Food Chemistry 169 (2015) 5–12

Contents lists available at ScienceDirect

Food Chemistry

journal homepage: www.elsevier.com/locate/foodchem

The impact of cold storage and ethylene on volatile ester production and aroma perception in 'Hort16A' kiwifruit

Catrin S. Günther [a,*], Ken B. Marsh [a], Robert A. Winz [a], Roger F. Harker [a], Mark W. Wohlers [a], Anne White [a], Matthew R. Goddard [b]

[a] The New Zealand Institute for Plant & Food Research Ltd, Private Bag 92169, Auckland 1142, New Zealand
[b] School of Biological Sciences, University of Auckland, Private Bag 92019, Auckland Mail Centre, Auckland 1142, New Zealand

ARTICLE INFO

Article history:
Received 23 April 2014
Received in revised form 10 July 2014
Accepted 12 July 2014
Available online 1 August 2014

Chemical compounds studied in this article:
Ethyl acetate (PubChem CID: 8857)
Ethyl propanoate (PubChem CID: 7749)
Ethyl 3-(methylsulfanyl)propanoate (PubChem CID: 61592)
Ethyl butanoate (PubChem CID: 7762)
Ethyl 3-hydroxybutanoate (PubChem CID: 62572)
3-Methylbutyl butanoate (PubChem CID: 7795)
Ethyl 3-hexenoate (PubChem CID: 5362622)
Ethyl octanoate (PubChem CID: 7799)
Ethyl benzoate (PubChem CID: 7165)
Ethyl furoate (PubChem CID: 11980)

Keywords:
Kiwifruit
Ethylene
Cold storage
Fruit flavour
Volatile esters
GC–MS
Postharvest
Sensory

ABSTRACT

Fruit esters are regarded as key volatiles for fruit aroma. In this study, the effects of cold storage on volatile ester levels of 'Hort16A' *(Actinidia chinensis* Planch. var *chinensis*) kiwifruit were examined and the changes in aroma perception investigated. Cold storage (1.5 °C) for two or four months of fruit matched for firmness and soluble solids concentration resulted in a significant reduction in aroma-related esters such as methyl/ethyl propanoate, methyl/ethyl butanoate and methyl/ethyl hexanoate. Levels of these esters, however, were restored by ethylene treatment (100 ppm, 24 h) before ripening. A sensory panel found that "tropical" and "fruit candy" aroma was stronger and "green" odour notes less intensively perceived in kiwifruit which were ethylene-treated after cold storage compared to untreated fruit.

The key findings presented in this study may lead to further work on the ethylene pathway, and innovative storage and marketing solutions for current and novel fruit cultivars.

1. Introduction

During ripening remarkable changes in fruit biochemistry and gene expression lead to major changes in skin colour, flesh colour, texture and flavour, and thus indicate a fruit's edibility (Alexander & Grierson, 2002). In comparison to fruit maturation this window of edibility is relatively short and timely presentation to consumers is crucial (Campbell, Smith, Jaeger, & Harker, 2009) as it can also mark the end of useful storage. In climacteric fruit, such as kiwifruit, these ripening-related changes are orchestrated by the ripening hormone ethylene in an autocatalytic manner. In response to exogenous ethylene, endogenous ethylene production is triggered and fruit respiration increases rapidly (Prasanna, Prabha, & Tharanathan, 2007). In kiwifruit, fruit softening processes are initiated by the perception of exogenous ethylene, whereas aroma production appears to be correlated to endogenous fruit ethylene biosynthesis (Atkinson et al., 2011).

* Corresponding author. Permanent address: School of Biological Sciences, University of Auckland, Private Bag 92019, Auckland Mail Centre, Auckland 1142, New Zealand. Tel.: +64 9 9232777; fax: +64 9 3737416.
E-mail address: c.guenther@auckland.ac.nz (C.S. Günther).

http://dx.doi.org/10.1016/j.foodchem.2014.07.070
0308-8146/

Postharvest treatments are applied to delay fruit softening in order to increase storage time and shelf life, thus allowing extended supply of seasonal fruits to a global market. Commonly cold storage, controlled atmosphere storage and/or ethylene-inhibiting postharvest techniques such as 1-methylcyclopropene (1-MCP) treatment are deployed (Watkins, 2006). However, these practises can have detrimental effects on aroma production in various climacteric fruits (Watkins, 2006) in particular volatile ester synthesis was shown to be affected. Fruit ester biosynthesis is catalysed by enzymes of the alcohol acyltransferase (AAT) family (D'Auria, 2006) and gene expression of key members is ethylene-dependent in climacteric fruits such as apple (Schaffer et al., 2007), melon (El-Sharkawy et al., 2005) and banana (Jayanty, Song, Rubinstein, Chong, & Beaudry, 2002). Obviously, some ethylene-regulated ripening events are desired (aroma-related volatile biosynthesis and changes in skin colour), but others (fruit softening and browning) are not, and would preferably be delayed. In addition, cultivar specific ethylene-dependent and independent ripening processes can coexist as described for cantaloupe melons (Pech, Bouzayen, & Latche, 2008). Therefore, in-depth studies of individual fruit crops and even cultivars are needed to discern physiological responses to ethylene in order to control its action and improve fruit quality standards.

The kiwifruit industry is an integral part of the New Zealand economy. Fruit is exported to distant markets such as Asia, USA and Europe, making low temperatures a common feature of transport and storage (Schotsmans, Mackay, & Mawson, 2008). The yellow-fleshed *Actinidia chinensis* 'Hort16A' kiwifruit (Patterson, Burdon, & Lallu, 2003) has proven popular as an addition to the more extensively grown green *Actinidia deliciosa* 'Hayward' kiwifruit. Tropical fruit flavour is a key attribute of 'Hort16A' which is characterised as being reminiscent of melon, mango and banana (Jaeger & Harker, 2005). The discovery that AAT gene expression and subsequent biosynthesis of methylsulfanyl-esters in 'Hort16A' kiwifruit was ethylene-dependent and modulated by cold temperatures (Günther, Matich, Marsh, & Nicolau, 2010; Günther, Heinemann, Laing, Nicolau, & Marsh, 2011) raises the question of whether the production of volatile esters is influenced by cold storage and ethylene treatment. We hypothesised that significant changes in the aroma-related fruit ester profile likely to be reflected in a consumer's perception of kiwifruit flavour. Therefore, we investigated odour and basic taste characteristics of cold-stored 'Hort16A' kiwifruit in response to ethylene. Conclusions of this study may provide further clues for innovations in postharvest handling of climacteric fruit such as kiwifruit.

2. Materials and methods

2.1. Plant material and postharvest handling

A total of 1200 *A. chinensis* 'Hort16A' fruit (40 trays) sourced from three different commercial growers (Satara Kiwifruit Supply Ltd., Bay of Plenty, New Zealand) the following harvest parameters: 18.1–19.1% average dry matter; 55–65N firmness; colour protocol B. A number was assigned to each individual fruit to aid standardisation. 180 kiwifruit (6 trays) were immediately ripened (20 °C) to the desired firmness (14 days), and remaining fruit were cold stored at 1.5 °C directly after harvest. After two months, 450 kiwifruit were removed from storage and 270 of these fruit were ethylene-treated with 100 ppm for 24 h before ripening (20 °C, six days). The remaining 180 kiwifruit were ripened without ethylene treatment (seven days). The same procedure was followed after four months of cold storage with the remaining 570 kiwifruit, half of which were ethylene-treated. A ripening (20 °C) period of six days was appropriate to yield a sufficient number of kiwifruit that met the parameters set for standardisation as outlined in 2.2.

2.2. Measurement of fruit quality parameters

Kiwifruit firmness was monitored non-destructively during ripening using an acoustic firmness sensor (AWETA® AFS; microphone gain: 80, tick power: 16). For each sample, fruit were selected to have firmness from 5.5 to 7 $\times$ 10^6 Hz^2 $g^{2/3}$. In addition, an intrusive fruit texture analyser (Güss, model GS14, South Africa; fitted with a 7.9 mm Effegi (Alfonsine, Italy) penetrometer probe, speed setting 20 mm sec) was used to match firmness (0.65–0.8 kgF) of ethylene-treated and untreated kiwifruit. Soluble solid concentrations (SSC) based on the juice sampled from both ends were measured with a digital refractometer (ATAGO^{TM}PAL-1) and firmness-matched fruit of 15–17% SSC selected. 18 kiwifruit (six per grower), standardised to these parameters per time point and treatment were then prepared for analysis by cutting each fruit into 12 equally-sized pieces. A random sample of six kiwifruit pieces per grower ($n = 3$) was then presented to panellists and a separate sample ($n = 3$) processed for GC–MS analysis.

Endogenous fruit ethylene production from 30 random, firmness-matched fruit (FI = 6 ± 0.7 10^6 Hz^2 $g^{2/3}$) per time point and treatment was assessed on the sampling day. For this purpose, individual kiwifruit were placed in a sealed 529-cm^3 plastic container for one hour, and the ethylene concentration of a 1 cm^3 headspace sample was measured by flame ionisation chromatography (PU 4500 Chromatograph; Phillips, UK).

2.3. Volatile ester analysis

2.3.1. Chemicals

All chemicals, including the following authentic reference compounds were purchased from Sigma–Aldrich New Zealand Ltd.: butyl formate, ethyl acetate, hexyl acetate, methyl propanoate, ethyl propanoate, butyl propanoate, methyl butanoate, ethyl butanoate, propyl butanoate, pentyl butanoate, hexyl butanoate, ethyl 2-methylbutanoate, 2-methylpropyl butanoate, ethyl E-2-butenoate, 3-methylbutyl butanoate, ethyl 3-hydroxy butanoate, methyl pentanoate, ethyl pentanoate, methyl hexanoate, ethyl hexanoate, propyl hexanoate, ethyl E-3-hexenoate, ethyl heptanoate, methyl octanoate, methyl 2-(methylsulfanyl)acetate, ethyl 2-(methylsulfanyl)acetate, methyl 3-(methylsulfanyl)propanoate, ethyl 3-(methylsulfanyl)propanoate, methyl benzoate, ethyl benzoate, methyl furoate, ethyl furoate, diethylcarbonate.

Propyl and butyl 2-(methylsulfanyl)acetate and propyl and butyl 3-(methylsulfanyl)propanoate were synthesised as described (Günther et al., 2010).

Deuterated aliphatic esters (ethyl [D11]-hexanoate, ethyl [D15]-octanoate, [D8] methyl benzoate) were used as internal standards and purchased from C/D/N Isotopes Inc. [D5]-ethyl 2-(methylsulfanyl)acetate, ethyl 3-([D3]-methylsulfanyl)propanoate, and [D5]-ethyl butanoate were synthesised as described (Günther et al., 2010, Günther, Heinemann, et al., 2011).

2.3.2. Dynamic headspace sampling of volatile esters

Each kiwifruit sample was snap-frozen with liquid nitrogen, pulverised using a stone crusher (Rocklabs, New Zealand) and stored at −80 °C. Ten grams of pulverised kiwifruit tissue was then mixed with 8 g $(NH_4)_2SO_4$ and 3 g NaCl in a 250 mL Erlenmeyer flask. Deuterated internal standards ([D5]-ethyl butanoate, 1 mg kg^{-1}; [D8] methyl benzoate, 50 μg kg^{-1}; ethyl [D11]-hexanoate, 0.2 mg kg^{-1}; ethyl [D15]-octanoate, 50 μg kg^{-1}; [D5]-ethyl 2-(methylsulfanyl)acetate, 20 μg kg^{-1}; ethyl 3-([D3]-methylsulfanyl)propanoate, 20 μg kg^{-1}) were added to the sample. Purified air (BOC) was purged (30 mL min^{-1}) for 2 h through the airtight