. The consignment was then transported by a refrigerated truck to produce markets in Beijing within 24 hours. During transport, the fruit cooled from 12°C to 8°C. The fruit were maintained at 4°C in the distributor's coldroom prior to distribution to a laboratory in Beijing where fruit were maintained at 21-22°C for 3 days to simulate retail display in an air-conditioned supermarket. Considerable condensation was observed to develop on fruit and cartons and contributed to carton collapse. Consumers in both Shanghai and Beijing rated all of the tracked mandarin fruit highly in terms of general appearance and flavour quality. The consumers preferred the appearance and lower acid flavour of the non-coated fruit over the wax-treated samples. Pre-treating fruit with wax reduced the shipment-related weight loss by 33% relative to the non-coated mandarins. Shipping fruit inside plastic bags was even more effective than waxing alone and reduced weight loss by 59-63%. Very few consumers commented on off-flavours in the non-coated and wax-coated fruit; the ethanol content in the test fruit was relatively low (0.4-0.8 g/L) by the end of a 3-day shelf life, and possibly below the threshold of detection.

Evaluation and Discussion

Objective 1. Consumer expectations of mandarin quality

Treating mandarins with waxes or resins is widely practiced to reduce water loss and enhance surface shine. However, these coatings can restrict gas exchange through the peel and trigger off-flavour development (Davis et al., 1973). In the present study, we found that coating 'Murcott' mandarins with a relatively permeable carnauba wax imparted considerable peel shine relative to non-coated fruit. While this wax coating did not affect fruit soluble solids content, titratable acidity and BrimA during simulated shipment and shelf life, ethanol concentrations in the juice of the wax-coated fruit were generally higher than in the non-coated mandarins. Wax-coated fruit harvested during the 2013 season that exhibited elevated ethanol levels were distinguished by a local Australian sensory panel as possessing considerably more "over-ripe" and "musty/mouldy" off-notes than juice from fruit without a wax coating. On average, juice samples containing ≥ 0.8 g/L ethanol were described by the panel as having discernible off-flavours. However, when the study was repeated during the 2014 season, different perceptions of fruit flavour were observed among ethnic Chinese consumers residing in Brisbane to that of the sensory panel. The consumer group preferred the flavour of wax-coated fruit stored at 10°C over non-wax-coated mandarins stored at 1.5°C.

The conflicting response between the trained panel and Chinese ethnic consumers highlight potentially interesting differences in perceptions of mandarin fruit flavour. Unlike the sensory analysis completed in the 2013 season, the flavour preference of Chinese consumers did not appear to be influenced by the ethanol content of the fruit juice. Nonetheless, both the trained panel and consumer group perceived an overall decrease in juice flavour acceptability with an increase in fruit storage temperature and time. Other international sensory panels have previously reported that mandarin fruit show a greater rate of flavour deterioration in association with an increase in off-flavour metabolites during postharvest storage (Tietel et al., 2010). Thus, it is possible that Chinese consumers surveyed in the present study may be detecting flavour defects other than ethanol. Obenland et al. (2013) reported that while ethanol is a key contributor to off-flavour in mandarins, other alcohols (e.g. 3-methyl-1-butanol) and ethyl esters can also build-up in stored fruit and reduce flavour quality. The accumulation of these off-flavour metabolites may also be linked with an increase in juice ethanol levels as ethanol can be metabolised into ethyl esters. Further R&D will be required to document the accumulation of these additional metabolites in Australian mandarins on the export market and how they relate to consumer preferences.

Our findings represent an opportunity to establish a model to predict consumer acceptability from sensory and scientific data. Our sensory and consumer data highlight that mandarin flavour quality is lost during postharvest life. Our scientific data confirm earlier reports that applying semi-permeable coatings such as carnauba wax to mandarin fruit can enhance ethanol development (Cohen et al., 1990; Hagenmaier, 2002; Tietel et al., 2011). Moreover, flavour quality loss is problematic when fruit are held at elevated temperatures (Hofman et al., 2013). Taken overall, these observations point to the possibility that flavour quality defects are present in Australian mandarins on the export market.

Objective 2. Biochemical markers to rapidly detect mandarin flavours

Based on our initial findings that a trained panel's perception of mandarin flavour loss was largely associated with elevated ethanol content in the fruit juice, we investigated methods for rapidly and accurately measuring fruit ethanol concentrations. Previous research by Obenland and Arpaia (2011) established that alcohol off-flavours in mandarins were more accurately determined by measuring the concentration of ethanol in fruit juice rather than quantifying ethanol volatiles produced by intact fruit. Therefore, in the present study, we utilised a multi-step enzyme-based assay (Ethanol kits; Megazyme International, Ireland) to measure the ethanol content in 'Murcott' mandarin fruit juice. We found that the kits were accurate and easy to use. The final quantification step of the assay requires the use of a spectrophotometer. Thus, the assay may be more suited for use by a central testing facility rather than by individual exporters or importers. The assay could potentially be used to monitor ethanol off-flavours in mandarin shipments. This data could be used to predict consumer acceptance, optimal storage conditions and/or remedial action to improve handling practices and fruit eating quality. As part of this project, we tested the kits to measure ethanol off-flavours in Australian mandarins sampled in the Chinese market, as described below. Our consumer data from above indicates that additional R&D may also be required to identify simple tests for detecting other off-flavour metabolites such as esters that may be important in flavour perception by consumers in different export markets.

Objective 3. Australian mandarin flavour in the Chinese market

Our study highlighted that Chinese consumers preferred the appearance of non-coated mandarins relative to carnauba-wax-treated fruit. While the wax coating imparted greater peel shine than the non-coated fruit, consumers apparently could not distinguish this difference under the relatively low light intensity found in retail stores. Pre-treating fruit with wax reduced weight loss from 1.9% to 1.3% during shipment but did not significantly affect weight loss during subsequent distribution and shelf life. Thus, the benefit of the wax treatment in reducing weight loss was not commercially significant. Shipping non-coated fruit in bags more effectively reduced weight loss. However, packing and cooling fruit inside cartons lined with plastic bags may not be practical. In addition, without adequate perforation, the bags may trap condensation and lead to moisture being absorbed into cartons. There is also a risk that elevated CO_2 concentrations will accumulate inside closed bags, particularly during unrefrigerated handling, and this may lead to off-flavours developing in fruit (Kader and Arpaia, 2002). We observed significant breaks in cool-chain handling of mandarins after marine shipment to China. There were often limited cold room facilities at the Shanghai and Beijing markets. Substantial volumes of condensation developed on cold-shipped fruit that were maintained under warm, humid ambient conditions. This condensation contributed to the wetting of cartons resulting in carton collapse and potentially could accelerate fruit flavour loss (Hofman et al., 2013).

Consumers in Shanghai and Beijing also preferred the low acid flavour of non-coated fruit. Higher acid levels and possible tainting with ethanol off-flavours may have reduced the eating quality of the wax-coated fruit. While the ethanol content in the test fruit was relatively low (0.4-0.8 g/L) by the end of a 3-day shelf life, considerably higher levels (> 1 g/L) were detected in other Australian mandarins that were randomly sampled from retailers. The glossy surface of the sampled fruit suggested that they had been treated with a wax or resin coating. While it was not possible to determine fruit history and handling, the juice ethanol concentrations were consistent with mandarins that had been shipped and/or stored at elevated temperature. Mandarins are best stored at 5-8°C to minimise flavour loss (Obenland et al., 2011, 2013). Thus, where refrigerated retail display is not feasible, returning unsold mandarins back into cold storage each night could extend fruit flavour life. Our findings point to the potential for off-flavours to develop in exported mandarins and the need for careful selection of fruit coatings and maintenance of the cool-chain. Our data also highlight the importance of developing a greater understanding of consumer attitudes towards wax- and non-wax-coated mandarins.

In conclusion, this project has established that flavour quality loss in Australian mandarins is a major issue for export markets. Our study revealed that significant breaks in the cool-chain exist during handling and distribution of Australian mandarins in the Chinese market. These extended breaks in refrigerated handling were associated with carton collapse and fruit flavour loss. We suggest that a greater understanding of the Australia-China fresh produce supply chain is required to identify opportunities for developing more strategic partnerships to improve efficiencies. We recommend that importers work closely with distributors to ensure refrigerated storage and transport are available for handling fruit consignments to help facilitate more orderly marketing. We also suggest this represents an ideal opportunity for the Australian industry to train and advise Chinese importers, distributors and retailers on best practices for handling mandarins. From a sensory and consumer science perspective, a greater understanding of the different metabolites that contribute to flavour quality loss as perceived by Chinese consumers is required.

Recommendations

This 1.5-year project characterised the nature and extent of flavour quality loss in Australian 'Murcott' mandarins in the Chinese market. Additional follow-on R&D will now be necessary to implement and evaluate practice change that enhances the export performance of Australian mandarin by improving flavour quality. The following are preliminary recommendations to aid future efforts to reduce the occurrence and commercial impact of off-flavours in mandarins. While the recommendations have a specific focus on 'Murcott' mandarins produced in Queensland for the Chinese market, resolving off-flavour issues will have wider benefits and enhance the export reputation of the whole citrus industry.

- Actively monitor and maintain the cool-chain during export handling using tools such as remote sensing technology with capacity for rapid response to correct temperatures should they exceed the recommended range. This activity will rely on good communication and information flow between all supply chain partners to encourage adoption of best practice.
- Develop, evaluate and careful selection of surface coatings that are sufficiently permeable to maintain adequate gas exchange through the fruit peel. This activity will need to find a balance between maintaining sufficient peel shine and a flavour profile that meets consumer expectations.
- Minimise time in the system such that fruit are picked, packed and exported with minimum delays to maximise the postharvest life and eating quality experience for consumers in export markets. Fruit flavour quality loss is a function of storage temperature and time.
- Routinely monitor fruit out-turn appearance and eating quality including measuring off-flavours by randomly sampling fruit upon arrival in export markets and off retail shelves. We recommend that ethanol kits are used to measure the ethanol content of fruit juice. This data should be related back to supply chain practices so that remedial action can be implemented where required.
- Train and advise chain partners in best handling practices for mandarins, including the importance of handling mandarins under refrigeration and minimising unnecessary storage times.
- Complete future sensory work on fresh fruit and not on frozen juice samples where study logistics allow. This should help eliminate potential confounding factors such as the metabolism of bitter compounds in juiced fruit.
- Undertake additional R&D to better understand the contribution of off-flavour metabolites other than ethanol on consumer and market preferences.

Taken overall, our observations suggest that flavour quality loss represents an impediment to maintaining and increasing the volume and value of Australian mandarin exports to lucrative markets. Our project findings provide the citrus industry with a better understanding of Chinese consumer attitudes towards flavour in Australian mandarins. This should lead to a greater capacity to optimise flavour in our exported fruit, thereby increasing consumer demand for Australian mandarins and greater whole of industry profitability.

Scientific Refereed Publications

None to report.

Intellectual Property/Commercialisation

No commercial IP generated.

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Appendices

Appendix 1. Sensory evaluation of ‘Murcott’ mandarin fruit flavour quality

Appendix 2. Consumer expectations of ‘Murcott’ mandarin fruit flavour

Appendix 3. Australian mandarin flavour quality in the Chinese market

Appendix 1.

Sensory evaluation of 'Murcott' mandarin fruit flavour quality

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Introduction

Mandarin (*Citrus reticulata*) fruit are becoming increasingly popular with consumers largely owing to their convenient easy-to-peel skin and sweet, fruity flavour. While the appearance quality of mandarin fruit is major determining factor in encouraging consumers to make a purchase, the eating quality, including the perception of flavour, is a key driver towards stimulating repeat sales. Sugars, organic acids and volatiles are among important components of mandarin fruit flavour. The balance of sugars (^{o}Brix) and acids are widely associated with good eating quality. However, the influence of other flavour attributes such as off-flavours on consumer purchasing behaviour are not well understood.

Despite their increasing popularity, mandarins are significantly more perishable than other citrus species (Kader and Arpaia, 2002). Mandarin fruit display relatively high rates of respiration and a peel that is less permeable to gas exchange compared to grapefruit (Shi et al., 2005, 2007). In addition, mandarin juice exhibits relatively high activities of pyruvate decarboxylase and alcohol dehydrogenase, the two enzymes required to produce acetaldehyde and ethanol off–flavours (Davis et al., 1973; Shi et al., 2005). Taken together, these features may explain the higher susceptibility of mandarins to lose flavour quality.

Previous international research using trained sensory panels has established that mandarin fruit acidity and flavour acceptability scores decrease substantially during postharvest storage (Tietel et al., 2010). Moreover, wax-coated fruit showed a greater rate of flavour deterioration in association with an increase in off-flavours. While the practice of coating mandarins after harvest with various waxes can enhance their marketability by imparting shine and reducing water loss, it can also restrict gas exchange through the peel (Hagenmaier and Baker, 1993). The resulting decrease in O_2 diffusion into fruit and concurrent increase in internal CO_2 levels can trigger anaerobic respiration and the accumulation of off-flavour metabolites such as ethanol and acetaldehyde in the fruit juice (Cohen et al., 1990; Hagenmaier, 2002; Tietel et al., 2011).

Off-flavour development is likely influenced by cultivar, surface coatings such as waxes, and the storage and holding conditions (temperature and time) often experienced during sea-freight and in-country distribution. This represents a potential impediment to increasing the value of Australian mandarin exports to lucrative markets. The primary purpose of the current study was to develop a better understanding of the flavour profile in Australian mandarins on the export market.

Materials and Methods

Plant material and general processing

Late-season 'Murcott' mandarin (*Citrus reticulata*) fruit were harvested at commercial maturity (15.1°Brix, 0.88% citric acid, 191 BrimA) in September 2013 from an orchard near Mundubbera in Queensland. 'Murcott' is the most commonly exported Australian mandarin variety. The fruit were prepared for export on the following day at a nearby packing shed following standard commercial procedures. Briefly, the fruit were loaded onto a packing line consisting of rotating brushes and overhead sprayers and graded for uniform quality. The fruit were washed in a sanitising solution and treated with postharvest fungicides. They were then treated with a proprietary carnauba wax-based surface coating at 700 mL/tonne to permit evaluation of this commonly applied treatment on fruit flavour quality. The precise composition of the coating is a trade secret and not publicly available. Additional fruit that were not coated with wax served as a control. All fruit were pattern-packed (100-count) into standard C6 (18 kg capacity) cardboard citrus cartons. Fruit in cartons were transported to the DAF Maroochy Research Facility laboratory in Nambour, Queensland within 4 hours.

Simulated shipment and handling

Upon arrival in the laboratory, six replicate cartons of wax and non-wax-treated fruit were randomly assigned to controlled environment rooms operating at 1.5, 5 and 10°C and 90% relative humidity. The fruit were maintained at each temperature for 21 days to simulate a range of export and domestic market shipping and handling regimes. Data loggers that recorded the air temperature and relative humidity were placed into each room. Thereafter, fruit in cartons were transferred to a 20°C room for shelf life evaluation. Fruit were sampled on days 0, 4, 6 and 8 of shelf life to generate a broad spectrum of eating quality responses.

Fruit appearance quality

Eight wax-coated and non-coated fruit were randomly selected and evaluated for general appearance, including peel shine, before, during and after simulated storage and shelf life. The fruit were rated using a 0-5 scale with 0 being of low quality and 5 of high quality.

Fruit juice quality

Twenty non-coated fruit were randomly selected from each carton before cold storage to furnish day 0 (at harvest) samples. Thereafter, 20 non-coated and 20 wax-coated fruit were sampled from each carton at the end of simulated cold storage and again at days 4, 6 and 8 of shelf life. At each sampling time, the 20 fruit from each carton were pooled together. The juice from each sub-sample of 20 fruit was extracted using a domestic juicer and filtered through two layers of cheesecloth to remove seeds and pulp. The juice was collected in 2 L plastic bottles and 15 mL plastic tubes and held in a freezer at -20°C pending analysis.

The frozen juice was thawed and analysed for total soluble solids (°Brix), titratable acidity (% citric acid), BrimA (calculated as °Brix – 4*titratable acidity*16.5; Jordan et al., 2001, Obenland et al., 2009) and ethanol content (g/L). The total soluble solids of juice samples were determined using an Atago PAL-1 digital handheld refractometer. The titratable acidity was determined by titrating a juice sample with 0.1 N NaOH to pH 8.2. The ethanol content of juice was determined using an enzymatic test kit (Megazyme International, Ireland) according to the manufacturer's instructions. Each juice sample was analysed in duplicate.

Fruit juice taste

Juice samples were evaluated by a 10-member experienced trained sensory panel. Briefly, the panellists were recruited from the DAF Health and Food Sciences Precinct at Coopers Plains, Queensland. They were selected on the basis of their interest, availability and capacity to evaluate sensory attributes of mandarins. The panellists attended six training sessions of 1 hour in duration to familiarise themselves with the sensory characteristics of mandarins. As part of the training, the panellists generated a list of appearance, flavour, aroma, texture and aftertaste terms were selected to rate during the formal evaluation sessions (Table 1). Key internal quality attributes to describe mandarins, including "on-notes" (sensory attributes expected in the product) and "off-notes" (potential sensory defects) were developed. The panellists were exposed to random juice samples to practice evaluating the attributes using a line scale on a score sheet and to achieve consensus for how to rate the attributes. The panellists also practiced evaluating juice samples in the sensory booths, using automated data collection (Compusense Five software, Ontario, Canada).

Seven formal evaluation sessions were held on consecutive weekdays, during which panellists evaluated the different juice samples in duplicate. Eight to ten samples were presented in total at each session. Panellists were provided with one sample at a time (50 mL) in individually blind coded plastic cups (100 mL) with lids. A randomised presentation design was used within each replicate for all trials, ensuring no two panellists received samples in the same order. Each juice sample was served with a cup of water and some unsalted saltine crackers as palate cleansers. Evaluation was performed in individual sensory booths at room temperature (20°C). The panelists were asked to evaluate the juice sensory attributes on an unstructured 15 cm line scale using Compusense Five software. At least five panellists were present for all evaluation sessions. The overall aim of this work was to establish a scale for flavour intensity and generate a sensory map to identify key attributes.

Table 1. Attributes used for the sensory analysis of 'Murcott' mandarin fruit juice.

Sensory attribute	**Descriptor term**
Appearance	Orange, Cloudiness
Aroma	Overall intensity, Fruit punch / Fruit salad, Passion fruit, Orange, Mango, Citrus peel, Overripe, Musty / Mouldy
Texture	Smooth, Viscous
Flavour	Overall intensity, Bitter, Sweet, Acidic, Fruit Punch / Fruit salad, Passion fruit, Orange, Mango, Citrus peel, Over-ripe, Musty / Mouldy
Aftertaste	Bitter, Sweet, Acidic, Astringent

Results and Discussion

'Murcott' fruit exhibited high flavour quality at harvest. On average, the soluble solids content of juice was 15.1°Brix while the titratable acidity was 0.88% citric acid equivalents (Figure 1). These relatively high soluble solids and low acidity data were consistent with fruit harvested later in the season. During mandarin fruit maturation, the total soluble solids content increases whereas the acidity decreases (Holland et al., 1999). As a consequence of the high soluble solids and low acids content, the BrimA at harvest was high at 191 and in excess of the minimum Australian maturity standard of 110 (CAL, 2015). The initial ethanol content in fruit juice was low (0.1 g/L), indicative of low levels of off-flavours at harvest.

Coating mandarins with carnauba wax imparted considerable peel shine relative to non-coated fruit (data not shown). There were no consistent effects of fruit wax coatings, shipment temperatures and time on the juice soluble solids, acids and BrimA during simulated shipment and shelf life (Figure 1). In contrast, fruit coated with carnauba wax developed 2- to 8-fold higher juice ethanol concentrations during shipment and shelf life than the non-coated mandarins (Figure 1). In wax-coated fruit, the ethanol content in juice increased substantially to moderate to high levels (0.4-1.2 g/L) during the 3-week simulated shipment. These ethanol concentrations rapidly increased to high levels (1.0-1.5 g/L) during the subsequent shelf life at 20°C. In non-coated fruit, the juice ethanol content remained low (< 0.2 g/L) during shipment but increased steadily to moderate levels (0.5-0.9 g/L) during shelf life. Maintaining fruit at 5 and 10°C during shipment resulted in a more rapid increase in juice ethanol concentrations during shelf life than for fruit shipped at 1.5°C.

These findings confirm earlier reports that applying semi-permeable coatings such as carnauba wax to mandarin fruit can enhance ethanol off-flavour development (Cohen et al., 1990; Hagenmaier, 2002; Tietel et al., 2011). Our data also highlight that ethanol off-flavour metabolism is particularly problematic when fruit are held at elevated temperatures (Hofman et al., 2013). Taken overall, these observations point to the very real possibility that that alcohol flavours could be present in Australian mandarins on the export market.

A trained sensory panel was able to distinguish differences in the flavour of the juice samples (Figure 2). While the samples were well distributed on the sensory map, those with similar flavour attributes could be grouped into four distinct clusters. The juice from fruit coated with carnauba wax exhibited considerably more "over-ripe" and "musty/mouldy" off-notes than juice from fruit without a wax coating. The intensity of juice flavour off-notes from wax-coated fruit increased with storage temperature (1.5°C to 10°C) and time (day 0 to day 8 of shelf life). The juice from non-wax-coated fruit was described as possessing a distinct and typical "citrus" smell and taste. Non-wax-coated fruit sampled immediately after simulated shipping exhibited a "citrus peel" flavour, while fruit stored at 5 and 10°C and sampled later during shelf life (e.g. days 6 and 8) developed a more complex "fruit punch" smell and taste. Other sensory panels have also established that the perception of mandarin fruit flavour can vary during postharvest storage (Tietel et al., 2010). Moreover, wax-coated fruit showed a greater rate of flavour deterioration in association with an increase in off-flavours.

There were no differences in the juice soluble solids, titratable acidity and BrimA among samples from the four flavour clusters (Table 2). However, elevated juice ethanol content was associated with over-ripe, musty, mouldy off-notes. On average, juice samples containing ≥ 0.8 g/L ethanol were described as having discernible off-flavours. This finding represents an opportunity to establish a model to predict consumer acceptability from sensory and scientific data. As a next step, the juice samples will be offered to Chinese consumers to determine if the trained sensory panel data trends match those of consumers.

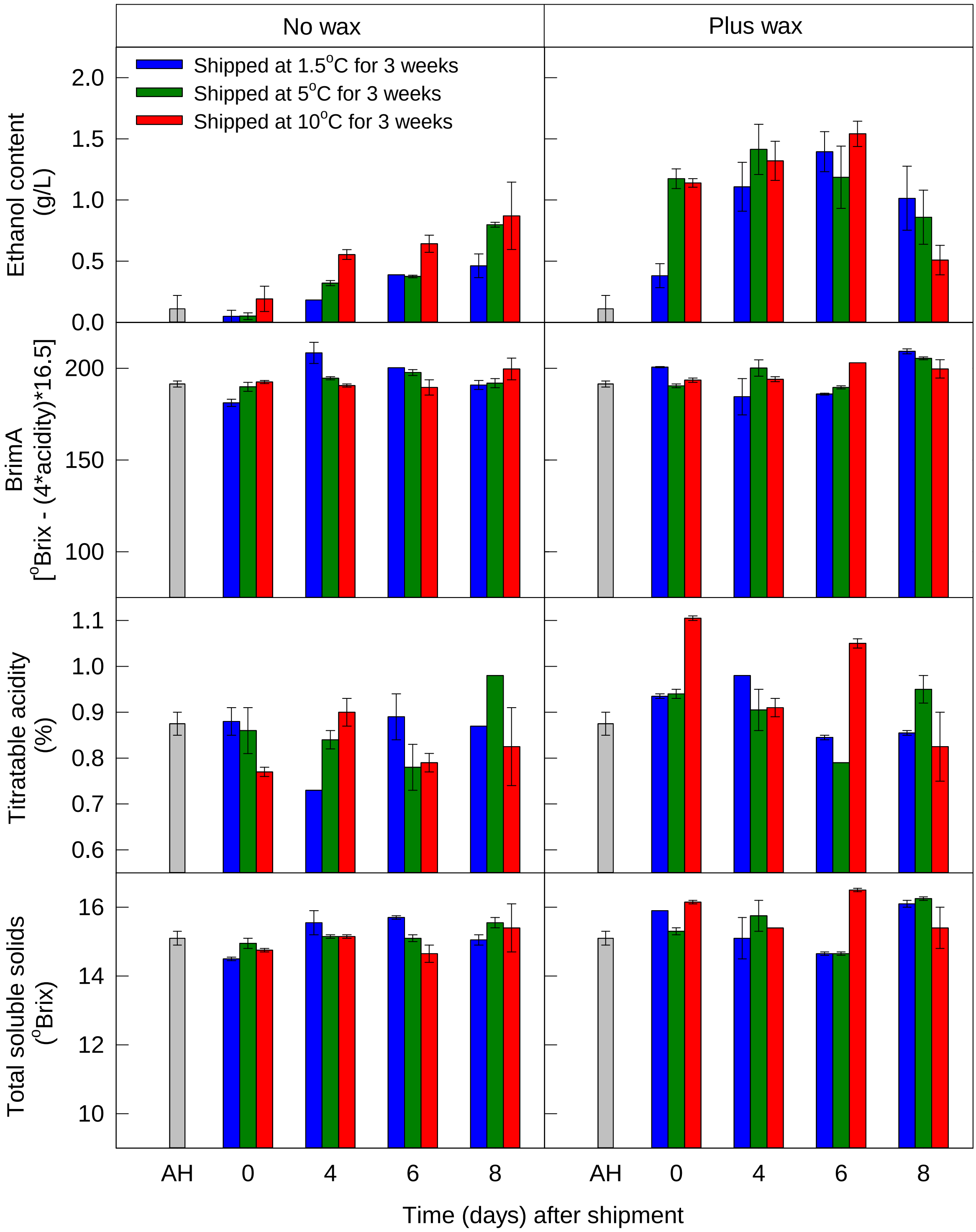

Figure 1. Changes in selected flavour properties of 'Murcott' mandarin fruit juice during and after simulated shipping (3 weeks at 1.5, 5, 10°C) and shelf life (8 days at 20°C). The fruit were pre-treated with or without a carnauba wax-based coating and maintained inside vented cardboard cartons. AH represents "at harvest" or the initial harvest data. Vertical bars represent the standard errors of means.

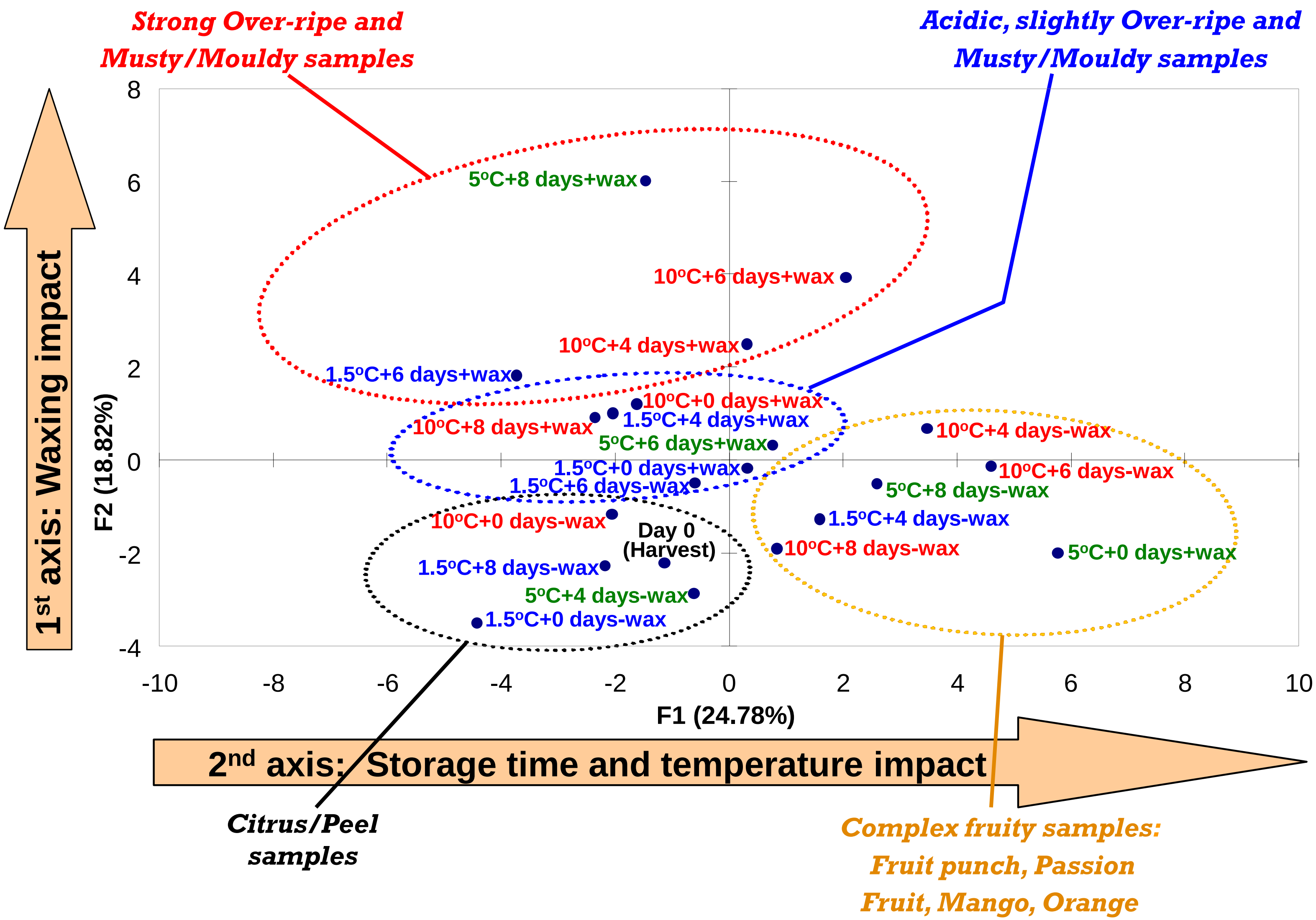

Figure 2. A sensory preference map for 'Murcott' mandarin fruit juice flavour showing the position of 21 samples as evaluated by a trained panel. Samples were grouped into four clusters on the basis of flavour attributes. Fruit were pre-treated with (+) or without (-) a carnauba wax-based coating. They were subjected to simulated shipping (1.5, 5, 10°C for 3 weeks) and shelf life (20°C for 8 days). Juice samples were evaluated at harvest (day 0) and during shelf life (0, 4, 8 days).

Table 2. Average total soluble solids, titratable acidity, BrimA and ethanol content of four clustered samples of 'Murcott' mandarin fruit juice. Individual fruit juice samples were assigned by a trained sensory panel to the clusters on the basis of flavour attributes (see Figure 2). The fruit were pre-treated with or without a carnauba wax-based coating. They were subjected to simulated shipping (1.5, 5, 10°C for 3 weeks) and shelf life (20°C for 8 days).

Fruit flavour cluster	Total soluble solids (°Brix)	Titratable acidity (%)	BrimA	Ethanol content (g/L)
Strong over-ripe, musty, mouldy	15.7 ± 0.4 NS	0.94 ± 0.04 NS	197 ± 4 NS	1.28 ± 0.15 a
Slightly over-ripe, musty, mouldy	15.4 ± 0.2	0.92 ± 0.05	195 ± 3	0.79 ± 0.16 b
Citrus peel	14.9 ± 0.1	0.82 ± 0.02	190 ± 2	0.23 ± 0.07 c
Complex fruity	15.3 ± 0.1	0.86 ± 0.04	195 ± 3	0.70 ± 0.14 b

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Appendix 2.

Consumer expectations of 'Murcott' mandarin fruit flavour

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Introduction

Mandarin (*Citrus reticulata*) fruit are increasing in popularity amongst consumers, largely because of their easy-to-peel skin. This rise in consumption has prompted increased plantings of mandarins in Australia and a greater focus on export marketing (ABS, 2013). Despite their popularity, the storage life of mandarins is often limited to 2-4 weeks (Kader and Arpaia, 2002). High rates of respiration, moisture loss and flavour deterioration are among factors that limit the performance of mandarins.

The loss of mandarin flavour quality after harvest is often associated with the build-up of anaerobic metabolites such as ethanol and acetaldehyde in the fruit juice (Davis et al., 1973). These fermentative off-flavours develop in response to reduced rates of O_2 influx and CO_2 efflux through the fruit surface (Cohen et al., 1990; Hagenmaier, 2002). While it is customary to apply wax or resin-based coatings to the surface of mandarins to increase peel shine and decrease moisture loss, these treatments can further restrict gas exchange and exacerbate off-flavours (Hagenmaier and Baker, 1993). Given the high respiration rate of mandarins, the build-up of internal CO_2 and associated off-flavours is an issue when fruit are handled at non-refrigerated temperatures (Hofman et al., 2013).

Anecdotal evidence suggests that alcohol flavours can develop in mandarins supplied to export markets where continuous refrigeration may not be available (Terry Campbell, pers. comm.). This potentially represents an impediment to increasing the volume and value of Australian mandarin exports to markets in Asia. Although Australian mandarins are popular with consumers, very little is known about the effects of off-flavours on consumer acceptance and purchasing behaviour. In our previous work (Appendix 1), we established that a trained panel could distinguish increased levels of 'off-notes' in wax-coated versus non-coated 'Murcott' mandarins following simulated export handling. The intensity of off-flavours was also highest for fruit handled at elevated (e.g. 10·C) temperatures.

The purpose of the present study was to identify and describe mandarin fruit off-notes and measure their impact on consumer liking.

The main objectives of this study were to:

1. Develop individual organoleptic profiles for different mandarin juice samples
2. Determine the desirable and non-desirable sensory properties of different mandarin juice samples among Asian consumers
3. Repeat 2013 season work so that a consumer test could be completed

Materials and Methods

Plant material and general processing

Mid-season 'Murcott' mandarin (*Citrus reticulata*) fruit were harvested at commercial maturity (12.1°Brix, 0.91% citric acid, 139 BrimA) in August 2014 from an orchard near Mundubbera in Queensland. The fruit were prepared for export at a nearby packing shed following standard commercial procedures employed during the 2013 season (Appendix 1). Briefly, the fruit were loaded onto a packing line consisting of rotating brushes and overhead sprayers and graded for uniform quality. They were washed in a sanitising solution, treated with fungicides and coated with a proprietary carnauba wax at 700 mL/tonne. Additional fruit that were not coated with wax served as a control. All fruit were pattern-packed (100-count) into standard C6 (18 kg capacity) cardboard cartons. Fruit in cartons were transported to the DAF Maroochy Research Facility laboratory in Nambour, Queensland within 4 hours.

Simulated shipment and handling

Upon arrival in the laboratory, eight replicate cartons of wax and non-wax-treated fruit were randomly assigned to controlled environment rooms operating at 1.5, 5 and 10°C and 90% relative humidity. The fruit were maintained at each temperature for 21 days to simulate a range of export and domestic market shipping and handling regimes. Data loggers that recorded the air temperature and relative humidity were placed into each room. Thereafter, fruit in cartons were transferred to a 20°C room for shelf life evaluation. Fruit were sampled on days 0, 2, 4, 6 and 8 of shelf life to generate different eating quality responses.

Fruit appearance quality

Eight wax-coated and non-coated fruit were randomly selected and evaluated for general appearance, including peel shine, before, during and after simulated storage and shelf life. The fruit were rated using a 0-5 scale with 0 being of low quality and 5 of high quality.

Fruit juice quality

Fifteen non-coated fruit were randomly selected from each carton at harvest to furnish day 0 samples. Thereafter, 15 non-coated and 15 wax-coated fruit were sampled from each carton at the end of simulated cold storage and again at days 2, 4, 6 and 8 of shelf life. At each sampling time, the 15 fruit from each carton were pooled together and their juice was extracted using a domestic juicer. The juice was filtered through a strainer and collected in 2 L plastic bottles and 15 mL plastic tubes and held in a freezer at -20°C pending analysis.

The frozen juice was thawed and analysed for total soluble solids (°Brix), titratable acidity (% citric acid), BrimA (calculated as °Brix – 4*titratable acidity*16.5; Jordan et al., 2001, Obenland et al., 2009) and ethanol content (g/L). The total soluble solids of juice samples were determined using an Atago PAL-1 digital handheld refractometer. The titratable acidity was determined by titrating a juice sample with 0.1 N NaOH to pH 8.2. The ethanol content of juice was determined using an enzymatic test kit (Megazyme International, Ireland) according to the manufacturer's instructions. Each juice sample was analysed in duplicate.

Descriptive sensory analysis of fruit juice

Conventional quantitative descriptive analysis was employed for the sensory evaluation of juice samples in line with procedures followed in the 2013 season (Appendix 1). In 2014, a 10-member experienced trained panel was recruited from the Health and Food Sciences Precinct, Coopers Plains.

A minimum of eight panellists were present at any one tasting, however, all 10 panellists attended every formal evaluation session. The panellists ranged in age from 31 to 54 years, comprising six female and four male panellists of a range of nationalities and ethnic backgrounds including Australian, Indian, English and Indonesian.

Six 45-60-minute training sessions were conducted over 2 weeks, and involved four discussion sessions and two individual practice sessions using the computers in the booths. Training sessions took place in an open board room where participants evaluated products both individually and as a group, followed by discussion led by the panel leader (Philippa Tyler). During this time, panellists generated a large set of descriptive terms derived from tasting the entire sample set. The terms collated by the panel were reduced to a short, accurate and relevant list, via panel discussion. By consensus, a list of aroma, appearance, flavour, texture/mouth-feel and aftertaste terms were selected for use during the formal evaluation sessions (Table 1). For each term developed, panellists were provided with a definition and a standard, where suitable. Sensory reference standards were developed using relevant food stuffs. The practice booth session ensured the panellists were familiar with the set-up and were confident in using the line scales and terms agreed upon.

Table 1. 'Murcott' mandarin juice samples used in the sensory evaluation.

Sample code	**Storage temperature (°C)**	**Shelf life (days)**	**Fruit surface coating**
950	At harvest	-	No wax
735	1.5	0	Plus wax
821	1.5	0	No wax
502	1.5	6	Plus wax
649	1.5	6	No wax
381	5	6	Plus wax
447	5	6	No wax
172	10	6	Plus wax
208	10	6	No wax

Formal evaluation sessions were held on consecutive days, during which panellists evaluated the juice samples in triplicate over three 60-minute sessions. Based on results from the 2013 season, nine samples were selected for evaluation (Table 2). The nine frozen juice samples, each in 1 L plastic bottles, were defrosted at room temperature (22°C) for 12 hours. Once defrosted, samples were individually prepared by straining through a muslin cloth into glass carafes. Formal evaluations were conducted in the sensory laboratory at the Health and Food Sciences Precinct, Coopers Plains. This facility contains twelve isolated booths equipped with computers, temperature control (22°C) and under day-light equivalent lighting (Figure 1).

During the formal evaluation sessions, panellists were provided with a set of freshly prepared reference standards, a printed attribute definition list (Table 1), and fresh drinking water and water crackers for palate cleansing. Panellists were provided with 50 mL of each sample at a time in individually blind coded 100 mL clear plastic cups with lids (Figure 2). A randomised presentation design was used within each replicate, ensuring no two panellists received samples in the same order. Panellists stirred each sample with a clean plastic spoon prior to assessment to ensure any sediment present was mixed throughout the sample. The panellists were asked to evaluate the juice sensory attributes on an unstructured 15 cm line scale using Compusense Five software. Panellists took a 10-minute break from evaluations after tasting three samples, where they were encouraged to cleanse their palates.

Table 2. Attributes used for the sensory analysis of 'Murcott' mandarin fruit juice.

Attribute	0	100	Description
Appearance			
Colour intensity	Low	High	Intensity of colour from low (lemon) to high (burnt orange, similar to apricot nectar)
Translucence	Low	High	How see-through a sample is, from low (opaque) to high (see a spoon through sample)
Aroma			
Intensity	Weak	Intense	
Fruit cup cordial	Not perceived	Intense	Aroma like Cottees fruit cup orange cordial. Also like orange flavoured ice block/concentrate, with notes of tropical/passion fruit
Citrus pith	Not perceived	Intense	Slightly pungent, bitter and pithy aroma with a citrus note
Soy/Beany	Not perceived	Intense	Aroma of hexanal – slightly rancid oil type, similar to silken tofu/beany
Flavour			
Intensity	Weak	Intense	
Acidity	Weak	Intense	Flavour associated with citric acid solution
Sweetness	Weak	Intense	Flavour associated with sucrose solution
Bitterness	Weak	Intense	Flavour associated with caffeine solution and citrus pith/seeds
Fruit cup cordial	Not perceived	Intense	Flavour like Cottees fruit cup orange cordial. Also like orange flavoured ice block/concentrate, with notes of tropical/passion fruit
Citrus pith	Not perceived	Intense	Slightly pungent, bitter and pithy flavour with a citrus note
Texture / mouth-feel			
Viscosity	Low	High	Degree to which sample has a nectar-like, coating characteristic
Tooth-stick	Not perceived	High	Presence of 'tackiness' on the teeth after consumption
Astringency	Not perceived	High	Degree to which sample causes mouth drying, roughness in the cheek pouch
Aftertaste			
Intensity	Weak	Intense	
Bitterness	Not perceived	Intense	After taste associated with caffeine solution and citrus pith/seeds

Figure 1. Individual sensory booths at the Coopers Plains laboratory.

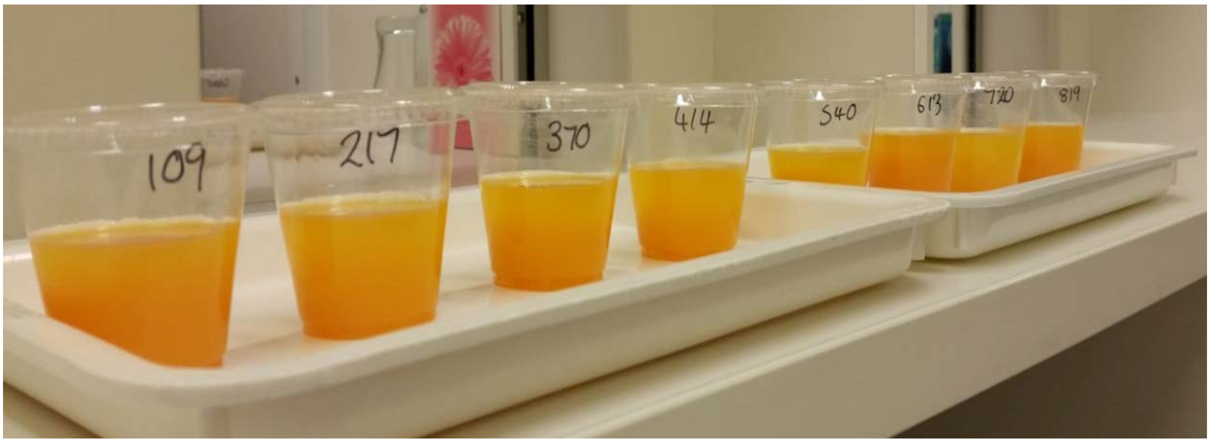

Figure 2. Photograph showing 50 mL servings of each 'Murcott' mandarin juice sample.

Consumer evaluation of fruit juice

One hundred and two consumers of Chinese ethnicity residing in Brisbane were recruited by staff at the Maroochy Research Facility in Nambour. Preference was given to consumers who had resided in Australia for less than 3 years. The consumer group was comprised of 54 males and 48 females with an average age of 32 years. The consumers were invited to evaluate mandarin juice samples at the Coopers Plains facility on 29 and 30 November, 2014. They were rewarded with a $50 EFTPOS card for participating in the juice evaluation.

Based on the descriptive sensory analysis above, five out of the nine juice samples were selected for evaluation by consumers (Table 3). These products were chosen to best represent the range of variability of flavour quality. The juice samples were defrosted and strained as described above. Aliquots of 30 mL of each sample were transferred into disposable clear 50 mL plastic cups covered with plastic lids. The cups were labelled with a series of three-digit blind codes. Five groups of 20 consumers each were randomly allocated to a 60-minute session. The consumers were presented with the five samples of juice together with a paper questionnaire (Figure 3; Supplementary material), water and water crackers for palate cleansing. Consumers were asked to rate the overall acceptability, appearance, aroma, flavour, texture/mouth-feel and aftertaste of each sample using a structured 1-9 hedonic line scale where 1 = dislike extremely, 5 = neither like nor dislike, and 9 = like extremely. Assessments were made under controlled conditions (Figure 4).

Figure 3. Photograph showing the five samples of mandarin juice and a paper questionnaire that was presented to each consumer.

Figure 4. Photograph of the mandarin juice sample evaluation by consumers under controlled conditions at the Health and Food Sciences Precinct, Coopers Plains.

Results and Discussion

Fruit appearance and juice quality

'Murcott' fruit exhibited high appearance and flavour quality at harvest. The soluble solids content of juice was 12.1°Brix and the titratable acidity was 0.91% citric acid equivalents (Figure 5). The BrimA at harvest was 139 and easily met the minimum Australian maturity standard of 110 (CAL, 2015). The initial ethanol content in fruit juice was just 0.02 g/L, indicative of very low levels of off-flavours at harvest.

As reported for the 2013 study, coating mandarins with carnauba wax imparted significantly more peel shine as compared to non-coated fruit. This enhanced level of peel shine was maintained throughout simulated shipment and shelf life (data not shown). The wax coating treatment did not affect fruit soluble solids but did appear to increase fruit acids content, particularly towards the end of the 6-day shelf life (Figure 5). Despite the differences in fruit acids content, there were no significant differences in BrimA during simulated shipment and shelf life (Figure 5). There were also no consistent effects of the simulated shipment temperature on fruit appearance and juice quality.

The ethanol content of fruit juice remained at low levels (i.e. < 0.2 g/L) during simulated shipment at 1.5, 5 or 10°C for 21 days for both wax- and non-wax-coated mandarins (Figure 5). Thereafter, juice ethanol increased to moderate to high levels (i.e. 0.5-1.2 g/L) upon transfer to shelf life display at 20°C. The ethanol content was only slightly more elevated in wax-coated fruit relative to non-coated mandarins. This observation differs from our findings in the 2013 season (Appendix 1) and numerous other reports that applying semi-permeable coatings to mandarin fruit can greatly enhance ethanol off-flavours (Cohen et al., 1990; Hagenmaier, 2002; Tietel et al., 2011). Of interest, the ethanol content in wax-coated and non-coated fruit shipped at 10°C reached a maximum by day 4 of shelf life before decreasing over time. A similar trend was observed for wax-coated fruit shipped at 1.5°C. This decrease in fruit juice ethanol concentration during later stages of shelf life may simply reflect ethanol metabolism or depletion (Pesis, 2005). For example, ethanol can be converted into other off-flavour metabolites such as ethyl esters. In contrast, both the wax- and non-wax-coated fruit shipped at 5°C exhibited a sustained increase in ethanol concentration throughout shelf life.

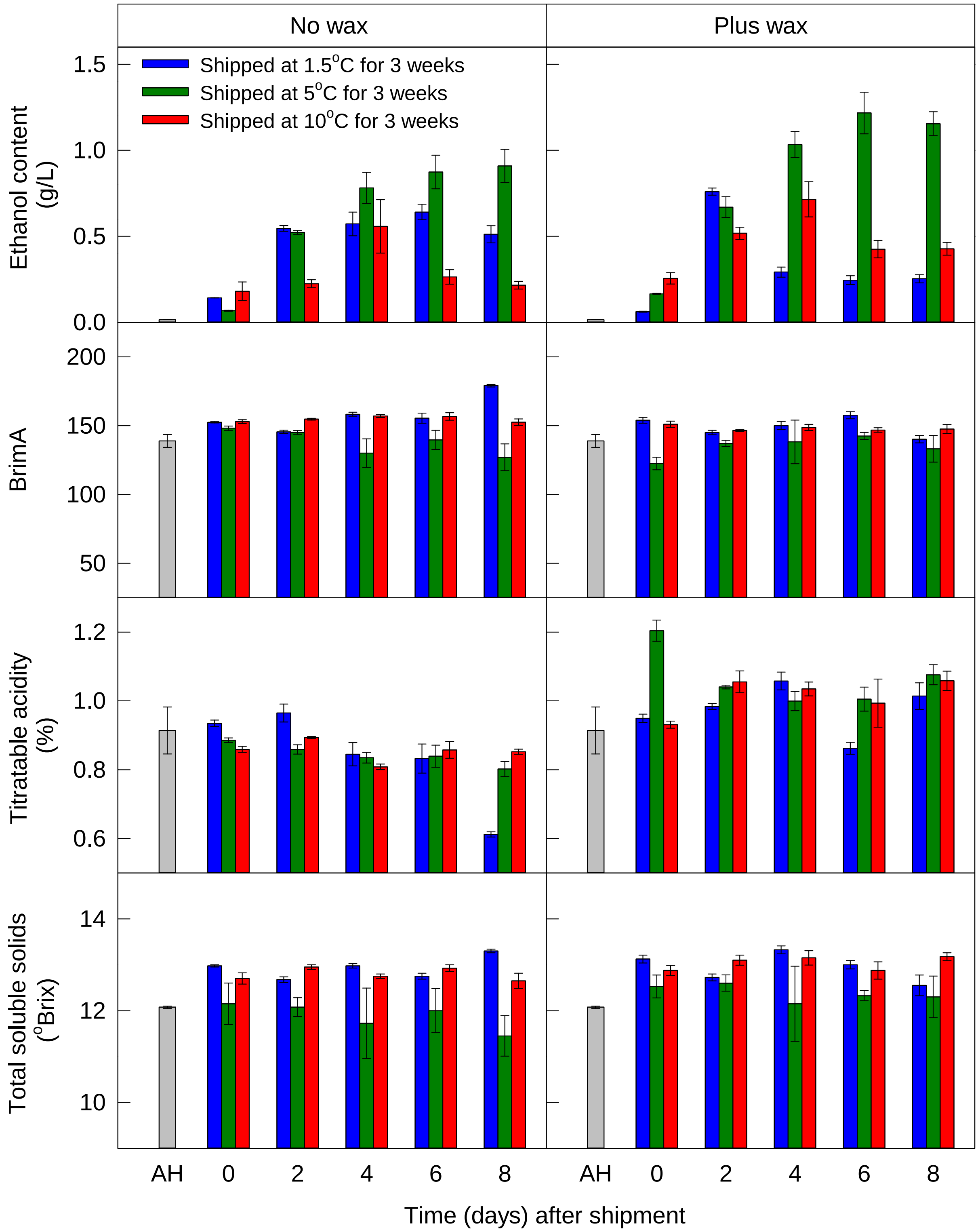

Figure 5. Changes in selected flavour properties of 'Murcott' mandarin fruit juice during and after simulated shipping (3 weeks at 1.5, 5, 10°C) and shelf life (8 days at 20°C). The fruit were pre-treated with or without a carnauba wax-based coating and maintained inside vented cardboard cartons. AH represents "at harvest" or the initial harvest data. Vertical bars represent the standard errors of means.

Sensory descriptive analysis

Nine out of the 31 fruit samples tested were selected for sensory analysis. These samples included a freshly harvested control and several wax-coated and non-coated fruit that were juiced immediately after simulated shipment at 1.5, 5 and 10·C or on day 6 of shelf life (Table 1). Fruit subjected to shipment and shelf life handling generally displayed higher total soluble solids and lower acids content relative to the control fruit (Table 3). Of notable exception, fruit shipped at 5·C exhibited relatively low soluble solids content, while wax-coated fruit shipped at 5 and 10·C maintained comparatively high titratable acidity. These attributes translated into a relatively high BrimA for all samples except the controls and fruit shipped at 5·C. The juice ethanol content was lowest in fruit sampled at harvest (control) and immediately after shipment (Table 3). Ethanol concentrations were moderate to high in all other samples, particularly for the wax-coated fruit.

Using standard descriptive analysis techniques, a panel of 10 assessors rated the nine samples for two appearance, four aroma, six flavour, three texture/mouth-feel and two aftertaste attributes (Table 2). The results of the descriptive analysis are shown in the Principal Component Analysis (PCA) bi-plot in Figure 6. Within the first two principals 71% of data variation was explained. Key findings from the sensory analysis included:

- Non-wax-coated fruit stored at 5°C for 3 weeks and sampled after a 6-day shelf life at 20°C were judged to exhibit the *sweetest* flavour of all the samples. These fruit also had the least *astringent texture* and no *soy/beany* notes.
- Wax-coated fruit stored at 1.5°C and 10°C and sampled on day 6 of shelf life displayed a distinctively more *viscous texture/mouth-feel* and a *fruit cup cordial aroma*.
- Comparatively, non-coated fruit stored at 1.5°C and sampled on day 0 of shelf life had a *bitter flavour and aftertaste* with a *soy/beany aroma.*
- Non-coated fruit held at 10°C and sampled after 6 days at 20°C were identified by the sensory panel as exhibiting the highest level of *citrus pith aroma* and *flavour.*
- Non-coated fruit stored at 1.5°C and wax-coated fruit stored at 5°C and sampled on day 6 of shelf life were less distinct in their organoleptic profiles. Both of these samples exhibited low intensity *flavours* and *aromas*, with a balance between the *sweet fruit cup cordial* characteristics and the *bitter* notes.
- Wax-coated fruit stored at 1.5°C and sampled on day 6 of shelf life had a similar profile in terms of *flavours* and *aromas*; however it was far more *viscous* in *texture/mouth-feel* than the non-wax-coated fruit, which were the least *viscous* of all samples.
- Juice from freshly harvested control fruit was described as having a *low intensity, fruit cup cordial flavour* and *aroma,* very little *tooth stick texture/mouth-feel* when consumed and a non-*viscous texture/mouth-feel.*
- Juice extracted from wax-coated and non-coated fruit that had been stored at 1.5·C possessed a distinctive *soy/beany aroma*. The panel associated the *soy/beany aroma* with rancid oil, akin to silken tofu, an unpleasant characteristic for mandarin juice.

Unlike the sensory analysis completed in the 2013 season (Appendix 1), the sensory profile of the samples in 2014 did not appear to be influenced by the ethanol content of the fruit juice. This study highlights the sometimes complex nature of matching instrumental measures of fruit flavour quality with human sensory descriptors.

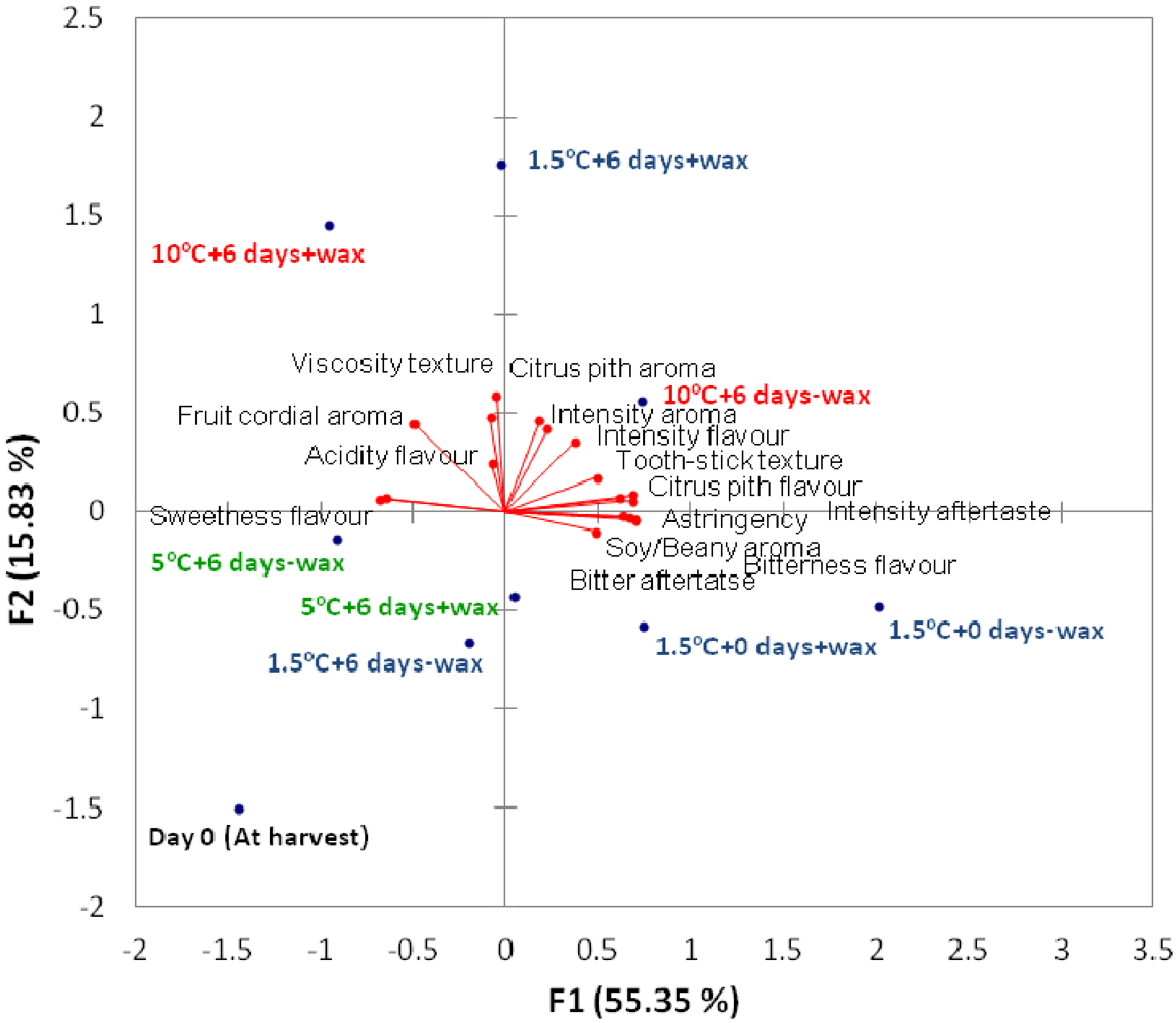

Figure 6. A Principal Component Analysis bi-plot of sensory descriptive data for 'Murcott' mandarin juice samples (PV1 v PV2). Mandarin fruit were pre-treated with or without a carnauba wax coating and stored at 1.5, 5 or 10°C for 21 days. The fruit were then transferred to 20°C for up to 6 days prior to juicing. A trained sensory panel evaluated the fruit juice for different aroma, aftertaste, flavour and texture attributes.

Nine mandarin juice samples were evaluated by a trained sensory panel. The samples represented wax-coated and non-coated mandarin fruit that had been stored at 1.5, 5 and 10°C for 21 days plus shelf life at 20°C for 0-6 days prior to juicing and evaluation.

Table 3. Average total soluble solids, titratable acidity, BrimA and ethanol content of nine samples of 'Murcott' mandarin fruit juice evaluated for flavour quality. The fruit were pre-treated with or without a carnauba wax-based coating. They were subjected to simulated shipping (1.5, 5, 10°C for 3 weeks) and shelf life (20°C for 0 or 6days). The fruit juice was then frozen and later analysed for selected flavour characteristics.

Storage temperature (°C)	**Shelf life (days)**	**Fruit coating**	**Total soluble solids (°Brix)**	**Titratable acidity (%)**	**BrimA**	**Ethanol (g/L)**
At harvest	-	No wax	12.1 ± 0.0 c[1]	0.91 ± 0.07 ab	139 ± 5 d	0.02 ± 0.00 f
1.5	0	Plus wax	13.1 ± 0.1 a	0.95 ± 0.01 ab	154 ± 2 ab	0.14 ± 0.00 ef
1.5	0	No wax	13.0 ± 0.0 a	0.93 ± 0.01 ab	152 ± 0 abc	0.06 ± 0.00 f
1.5	6	Plus wax	13.0 ± 0.1 a	0.86 ± 0.02 b	158 ± 3 a	0.64 ± 0.05 c
1.5	6	No wax	12.8 ± 0.1 ab	0.83 ± 0.04 b	155 ± 4 ab	0.25 ± 0.03 e
5	6	Plus wax	12.3 ± 0.1 bc	1.00 ± 0.03 a	142 ± 3 cd	1.22 ± 0.12 a
5	6	No wax	12.0 ± 0.5 c	0.84 ± 0.03 b	140 ± 7 d	0.87 ± 0.10 b
10	6	Plus wax	12.9 ± 0.2 a	0.99 ± 0.07 a	147 ± 2 bcd	0.43 ± 0.05 d
10	6	No wax	12.9 ± 0.1 a	0.86 ± 0.02 b	157 ± 3 ab	0.26 ± 0.04 de

[1] Data followed by different letters are significantly different at $P = 0.05$.

Consumer evaluation

Of the nine mandarin samples evaluated by the trained panel, five samples that broadly represented the full spectrum of flavour attributes were selected for evaluation by a consumer group that consisted of 102 ethnic Chinese (Table 4). Key findings included:

- Consumers showed the greatest degree of liking for juice from the freshly harvested control fruit.
- Consumers also strongly preferred the flavour of wax-coated fruit that had been stored at 10°C and juiced after a 6-day shelf life.
- Both of the control and wax-coated fruit scored a 6 or above for all attributes on the 9-point scale, including *overall*, *appearance*, *aroma*, *flavour*, *texture/mouth-feel* and *aftertaste*.
- Non-wax-coated fruit that had been stored at 1.5°C prior to juicing were the least preferred sample overall, scoring on the 'dislike' scale (less than 5) for the attributes *flavour*, *texture/mouth-feel*, *aftertaste* and *overall liking*.

The consumer data was subjected to a hierarchical cluster analysis to determine groups of consumers with similar preference patterns. This test was conducted with the aim of defining groups of consumers who preferred the same types of juice samples and who also shared similar demographics. Three groups or clusters of consumers were identified with similar preferences across the five juice samples tasted in this study. The results of the preferences of these three clusters are summarised in Figure 7. The differentiating features (in terms of preferences) between the consumer clusters are presented in Table 5 and the corresponding cluster demographics are listed in Table 6.

Consumers in cluster 1 comprised 50% of the population sampled in this study (Table 6). These consumers showed a strong overall liking for the juice from freshly harvested control fruit (Table 5). Wax-coated fruit that had been stored at 10°C and juiced after a 6-day shelf life were also preferred by cluster 1 consumers. Both of these samples were judged to exhibit a high level of *fruit cup cordial flavour* and *sweetness*. Cluster 1 consumers particularly disliked the *flavour* and *aftertaste* attributes of the juice from non-wax-coated fruit that had been stored at 1.5°C or 10°C. These two samples were described by the sensory panel as having a *bitter*, *citrus pith flavour* and *soy/beany aroma*. Cluster 1 consumers were relatively young in age (29 years) and had resided in Australia for 3 years.

Consumers in cluster 2 also showed a very strong preference for the juice from freshly harvested control fruit (mean score of 7.6) relative to all other samples (Table 5). Cluster 2 consumers displayed a particularly strong dislike for wax-coated fruit that had been stored at 10°C and juiced after a 6-day shelf life, with *flavour*, *texture/mouth-feel* and *aftertaste* seemingly responsible for this response. In contrast, this wax-coated sample was generally favoured by the consumer population as a whole. Cluster 2 accounted for 20% of the population sampled, had an average age of 30 years, and had lived in Australia for the shortest period of time; 2 years on average.

Consumers in cluster 3 were of the oldest average age of 38 years. Cluster 3 consumers showed a distinct preference for the juice from wax-coated and non-coated fruit that had been stored at 10°C (Table 5). The consumers considered that these juice samples exhibited high *aroma intensity* and a high level of *citrus pith*. These two samples were, however, distinctly different from one another in terms of their organoleptic profile, indicating that consumers within cluster 3 did not have a particular flavour preference.

Table 4. Consumer acceptance of five different 'Murcott' mandarin juice samples. Mandarin fruit were pre-treated with or without a carnauba wax coating and stored at 1.5, 5 or 10°C for 21 days. The fruit were then transferred to 20°C for up to 6 days prior to juicing. Five fruit juice samples were then evaluated for flavour quality by 102 consumers of Chinese ethnicity. A hedonic scale was used to rate samples where 1 = strongly dislike and 9 = strongly like.

Fruit treatments			**Juice quality parameters**					
Storage temperature	**Shelf life at 20°C**	**Coating**	**Appearance**	**Aroma**	**Flavour**	**Texture**	**Aftertaste**	**Overall liking**
10°C	6 days	Plus wax	6.7 ab*	6.3 a	6.1 a	6.4 a	6.1 a	6.2 a
10°C	6 days	No wax	6.7 ab	6.0 ab	4.7 b	5.4 c	4.9 b	5.0 b
5°C	6 days	Plus wax	6.7 ab	6.3 a	5.2 b	5.6 bc	5.0 b	5.4 b
1.5°C	0 days	No wax	6.3 b	5.6 b	4.0 c	4.7 d	4.1 c	4.1 c
At harvest	-	-	7.0 a	6.5 a	6.0 a	6.3 ab	6.2 a	6.3 a

* Letters within columns indicate a significant difference by Tukey LDS ranking test. Significant differences were found between samples for all variables (*p<0.0001*).

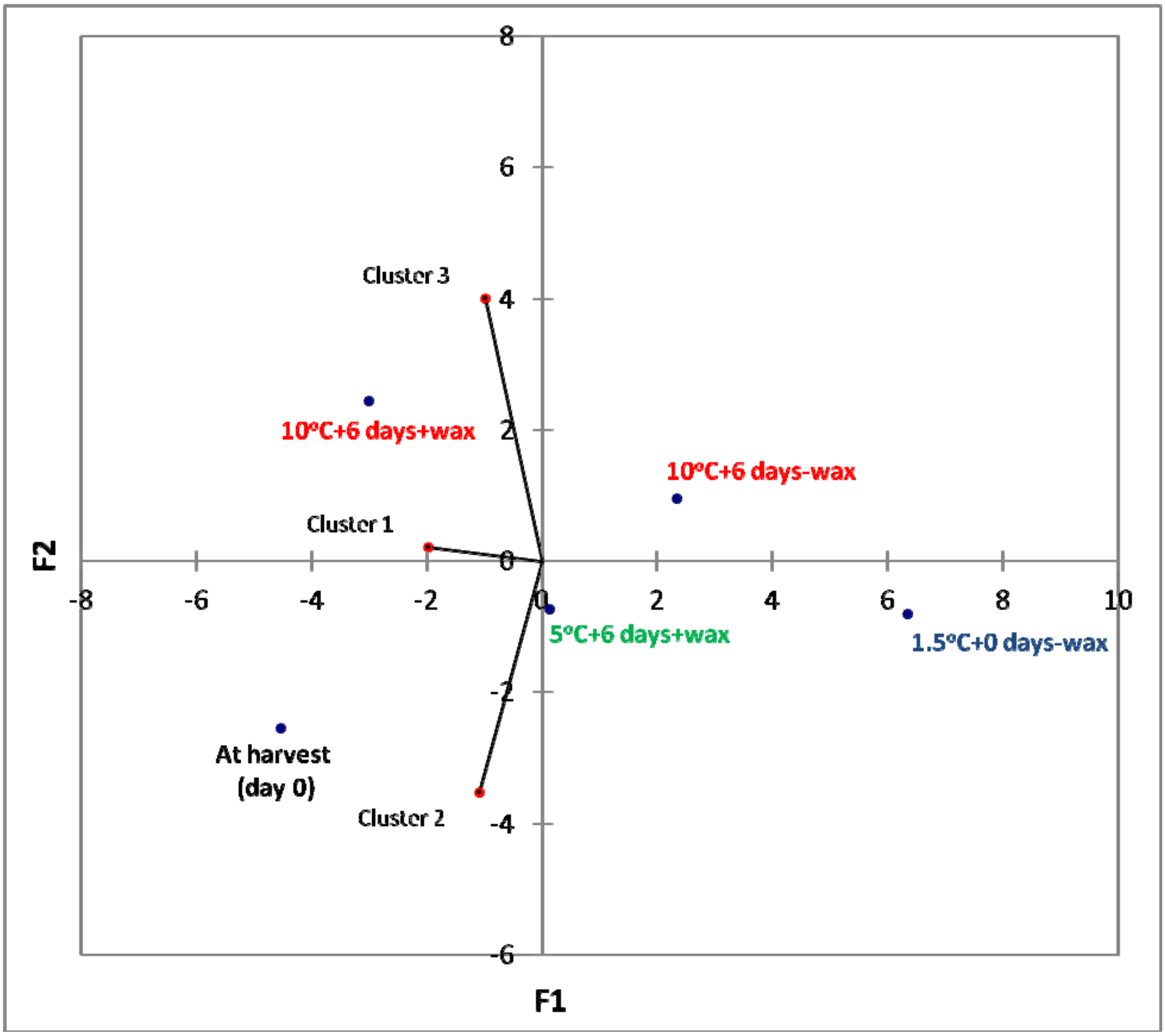

Figure 7. Preference map of three consumer clusters for 'Murcott' mandarin juice samples. Mandarin fruit were pre-treated with or without a carnauba wax coating and stored at 1.5, 5 or 10°C for 21 days. The fruit were then transferred to 20°C for up to 6 days prior to juicing. Five fruit juice samples were then evaluated for flavour quality by 102 consumers of Chinese ethnicity. Cluster 1 (n=50), cluster 2 (n=20), cluster 3 (n=32).

Table 5. Hedonic scores assigned by three clusters of consumers of Chinese ethnicity to describe flavour quality attributes of five 'Murcott' mandarin juice samples. Mandarin fruit were pre-treated with or without a carnauba wax coating and stored at 1.5, 5 or 10°C for 21 days. The fruit were then transferred to 20°C for up to 6 days prior to juicing. Five fruit juice samples were then evaluated for flavour quality.

Consumer cluster	Fruit treatments	Juice quality parameters					
		Appearance	Aroma	Flavour	Texture	Aftertaste	Overall liking
Cluster 1	**10°C+6 days+wax**	6.7	6.4	6.3	6.6	6.5	6.6
	10°C+6 days-wax	6.6	5.6	3.9	4.7	4.1	4.1
	5°C+6 days+wax	6.7	6.4	5.5	5.8	5.2	5.6
	1.5°C+0 days-wax	6.1	5.5	3.8	4.5	3.8	3.8
	At harvest control	6.9	6.6	6.2	6.4	6.3	6.5
Cluster 2	**10°C+6 days+wax**	6.4	5.9	4.5	5.4	4.6	4.4
	10°C+6 days-wax	6.7	6.1	5.3	5.7	5.3	5.5
	5°C+6 days+wax	6.5	6.3	5.4	5.5	5.2	5.7
	1.5°C+0 days-wax	6.2	5.6	4.6	5.0	4.4	4.6
	At harvest control	7.4	7.1	7.6	7.4	7.5	7.6
Cluster 3	**10°C+6 days+wax**	6.9	6.3	6.6	6.6	6.4	6.8
	10°C+6 days-wax	6.9	6.5	5.7	6.3	5.9	6.2
	5°C+6 days+wax	6.8	6.2	4.8	5.5	4.8	4.9
	1.5°C+0 days-wax	6.5	5.8	4.0	4.8	4.2	4.4
	At harvest control	6.8	6.0	4.8	5.3	5.1	5.1

Table 6. Demographics of the three clusters of Chinese consumers that participated in the evaluation of 'Murcott' mandarin juice samples.

Consumer characteristics	Cluster 1	Cluster 2	Cluster 3
Male	24	12	18
Female	26	8	14
Average age	29 years	30 years	38 years
Average length of residency in Australia	3 years	2 years	2.5 years

Preference mapping

Preference mapping was completed to model consumer and sensory data across the five mandarin juice samples (Figure 8). Typically, consumers liked mandarin juices with a *fruit cup cordial flavour* and *aroma,* together with *sweetness* and *acidity.* They also liked samples with a more *translucent appearance* and a less *viscous texture/mouth-feel*, namely the freshly harvested control fruit and wax-coated fruit that had been stored at 10°C. Consumers did not like mandarin juices with a *bitter, citrus pith flavour, soy/beany aroma* and a *tooth-stick texture/mouth-feel*, in particular non-coated fruit stored at 1.5°C.

Overall we found that consumers show a strong preference for the sweet, tropical-like flavours and those with less bitterness and no soy/beany aroma. While the soy/beany unpleasant characteristic was not associated with juice ethanol content, it was detected in fruit stored at 1.5°C. As mandarins must presently be shipped from Australia to Asia at ≤ 2°C to meet quarantine requirements, it would be desirable to gain a better understanding of the soy/beany flavour attribute. The non-wax-coated samples held at 1.5°C or 5°C exhibited sweeter notes and a juice sample that was less viscous in texture/mouth-feel. In comparison, the most viscous juice samples, with a balance of citrus pith and sweet characteristics were wax-coated and shipped at 10°C. Taken together, these results highlight the importance of understanding consumer preferences and modifying or improving postharvest practice to deliver fruit that meet customer expectations.

The observation that some of the non-wax-coated fruit samples were quite bitter in taste may reflect the presence of naringin or limonin, naturally occurring bitter compounds in citrus (Puri et al., 1996). These compounds are typically only metabolised once fruit juice sacs are ruptured during processing. Because it is possible that the presence of these bitter compounds in juiced fruit may have confounded the perception of other flavour attributes, we recommend that future mandarin sensory analysis be completed on fresh fruit samples where study logistics allow.

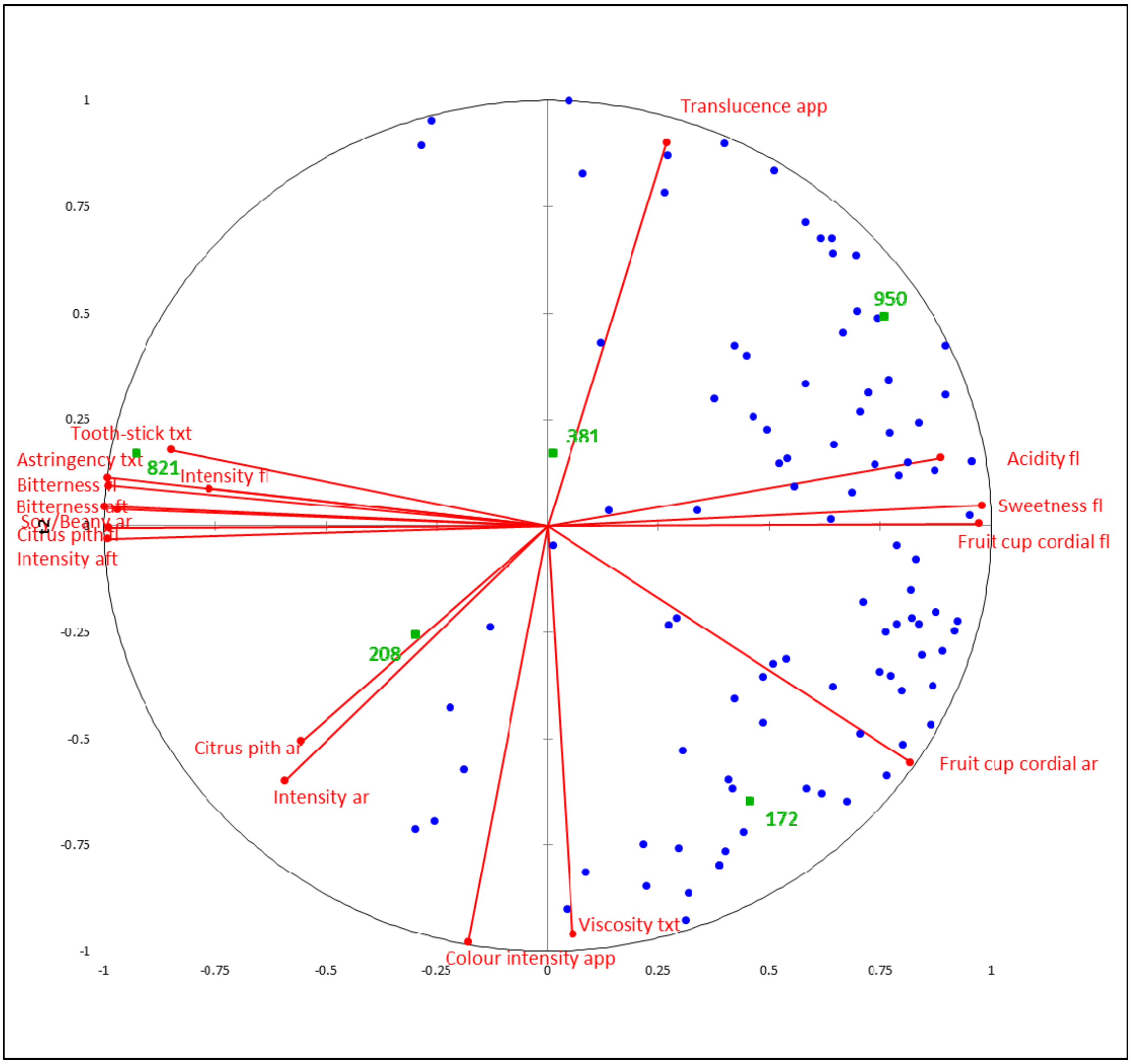

Figure 8. Preference map of consumer clusters for five different 'Murcott' mandarin juice samples. Mandarin fruit were pre-treated with or without a carnauba wax coating and stored at 1.5, 5 or 10°C for 21 days. The fruit were transferred to 20°C for up to 6 days prior to juicing. Juice samples were frozen and later thawed and evaluated for flavour quality by 102 consumers of Chinese ethnicity. Refer to Table 1 for sample (green codes) information.

Acknowledgements

This project was funded by Horticulture Innovation Australia Ltd using voluntary contributions from the Department of Agriculture and Fisheries, Queensland and matched funds from the Australian Government. We gratefully acknowledge Sweetee Citrus, Central Fruit Packers and FAVCO for providing fruit for testing. In particular, we record special thanks to Greg Parr (Sweetee Citrus), Brent Chambers (Central Fruit Packers), and Darryl Lowe (FAVCO) for their professional advice and assistance.

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Supplementary material. An example of the consumer taste testing questionnaire.

Consumer Taste Testing

Mandarin Juice

Hello and welcome to today's taste testing. Thank you for agreeing to take part and help us with this study.

Today you have been provided with 5 samples of mandarin juice, each with their own 3-digit code.

You will be asked to taste each of these samples individually and complete a simple questionnaire relating to your liking of these samples.

You shall be asked how much you like the Aroma, the Appearance, the Flavour, the Texture and the Aftertaste, as well as how much you like the sample Overall.

There are no right or wrong answers today; I am looking for your own personal opinion ☺

Firstly, you will be asked to answer a few demographic questions, relating to age, gender and ethnic background.

Please begin when you are ready and feel free to raise your hand should you have any questions.

Demographic questions:

1. Age *(please underline the correct answer)*
    a. Under 18 years
    b. 18-24 years
    c. 25-34 years
    d. 35-44 years
    e. 45-54 years
    f. 55-64 years
    g. 65+ years

2. Gender *(please underline correct answer)*
    a. Male
    b. Female

3. What would you consider to be your primary language? *(please detail below)*
   ....
   ....

4. How long have you lived in Australia? *(please detail below)*
   ....
   ....

5. What was your country of birth? *(please detail below)*
   ....
   ....

6. What is your ethnic background? *(please detail below)*
   ....
   ....

Thank you
Please turn over to begin your sample assessments

Please ensure you are assessing sample 381

1. How much do you like the **APPEARANCE** of this sample? *(please tick one box)*

Dislike extremely	Dislike very much	Dislike moderately	Dislike slightly	Neither like nor dislike	Like slightly	Like moderately	Like very much	Like extremely
☐	☐	☐	☐	☐	☐	☐	☐	☐

2. How much do you like the **AROMA** of this sample? *(please tick one box)*

Dislike extremely	Dislike very much	Dislike moderately	Dislike slightly	Neither like nor dislike	Like slightly	Like moderately	Like very much	Like extremely
☐	☐	☐	☐	☐	☐	☐	☐	☐

3. How much do you like the **FLAVOUR** of this sample? *(please tick one box)*

Dislike extremely	Dislike very much	Dislike moderately	Dislike slightly	Neither like nor dislike	Like slightly	Like moderately	Like very much	Like extremely
☐	☐	☐	☐	☐	☐	☐	☐	☐

4. How much do you like the **TEXTURE** of this sample? *(please tick one box)*

Dislike extremely	Dislike very much	Dislike moderately	Dislike slightly	Neither like nor dislike	Like slightly	Like moderately	Like very much	Like extremely
☐	☐	☐	☐	☐	☐	☐	☐	☐

5. How much do you like the **AFTERTASTE** of this sample? *(please tick one box)*

Dislike extremely	Dislike very much	Dislike moderately	Dislike slightly	Neither like nor dislike	Like slightly	Like moderately	Like very much	Like extremely
☐	☐	☐	☐	☐	☐	☐	☐	☐

6. How much do you like this sample **OVERALL**? *(please tick one box)*

Dislike extremely	Dislike very much	Dislike moderately	Dislike slightly	Neither like nor dislike	Like slightly	Like moderately	Like very much	Like extremely
☐	☐	☐	☐	☐	☐	☐	☐	☐

Appendix 3.

Australian mandarin flavour quality in the Chinese market

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Introduction

Mandarin fruit are becoming increasingly popular with consumers owing to their easy-to-peel skin and sweet, fruity flavour. This rise in consumption has prompted increased plantings of mandarins in Australia (ABS, 2013). Queensland presently accounts for about 50% of Australian mandarin production. China, Indonesia, New Zealand, Thailand and the UAE are among the main export markets. China is the most valuable and fastest growing market for Australian mandarins (QGSO, 2015).

The export performance of mandarin fruit is sometimes limited by internal (e.g. off-flavours) fruit quality defects. Mandarins are inherently more perishable than oranges and display higher rates of respiration, water loss, decay and flavour loss during storage and shelf life (Kader and Arpaia, 2002). It is also customary to apply waxes or resins to the peel of mandarins after harvest to increase fruit surface shine and decrease water loss. Treating fruit with coatings can, however, lead to the development of off-flavours, particularly for fruit coated with relatively impermeable waxes or resins (Cohen et al., 1990; Hagenmaier, 2002; Tietel et al., 2011).

We previously reported that considerable levels of ethanol off-flavour metabolites can develop in Australian 'Murcott' mandarin fruit during and after simulated intercontinental marine shipment (see Appendix 1). Off-flavour development is likely influenced by surface wax coatings and the storage and holding temperature during sea-freight and in-country distribution. Accordingly, fruit off-flavour issues represent a potential impediment to maintaining and increasing the volume and value of Australian mandarin exports to lucrative markets such as China.

The general purpose of the current study was to document the flavour profile of 'Murcott' mandarin fruit and the consumer reaction to fruit following export from Australia to China. The key research activities were:

1. Determine the impact of surface coatings and packaging systems on fruit flavour quality, including off-flavour development

2. Document key supply chain parameters (e.g. temperature, time) with a view to identifying potential improvements to maximise fruit flavour quality

3. Survey the flavour quality of Australian mandarins sampled from retail shelves in China

Materials and Methods

Fruit material

'Murcott' mandarins were harvested at commercial maturity on 7 August 2014 from an orchard near Mundubbera, Australia. The fruit were loaded onto a packing line consisting of rotating brushes and overhead sprayers and graded for uniform quality. The graded fruit were washed in a sanitising solution and treated with fungicides at the label rate. Half of the fruit were coated with a relatively permeable carnauba wax at 700 mL/tonne in line with commercial practice. The remaining fruit were not treated with a wax coating.

Coating and packaging treatments

We hypothesised that non-wax-treated fruit would be less prone to developing off-flavours after export as compared to fruit coated with carnauba wax. We anticipated that the non-wax-treated fruit would, however, lose more water during and after export than the coated fruit. To this end, we evaluated the potential of shipping fruit inside a closed plastic bag within cartons as a means to reducing weight loss. With a view towards finding a balanced approach to enhancing the appearance and eating quality of mandarins, the following surface coating and packaging treatments were tested:

1. Wax-treated fruit in conventional two-piece C6 cardboard cartons.
2. Non-wax-treated fruit in conventional two-piece C6 cardboard cartons.
3. Wax-treated fruit in conventional two-piece C6 cardboard cartons plus a plastic bag.
4. Non-waxed fruit in conventional two-piece C6 cardboard cartons plus a plastic bag.

Each C6 carton was comprised of corrugated cardboard and designed to hold 18 kg of fruit.

General processing and shipment

Wax and non-wax-coated fruit (100 count) were pattern-packed into five replicate cartons per treatment. The plastic bag that lined half of the conventional cartons was closed to establish a modified atmosphere of elevated CO_2 and reduced O_2 concentrations around fruit. A datalogger that recorded temperature and relative humidity was inserted into randomly selected cartons. Representative cartons from each wax coating and packaging treatment were randomly built into the bottom two layers of three pallets (Figure 1). The pallets were then transferred to a cool room at 1.5°C for 72 hours.

Commercial shipment

The three test pallets were loaded at the front, middle and rear of a 40-foot-long refrigerated marine container. Additional pallets of pre-cooled mandarins were loaded into the container to ensure a full load. The container was transported by truck from Mundubbera to the Brisbane seaport within 6 hours. The consignment was then sea-freighted from Brisbane, Australia, to Shanghai, China in 21 days. Upon arrival at the importer's facility in Shanghai, the pallets were inspected by Customs and transferred to a refrigerated truck within 48 hours. The pallets were transported by road to Beijing, China within 24 hours as per commercial protocols. They were maintained in a cool room for 24 hours. The pallets were broken down and the experimental cartons were transported in an air-conditioned van to the COFCO laboratory near Beijing in 1.5 hours. The cartons were then maintained at 20°C for 3 days to simulate retail display.

Figure 1. Photograph of a representative pallet prior to shipment showing the experimental cartons on the bottom two layers.

Carton atmosphere

The concentration of CO_2 and O_2 inside cartons and closed plastic bags was determined upon arrival of the shipment in Shanghai and Beijing using a gas analyser. Following CO_2 and O_2 measurement in Beijing, the bags were opened.

Fruit appearance quality

Ten fruit from each carton were randomly selected, labelled and rated for general appearance quality, including peel shine, using the following 5-point scale: 5 = excellent, 4 = good, 3 = moderate, 2 = slight, 1 = poor. Fruit appearance was determined at the beginning of shipment in Mundubbera, upon arrival in Shanghai, and at the end of shelf life in Beijing.

Fruit weight loss

The same 10 fruit from each carton were combined and weighed at packing in Mundubbera, upon arrival in Shanghai and after shelf life in Beijing to enable calculation of weight loss.

Fruit total soluble solids, acids and ethanol content

An additional 10 fruit from each test carton were selected at random. The peel was removed and fruit segments were separated. The juice from two segments per fruit was extracted by hand and collected in 15 mL plastic tubes. The juice in tubes was frozen immediately and maintained at -20ºC for chemical analysis at the COFCO laboratory. Briefly, the total soluble solids content (expressed as ^{o}Brix) of each juice sample was measured using a digital refractometer. The acids content or titratable acidity (expressed as % citric acid) of juice was determined by titrating a diluted sample of juice with 0.1 N NaOH to pH 8.2. The ethanol content (expressed as g/L) of juice was determined using an enzymatic test kit (Megazyme International, Ireland) according to the manufacturer's instructions.

Consumer tests

The consumer acceptance of Australian mandarin appearance and flavour quality was determined for fruit sampled upon arrival in Shanghai and after a 3-day shelf life in Beijing. In-store consumer tests were completed on fruit samples from the four coating and packaging treatments described above. Groups of 100 consumers of Chinese ethnicity representing a diverse combination of age and gender were surveyed in a major supermarket in Shanghai and at a leading green grocer in Beijing that both sold imported mandarin fruit (Figure 2). Freshly peeled fruit segments selected from the 10 fruit described above were placed into labeled plastic containers for taste testing. Representative non-peeled fruit from each treatment were also displayed beside each segment for assessment of appearance. Consumers who responded that they ate fresh mandarins were surveyed. For each sample, the consumer was asked to score the four samples on a 9-point hedonic scale where 1 = dislike extremely and 9 = like extremely. The consumers were offered a water cracker and a cup of water to cleanse their palates between tasting each sample.

Figure 2. Photographs showing the in-store consumer tests in a supermarket in Shanghai (A, B) and a green grocer in Beijing (C, D).

Survey of Australian mandarins at retail in China

To document the flavour profile of fruit following export and distribution, Australian 'Murcott' mandarins were sampled from retail shelves in Shanghai and Beijing following typical sea-freight and in-country transport and storage. Five replicate fruit from four different growers, as determined by the PLU sticker, were randomly selected from two retail outlets in Shanghai and three retailers in Beijing. On the same day of sampling, the peel of each fruit was removed. The juice from two flesh segments per fruit was extracted by hand and collected in 15 mL plastic tubes. The tubes of juice were immediately stored at -20·C pending chemical analysis. The juice was analysed in-country at the COFCO laboratory for total soluble solids, acids and ethanol content as described above.

Results and Discussion

Shipment conditions and supply chain capabilities

The consignment of mandarin fruit tracked from Australia to China was maintained at a relatively constant 0.6°C during the 21-day marine shipment (Figure 3). This is slightly below the recommended 2.1°C for intercontinental citrus shipments. However, no chilling injury symptoms were observed on fruit. The packed fruit were also maintained at 93-97% relative humidity (RH) during sea-freight (Figure 3). Upon arrival at the Shanghai seaport, the container was transported by road to the Huizhan fruit markets, the main fruit importing facility in Shanghai. The container was opened and inspected by Customs agents. The fruit were exposed to fluctuating temperatures of 2-12°C and 95-100% RH during the 1-day inspection and handling process in Shanghai.

There were limited cold room facilities at the Huizhan market. Most fruit consignments remained inside numerous refrigerated containers until being transferred to trucks for distribution to wholesalers and retailers across China (Figure 4). However, some consignments were removed from the refrigerated containers and maintained at ambient conditions (ca. 28-32°C, 70-90% RH) for several minutes to hours before being loaded into trucks and dispatched. Selected pallets from different shipments were also often partially broken down and displayed by wholesale agents on the market floor at ambient conditions (Figure 4). Substantial volumes of condensation developed on cold-shipped apples, avocados, mandarins, oranges and pears that were maintained at ambient conditions (Figure 5). While the handling practice was variable, considerable carton bulging and partial collapse was associated with fruit held outside the cool-chain (Figure 6).

Following inspection and handling, the tracked mandarin consignment was loaded into a refrigerated truck and transported to the Xinfadi produce markets in Beijing within 24 hours. During transport, the fruit cooled from 12°C to 8°C (Figure 3). Upon arrival in Beijing, the fruit were maintained at 4°C in the distributor's coldroom. While the Xinfadi markets had greater cold room capacity than the Huizhan facility, many fruit consignments were still maintained at ambient conditions (ca. 25-30°C) prior to distribution to retailers. For pallets and cartons maintained under ambient conditions, substantial damage was observed, in part due to weakened pallet strapping and shifting of cartons on pallets. Where cartons on the bottom layer of pallets had either fully or partially collapsed, severe fruit compression injury was evident. Following further handling and storage, the tracked mandarin consignment was broken down and individual cartons were transported to the COFCO laboratory in Beijing with 1.5 hours. The fruit were maintained at 21-22°C and 55-85% relative humidity for 3 days in the laboratory to simulate retail display in an air-conditioned supermarket (Figure 3).

Taken overall, our study revealed that significant breaks in the cool-chain exist during handling and distribution of Australian mandarins in China. These extended breaks in refrigerated handling were associated with substantial condensation developing on fruit and packaging and led to carton collapse. We suggest that a greater understanding of the Australia-China produce supply chain is required to identify opportunities for developing more strategic partnerships to improve efficiencies. We recommend that importers work closely with distributors to ensure refrigerated storage and transport are available for handling fruit consignments to help facilitate more orderly marketing. We also suggest this represents an ideal opportunity for the Australian industry to train and advise Chinese importers, distributors and retailers on best practices for handling mandarins.

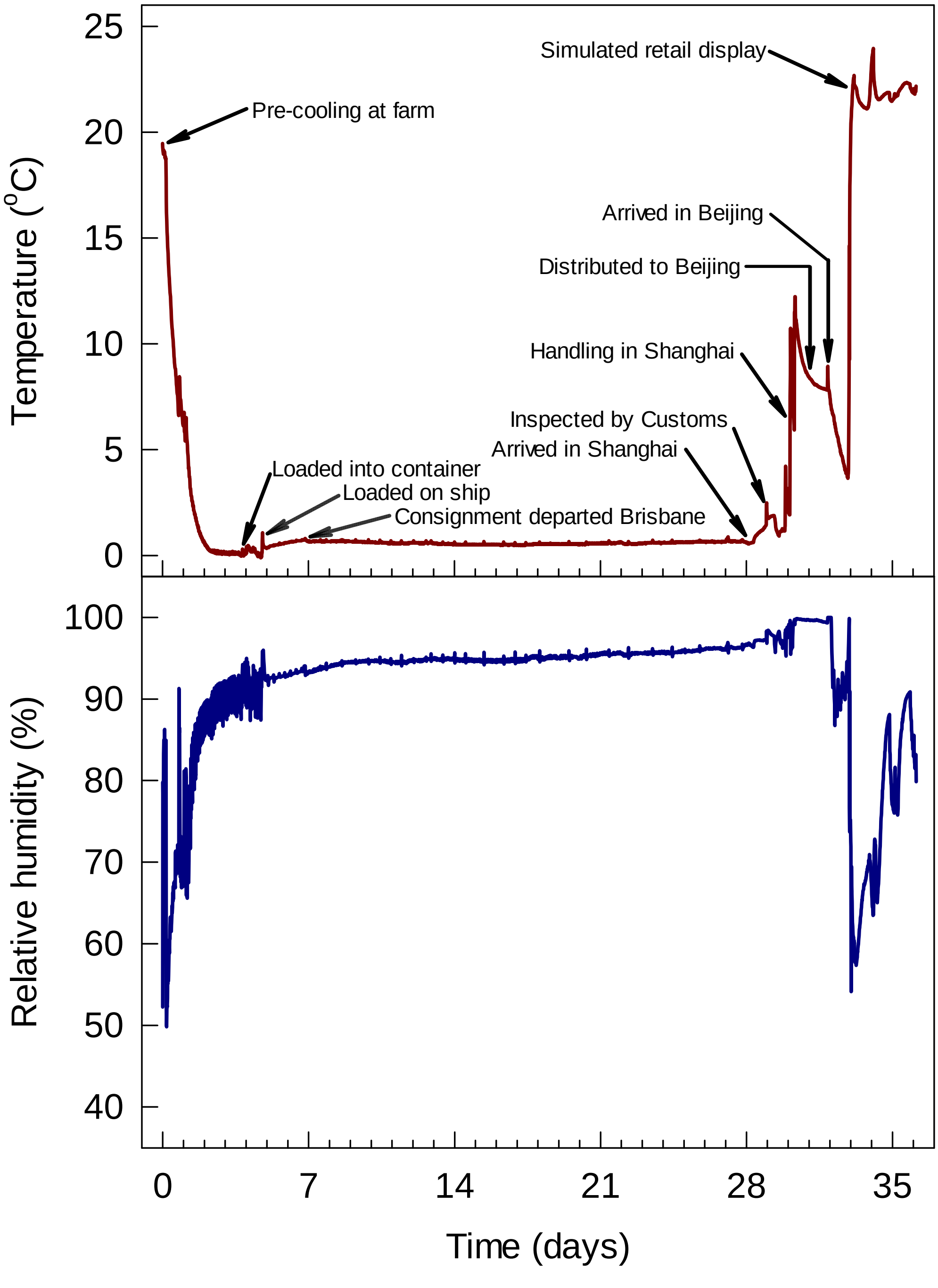

Figure 3. Average temperature and relative humidity inside palletised cartons of 'Murcott' mandarin fruit during export from Mundubbera, Australia to Beijing, China. Dataloggers that recorded temperature and relative humidity were positioned in three pallets that were randomly loaded into the marine container.

Figure 4. Photographs of the Huizhan market in Shanghai showing refrigerated containers that held palletised fruit (A), a pallet of citrus fruit being held outside at ambient conditions (B), and palletised fruit being stored inside the market at ambient conditions (C).

Figure 5. Photographs of cold-shipped mandarin (A), orange (B) and avocado (C) fruit exhibiting condensation during display at ambient at the Huizhan market, Shanghai.

Figure 6. Photographs of partially palletised Australian citrus fruit that were displayed under ambient conditions at the Huizhan market in Shanghai, China. The photographs highlight (A) carton bulging, (B) carton sagging and partial collapse, (C) absence of carton damage.

Carton atmosphere

The concentration of CO_2 that accumulated around mandarins that were shipped inside closed plastic bags at 0.6°C was 2.0 ± 0.5% upon arrival in Shanghai. The CO_2 level in bags increased to 6.5 ± 0.2% following distribution at 8-12°C to Beijing. There was no significant difference in the atmosphere that developed around wax-treated and non-wax-treated fruit. The CO_2 concentration inside cartons without plastic bags was below the limit of detection (i.e. >0.1%) in both Shanghai and Beijing. In general, mandarins can tolerate short-term exposure to 5% CO_2 without negative on fruit quality (Kader and Arpaia, 2002).

Fruit appearance quality

Both the wax-treated and non-wax-treated mandarins displayed a high level of appearance quality following marine shipment from Australia to China (Figure 7). Fruit treated with the carnauba wax coating exhibited a statistically superior peel shine at each step along the supply chain. Fruit that did not receive the wax coating still displayed a high degree of cosmetic appeal. There was no effect of shipping fruit in closed plastic bags on the appearance quality or fruit decay levels.

Fruit weight loss

Non-coated mandarins lost 1.9 ± 0.1% weight during the 21-day shipment at 0.6°C from Australia to China (Figure 8). Pre-treating fruit with a carnauba wax reduced this shipment-related weight loss by 33%. Shipping fruit inside plastic bags was more effective than waxing alone and reduced weight loss by 59-63% (Figure 8). Despite the positive effects during shipment, the wax treatment did not reduce weight loss during distribution and shelf life as compared to the non-coated fruit. However, packing and cooling fruit inside cartons lined with plastic bags may not be practical. In addition, without adequate perforation, the bags may trap condensation and lead to moisture being absorbed into cartons. There is also a risk that elevated CO_2 concentrations will accumulate inside closed bags, particularly during unrefrigerated handling, and this may lead to off-flavours developing in fruit.

Fruit total soluble solids, acids and ethanol content

The total soluble solids of both wax-treated and non-coated fruit varied from 12.8 to 13.5 °Brix immediately after shipment (Figure 9). There was no change in °Brix during subsequent distribution and shelf life. There was also no effect of the wax and packaging treatments on the total soluble solids. Fruit treated with a wax coating consistently exhibited a higher titratable acid content (Figure 9). Shipping fruit in closed plastic bags did not affect the final titratable acidity. As a consequence, the BrimA for wax-coated fruit was significantly lower than the non-coated fruit (Figure 9). Nevertheless, all fruit samples had a BrimA of >135. Pre-treating fruit with wax resulted in a higher concentration of ethanol off-flavours developing, particularly during shelf life (Figure 9). The ethanol content was highest for wax-treated fruit that were shipped in bags. Exporting non-coated fruit in bags did not elevate final ethanol levels. Overall, the level of ethanol in the fruit juice was relatively low by day 3 of shelf life.

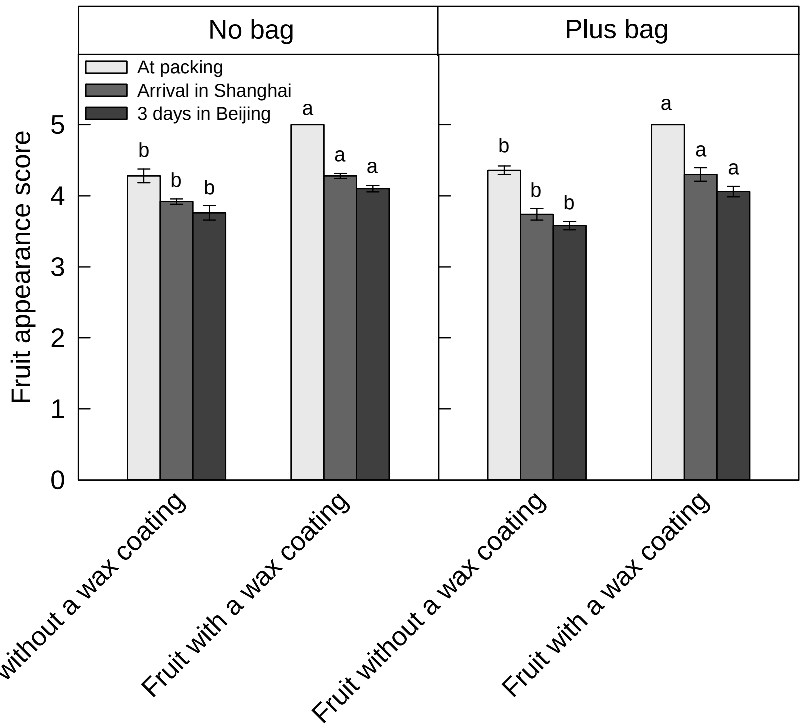

Figure 7. Changes in 'Murcott' mandarin fruit appearance during and after commercial sea-freight from Australia to China. The fruit were pre-treated with or without a carnauba wax coating. Half of the fruit were exported in cartons lined with a closed plastic bag. The remaining fruit were exported in cartons without a plastic bag lining.

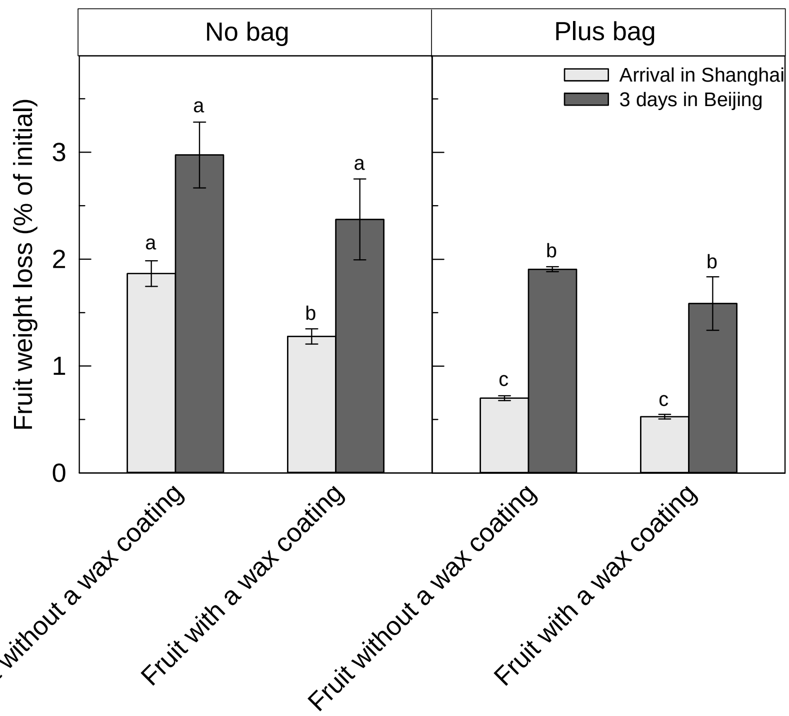

Figure 8. 'Murcott' mandarin fruit weight loss during and after commercial sea-freight from Australia to China. The fruit were pre-treated with or without a carnauba wax coating. Half of the fruit were exported in cartons lined with a closed plastic bag. The remaining fruit were exported in cartons without a plastic bag lining.

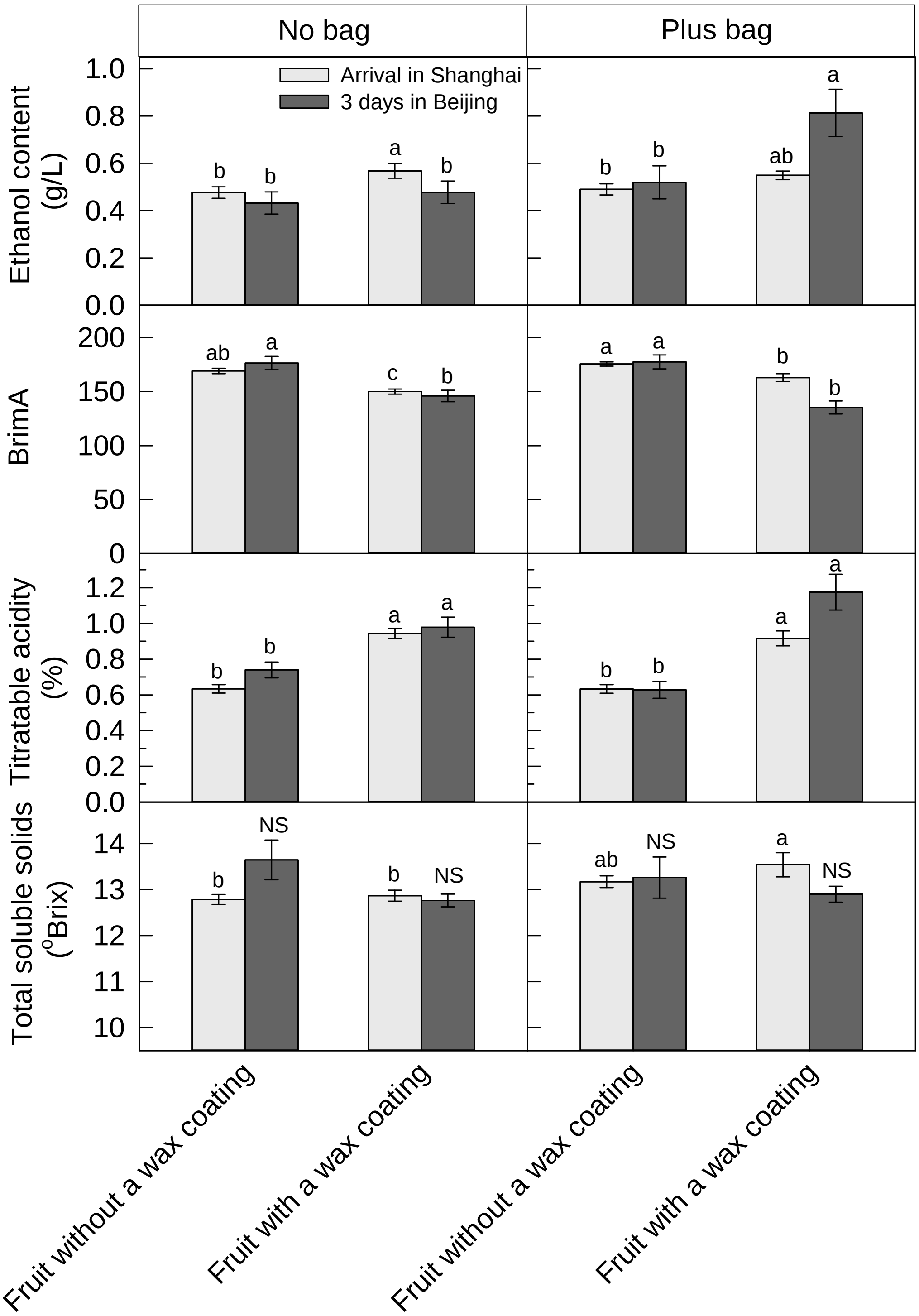

Figure 9. Changes in selected flavor properties of 'Murcott' mandarin fruit during and after commercial sea-freight from Australia to China. The fruit were pre-treated with or without a carnauba wax coating. Half of the fruit were exported in cartons lined with a closed plastic bag. The remaining fruit were exported in cartons without a plastic bag lining.

Consumer tests

Consumers in both Shanghai and Beijing rated all of the tracked mandarin fruit highly in terms of general appearance and flavour quality (Figure 10). Of interest, the consumers preferred the appearance and lower acid flavour of the non-coated fruit over the wax-treated samples. While the wax coating imparted greater peel shine than the non-coated fruit, consumers apparently could not distinguish this difference under the relatively low light intensity found in retail stores. In general, the consumers slightly downgraded the appearance of fruit that had been shipped in closed plastic bags relative to mandarins shipped in conventional vented cartons. However, they did not discern differences in the flavour of bagged and non-bagged fruit. Very few consumers commented on off-flavours in the wax-coated fruit; the ethanol content in the test fruit was relatively low (< 0.8 g/L) by the end of a 3-day shelf life and possibly below the threshold of detection (see Appendix 1).

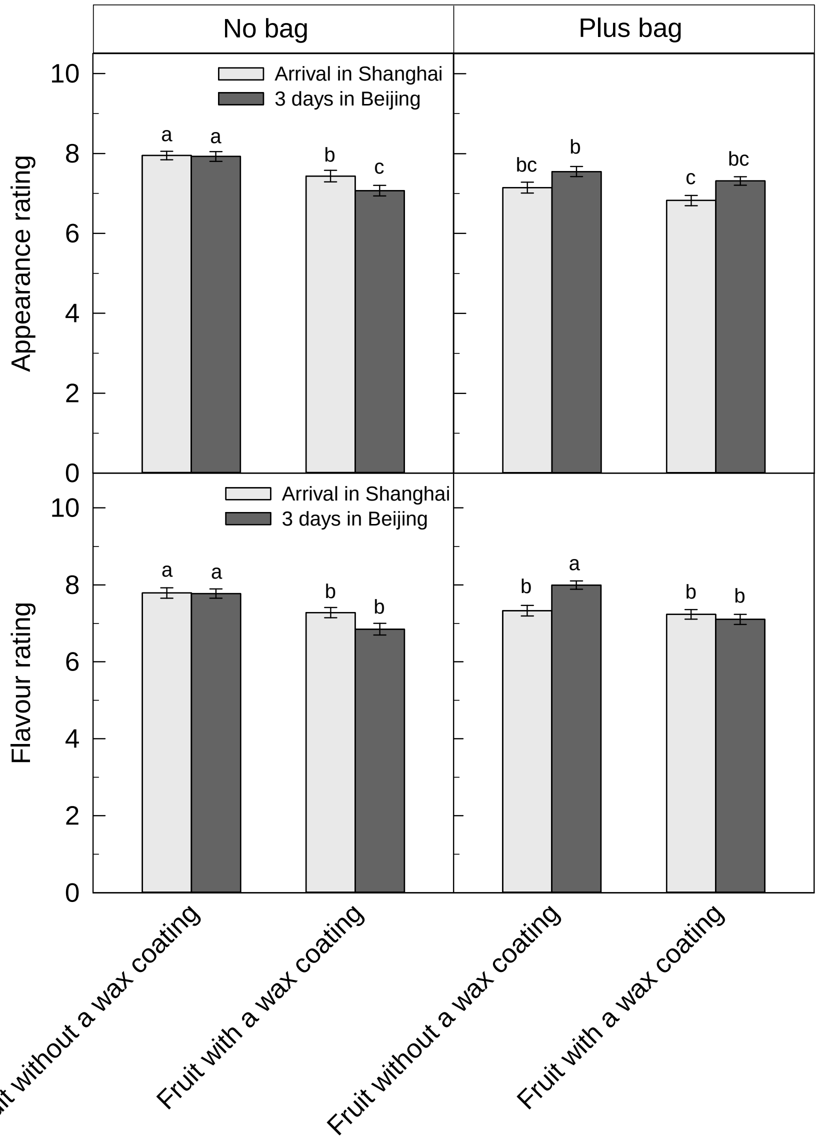

Figure 10. Consumer preference for the appearance and flavor quality of 'Murcott' mandarin fruit immediately following commercial sea-freight from Australia to China and again after a 3-day shelf life. The fruit were pre-treated with or without a carnauba wax coating. Half of the fruit were exported in cartons lined with a closed plastic bag. The remaining fruit were exported in cartons without a plastic bag lining.

Survey of Australian mandarins at retail in China

Australian 'Murcott' mandarin fruit were randomly sampled from retail outlets in Shanghai and Beijing during September, 2014. Fruit were surveyed for appearance and flavour quality. This survey coincided with the Chinese Moon Festival, a national holiday during which stores sell large quantities of imported mandarins. Fruit were sampled from five retailers; a green grocer and supermarket in Shanghai, and two supermarkets and a green grocer in Beijing. The fruit were sampled from refrigerated (10°C), air-conditioned (20-22°C) and ambient (25-30°C) display environments (Figure 11, Table 1). Produce managers indicated that imported Australian mandarin fruit were normally stored under refrigeration and then displayed on shelves for 1-3 days. The survey captured fruit from four Australian growers.

The appearance quality of all sampled fruit was of a high standard. Fruit were generally free of blemish. While most fruit were of a smooth skin, some rough skin fruit were observed. All fruit displayed considerable peel shine which suggested they were coated with wax or resin during packing in Australia. However, it was not possible to determine the type or permeability of the surface coatings.

The total soluble solids content of the sampled fruit juice was similar (i.e. 11.4-12.7° Brix) for all four brands tested (Table 1). Accordingly, each sample met the nominal harvest maturity standard of 11°Brix for mandarins. The titratable acidity of samples varied from 0.52% to 0.79% citric acid equivalents (Table 1). These slightly low acidity levels are consistent with mandarin fruit that have been shipped and/or stored for several weeks. While mandarins are recommended be harvested at 1.0% citric acid for maximum flavour, the titratable acidity typically decreases during postharvest life. The variation in °Brix and titratable acidity for each sample was also reflected in the BrimA, a relatively new measure that takes into account the balance of juice sugar and acid (Jordan et al., 2001). Citrus Australia Ltd recently established the Australian Citrus Quality Standard of a BrimA of ≥110 at harvest for mandarins (CAL, 2015). All fruit sampled at retail exceeded this minimum standard (Table 1).

There was considerable variation in the juice ethanol content among the fruit samples (Table 1). Six out of the seven fruit samples exhibited an ethanol content of > 1 g/L. While it was not possible to determine fruit history and handling, the juice ethanol concentrations were consistent with mandarins that had been shipped and/or stored at elevated temperature. Our previous research (see Appendix 1) suggests that fruit containing ≥ 0.8 g/L ethanol would be likely detected by consumers as possessing off-flavours. The ethanol content was not related to ·Brix, titratable acidity or BrimA. While display at a lower temperature would be expected to reduce the accumulation of off-flavours in fruit (Hofman et al., 2013), there was no consistent relationship between the retail display temperature and the ethanol content. This may simply reflect that other factors such as the type of surface coating and the shipment/storage time may have confounded the off-flavour response.

Previous research suggests that mandarins are best stored at 5-10°C to minimise flavour loss (Obenland et al., 2011, 2013). Thus, where refrigerated retail display is not feasible, returning unsold mandarins back into cold storage each night could extend the shelf and flavour life of fruit. Our findings point to the potential for off-flavours to develop in exported mandarins and the need for careful selection of fruit surface coatings and maintenance of the cool-chain. It also highlights the importance of developing a greater understanding of consumer attitudes towards wax-treated and non-coated mandarins. Taken overall, further research is clearly needed to optimise flavour in export mandarins and other citrus with a view towards increasing the volume and value of exports to lucrative Asian markets.

Figure 11. Photographs showing two common systems used to display mandarin fruit at retail in China. (A) A refrigerated display cabinet at a green grocer, (B) Open display in an air-conditioned supermarket.

Table 1. Selected flavour properties of four brands (A, B, C, D) of Australian 'Murcott' mandarin fruit that were randomly sampled from retail shelves in Shanghai and Beijing, China during September 2014.

Location	Display conditions	Brand	Soluble solids (°Brix)	Titratable acidity (%)	BrimA	Ethanol content (g/L)
Shanghai						
Green grocer	Refrigerated; 10°C	A	11.8 ± 0.1	0.69 ± 0.05	149 ± 3	0.72 ± 0.05
Supermarket A	Air-conditioned; 22°C	A	12.2 ± 0.4	0.61 ± 0.09	161 ± 4	1.40 ± 0.19
Supermarket A	Air-conditioned; 22°C	B	11.7 ± 0.5	0.70 ± 0.06	147 ± 10	1.11 ± 0.12
Beijing						
Supermarket A	Air-conditioned; 20°C	C	11.4 ± 0.4	0.64 ± 0.03	141 ± 6	1.72 ± 0.10
Supermarket A	Air-conditioned; 20°C	A	11.5 ± 0.4	0.52 ± 0.06	156 ± 5	1.24 ± 0.11
Supermarket B	Air-conditioned; 22°C	D	12.7 ± 0.1	0.68 ± 0.03	164 ± 2	1.56 ± 0.09
Green grocer	Ambient; 25-30°C	A	12.3 ± 0.4	0.79 ± 0.14	150 ± 11	1.11 ± 0.13

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