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Research note

Impact of postharvest ripening strategies on 'Hass' avocado fatty acid profiles

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ABSTRACT

Persea americana Mill. cv 'Hass' is a subtropical fruit highly appreciated as a rich source of fatty acids mostly of the monounsaturated type. Commonly commercially applied postharvest ripening strategies for the ready to eat market based on high temperature (15 and 20 °C) and external ethylene (0 or 100 ppm applied for 24 h) application did not have a detrimental effect on the fatty acid profile or composition and total amount of oil recovered at edible ripeness. The results of this study have important implications for the fresh fruit and avocado oil industry. The composition of the fatty acid profile in 'Hass' avocados was mostly influenced by growing and environmental conditions. Commercially applied postharvest ripening strategies based on temperature and ethylene did not affect negatively the fatty acid composition of the fruit.

1. Introduction

'Hass' avocado is enjoyed by consumers worldwide due to its rich flavour, high overall quality and health related attributes. The nutritional and dense phytochemical composition of avocado is attracting more consumers. The mesocarp tissue of the fruit is composed of 72 g/100 g water, 15.4 g/100 g total lipids, 1.96 g/100 g protein, 6.8 g/100 g fiber, total sugars 0.3 g/100 g, 8.64 g/100 g carbohydrates, 1.66 g/100 g ash, besides the presence of almost all important vitamins and minerals (Dreher and Davenport, 2013). The high lipid content of the edible portion of the fruit is composed of high amount of fatty acids (oleic, palmitic, palmitoleic, linoleic, linolenic) especially of the unsaturated type: monounsaturated (MUFA) and polyunsaturated (PUFA) fatty acids (Villa-Rodriguez et al., 2011; Dreher and Davenport, 2013). High dietary uptake of unsaturated fatty acids is associated with decreased risk of cardiovascular disease (Lopez-Ledesma et al., 1996; Mendez and Hernandez, 2007). The high content of MUFAs (~71%) together with the high dietary fiber content in avocados (~6.8%) seem to have positive effects on weight control (Bes-Rastrollo et al., 2008). In addition, avocado fruit mesocarp is an extremely rich source of bioactive phytochemicals including the C_7 sugars (e.g., mannoheptulose and perseitol) which have been mentioned to possess anticancer activity (Board et al., 1995); vitamin E, carotenoids, sterols, among others which have been shown to possess antioxidant and radical scavenging activities (Villa-Rodriguez et al., 2011).

Oil content is the best harvest maturity index for avocado (Hofman et al., 2002). Oil accumulates until harvest, thus, within a location late season fruit have much higher concentration of oil than early season fruit, ranging between 9 and 14%. Structural lipids are part of the cell membrane (phospholipids and glycolipids) and storage lipids (triglycerides) are in the idioblasts (Requejo-Tapia, 1999). The fatty acid profile is the result of the adaptation to the environment (Blakey, 2011) and even they have recently been proposed as potential biomarkers to distinguish avocado fruit growing areas (Donetti and Terry, 2014). Avocados grown in cooler climates present a higher proportion of monounsaturated to saturated fatty acids (Requejo-Tapia, 1999). Minimal change in the fatty acid composition has been reported postharvest (Luza et al., 1990). The increased oil concentration reported during storage and ripening were related to postharvest dehydration and the increased oil concentration during ripening was attributed to increased lipid recovery due to partial cell wall breakdown (Mostert et al., 2007; Meyer and Terry, 2008).

There is a huge gap in the understanding of lipid metabolism in avocados. Blakey (2011) postulated that lipids are not inert and that postharvest handling will affect the lipids present in the fruit. To our knowledge, there is no study focused on the effect of postharvest

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commercial ripening strategies on the fatty acid profiles of Hass avocados. The main objective of this work was to evaluate the impact of postharvest ripening protocols on fatty acid profiles. Secondly, we considered the effect of growing conditions (sourcing) on the fatty acid profiles. The results of this work provide facts relevant not only for the fresh fruit supply chain but also to the avocado oil industry.

2. Materials and methods

2.1. Fruit material, storage and transport conditions

'Hass' avocado middle season fruit (*Persea americana* Mill.) was sourced from 4 different growers and labelled as: A, B, C and D from Chile and transported in a refrigerated truck at 5 °C to Instituto de Investigaciones Agropecuarias (INIA – La Platina - Santiago) and further stored for 1 day at 5 °C until fruit was prepared to be sent overseas to the Netherlands in a commercial container. A total of 180 avocados of size 16 (227 – 274 g) per grower were sampled. Ten independently sampled fruits per grower were used for dry matter 'average' calculations. The fruit was sea transported to the Netherlands at 5 °C and under commercial controlled atmosphere (CA) conditions (4% O_2 and 6% CO_2) for 21 days. Temperature and CA conditions were monitored during the 21 d transport. The container with the fruit at destination was retrieved and the fruit transported to Food & Biobased Research, Wageningen University (The Netherlands) in a refrigerated truck at 5 °C and kept at that temperature for one day until the start of the postharvest ripening experiments. Environmental and growing conditions data for the 4 different growers are presented in Table 1.

2.2. Postharvest ripening conditions

Fruit were forced ripened with and without exogenous ethylene (0 or 100 ppm) applied at 18 °C for 24 h and after the fruit was ripened at two different temperatures (15 and 20 °C, respectively). Per condition, a total of 30 fruits were used and ripening through firmness assessment was followed non-destructively independently in each fruit. Vapour pressure deficit was maintained constant for all treatments. The ready to eat stage (RTE) or edible ripeness was reached at a fruit firmness of 4.4-13.3 N on each independent fruit.

2.3. Lipid extraction and fatty acid identification and quantification

Powder freeze dried avocado mesocarp tissue at edible ripeness (~1 g) was mixed with 30 mL hexane for 30 s using an Ultra-Turrax homogenizer according to Meyer and Terry (2008) with some modifications. The mixture was let to stand for 1 min at room temperature and then vacuum filtered, using a Büchner and a funnel, through a Whatman™ filter # 1 of 5.5 cm diameter. The powdered residue was recovered from the filter and re-extracted with 20 mL of hexane and let to stand for 1 min at room temperature before being vacuum filtered. Ten extra mL hexane were used to rinse the filter and funnel. The total mixture of 60 mL was vacuum concentrated in a rotary evaporator at 40 °C. The recovered oil was weighed, kept in an amber glass vial, nitrogen flushed and stored at -20 °C until fatty acid analysis.

Table 1
Environmental and growing conditions for the four growers sampled.

Variable	Grower A	Grower B	Grower C	Grower D
Annual solar radiation (W/m^2)	379	368	374	374
Minimum absolute temperature (°C)	-2.8	1.2	1.1	0.7
Maximum absolute temperature (°C)	33	34	34	37
Average Temperature (°C)	15.6	14.7	15.2	14.3
Relative humidity	72	72	69	77
Reference Evapotranspiration (mm/year)	1118	964	829	917

Fatty acid methyl esters (FAMEs) were produced using a trimethyl-sulphonium hydroxide (TMSH) solution of 0.25 M in methanol from Sigma-Aldrich. Oil samples (~0.3 g) were dissolved in 25 mL of $CHCl_3$:MeOH (2:1) and an aliquot of it (150 μL sample) was mixed with 100 μL internal standard (tetradecane), 150 μL TMSH and 200 μL chloroform. Similarly, calibration curves and response factors were calculated for the different fatty acids already reported in avocado (oleic 18:1, palmitic 16:0, palmitoleic 16:1, stearic 18:0, linoleic 18:2, linolenic 18:3). All standards were obtained from Sigma Aldrich. The fatty acid mixture identification and quantification were performed on a Thermo Focus Gas Chromatograph (GC) equipped with an automatic injection system (AS3000 auto-sampler). The following parameters/settings were used: injection volume 1 μl, split ratio 1:20, column pressure 150 kPa helium, GC column: Varian CP-FFAP (free fatty acids), 25 m x 0.32 mm x 0.25 μm, detector: FID at 280 °C, injection port temperature 250 °C. GC program: hold 1 min at 50 °C, ramp 7 °C/min to 150 °C, then ramp 4 °C/min to 250 °C and hold at 250 °C for 20 min. Identification and quantification was carried out by comparing peak areas of fatty acid standards and injection inaccuracies were corrected by the use of an internal standard (tetradecane) in each sample injected.

2.4. Experimental design and Statistical analysis

All statistical analysis were carried out in IBM SPSS Statistics 19 (New York, USA). A non-parametric test, Kruskal Wallis, was used to assess for significant differences ($p < 0.05$) among treatments.

3. Results and discussion

3.1. Effect of postharvest ripening strategies on fatty acid profiles

Previous studies have reported fatty acid composition in relation to changes in dry matter content and maturity stage (Ozdemir and Topuz, 2004; Villa-Rodriguez et al., 2011), growing conditions (Landahl et al., 2009; Donetti and Terry, 2014) and at different points during postharvest ripening (Ozdemir and Topuz, 2004). To our knowledge, the effect of different postharvest ripening strategies (temperature and ethylene application) on the fatty acid profile obtained at edible ripeness at an individual fruit level has not been reported before. The effect of the different ripening temperatures (15 and 20 °C) and ethylene (0 or 100 ppm) on the final fatty acid profiles of avocadoes was assessed only with the samples with the lowest average dry matter content (26.4 ± 2.1) from grower D (Table 2) and later confirmed with the other growers (A, B and C) at temperatures of 15 and 20 °C with and without ethylene application (Table 3).

Non-significant differences in fatty composition (Table 2) and total amounts of FA (data not shown) were found for the different ripening strategies. Oleic acid (C18:1) was the most abundant fatty acid found representing ~ 68% of the total fatty acid amount, followed by palmitic acid (C16:0) ~ 13.4%, linoleic (C18:2) ~ 14%, palmitoleic (C16:1) ~ 3.4%, linolenic (C18:3) ~ 0.97% and stearic (C18:0) ~ 0.2%. The proportion of monounsaturated fatty acids (MUFAs), polyunsaturated fatty acids (PUFAs) and saturated fatty acids (SFAs) accounted for 71.3%, 15.0% and 13.3%, respectively. Values of 71% MUFAs, 13% PUFAs and 16% SFAs have been reported before for 'Hass' avocado (Dreher and Davenport, 2013), however, the ripeness stage has not been indicated. An increase in PUFAs has been reported during postharvest ripening by (Ozdemir and Topuz, 2004) and this might explain the difference between our results at edible ripeness and those reported by (Dreher and Davenport, 2013). However, at a ripening temperature of 15 °C, Villa-Rodriguez et al. (2011) reported a decrease on PUFAs during postharvest ripening at ready to eat stage with relative values of 73.7%, 14.9% and 11.4% for MUFA, PUFAs and SFAs respectively similar to our results. Fatty acids are precursors of aroma volatiles and temperature has been previously mentioned (Salas et al., 2000) to influence the