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Full Length Research Paper

Characterization of aromatic volatile constituents in 11 Asian pear cultivars belonging to different species

Guopeng Li, Huijuan Jia, Ruiyuan Wu, Sayed Hussain and Yuanwen Teng*

Department of Horticulture, State Agricultural Ministry Key Laboratory of Horticultural Plant Growth, Development and Quality Improvement, Zhejiang University, Hangzhou 310058, Zhejiang Province, People's Republic of China.

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Aroma is an important fruit sensory attributes that is influenced by the volatile compounds present in the fruit, and it determines whether a fruit is acceptable to consumers. The volatile compounds in 11 pear cultivars from different species were investigated by headspace solid phase microextraction and gas chromatography-mass spectrometry. In total, 70 volatile compounds, including three alcohols, six aldehydes, fifty-two esters, three acids, three terpenes and three ketones were identified and quantified. Ester was the dominant chemical class of which a total of 52 compounds were detected. Hexanal, hexyl acetate, ethyl hexanoate, ethyl 2-methylbutanoate and ethyl butanoate were the major volatile compounds. *Pyrus ussuriensis* Maxim showed a higher concentration of volatiles, and higher ester content than in the other Asian pear species studied. Cultivars were clearly grouped into four clusters based on the concentration and number of different volatile compounds present in each cultivar. The clusters of different cultivars could be discriminated from each others using the first two principal components. The volatile profiles of the pears were qualitatively and quantitatively influenced by the cultivar.

Key words: *Pyrus*, Asian pear, volatile compound, gas chromatography mass spectrometry (GC-MS), solid phase microextraction (SPME), multivariate analysis.

INTRODUCTION

The pear (*Pyrus* L.) is an important fruit crop that is cultivated worldwide. There are two kinds of cultivated pears: Asian pears and European pears (*Pyrus communis* L.). European pears are mainly cultivated in Europe and America, and require a period of chilling and/or ethylene exposure for ripening. During ripening, *P. communis* fruit has a strong aroma. Most cultivated Asian pears belong to one of four groups of commercial pear cultivars, namely *Pyrus pyrifolia* Nakai White Pear Group (formerly referred to as *Pyrus bretschneideri* Rehd.), *Pyrus ussuriensis* Maxim, *Pyrus sinkangensis* Yu and *P. pyrifolia* Nakai. *P. ussuriensis* is more similar to *P. communis* than the other Oriental pears, in that the fruit has a strong aroma and requires ethylene exposure during ripening (Teng and Tanabe, 2004). However, the fruits of other Oriental species do not need a postharvest ripening stage and are popular for their sweetness, crispness and subtle aroma.

Fruit aroma is an important sensory attributes that it is particularly sensitive to changes in the chemical composition. It is a complex quality because of the diversity of biosynthetic routes. To date, more than 300 compounds, including alcohols, aldehydes, esters, acids, ketones and hydrocarbons, have been identified in different pear cultivars, mainly from *P. communis* (Rapparini and Predieris, 2003). The composition and quantity of aroma volatiles vary considerably depending on the cultivar, maturity, and storage conditions (Chervin et al., 2000; Gorny et al., 2000; Argenta et al., 2003; Kondo et al., 2006; Braniste and Heroiu, 2007). Esters, especially hexanoate esters, are the largest contributors to pear aroma, and decanoate esters are important in determining the overall aroma of *P. communis.*

Although there are many published studies of *Pyrus* species, to the best of our knowledge, there have been few studies on the aroma components of pear cultivars

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*Corresponding author. E-mail: ywteng@zju.edu.cn. Tel: +86 571 8898 2803.

Table 1. Pear cultivars used in this study.

Cultivar	Species	Source	Harvest time
Hongxiangsu (HXS)	*P. sinkiangensis* × *P. pyrifolia* White Pear Group	Zhengzhou, Henan Province	2010-8-24
Zaobaimi (ZBM)	*P. pyrifolia*	Zhengzhou, Henan Province	2010-8-24
Kuerle Xiangli (KEL)	*P. sinkiangensis*	Kuerle, Xinjiang Province	2010-10-4
Dongguoli (DGL)	*P. pyrifolia* White Pear Group	Lanzhou, Gansu Province	2010-10-1
Pingxiangli (PXL)	*P. ussuriensis*	Gongzhuling, Jilin Province	2010-9-19
Daxiangshu i(DXS)	*P. ussuriensis*	Gongzhuling, Jilin Province	2010-9-19
Ruanerli(REL)	*P. ussuriensis*	Lanzhou, Gansu Province	2010-10-1
Youhongli (YHL)	*P. ussuriensis*	Gongzhuling, Jilin Province	2010-9-19
Xiaoxiangshui (XXS)	*P. ussuriensis*	Gongzhuling, Jilin Province	2010-9-19
Nanguoli (NGL)	*P. ussuriensis*	Baicheng, Liaoning Province	2010-10-4
Bartlett (BAL)	*P. communis*	Zhengzhou, Henan Province	2010-8-24

native to Asia. Takeoka et al. (1992) identified 72 components in the headspace of intact Asian pears (Seuri cultivar), and esters were the dominant constituents (Takeoka et al., 1992). Hexanal, ethyl hexanoate, ethyl butanoate, ethyl acetate, hexyl acetate, and α-farnesene have been identified as dominant components in 'Kuerle Xiangli' (*P. sinkiangensis* Yu) and 'Yali' (*P. pyrifolia* White Pear Group) (Chen et al., 2006a, b). The lack of information about the aroma components of pear cultivars in China limits their use. The diversity of aromatic volatiles among different species needs to be investigated. The aim of our research was to compare the volatile compounds in 11 pear cultivars from different species.

MATERIALS AND METHODS

Eleven pear cultivars were obtained from different fruit institutes in China from August to October, 2010. The cultivar names and abbreviations are detailed in Table 1. The *P. ussuriensis* and *P. communis* ('Bartlett') were incubated at room temperature (about 20°C) for ripening. Conventional indices such as skin color, fruit firmness, and ethylene production were used to determine the ripeness of the fruit. Flesh cut from the ripe fruits was immediately frozen in liquid nitrogen and stored at -80°C for further use.

Extraction of volatile compounds

10 g of frozen flesh was ground to fine powder in liquid nitrogen and then transferred to a 20 mL vial. To avoid browning, 4 mL of saturated sodium chloride solution was added to each vial. 2-Octanol (5 µL) was added as an internal standard. A stirring bar was used to maximize production of volatiles and then the vial was sealed with a Teflon septum and aluminum cap. The mixture was homogenized using a vortex oscillator.

Headspace solid-phase microextraction manual sampling equipment and fibers used in the analysis were purchased from Supelco (Sigma-Aldrich, St. Louis, MO). The fibers coated with 65 µm of polydimethylsiloxane/divinylbenzene were used in our study as described previously (Li et al., 2012). The fibers were conditioned before use according to the manufacture's direction at 250°C for 30 min to prevent contamination. Each fiber was exposed to the headspace of the vial for equilibration for 45 min.

Gas chromatography/mass spectrometry conditions

Gas chromatography/mass spectrometry was conducted with a Hewlett-Packard 6890 (Palo Alto, CA) equipped with a flame ionization detector. The injector and detector temperatures were maintained at 250 and 270°C, respectively. An Innowax capillary column (30 m × 0.32 mm I.D., 0.5 µm film thickness, Agilent Technologies, Santa Clara, CA) was used for all analyses. The column temperature was held at 40°C for 2 min, increased to 220°C at 4°C/min and 250°C at 15°C/min and then held at this temperature for 2 min. The sample was injected in splitless mode. Helium was used as the carrier gas at a flow rate of 1 mL/min. Mass spectra were obtained using a HP-5973 mass selective detector (Hewlett-Packard) at 70 eV in scan mode. Mass spectra were scanned from *m/z* 30 to 500. The temperature of the ion source and connecting parts were set at 230 and 280°C, respectively. Compounds were identified using a NIST/WILEY mass spectra library search. The identities of most of the compounds were then confirmed by comparison of their linear retention indices and electron ionization mass spectra with those of reference compounds. Semi-quantification was carried out by the internal standard method, where the concentrations of different volatile compounds were normalized with that of 2-octanol.

Statistical analysis

Compounds that were not detected were given a value of zero for the statistical analysis. Principal component analysis (PCA) (AlphaSoft, Version 11.0, Alpha MOS, Toulouse, France) was used to cluster the cultivars in groups according to their volatile composition.

RESULTS AND DISCUSSION

Gross quantitative variations

In the 11 cultivars, 70 volatile compounds were identified and quantified, including three alcohols, 6 aldehydes, 52 esters, three acids, three terpenes and three ketones (Table 2). The aromatic profiles and volatile concentrations varied largely among the cultivars. The 'Bartlett' pears (*P. communis*) had the highest total volatile concentration (6085.0 µg/kg) among the 11 cultivars

Table 2. Volatile compounds and their concentrations (μg/kg) in the ripe fruit from 11 different pear cultivars.

Volatile compound	Different species and cultivars										
	P. pyrifolia	*P. sinkiangensis*	*P. pyrifolia* White Pear Group	*P. ussuriensis*						*P. sinkiangensis* × *P. pyrifolia* White Pear Group	*P. communis*
	ZBM	**KEL**	**DGL**	**PXL**	**DXS**	**REL**	**NGL**	**YHL**	**XXS**	**HXS**	**BAL**
Alcohol											
1-Hexanol	1.5[a]	3.1	ND[b]	ND	2.8	ND	ND	3.9	ND	3.6	13.0
2-Ethyl-1-hexanol	ND	ND	ND	ND	ND	2.7	2.7	ND	ND	ND	ND
Ethanol	ND	ND	ND	37.2	ND	ND	ND	16.2	24.3	ND	67.7
Aldehyde											
Hexanal	35.8	22.6	48.8	307.7	140.3	124.0	80.4	211.2	223.1	44.0	ND
4-Methyl-3-pentenal	ND	ND	0.7	ND	ND	ND	ND	ND	ND	ND	ND
E-2-hexenal	2.3	2.8	9.4	39.3	16.0	15.9	11.1	25.8	29.9	24.6	ND
Nonanal	0.3	0.2	1.8	ND	ND	ND	ND	ND	ND	ND	ND
E-2-Nonenal	ND	ND	ND	ND	ND	ND	ND	0.5	ND	ND	0.9
Z-3-Hexenal	ND	ND	1.6	ND	ND	ND	ND	ND	ND	ND	ND
Ester											
Methyl butanoate	ND	ND	ND	6.4	4.6	25.8	13.0	3.8	15.7	ND	ND
Ethyl butanoate	ND	ND	ND	76.4	57.4	19.7	125.2	73.1	190.6	ND	ND
Methyl 2-methylbutanoate	ND	ND	ND	ND	ND	ND	ND	ND	6.3	ND	ND
Ethyl 2-methylbutanoate	ND	ND	ND	53.6	16.1	1.7	33.3	46.3	66.8	ND	ND
Methyl 2-methylhexanoate	ND	ND	ND	3.5	ND	ND	4.8	1.5	ND	ND	ND
Ethyl hexanoate	0.6	0.3	1.2	126.9	79.7	153.6	951.8	498.9	279.6	ND	ND
Pentyl acetate	ND	ND	ND	ND	2.0	ND	4.5	ND	ND	ND	30.7
Butyl acetate	0.3	0.8	1.1	ND	3.8	2.8	24.0	4.3	6.2	ND	326.9
3-Methylbutyl acetate	ND	ND	ND	ND	ND	ND	3.6	1.5	3.7	ND	3.3
Butyl 2-methylbutanoate	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	4.4
Ethyl pentanoate	ND	ND	ND	2.3	1.2	ND	6.7	4.5	2.9	ND	ND
Ethyl 2-butenoate	ND	ND	ND	1.2	3.4	ND	ND	1.1	7.9	ND	ND
methyl hexanoate	ND	ND	ND	9.7	5.8	288.1	66.5	13.8	41.0	ND	8.5
Ethyl tiglate	ND	ND	ND	1.4	2.3	ND	ND	ND	10.2	ND	ND
Hexyl acetate	ND	1.6	9.8	9.8	48.9	36.5	457.7	147.3	55.6	0.3	2755.6
Ethyl 3-hexenoate	ND	ND	ND	ND	0.7	ND	4.3	2.0	3.9	ND	ND
Ethyl 2-hexenoate	ND	ND	ND	1.5	4.6	ND	1.3	6.8	15.6	ND	ND
Ethyl heptanoate	ND	ND	ND	ND	1.2	2.4	3.9	2.4	5.2	ND	ND
Heptyl acetate	ND	ND	ND	ND	5.7	5.5	ND	6.1	5.3	ND	17.2
Methyl 4-methylhexanoate	ND	ND	ND	ND	0.3	10.4	ND	ND	ND	ND	ND
Methyl heptanoate	ND	ND	ND	ND	ND	9.4	22.7	ND	ND	ND	ND
2-Hexen-1-ol acetate	ND	ND	2.5	ND	ND	1.7	5.5	ND	ND	ND	ND
2-Ethylhexyl acetate	ND	ND	1.1	ND	ND	ND	0.7	ND	ND	ND	1.9
Hexyl 2-methyl-propanoate	ND	ND	0.4	ND	ND	4.3	5.1	1.1	ND	ND	2.7
Methyl acetate	ND	0.3	ND	ND	ND	21.8	ND	ND	ND	ND	ND
Methyl octanoate	ND	ND	ND	ND	ND	ND	ND	ND	3.1	ND	ND
Octyl formate	ND	0.2	0.6	ND	10.4	ND	5.5	1.7	5.0	ND	3.1
Ethyl octanoate	ND	ND	ND	0.5	2.1	ND	ND	2.0	20.6	ND	7.8
Ethyl 4-octenoate	ND	ND	ND	ND	ND	3.4	10.1	ND	ND	ND	ND
Methyl 2-octenoate	ND	ND	ND	ND	ND	0.6	1.7	0.5	1.5	ND	ND

Table 2. Contd.

Volatile compound	Different species and cultivars										
	P. pyrifolia	*P. sinkiangensis*	*P. pyrifolia* White Pear Group	*P. ussuriensis*						*P. sinkiangensis* × *P. pyrifolia* White Pear Group	*P. communis*
	ZBM	**KEL**	**DGL**	**PXL**	**DXS**	**REL**	**NGL**	**YHL**	**XXS**	**HXS**	**BAL**
Octyl acetate	ND	ND	ND	ND	5.0	ND	54.0	7.1	13.0	ND	47.1
Ethyl 2-octenoate	ND	ND	ND	2.7	2.2	ND	15.2	16.1	6.6	ND	8.6
Ethyl 3-(methylthio)propanoate	ND	ND	ND	ND	ND	ND	2.6	2.1	ND	ND	ND
Hexyl hexanoate	ND	ND	ND	ND	ND	ND	7.9	1.0	ND	ND	ND
Ethyl benzeneacetate	ND	ND	ND	ND	0.5	ND	6.2	1.6	ND	ND	ND
Ethyl 3-hydroxyhexanoate	ND	ND	ND	ND	1.2	ND	ND	1.7	6.0	ND	ND
Ethyl benzonoate	ND	ND	ND	ND	2.4	ND	ND	ND	ND	ND	ND
Methyl 8-methyldecanoate	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	1.0
Methyl 4-decenoate	ND	ND	ND	ND	ND	6.7	1.4	ND	ND	ND	6.9
Ethyl decanoate	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	12.6
Ethyl 4-decenoate	ND	ND	ND	ND	ND	ND	12.2	ND	ND	ND	41.7
Methyl 2-decenoate	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	1.4
Decyl acetate	ND	ND	ND	ND	ND	ND	6.4	ND	ND	ND	ND
Ethyl trans-2-decenoate	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	12.2
Methyl 2,4-decadienoate(Z,E)	ND	ND	ND	ND	1.1	10.5	ND	ND	2.3	ND	8.4
Methyl 2,4-decadienoate(E,Z)	ND	ND	ND	ND	ND	ND	12.4	8.4	ND	ND	442.3
2-Phenylethyl acetate	ND	ND	ND	ND	2.0	6.8	7.1	1.8	ND	ND	ND
Ethyl 2,4-decadienoate	ND	ND	ND	5.2	14.5	3.3	64.0	100.9	6.3	ND	2079.6
Methyl 3-hydroxyoctanoate	ND	ND	ND	ND	ND	17.0	ND	ND	ND	ND	ND
Isopentyl hexanoate	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	13.3
Ethyl 2,4-trans,cis-decadienoate	ND	ND	ND	ND	ND	ND	1.9	1.8	ND	ND	4.9
Acid											
Acetic acid	2.0	1.1	2.1	16.2	5.5	27.7	8.2	8.8	11.6	2.6	11.5
Hexanoic acid, 2-methyl-	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	1.7
Hexanoic acid	ND	ND	ND	ND	ND	ND	ND	12.6	ND	ND	ND
Terpene											
α-Farnesene	ND	0.1	0.3	50.1	22.8	26.8	822.9	317.2	1.7	ND	126.2
(Z,E)-α-Farnesene	ND	ND	ND	2.1	1.6	3.1	34.9	12.7	ND	ND	22.0
Limonene	ND	ND	ND	ND	ND	ND	ND	ND	1.7	0.5	ND
Ketone											
1-Hydroxy-2-propanone	ND	ND	ND	ND	ND	ND	ND	ND	ND	0.2	ND
2-Octanone	0.3	0.4	ND	ND	ND	ND	ND	ND	ND	ND	ND
6-Methyl 5-hepten-2-one	ND	ND	ND	1.2	0.6	ND	2.5	4.5	ND	ND	ND

[a] Values are the means of three replicates, and data are presented as 2-octanol equivalents;[b] ND, Not detected.

(Table 3). Among the Asian pears, 'Nanguoli' pears had the highest volatile concentration (2901.9 μg/kg), and 'Kuerle Xiangli' pears had the lowest volatile content (33.4 μg/kg). Twelve volatile compounds were identified in the 'Kuerle Xiangli' pears. In an independent study, Chen et al. identified 43 volatile compounds in 'Kuerle Xiangli' pears (Chen et al., 2006a). The differences in the number of volatile compounds detected could be attributed to the

Table 3. Absolute concentrations (µg/kg) and relative contents (%) of different classes of chemicals in 11 pear cultivars.

Aromatic profile	Different species and cultivars										
	P. pyrifolia Nakai	*P. sinkiangensis* Yu	*P. pyrifolia* White Pear Group	*P. ussuriensis* Maxim						*P. sinkiangensis* Yu × *P. pyrifolia* White Pear Group	*P. communis* L
	ZBM	KEL	DGL	PXL	DXS	REL	NGL	YHL	XXS	HXS	BAL
Alcohol	1.5(3.50)	3.1 (9.30)	0.0(0.00)	37.2(4.93)	2.8(0.59)	2.7(0.32)	2.7(0.09)	20.1(1.28)	24.3 (2.27)	3.6(4.71)	80.7(1.33)
Aldehyde	38.4(89.19)	25.6(76.61)	62.3(76.71)	347.0(45.97)	156.3(33.37)	140.0(16.74)	91.5(3.15)	237.5(15.08)	253.0 (23.58)	68.6(90.58)	0.9(0.01)
Ester	0.9(2.09)	3.1 (9.40)	16.5(20.35)	301.0(39.88)	278.9(59.54)	635.9(76.05)	1939.2 (66.83)	961.1 (61.04)	780.6(72.75)	0.3(0.44)	5842.1(96.01)
Acid	2.0(4.61)	1.1 (3.37)	2.1(2.54)	16.2(2.15)	5.5(1.18)	27.7 (3.32)	8.2(0.28)	21.4(1.36)	11.6 (1.08)	2.6(3.37)	13.2(0.22)
Terpene	0.0(0.00)	0.1(0.28)	0.3 (0.40)	52.1(6.91)	24.4(5.21)	29.8 (3.57)	857.8(29.56)	330.0(20.95)	3.4(0.31)	0.5(0.64)	148.1 (2.43)
Ketone	0.3(0.61)	0.4(1.05)	0.0(0.00)	1.2(0.16)	0.6(0.12)	0.0(0.00)	2.5(0.09)	4.5 (0.29)	0.0(0.00)	0.2(0.26)	0.0(0.00)
Total	43.1(100.00)	33.4(100.00)	81.2(100.00)	754.7(100.00)	468.5(100.00)	836.1(100.00)	2901.9(100.00)	1574.6(100.00)	1072.9(100.00)	75.8(100.00)	6085.0(100.00)

different kind of fibers used in their analysis and ours (IOFI Working Group on Methods of Analysis, 2010). Thirty-nine volatile compounds were identified in Nanguoli and 'Youhongli' pears, 33 in 'Daxiangshui', and 31 in 'Xiaoxiangshui'. *P. pyrifolia* cultivars had the lowest number of volatile compounds, for example, 'Hongxiangsu' contained seven volatiles and 'Zaobaimi' contained eight. 'Dongguoli' (*P. pyrifolia* White Pear Group) contained 14 volatile compounds and showed had a moderate total volatiles concentration (81.2 µg/kg). Our results agree with those from earlier studies. Tian et al. (2009) compared the aroma profiles of three cultivars of *P. pyrifolia* White Pear Group and three cultivars of *P. pyrifolia*. They found fewer volatiles and a lower total volatiles concentration in *P. pyrifolia* than in *P. pyrifolia* White Pear Group.

Relative contents of volatiles in different cultivars

The volatiles were grouped according to their chemical class, and included alcohols, aldehydes, acids, esters, terpenes and ketones. The relative contents of the different volatiles were dependent on the cultivar. Table 3 shows the volatile composition of the different cultivars.

Three alcohols, 1-hexanol, 2-ethylhexanol, and ethanol, were identified. Alcohols were present in ten of the 11 cultivars. These alcohols accounted for 0.09 ('Nanguoli') to 9.30% ('Kuerle Xiangli') of the total volatile concentration in the ten cultivars (Table 2). 'Bartlett', 'Pingxiangli', 'Xiaoxiangshui' and 'Youhongli' contained alcohols, while no alcohols were detected in 'Dongguoli'. Alcohols are formed from aldehyde by alcohol dehydrogenase reduction (Chervin et al., 1999; Prestage et al., 1999; Lara et al., 2003; Moya-leon et al., 2006). They are acceptors in an alcohol acyltransferase reaction that links an acetyl moiety from coenzyme A to the alcohol. The activity of alcohol acyltransferase is a key limiting factor in the formation of esters (Perez et al., 1996; Shalit et al., 2001; Defilippi et al., 2005; Li et al., 2006; Lara et al., 2007; Villatoro et al., 2008a; Zhu et al., 2008). 1-Hexanol, the precursor of hexyl acetate, was found in many of the cultivars, and hexyl acetate itself was an important ester in many of the cultivars.

Aldehydes give fruit green and herbaceous flavor notes (Nicolas et al., 2009). The relative content of aldehydes varied among the cultivars. 'Pingxiangli' had the highest aldehyde content (347.0 µg/kg) amongst the cultivars, while the

'Bartlett' (0.9 µg/kg) and 'Kuerle Xiangli' (25.6 µg/kg) cultivars had the lowest. Among the Asian pears, the absolute aldehyde contents in the *P. ussuriensis* cultivars were four-to-five times higher than in the cultivars from other species. Interestingly, the relative aldehyde contents were lower in the *P. ussuriensis* cultivars than the cultivars from the other species. For example, the absolute and relative aldehydes contents in 'Hongxiangsu' (*P. pyrifolia*) were 68.6 µg/kg and 90.58%, respectively. By contrast, the absolute and relative aldehydes contents in 'Ruanerli' (*P. ussuriensis*) were 140.0 µg/kg and 16.74%, respectively. This phenomenon could be explained by the higher total volatiles content in *P. ussuriensis* compared to the other Asian pear species. Among these aldehydes, hexanal and (*E*)-2-hexenal were prevalent in all cultivars, and gave typical green odor notes with low odor threshold values (17 and 5 µg/kg for hexanal and (*E*)-2-hexenal, respectively). Four aldehydes, namely hexanal, (*E*)-2-hexenal, nonanal and (*Z*)-3-hexenal were identified in 'Dongguoli'. Nonanal was also identified in 'Zaobaimi', 'Kuerle Xiangli', 'Youhongli' and 'Bartlett'.

Both qualitatively and quantitatively, esters were the predominant volatile compounds in the cultivars. Like aldehydes, the ester concentrations and number of esters in the *P. ussuriensis* cultivars were higher than those in cultivars from the other Asian species. The ester contents ranged from 0.3 µg/kg ('Hongxiangsu') to 5842.1 µg/kg ('Bartlett'). Except for in 'Pingxiangli', esters accounted for more than 55% of the total volatiles content in the *P. ussuriensis* cultivars. Esters are known to impart characteristic fruity or sweet odors in apples and European pear cultivars (Echeverria et al., 2008; Villatoro et al., 2008b). Among the Asian pear cultivars, the highest concentration of esters was in 'Nanguoli' (1939.2 µg/kg). The total esters concentrations in 'Hongxiangsu', 'Zaobaimi', Kuerle Xiangli, and 'Dongguoli' were 0.3, 0.9, 3.1 and 16.5 µg/kg, respectively. Ten esters, including methyl butanoate, ethyl butanoate, ethyl 2-methylbutanoate, ethyl hexanoate, butyl acetate, methyl hexanoate, hexyl acetate, heptyl acetate, ethyl octanoate and ethyl 2,4-decadienoate, were present in most of the *P. ussuriensis* cultivars. Ethyl hexanoate and hexyl acetate were the two most common esters in the cultivars studied. These two compounds are dominant in pear (Chen et al., 2005), apple (Echeverria et al., 2008), strawberry (Menager et al., 2004) and melon (Senesi et al., 2005), and provide sweet or fruity notes with low odor threshold values (1 and 2 µg/kg for ethyl hexanoate and hexyl acetate, respectively). Ethyl 2,4-decadienoate, which is an important volatile esters in the European pear cultivars (Russell et al., 1981; Quamme 1984; Eccher et al., 1993), was detected in seven of the cultivars, and probably contributed to the aroma in 'Youhongli' and 'Bartlett' because of its high odor threshold (100 µg/kg).

Terpene hydrocarbons were present in all the cultivars except for 'Zaobaimi'. However, their concentrations were low in most cultivars. The relative contents of the terpenes ranged from 0.28% in 'Kuerle Xiangli' to 29.56% in 'Nanguoli'. α-Farnesene, a sesquiterpene biosynthesized via the mevalonate pathway (Pechous and Whitaker 2004), was the main terpene. The product of α-farnesene oxidation is thought to play a key role in induction of superficial scald, which is a commercially important physiological storage disorder in apples and pears (Rowan et al., 2001; Green et al., 2007; Whitaker et al., 2009). α-Farnesene synthase, which catalyzes the final step in the α-farnesene biosynthetic pathway, has been characterized (Tsantili et al., 2007) and cloned in apples (Rupasinghe et al., 2000) and pears (GeneBank Accession No. AY566286). In our study, the concentration of α-farnesene in 'Nanguoli' was much higher than in the other cultivars, which implies that 'Nanguoli' will be susceptible to superficial scald.

Principle volatile components and odor activity values (OAVs)

The principle volatile components (PVCs) are those compounds that occur in large quantities of at least three times the concentration of any other constituents (Pandit et al., 2009). Nine volatiles, including two aldehydes, six esters and one terpene, were PVCs in the pear cultivars studied (Table 2). Hexanal was a PVC in all cultivars except for 'Bartlett' and 'Nanguoli'. The relative content of this compound ranged from 13.14% ('Youhongli') to 83.19% ('Zaobaimi'). Cultivars from the *P. ussuriensis* species than in cultivars had lower relative contents and higher absolute contents of hexanal than cultivars from other species. The aldehyde (*E*)-2-hexenal was a PVC in 'Hongxiangsu', and accounted for 32.46 % of the total volatiles. Ethyl hexanoate was the PVC in the 'Pingxiangli' (16.81%), 'Ruanerli' (18.37%), 'Youhongli' (31.69%), 'Xiaoxiangshui' (26.06%), and 'Nanguoli' (32.80%) cultivars. The contribution of this compound to the total aroma of each cultivar varied greatly. Many compounds, such as ethyl butanoate, methyl hexanoate and ethyl 2,4-decadienoate, were not common as PVCs, but were PVCs in single cultivars. α-farnesene was a PVC in the 'Youhongli' (20.15%) and 'Nanguoli' (28.36 %) cultivars.

The impact of a compound on flavor perception is determined by its concentration and the odor threshold (Goff and Klee 2006). OAVs were calculated to further elucidate the aroma contributions of these compounds (Table 4). The OAV is defined as the ratio of a volatile's concentration to its threshold (Qian and Wang 2005). Fourteen volatile compounds, including two aldehydes and 12 esters, had OAVs greater than one. The OAVs of these volatiles varied widely among the cultivars and constituents. For example, the OAVs of ethyl hexanoate in 'Nanguoli' and 'Dongguoli' were 951.76 and 1.19, respectively. While the OAVs of octyl acetate and ethyl 2-methylbutanoate in 'Xiaoxiangshui' were 1.08 and 668.10, respectively. The OAVs of hexanal were all

Table 4. Odor activity values of different volatile compounds in the 11 pear cultivars.

Volatile compound	Odor threshold (μg/kg)[a]	Different species and cultivars										
		P. pyrifolia	*P. sinkiangensis*	*P. pyrifolia* White Pear Group	*P. ussuriensis*						*P. sinkiangensis* × *P. pyrifolia* White Pear Group	*P. communis*
		ZBM	KEL	DGL	PXL	DXS	REL	NGL	YHL	XXS	HXS	BAL
Hexanal (H)	5	7.17	4.51	9.77	61.55	28.07	24.81	16.08	42.23	44.63	8.8[b]	ND
E-2-hexenal (He)	17	0.14	0.17	0.55	2.31	0.94	0.94	0.65	1.52	1.76	1.45	ND
Methyl butanoate (MB)	5[c]	ND	ND	ND	1.28	0.91	5.16	2.6	0.75	3.13	ND[d]	ND
Ethyl butanoate (EB)	1	ND	ND	ND	76.35	57.35	19.7	125.21	73.05	190.6	ND	ND
Ethyl 2-methylbutanoate (EMB)	0.1[e]	ND	ND	ND	536.3	161.3	17.4	333.2	463	668.1	ND	ND
Ethyl hexanoate (EH)	1	0.59	0.29	1.19	126.88	79.69	153.56	951.76	498.9	279.58	ND	ND
Pentyl acetate (PA)	5[e]	ND	ND	ND	ND	0.39	ND	0.9	ND	ND	ND	6.14
Butyl acetate (BA)	66	ND	0.01	0.02	ND	0.06	0.04	0.36	0.07	0.09	ND	4.95
3-Methylbutyl acetate (MBA)	2	ND	ND	ND	ND	ND	ND	1.79	0.74	1.84	ND	1.64
Methyl hexanoate (MH)	84	ND	ND	ND	0.12	0.07	3.43	0.79	0.16	0.49	ND	0.1
Hexyl acetate (HA)	2	ND	0.78	4.88	4.9	24.45	18.26	228.86	73.65	27.78	0.17	1377.81
Ethyl octanoate (EO)	15[f]	ND	ND	ND	0.03	0.14	0.37	0.68	0.13	1.37	ND	0.52
Octyl acetate (OA)	12	ND	ND	ND	ND	0.42	ND	4.5	0.6	1.08	ND	3.93
Ethyl 2,4-decadienoate (E,Z) (ED)	100	ND	ND	ND	0.05	0.14	0.03	0.64	1.01	0.06	ND	20.8

[a] Odor thresholds of different compounds from Takeoka et al.,1992 unless noted otherwise;[b] Values from the ratio of the volatile concentration to threshold;[c] Schieberle and Hofmann, 1997;[d] Not detected;[e] Bianco et al., 2008;[f] Moya-leon et al., 2006.

greater than one in the Asian cultivars. Cultivars from *P. ussuriensis* showed higher OAVs than the other Asian cultivars studied. For example, hexanal was present in all cultivars, and the OAV of this compound in ‘Pingxiangli’ was 12 times higher than in ‘Kuerle Xiangli’.

Some cultivars, including ‘Zaobaimi’, ‘Kuerle Xiangli’ and ‘Dongguoli’, had only hexanal as a contributor to their aroma. No esters contributed to the fruit aroma because of their low OAVs in the ‘Hongxiangsu’, ‘Zaobaimi’, ‘Kuerle Xiangli’, and ‘Dongguoli’. Compared with the other Asian species, *P. ussuriensis* contained more volatile compounds with OAVs greater than one, and had higher OAVs, particularly for the esters. The OAVs of ethyl butanoate, ethyl 2-methylbutanoate, ethyl hexanoate, and hexyl acetate in *P. ussuriensis* were all greater than one. Although the OAVs of these compounds were different among the cultivars, they contributed to the overall aroma of these cultivars and gave them a fruity odor. The contributions of these thirteen esters to the overall aroma varied depending on their OAVs. Between one and four of the esters were the main contributors to the overall aroma. For example, ethyl butanoate (125.21), ethyl 2-methylbutanoate (333.20), ethyl hexanoate (951.76) and hexyl acetate (228.86) were the main contributors to the overall aroma of ‘Nanguoli’, while ethyl hexanoate (153.56) was the main contributor to the aroma of ‘Ruanerli’. For ‘Bartlett’, ethyl (*E*,*Z*)-2,4-decadienoate and six acetate esters, including pentyl acetate, butyl acetate, 3-methylbutyl acetate, hexyl acetate and octyl acetate, were as the main contributors to the fruit aroma. The OAV of ethyl (*E*,*Z*)-2,4-decadienoate was 20.80, and this compound was not noted in any of the Asian cultivars in this study.

PCA of the volatile compounds in the different cultivars

PCA was used to visualize the complete data set in a reduced dimension plot. In this study, 15 factors were used for PCA, including 14 likely contributors to the fruit aroma and α-farnesene, which was one of the PVCs in the cultivars. Principal components 1 (PC1) and 2 (PC2) accounted for 87.69 % of the total variability. ‘Bartlett’ and ‘Nanguoli’ were clearly separated from the other cultivars, and could be discriminated using the PC1. Discrimination of these two cultivars from the others indicated that they might have very high contents of some compounds. ‘Nanguoli’ was characterized by high concentrations of ethyl hexanoate and α-farnesene, while ‘Bartlett’ had high concentrations of hexyl acetate, butyl acetate and ethyl (*E*,*Z*)-2,4-decadienoate. As indicated in Figure 1A, PC1 mainly discriminated ‘Bartlett’ from all the other cultivars, whereas PC2 mainly discriminated the cultivars of in

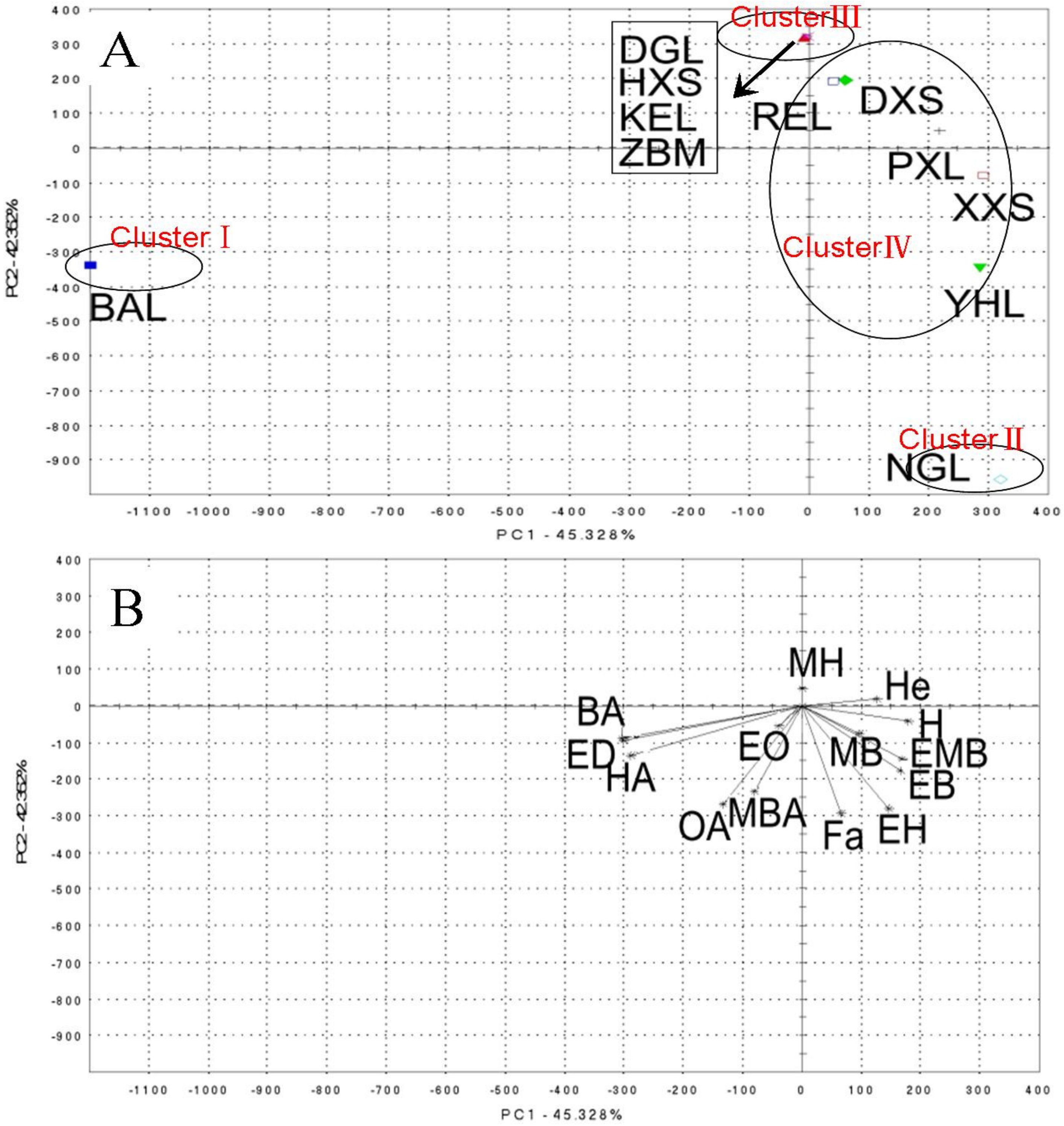

Figure 1. (A) Score and (B) loading plot of PC1 vs. PC2 corresponding to a PCA model for emission of aromatic volatile compounds from the different pear cultivars.

clusters II, III and IV from each other. Cluster III included 'Hongxiangsu', 'Zaobaimi', 'Kuerle Xiangli' and 'Dongguoli', which were positively correlated with the two PCs and had low volatile concentrations. The other clusters were mainly characterized by esters or terpenes. The cultivars in cluster IV contained high concentrations of butanoate esters and ethyl hexanoate.

Conclusion

In the 11 pear cultivars studied, qualitative and quantitative differences were found in their volatile compositions. They also showed large differences in their PVCs, particularly for esters and aldehydes. On the qualitative basis, esters were the main volatile compounds present in the cultivars. When quantitative data and odor threshold values were taken into account, hexanal was the most common compound with a high OAV in Asian pears. In *P. ussuriensis*, hexyl acetate, ethyl hexanoate, ethyl 2-methylbutanoate and ethyl butanoate were the major compounds. Among the different Asian species, *P. ussuriensis* had a higher concentration of volatiles and ester content was than in the other three species. The relative aldehyde contents were >75% in *P. pyrifolia* White Pear Group, *P. sinkangensis* and *P. pyrifolia*. The relative esters content was slightly higher in White Pear Group than in *P. sinkangensis* or *P. pyrifolia*. Different species differed markedly in their volatile profiles and could be divided into four clusters based on the first two PCs.

This research is very important for the pear industry, particularly for the selection of cultivars in breeding programs. Although our study has revealed obvious differences between different species and cultivars, the following two aspects will be investigated further in our future work. Firstly, the cause of the differences in the volatile profiles among the different species, and the contribution of the volatiles' biosynthetic routes, will be studied. Secondly, because fruit aroma is an important sensory attribute of fruit, these cultivars will be characterized according to consumer preferences.

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