

Study On The Volatile Components Extracted From Litchi Fruit (Litchi Chinensis) Of Viet Nam

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Abstract: Litchi fruit (*Litchi chinensis*) grown in Thanh Ha (Hai Duong, Viet Nam) was subjected in this study. The odor concentrates isolated from the edible parts (juice and pulp) of this fruit were obtained by solvent extracted before being subjected to chromatography in a column packed with Porapak Q resin (Supelco, 50/80 mesh). The odor components were analyzed by Gas Chromatography-Mass Spectrometry (GC-MS). Besides, the volatile composition of the peel and the seed of this litchi fruit were also investigated. The results showed that hexadecanoic acid methyl ester (23.80 %), acetoin (19.19 %), 2-butoxy-ethanol (4.35 %); 9,12,15-octadecatrienoic acid methyl ester (4.46 %), benzenethanol (4.23 %) and linalool (2.25 %) were the major compounds in the concentrate isolated from the juice. However, hexadecanoic acid methyl ester and 9,12,15-octadecatrienoic acid methyl ester were the major components of the concentrate obtained from the pulp of this fruit. Moreover, the predominant volatile constituents of the aroma concentrate isolated from the litchi seed were β -caryophyllene (15.84 %), (E)-anethole (10.04 %), germacrene-B (7.05 %) α -humulene (7.54 %), and α -copaene (4.33 %). Meanwhile, terpenes and terpene derivatives such as β -caryophyllene, β -bisabolene, α -cubebene, copaene, α -humulene were found as the major volatile constituents of the essential oil and the aroma concentrate from the peel of the litchi from Thanh Ha province.

Keywords: *Litchi chinensis*, volatile compounds, porapak Q, litchi of Thanh Ha

1. INTRODUCTION

The litchi (*Litchi chinensis*) is native from China, but it is also grown in some other tropical countries such as Vietnam, Thailand, Indonesia, India, and Bangladesh... The fruit of the litchi has a white, juicy aril and contains a shiny brown, usually large seed. Because there is only a short harvest period and the fruit has a short shelf life, the fresh fruit is only available during a month in Vietnam. However, there are some types of canned litchi, frozen litchi or dried litchi available all year but they have a different taste.

There have been studies about the improvement of the storage and cultivation, the antioxidant effects of compounds in the fruit of the litchi. However, the amount of studies that identifies the volatile components in the different parts (flesh, seed and peel) of the litchi is limited. Johnston *et al.* [1] identified the volatile compounds in the fresh fruit and confirmed that limonene, geranial and neral, β -phenethylalcohol were the key odor compounds of the litchi. Chyau *et al.* [2] made a comparison between free and glycosidically bound aroma compounds and found that the free volatile fraction had a fresh-fruity, litchi-like aroma whereas the glycosylated fraction was odorless. Mahattanatawee *et al.* [3] compared three major litchi cultivars and determined twenty-four common volatile compounds. Y. Wu *et al.* [4] studied the volatile profiles of nine litchi cultivars from Southern China by gas chromatography – mass spectrometry (GC-MS) combined with headspace solid phase micro-extraction. Seventeen common volatiles were investigated in all samples such as linalool, *cis*-rose oxide, α -terpineol, geraniol, 3-methyl-3-buten-1-ol, 3-methyl-2-buten-1-ol, hexanol... The study of Nguyen Tuan Duong *et al.* [5] examined the volatile components extracted from Vietnamese litchi seed and identified caryophyllene, α -copaene, α -humulene, naphthalene, δ -cadinene as the major compounds of the litchi aroma concentration.

Despite a number of research on the volatile composition of the litchi fruits in the world, so little is there information available on the volatile components of the different parts of Vietnamese ones. Therefore, the aim of this study is not only to investigate the volatile compounds in the edible parts (pulp and juice) of the Vietnamese litchi but also the other part that are considered waste, like the seed and peel.

2. MATERIALS AND METHODS

2.1. Litchi fruit and reagents

Litchi fruits grown at litchi garden in Thanh Ha (Hai Duong, Vietnam) were purchased and immediately stripped into their different parts: peel, seed and flesh (edible part). The edible part was blended and treated with the pectinase enzyme of 0.0176 % wt., at 30 °C in 90 min. The mixture was then filtered, centrifuged (at 3000 \times g at 4 °C for 20 min) and separated into the pulp and the juice.

Methanol, diethyl ether, hexane and pentane were purchased from Prolabo (France) while Porapak Q resin (50/80 mesh)

was obtained commercially from Supelco (Japan). Commercial pectinase enzyme named Pectinex Ultra SP-L was purchased from Novozymes (Denmark). All other chemicals used were of analytical grade.

2.2. Isolation of the aroma concentrates

The pulp of litchi fruit (100 g) was immersed in methanol in 24 h to extract the organic compounds. The obtained extract was centrifuged at $3000 \times g$ at 4°C for 10 min, filtered and adjusted to a 5 % methanol aqueous solution with purified water, before being subjected to chromatography on a column packed with 20 g of Porapak Q resin (Supelco, 50/80 mesh) [6]. After washing sugars, acids and other water-soluble compounds with 200 mL of purified water, the absorbed odor compounds were eluted with 200 mL of a mixture of pentane and diethyl ether (1:1, v/v). The eluate was dehydrated with anhydrous sodium sulfate and filtered, and the solvent was evaporated at 39.5°C at atmospheric pressure to obtain the aroma concentrate. The volatile compounds were then concentrated before injecting to GC-MS.

The juice of litchi fruit (100 ml) was added by purified water to make of 1 L and then subjected to the Porapak Q column chromatography according to the mentioned-above extraction method. The volatile compounds in the obtained aroma concentrate were also analyzed by GC-MS.

The seed was ground to powder and then isolated with hexane for 4 weeks to extract the aroma concentrate. The extracts were dried over anhydrous sodium sulfate and concentrated by a rotary evaporator. The obtained aroma concentrate from the seed was analyzed by GC-MS.

After washing with clean water, the shredded peel of litchi fruit (100 g) was completely immersed in hexane for 7 days. The extracts were dried over anhydrous sodium sulfate and filtered. The solvent was removed under vacuum using a rotary evaporator. The obtained aroma concentrate from the peel was subjected to analyze by GC-MS. Moreover, the essential oil from the litchi peel was also prepared by steam distillation in Clevenger-type apparatus in 4 h with the ratio of the peel and water was fixed at 1:3.

2.3. GC-MS analysis

Volatile compounds of the samples were analyzed by GC-MS QP 2010 instrument equipped with a flame-ionization detector (FID) and a fused silica capillary column: DB-5 column, $30\text{ m} \times 0.25\text{ mm i.d.}$, and $0.25\text{ }\mu\text{m}$ film thickness (Shimadzu, Japan). Helium was used as the carrier gas at a flow rate of 1.5 mL/min . The oven temperature was held at 60°C for 4 min and increased to 230°C at the rate of 3°C/min and then held for 15 min. The injector and detector temperatures were set at 200°C and 230°C , respectively. Each compound was identified by the agreement of mass spectrum with those of the authentic compound in the GC-MS library (Willey/Chemstation).

3. RESULTS AND DISCUSSION

3.1. Volatile components in the aroma concentrate from the edible parts of litchi fruit

Agreement of the mass spectra in the GS-MS library enabled 46 and 16 compounds to be identified as the volatiles in the aroma concentrate of the juice and the pulp of litchi fruit, respectively. The peak area relative of each compound to the total peak area (%) is expressed in Table 1. The results showed that hexadecanoic acid methyl ester (23.80 %), acetoin (19.19 %), 2-butoxy-ethanol (4.35 %), 9,12,15-octadecatrienoic acid methyl ester (4.46 %), benzenethanol (4.23 %) and linalool (2.25 %) were the major components of the aroma concentrate of the litchi juice. In Chinese litchi, acetoin (30.1 %) and geraniol (15.6 %) were the predominant volatile components of the flesh of this fruit [2]. Moreover, in the aroma concentrate obtained from the juice of Thanh Ha litchi, ester group accounted for the major proportions of total peak area whereas geraniol was detected as the minor compound, accounting for only 1.10 %. In the pulp, hexadecanoic acid methyl ester and 9,12,15-octadecatrienoic acid methyl ester were the major components. Especially, some volatile compounds with flower, pleasant, fruity characteristic odors such as linalool, benzenethanol, geraniol, benzaldehyde, limonene, methyl β -phenethyl ether, germacrene D, α -muroloene... accounting for nearly 17 % of total peak area were found in the juice, but these components being absent in the pulp.

Table 1. Volatile compounds identified in the aroma concentrate obtained from the edible parts (the juice and the pulp) of litchi fruit.

No	Compounds	Relative area* (%)	
		In the juice	In the pulp

1	isoamyl alcohol	0.82	---
2	3-methyl-3-buten-1-ol	2.24	---
3	ethyl linalool	0.77	---
4	<i>n</i> -octane	0.35	0.76
5	propane, 1-(1-methylethoxy)	0.54	0.30
6	acetoin	19.19	12.11
7	hexanol	0.54	---
8	propane, 1,1-dipropoxy	0.43	---
9	3-methyl-2-buten-1-ol	0.47	0.47
10	2,6-dimethyl-4-heptanol	0.27	---
11	nonane	0.80	4.10
12	ethanol, 2-butoxy-	4.35	---
13	3-methyl nonate	0.17	---
14	decane, 2,2-dimethyl	2.18	---
15	decane	2.56	0.33
16	hexane, 2,2,3-trimethyl	0.29	---
17	2-ethyl hexanol	1.29	---
18	benzyl alcohol	0.52	---
19	benzaldehyde	0.63	---
20	limonene	1.04	---
21	heptane, 2, 2-dimethyl	0.31	---
22	methyl β -phenethyl ether	2.59	---
23	linalool	2.25	---
24	undecane	0.34	---
25	benzenethanol	4.23	---
26	dodecane	2.16	---
27	(<i>R</i>)-(+)-1-phenyl-1-propanol	1.53	---
28	(<i>E</i>)-geraniol	1.10	---
29	β -cedrene	0.82	0.23
30	tetradecane	2.05	---
31	(<i>E</i>)-linalool oxide	0.71	---
32	germacrene D	1.76	1.10
33	α -muroloene	2.11	1.02
34	hexadecane	1.33	---
35	dodecanol	0.46	---
36	(<i>E</i>)-14-hexadecenal	0.94	---

37	cyclopentane, decyl	0.41	---
38	tetradecanol	0.47	---
39	9-octadecenoic acid	0.40	1.45
40	hexadecanoic acid, methyl ester	23.80	41.60
41	butyl phthalate	0.67	0.49
42	hexadecanoic acid	1.07	0.47
43	linolelaidic acid methyl ester	1.78	6.01
44	9,12,15-octadecatrienoic acid, methyl ester	4.46	24.50
45	11-octadecenoic acid, methyl ester	2.23	4.88
46	9-hexadecenoic acid	0.58	---
	Total	99.72	99.82

* Peak area relative to the total peak area (%) on GC-MS

These above results might be applied in the membrane concentration technology, where the juice and the pulp are firstly separated to concentrate the juice, then combined with each other in the final step to enhance aroma of this fruit. Therefore, depending on the different technology purposes, the juice or the pulp or both of these edible parts are used.

3.2. Volatile components in the aroma concentrate obtained from seed of litchi fruit

Table 2. Volatile compounds identified in the aroma concentrate of the seed of litchi fruit.

No	Compounds	Relative area* (%)	No	Compounds	Relative area* (%)
1	2-hexanone	2.16	15	dodecanol	2.53
2	3-hexanol	2.34	16	α -copaene	4.33
3	2-hexanol	2.75	17	β - elemene	1.28
4	n-nonane	4.08	18	β -caryophyllene	15.84
5	α - pinene	1.03	19	α -humulene	7.54
6	benzene 1,3,5 trimethyl	1.70	20	germacrene -D	2.98
7	decane	5.19	21	γ - gurjunene	0.60
8	β -pinene	0.72	22	germacrene-B	7.05
9	l-limonene	0.98	23	α - muurollene	2.28
10	nonanol	1.39	24	δ - cadinene	4.30
11	undecane	3.00	25	pathulenol	0.85
12	(E)-anethole	10.04	26	isobutyl o-phthalate	3.67
13	decanol	2.44	27	dibutyl phthalate	5.47
14	undecanol	3.47			

* Peak area relative to the total peak area (%) on GC-MS

The aroma concentrate from the seed extracted by hexane for 4 weeks was analyzed by GC-MS (Table 2). The results showed that the predominant volatile constituents of the aroma concentrate included β -caryophyllene (15.84 %), (E)-anethole (10.04 %), germacrene-B (7.05 %) α -humulene (7.54 %), and α -copaene (4.33 %) with characteristic pleasant fruity, woody and herb-like odors. These findings are quite in agreement with those of the previous study [5] in which β -caryophyllene, α -copaene,

α -humulene...were the major components of the aroma concentrate of the seed of litchi.

3.3. Volatile components in the essential oil and in the aroma concentrate obtained from peel of litchi fruit

By distillation method, the twenty-four compounds were identified as the volatile compounds in the essential oil of the litchi peel (Table 3). Some terpenes and their derivatives such as β -caryophyllene (20.64 %), β -bisabolene (17.74 %), α -cubebene (14.6 %), and δ -cadinene (11.15 %) were considered as the major compounds in this essential oil.

Table 3. Volatile compounds identified in the essential oil of the peel of litchi fruit.

No	Compounds	Relative area* (%)	No	Compounds	Relative area* (%)
1	3- hexanone	0.01	15	α -muurolene	1.72
2	β -cubebene	4.44	16	α -copaene	0.31
3	γ -cadinene	0.10	17	β -bisabolene	17.74
4	α -cubebene	14.6	18	δ -cadinene	11.15
5	α -gurjunene	0.36	19	β -cedrene	0.64
6	β -caryophyllene	20.64	20	(E) – α - bisabolene	0.41
7	allocerromadendrene	1.19	21	(-)- caryophyllene oxide	0.29
8	germacrene – D	7.61	22	veridiflorol	0.15
9	α – humulene	7.74	23	cubenol	1.85
10	aromadrene	0.32	24	α - cactinol	1.64

* Peak area relative to the total peak area (%) on GC-MS.

On the other hand, 24 compounds were identified as the volatiles in the aroma concentrate of the litchi peel obtained by hexane extraction for 7 days (Table 4). The results suggested that some terpenes and their derivatives such as β -caryophyllene (39.44%), α -amorphene (15.29 %), β -copaene (13.21 %), and α -humulene (10.26 %) were identified as the major compounds in the peel of litchi isolated by hexane.

Table 4. Volatile compounds identified in the aroma concentrate of the peel of litchi fruit.

No	Compounds	Relative area* (%)	No	Compounds	Relative area* (%)
1	3-hexanone	0.15	13	α -cedrenal	0.12
2	2-hexanone	0.11	14	α -humulene	10.26
3	2-methyl 4-pentanone	0.19	15	germacrene – B	7.90
4	3-hexanol	0.43	16	β -costol	0.18
5	2-hexanol	0.47	17	β - elemene	3.82
6	hexane	0.05	18	α -muurolene	3.63
7	nonane	0.17	19	β - mycrene	0.09
8	(+)- cycloisotaveve	0.90	20	γ -cadinene	1.16
9	β -copaene	13.21	21	δ -cactinene	9.15
10	α -amorphene	15.29	22	γ -elemene	0.73
11	β -caryophyllene	39.44	23	(E) - α - bergamotene	0.23
12	germacrene – D	3.05	24	(-)-carophyllene oxide	0.65

* Peak area relative to the total peak area (%) on GC-MS

4. CONCLUSIONS

Although a number of researches have been reported on the odor of litchi fruit, the overall volatile composition of different parts (the edible part, peel and seed) of this fruit grown in Thanh Ha were analyzed in this study. The results showed that the predominant components of the edible parts (the juice and the pulp) were mainly ester groups, acetoin, terpenes and their derivatives. Furthermore, it could be concluded that terpenes and its derivatives were the main compounds of the peel and the seed of this fruit. These obtained results should contribute to evaluate the odor compounds and the odor quality of these different parts of Thanh Ha litchi fruit, to extent the utilization of this fruit in food industry as well as in pharmaceuticals, cosmetics, and would be useful for further aromatic studies.

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